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Novel cost of a sexually selected trait in the rubyspot damselfly *Hetaerina americana*: conspicuousness to prey

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Conspicuousness to predators frequently has been invoked as a cost of sexually selected traits, but conspicuousness to prey has not. We tested for the latter using rubyspot damselflies (*Hetaerina americana*) as the predator. Previous work on this species showed that the red spots on male wings are intrasexually selected and reduce survival. Since female wings lack red spots, we first compared male and female weight gain rates per unit hunting time. Females gained weight significantly faster than males in both mg per hour and relative to body weight. We then compared the weight gain rates of females painted with red wing spots to those of control females painted with clear ink or not manipulated. Controls gained weight significantly faster than red-painted females. Behavioral observations revealed that red females attempted to capture prey at normal rates and experienced normal rates of agonistic interference from conspecifics of both sexes. Nevertheless, red females captured fewer prey per minute and per capture attempt than did sham-manipulated and unmanipulated controls. We infer that the red spots reduced female weight gain rates by increasing their visibility to prey. Close similarity between male and red female weight gain rates relative to unmanipulated females suggests that red spots may also be a hunting handicap for males. *Key words:* coloration, conspicuousness, crypsis, hunting, natural selection, Odonata, predation, predator, prey, sexual selection. *[Behav Ecol 7:465–473 (1996)]*

Sexual selection apparently has produced an astounding diversity of secondary sexual characters, from the ponderous horns of stag beetles to the ornate plumes of peacocks (Darwin, 1871). Recent efforts to explain this diversity have emphasized the value of these traits as weapons (Conner, 1989), claspers (Arnqvist, 1989), quality indicators (Grafen, 1990; Hasson, 1991; Kodric-Brown and Brown, 1984; Zahavi, 1978, 1982), and sensory system stimulants (Eberhard, 1985; Endler, 1992; Enquist and Arak, 1993; Ryan, 1990). But selective benefits are only half the puzzle. In theory, sexually selected traits evolve to an equilibrium where their costs and benefits balance (Fisher, 1930; Kirkpatrick, 1982; Lande, 1981; Pomiankowski, 1988; Seger, 1985). To understand why certain secondary sexual characters evolve instead of others, or why different taxa evolve different traits, we need to learn more about costs (Andersson, 1994; Arnqvist, 1994; Balmford et al., 1993; Harvey and Bradbury, 1991; Meyer et al., 1994; Partridge and Endler, 1987). Hypothesized costs include the physiological demands of trait development, maintenance, or production (e.g., Clutton-Brock et al., 1985; Halliday, 1987; Hill, 1994; Vehrencamp et al., 1989), attraction of parasites (Cade, 1975, 1979), vulnerability to cannibals (Arnqvist, 1994), and interference with parental care (Møller, 1989; Wingfield et al., 1990). By far the most frequently invoked and convincingly demonstrated cost is conspicuousness to predators (e.g., Endler, 1980, 1983; Lloyd and Wing, 1983; Ryan, 1985; Ryan et al., 1982). Oddly, the reverse cost, conspicuousness to prey, has seldom if ever been reported.

We tested for conspicuous-to-prey effects using rubyspot damselflies (*Hetaerina americana*) as the predator. Sexually mature males of this species have metallic red exoskeletons, dark reddish eyes, and a large red spot at the base of each wing. Females, in contrast, have small faint amber wing spots, pale brown eyes, and cryptically patterned bodies that vary from brown to green (Dunkle, 1990; Grether, 1995). Previous work indicated that male wing spots are subject to sexual selection for increased size (Grether GF, in press) and survival selection for decreased size (Grether GF, in preparation). Large spots apparently provide an advantage in competition for mating territories (Grether GF, in press), but their costs remain unknown.

The chief advantages of this system for the current study are that hunting occurs at predictable times and locations, predatory attacks are directly observable, and capture rates can be measured precisely.

METHODS

Study site

We studied rubyspots at Bear Creek, a perennial stream running through pine-oak woodland in the coastal range of Colusa County, California (39°01′ N, 122°23′ W, elevation 260 m). *H. americana* is the most abundant odonate and the only calopterygid species at this site. Adults emerged continuously from mid-April through November. Prey at this site include small species of the insect orders Diptera, Ephemeroptera, Homoptera, Lepidoptera, Plecoptera, and Trichoptera.

Overview and natural history

Rubyspots at Bear Creek hunt mainly during two daily periods, one in the morning and one in the late afternoon. All data reported here were obtained during the morning hunting period. This period begins soon after the sun warms the animals directly or raises the air temperature above about 21°C. Until then, rubyspots remain immobile at roosts along

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the bank. Hunting is the sole activity of both sexes until males become territorial in the late morning (Grether GF, in press).

Hunting rubyspots perch along the creek bank facing the sun and launch attacks at passing insects. Most attacks fail because of evasive maneuvers by the prey. Conveniently, rubyspots tend to return to the same perch after each attack. This enabled us to sample capture rates by direct observation. We also measured rates of weight gain during hunting, by weighing animals before they began hunting, letting them hunt, and then re-weighing them before they stopped.

In 1992, we compared the weight gain rates and prey capture rates of males to those of females. In 1993, we compared the weight gain rates and prey capture rates of females painted with red wing spots to those of unmanipulated and sham-manipulated female controls. Except for this, and where otherwise noted, the methods used in the two years were identical.

Marking and aging

We marked animals on the left hindwing with unique combinations of three letters and digits using a black Sharpie pen. Our method of aging males is described elsewhere (Grether GF, in press), here we briefly describe our method of aging females. We obtained a sample of known-age females by marking newly emerged adults (N = 97). We later recaptured and scored these females for four characters that change with age: wing stiffness (two levels), thorax sheen (two levels), mid-dorsal carina darkening (11 levels; expanded from Weichsel, 1987), and body silt (three levels; silt accumulates when females submerge to oviposit). By regressing the ages at the last recapture of the known-age females on their dummy-coded character values, we obtained a multiple-regression equation for estimating the ages of females first captured after the day of emergence. We checked the reliability of these estimates by using an equation derived from data on a random half of the recaptured known-age females (N = 48) to predict the ages at last capture of the other half (N = 49). The correlation between actual and predicted ages was 0.98, the mean ± SE absolute difference between actual and predicted ages was 1.12 ± 0.12 days, and the maximum difference was 3.16 days. Given that the actual ages of these females ranged from 2 to 32 days, with a mean ± SE of 7.39 ± 0.64 days, this aging method is reasonably accurate. Hereafter, we refer to both known and estimated ages simply as age.

Female wing color manipulation

The purpose of this experiment was to determine whether putting red wing spots on females would reduce their capture success rates, as predicted by the conspicuous-to-prey hypothesis. We replicated this experiment on 23 days in 1993, using a different group of females each day.

Females were assigned to treatment groups before capture, in a predetermined order that varied from day to day. Three standard caliper measurements, with repeatabilities of 0.95 or higher (Grether GF, in press), were taken on each female: wing length, wing width, and thorax width. All four wings of females assigned to the red treatment group were then painted with male-sized wing spots using red Berol Prismacolor Marker-3. PM-3 provides a close match to the natural color of male wing spots, as judged by both human eyes and spectroradiometry (Grether GF, in press). To control for the extra handling and the addition of weight to the wings of red females, sham controls' wings were painted with clear ink PM-121. Both markers contain a normal propanol base and meleic modified resin binder; the red marker also contains solvent dyes (DeBietro T, Empire-Berol USA, personal communication). Females assigned to the unmanipulated group were left unpainted but otherwise were treated identically.

Artificial wing spots of females in this experiment were designed to mimic the natural wing spots of males. The mean ± SE wing spot length of fully developed males was 10.00 ± 0.03 mm (or 39.1% ± 0.1% of wing length; N = 658), versus 9.98 ± 0.09 mm (38.0 ± 0.3%; N = 61) for red females, and 10.33 ± 0.09 mm (39.4 ± 0.3%; N = 61) for clear females.

To determine whether the red and clear treatments added different amounts of weight to the wings, we weighed eight pairs of wings before and after painting one member of the pair red and the other clear. Wings lose weight rapidly after death, so it was necessary to dry them to a constant weight before weighing the ink. To express ink weight as a percentage of fresh wing weight, we weighed eight wings immediately after clipping them off two decapitated females. Another eight wings were clipped off four females that died naturally. We weighed each wing twice at each step and used mean values in the analysis. Both dry wing weight and ink weight were highly repeatable (r = .999 and r = .978, respectively, N = 16).

Weight gain measurements

To measure rates of weight gain, we took animals from their roosts just before dawn, marked them, weighed them, and placed them at a common roost site. Females in the color manipulation experiment were painted just before being weighed. Placing all animals at a common roost site helped to synchronize hunting start times and made it easier for us to record the start times of individuals. About one hour after hunting began, we netted and re-weighed all animals that we found still hunting, subject to the restriction that we alternated sexes (or treatment groups) between captures. Rubyspots usually hunt in a characteristic fashion: perched at the tops of emergent objects, facing the sun, with their wings held low, and their abdomens parallel to the horizon. When we were in doubt about whether an animal was still hunting, we waited to see whether it attempted to capture passing prey. We repeated this procedure on six days in 1992 and 23 days in 1993 with groups of 6-21 animals per day. In total, we re-weighed 30 of 50 females and 33 of 53 males in 1992. In 1993, we re-weighed 55 of 102 unmanipulated controls, 61 of 105 sham controls, and 61 of 105 red females. Weight gain rates were calculated as the change in weight between the first and second weighing, divided by the time spent hunting. Relative weight gain rates were calculated as weight gain rate divided by the average of pre- and post-hunting weights.

To examine long term effects of the wing color manipulation on female condition, we opportunistically re-weighed some experimental females at later dates. We also obtained a second weight gain rate estimate for 52 of these females (using the protocol described above), to examine changes in weight gain rates over time. Repeat weight gain rates were only used for this purpose; all other analyses are restricted to gain rates measured on the morning females were added to the experiment.

Behavioral observations

In both years, we made focal observations (Altmann, 1974) after finding significant differences between the sexes and treatment groups in weight gain. We made more detailed observations in 1993, so we describe the 1992 protocol first and then explain the changes made in 1993.

In 1992, we gathered weight gain and behavioral data on separate sets of animals. We began making focal observations near the onset of the morning hunting period (0850 h) and continued until the majority of males ceased hunting (females usually hunt later in the day than males; personal observation). Focal animals were selected by taking the male or female hunting closest to a randomly selected location and alternating sexes between 20-min samples. If we lost sight of a focal animal before 20 min elapsed, we tried to observe it again on the same morning. Focal observations on the same animal were merged; observations shorter than 15 min were discarded.

We recorded focal observations on Tandy 102 computers programmed as event recorders. Rubyspots hunt by flying out and back from a consistent perch. We recorded each flight as an event and with the help of binoculars, categorized it by outcome. Most flights were obvious attempts to capture prey; the rest were agonistic chases. Agonistic chases were usually brief (< 2 s) and involved animals hunting near each other; the usual outcome was for one animal to perch elsewhere. Cases in which a perched animal fluttered its wings when another animal approached were not considered agonistic unless the perched animal took flight. We recognized two prey classes in 1992: minute prey, detected only by movement of the mouth parts, and visible prey, which temporarily protruded from the mouth. Two rate variables were calculated for each prey size class: (1) number of prey per min, and (2) number of prey per capture attempt.

In 1993, we tried to obtain weight gain rates for each of the focal females by weighing them before they began hunting and re-weighing them immediately after their focal sample (5 of 86 focal females eluded recapture). Focal females were selected using the criteria described above for re-weighing. We recorded hunting flights as in 1992, but classified them in greater detail. Visible prey (in the mouth of captors) were categorized as follows: (1) small: only wings, legs, or antennae of prey showing; (2) medium: body showing but smaller than a rubyspot head; and (3) large: visible portion larger than a rubyspot head. To incorporate information on prey size into a single weighted capture rate, we used the following estimated mean prey weights: minute, 0.33 mg; small, 1 mg; medium, 3 mg; large, 9 mg. These estimates were obtained by weighing rubyspots before and after hand-feeding them natural prey (Grether GF and Veldhuizen TC, submitted). The results were not sensitive to the magnitude of these weights at $\alpha = 0.05$. To present all prey capture rates in the same units, we divided the weighted prey capture rates by the overall mean prey weight (a constant). This transformation merely scaled weighted prey to the equivalent number of averaged-sized prey.

In 1993 we classified agonistic chases by sex of the interactants and the direction of attack, for six categories: (1) attacks by the focal female on other females, (2) attacks by other females on the focal female, (3) two-way chases with females, (4) attacks by the focal female on males, (5) attacks by males on the focal female, and (6) two-way chases with males. We had no a priori expectations regarding treatment group differences in agonism rates, so we examined all six agonism rate categories. Most of these rates contained mostly zeros, however, so we also examined five combinations of categories: (7) all agonistic interactions with females, (8) all agonistic interactions with males, (9) all attacks by the focal female, (10) all attacks on the focal female, and (11) all agonistic interactions of any kind. We are aware that repeatedly testing the same null hypothesis inflates the probability of a type I error, but for our purposes, this was the most conservative approach.

Statistical procedures

We used parametric tests when the original variables could be transformed to skewness and kurtosis < 1.0 with no significant heteroscedasticity (Neter et al., 1985) and used non-parametric tests otherwise. Time was converted to serial format and ln-transformed, absolute gain rate was $(X + 2)^{0.5}$-transformed, relative gain rate was $\ln(X + 0.1)$-transformed, and all behavioral variables were square root–transformed. We used approximate t tests (Sokal and Rohlf, 1981) in paired comparisons for which an F test showed that the sample variances differed at $\alpha = 0.05$ (t' identifies approximate t values).

Before analyzing the data, we identified age and time of day at the start of the focal samples as potentially useful covariates (Neter et al., 1985). Where these variables were significantly correlated with the dependent variable, and did not interact with the factor, we controlled for their effects via ANCOVA. Throughout this paper, p values are two-tailed and means are presented with standard errors.

RESULTS

Sex differences in weight gain rates

Females in 1992 gained weight faster than males, per unit hunting time, whether weight gain rate was measured in mg per hour (approximate t test, $t' = 2.96$, $df_f = 29$, $df_m = 32$, $p < .01$) or relative to body weight ($t' = 2.49$, $p < .02$), as predicted by the conspicuous-to-prey hypothesis. Female gain rates were also more variable than male gain rates (absolute gain rate $F_{29,32} = 2.84$, $p < .01$; relative gain rate $F_{29,32} = 2.33$, $p = .02$).

Could the sex difference in weight gain rate be because of a sex difference in size? The sexes overlap broadly in size, but females are statistically larger in some dimensions (Grether, 1995). The use of relative weight gain rates removed proportional size effects, but weight gain rates could increase exponentially or as a step function of body size. Rank correlations between body weight and relative weight gain rate were not significant, however, for females ($r_s = .16$, $N = 30$, $p > .4$) or males ($r_s = -.18$, $N = 33$, $p > .3$). None of the body size variables we measured in 1993 correlated significantly with relative weight gain rate (all $r_s < 0.04$, $N = 116$ control females, $p > .7$). Thus, the sex difference in relative weight gain rate was not caused by a sex difference in size.

There are also slight but significant mean differences between the sexes in wing shape. The wing width to wing length ratio of males exceeded that of females by 4.1% in 1992 (two-way ANOVA with emergence month as a factor to control for seasonal variation in wing shape; sex effect: $F_{1,629} = 124.14$, $p < .0001$; month effect: $F_{5,629} = 42.57$, $p < .0001$; interaction $F_{5,629} = 1.69$, $p > .1$) and by 3.5% in 1993 (sex effect: $F_{1,459} = 30.10$, $p < .0001$; month effect: $F_{5,459} = 42.38$, $p < .0001$; interaction $F_{5,459} = 1.64$, $p > .1$). If the mean sex difference in wing shape caused the mean sex difference in weight gain rate, the correlation between wing shape ratio and weight gain rate should be negative. Instead, the correlation was positive and nonsignificant ($r_s = .13$, $N = 116$ control females, $p > .15$).

The males in our weight gain sample were older than the females (mean age of males: 17.3 ± 1.5 days; of females: 10.8 ± 1.1 days; $t = 3.40$, $df = 61$, $p = .001$). This could explain the sex difference in weight gain rate if gain rates decreased with age, but female gain rates increased with age (absolute gain rate: $r = .38$, $N = 30$, $p = .04$; relative gain rate: $r = .29$, $N = 30$, $p = .12$), and there was no significant relationship between weight gain rates and age in males (absolute gain rate: $r = -.06$, $N = 33$, $p > .7$; relative gain rate: $r = -.002$, $N = 33$, $p > .9$), despite the wider range of ages among males (4.7–35.5 days) than among females (5.0–24.4 days). As shown in Figure 1, the mean gain rate of males was lower than that of the youngest females; gain rates of the oldest females were more than twice the male mean.

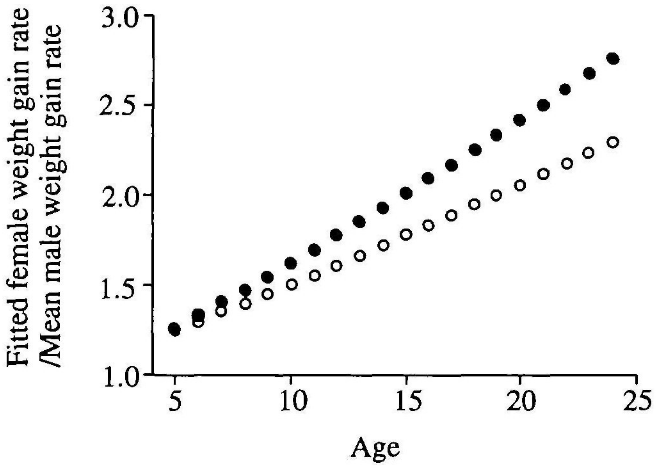

Figure 1
Sex differences in weight gain per unit time hunting. Each dot represents the ratio of female and male weight gain rates at a particular age. Filled dots represent absolute weight gain rate ratios; unfilled dots represent relative weight gain rate ratios. Age-specific female weight gain rates were obtained from the linear regression of weight gain rates on age (using transformations given in Methods). Ratios were calculated by dividing back-transformed fitted female weight gain rates by the back-transformed mean male weight gain rate.

Sex differences in agonism and prey capture rates

The purpose of the behavioral observations was to examine factors other than prey capture that might account for the sex difference in weight gain rate, such as hunting effort and agonistic interference. There were no significant sex differences in the number capture attempts per min ($t = 1.11$, $df = 44$, $p > .2$) or in the rate of agonistic interactions (Mann-Whitney *U* test, $p > .6$, both per min and per flight). Thus, females did not hunt more vigorously than males or experience lower rates of agonistic interference.

Females captured more visible prey per minute, and fewer minute prey per min, than males (Figure 2a), but neither difference was statistically significant (minute prey: $t = 0.70$, $df = 44$, $p > .4$; visible prey: $t' = 1.36$, $N_f = 23$, $N_m = 23$, $p > .1$). The same trends appeared in the number of prey captured per attempt (Figure 2b); the difference was significant for minute prey ($t = 2.31$, $df = 43$, $p = .03$) but not for visible prey ($t' = 1.16$, $N_f = 23$, $N_m = 22$, $p > .2$). Consistent with the greater variance in female weight gain rates, visible prey capture rates were more variable among females than among males (per min: $F_{22,22} = 2.86$, $p = .02$; per attempt: $F_{22,21} = 2.47$, $p = .04$).

Effects of artificial red wing spots on female weight gain rates

Red and clear treatments did not differ significantly in the amount of weight they added to female wings (red: 0.081 ± 0.027 mg; clear: 0.089 ± 0.021 mg; paired *t* test, $t = 0.28$, $N = 8$, $p > .7$). On average, the ink weighed 4.86 ± 1.00% ($N = 8$) of fresh wing weight, or 0.23 ± 0.05% ($N = 8$) of live body weight (extrapolated to four painted wings).

As predicted by the conspicuous to prey hypothesis, females painted with red wing spots gained weight at significantly lower rates than both sham–manipulated and unmanipulated controls, whether weight gain was measured in mg per hour (overall $F_{2,173} = 9.78$, $p < .0001$; both Bonferroni $p < .01$), or relative to body weight ($F_{2,173} = 10.16$, $p < .0001$; both Bonferroni $p < .01$) (Figure 3). Weight gain rates of the control

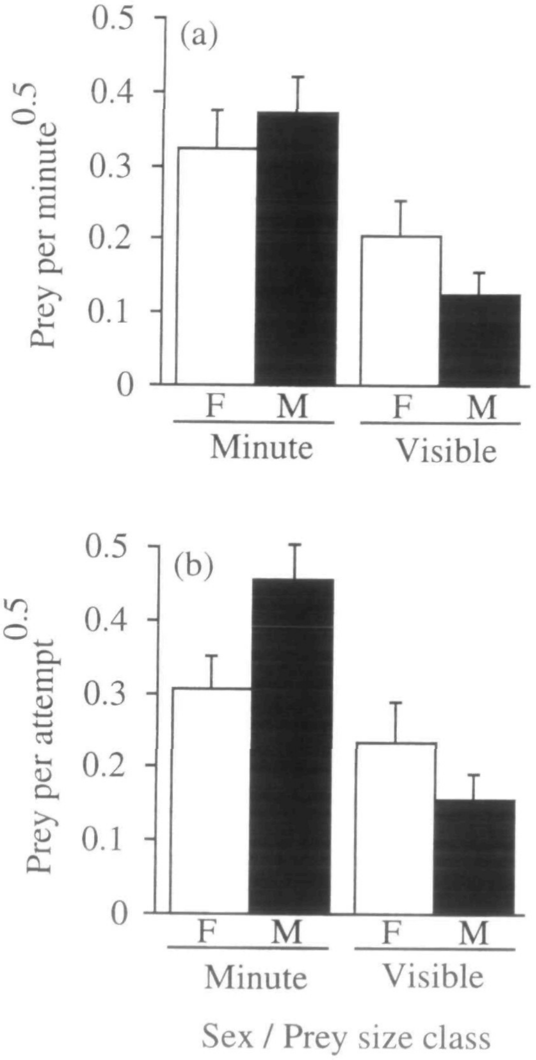

Figure 2
Sex differences in prey capture rates (mean + SE). (a) Prey captured per min; and (b) prey captured per capture attempt. F = female; M = male. The square root transform was used to reduce heteroscedasticity. See text for prey size class definitions.

groups did not differ significantly (Bonferroni $p > .5$). We used age as a covariate in this analysis because weight gain rates increased linearly with age (absolute gain rate: $r = .22$, $N = 177$, $p = .003$; relative gain rate: $r = .17$, $N = 177$, $p = .03$), the treatment groups did not differ in age ($F_{2,174} = 0.30$, $p > .7$), and the age by treatment group interaction was not significant ($p > .6$).

Effects of artificial red wing spots on female agonism and prey capture rates

The focal data revealed that red females attempted to capture prey at normal rates and experienced normal rates of agonistic interference. Capture attempt rates decreased with time of day ($r = -.36$, $N = 86$, $p < .001$) and did not vary with age ($r = .03$, $N = 86$, $p > .8$). ANCOVA controlling for time of day, revealed no significant effect of the wing color manipulation on the rate of capture attempts ($F_{2,82} = 0.75$, $p > .4$). Out of all 22 agonism rate variables examined, significant treatment group differences were found only for the number of attacks by males on the focal female (per min: Kruskal-

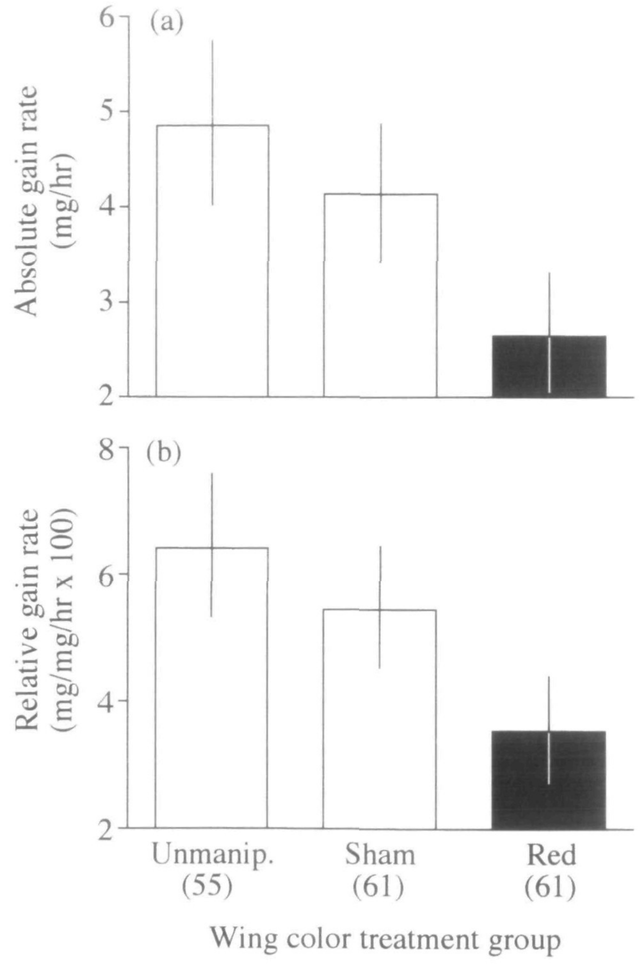

Figure 3
Effect of the wing color manipulation on weight gain per unit hunting time (mean ± 95% confidence limits). (a) Absolute weight gain rate, and (b) relative weight gain rate. Mean weight gain rates were back-transformed (see Methods) after being adjusted to a common mean age by ANCOVA. Sample sizes are shown below the treatment group labels.

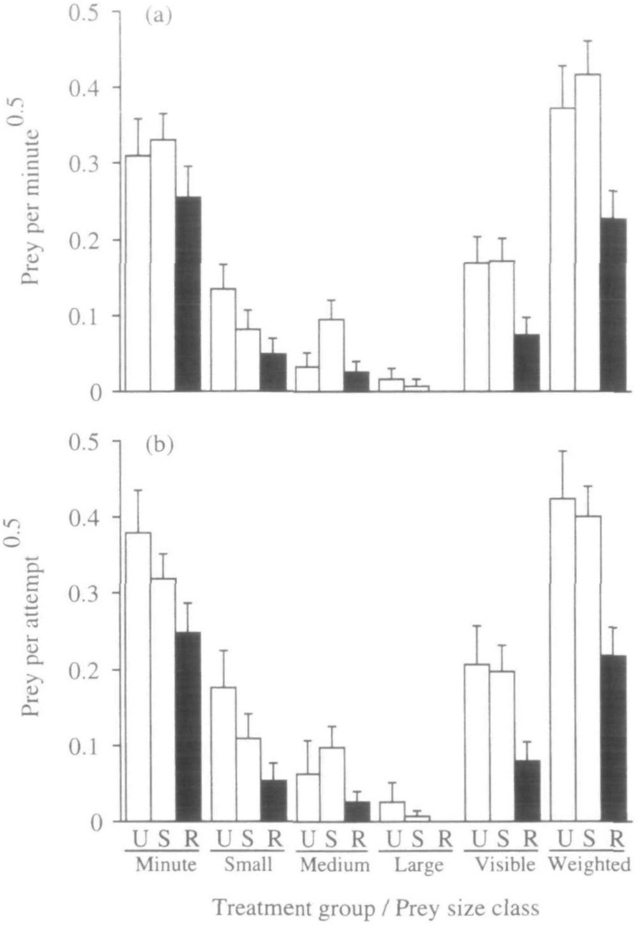

Figure 4
Effect of the wing color manipulation on prey capture rates of hunting females (mean + SE). (a) Prey captured per min, and (b) prey captured per capture attempt. U = unmanipulated control; S = sham control; R = red-painted. Visible prey include all but minute prey; weighted capture rates incorporate estimated prey weights (see Methods). The square root transform helped reduce heteroscedasticity. See Table 1 for statistics.

Wallis $H = 7.93$, $p < .02$; per flight: $H = 7.60$, $p = .02$). Sham controls were attacked at the highest rate by males and red females were attacked the least, but post hoc tests revealed no significant pairwise differences (Dunn's procedure, $p > .05$). Thus, the low weight gain rates of red females do not seem to have been caused by reduced hunting effort or increased agonistic interference.

Could subtle (undetected) effects of the wing color manipulation on agonism rates have caused large treatment group differences in weight gain rates? We screened all 22 agonism rate variables for correlations with the absolute rate of weight gain. Two correlations were marginally significant. Weight gain rate correlated positively with the rate of attacks by the focal on other females per flight ($r_s = 0.22$, $N = 80$, $p = .05$) and negatively with the rate of two-way chases with males per flight ($r_s = -.22$, $N = 80$, $p = .05$). None of the other 20 agonism rates were significantly correlated with the rate of weight gain. Agonistic interference therefore appears to have negligible effects on hunting success and could not logically have caused the treatment group differences in weight gain rates.

Red females captured fewer prey per minute, and per capture attempt, than controls on all four prey size classes (Figure 4). We tested for treatment effects in ANCOVAs controlling for time of day, because rates of prey capture decreased over time (per min: minute $r = -.32$, $N = 86$, $p = .003$, visible $r = -.25$, $p = .02$, weighted $r = -.32$, $p = .002$; per attempt: minute $r = -.23$, $N = 85$, $p = .04$, visible $r = -.26$, $p = 0.02$, weighted $r = -.24$, $p = .03$). With time of day held constant, the wing color manipulation had no significant effect on the rate of capture of minute prey, but it had significant effects on both visible and weighted prey capture rates (Table 1). Pairwise tests showed that sham and unmanipulated controls captured significantly more prey per minute, and per capture attempt, than red females (Table 1). The capture rates of unmanipulated and sham controls did not differ significantly. Some differences between red females and controls became non–significant after the correction for multiple comparisons (Table 1).

How well were weight gain rates predicted by rates of prey capture? The best predictor, weighted prey per capture attempt, explained 18% of the variance in the absolute rate of weight gain (Table 2). In general, rates of prey capture per attempt were better predictors than rates of prey capture per minute (Table 2). This suggests that hunting is energetically demanding.

Table 1

Effect of the female wing color manipulation on prey capture rates

Dependent variable	Treatment F test			Pairwise comparisons[a]					
				U versus S		U versus R		S versus R	
	F	df[b]	p	p_t	p_b	p_t	p_b	p_t	p_b
Minute prey per min	0.87	2, 82	.425	.986	>.9	.285	.855	.243	.728
Visible prey per min	3.80	2, 82	.026	.835	>.9	.019	.058	.021	.064
Weighted prey per min	5.24	2, 82	.007	.721	>.9	.016	.048	.003	.010
Minute prey per attempt	2.61	2, 81	.079	.211	.632	.025	.075	.266	.798
Visible prey per attempt	3.77	2, 81	.015	.647	>.9	.015	.044	.031	.093
Weighted prey per attempt	5.23	2, 81	.007	.578	>.9	.004	.012	.012	.035

U, unmanipulated female; S, sham-manipulated female; R, red-painted female.

For variable descriptions, see Methods.

[a] p_t = t test p values, p_b = Bonferroni p values (adjusted for multiple comparisons).

[b] Success rates have one less df than per min rates because one female made no capture attempts.

Comparison of sex and treatment group differences

Here we compare key results of the sex difference and female wing color manipulation studies. Weight gain rates of males and red females are not directly comparable because they were measured in different years. Nevertheless, they can be expressed in relation to the weight gain rates of unmanipulated females in their respective years, and thereby compared indirectly. Unmanipulated controls in 1993 gained weight 1.81 times faster than red females (Figure 3). At the mean age of females in 1993 (12.38 ± 0.41 days), females in 1992 gained weight 1.80 times faster than males (Figure 1). Thus, relative to unmanipulated females, males and red females had quite similar rates of weight gain.

The mechanism behind the reduced weight gain rates of red females was clear: they attacked prey at the same rate as controls but were less successful per attack (Figure 4; Table 1). The reason for the sex difference in weight gain rates was less apparent. Males attacked prey at the same rate as females but were better at catching minute prey and not significantly worse at catching larger prey (Figure 2). There are two plausible reasons for this discrepancy: (1) lower statistical power in the focal observation data, and (2) females caught larger prey than males. Consistent with the first option, the sex difference in visible prey capture rate was actually greater than the sex difference in weight gain rate. Females gained weight 1.69 times faster than males but caught 2.62 times more visible prey per min. Moreover, in both years, capture rates were more variable than weight gain rates (visible prey capture rate coefficient of variation: 1.79 in 1992, 1.50 in 1993; absolute weight gain rate CV: 0.68 in 1992, 0.76 in 1993). The second possibility, that males specialized on smaller (slower) prey, is consistent with the trends in Figure 2.

Table 2

Correlations between prey capture rates and the absolute rate of weight gain (mg/hr)

Capture rate variable	r	N	p
Prey per minute			
Minute prey	.21	81	.061
Visible prey	.21	81	.055
Weighted prey	.29	81	.008
Prey per capture attempt			
Minute prey	.41	80	.0001
Visible prey	.33	80	.002
Weighted prey	.43	80	<.0001

Long term effects of the female wing color manipulation

The results presented thus far were based on a single morning of hunting for each animal. Here we examine whether the wing color manipulation had lasting effects on female condition. Age is a complicating factor because females normally gain weight with age. As shown in Figure 5, red females initially lost weight while controls gained weight steadily. Qualitatively, this is what we would expect if the reduced hunting success of red females combined additively with the normal increase in weight with age. But the steep weight gains of red females after three days (Figure 5) suggest that they compensated for their initial losses. A comparison of weight changes for the first three days revealed significant treatment group differences (ANOVA $F_{2,91} = 3.90$, $p = .02$). Red females lost 1.55 ± 0.95 mg ($N = 41$), on the average, while unmanipulated and sham controls gained 1.59 ± 0.72 ($N = 38$) and 2.03 ± 1.81 mg ($N = 15$), respectively. After the first three days, however, treatment group differences vanished (ANOVA $F_{2,52} = 0.32$, $p > .7$).

Red females might have compensated by hunting longer each day, or by learning to hunt more efficiently. We have no data on total hunting duration, but increases in efficiency should be reflected by increases in weight gain rates over time. In our sample of repeat weight gain rates, we found no correlation between the change in rate and the time elapsed between measurements among red females (absolute gain rate: $r = -.01$, $N = 12$, $p > .9$; relative gain rate: $r = -.03$, $N = 12$, $p > .9$). These correlations were positive for both control groups, but not significant; pooling the controls produced marginally significant correlations (absolute gain rate: $r = .42$, $N = 20$, $p = .06$; relative gain rate: $r = 0.46$, $N = 20$, $p = .04$).

In summary, red females lost weight for a few days after the manipulation, but regained it more rapidly than expected from the normal increase in weight with age (Figure 5). We have no evidence, however, that red females compensated by learning to hunt more efficiently.

DISCUSSION

The results show that red wing spots would be costly to females, and may, in part, explain the reduced survival of males with experimentally enlarged wing spots (Grether GF, in prep-

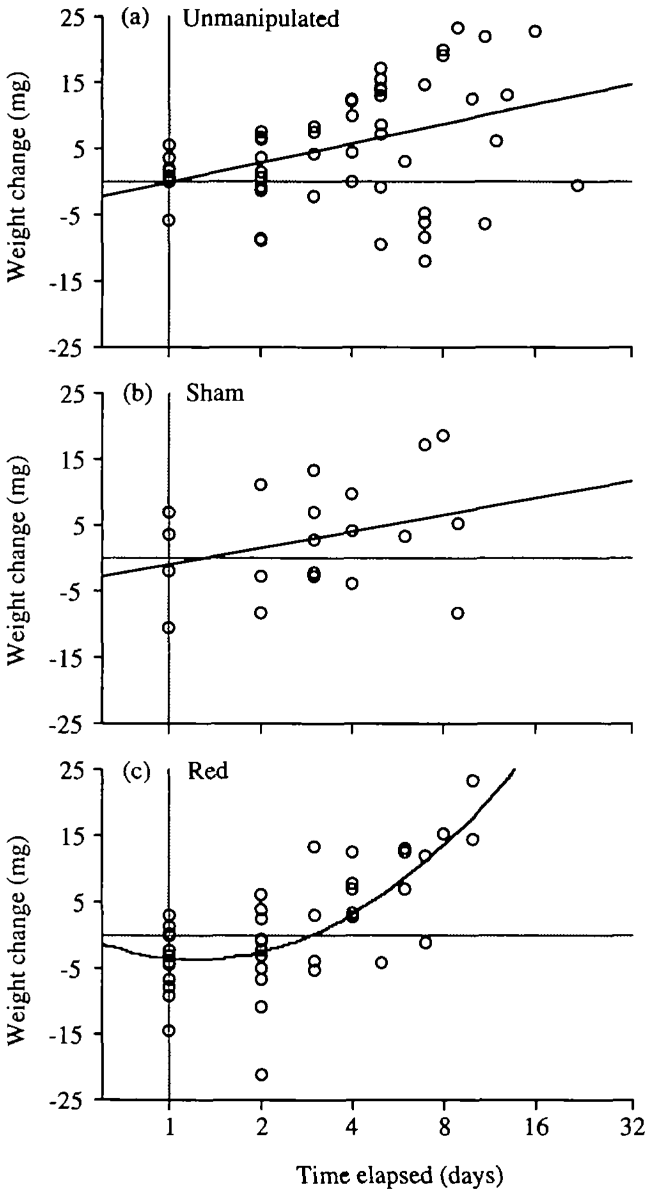

Figure 5
Change in pre-hunting weight (y) versus time elapsed (x, ln–scale) for females in the wing color manipulation experiment. Each point represents the difference between final and initial pre-hunting weights of a single female. The lines are from least squares regressions; quadratic regression was only used for red females because the squared term was significant only for this group. (a) Unmanipulated controls: $y = -0.12\ (\pm\ 2.28) + 4.22\ (\pm\ 1.45)\ x$, $F_{1,52} = 8.40$, $r^2 = .14$, $p < .01$. (b) Sham controls: $y = -0.85\ (\pm\ 3.33) + 3.60\ (\pm\ 2.55)\ x$, $F_{1,19} = 2.00$, $r^2 = .10$, $p = .17$. (c) Red-painted females: $y = -3.44\ (\pm\ 1.56) - 1.92\ (\pm\ 3.83)\ x + 4.85\ (\pm\ 1.88)\ x^2$, $F_{2,44} = 20.97$, $R^2 = 0.50$, $p < .0001$, coefficient t tests: x, $t = 0.50$, $p > .6$; x^2, $t = 2.58$, $p = 0.01$.

aration). We think the most plausible mechanism for the reduced hunting success of red females is that the artificial red spots made them more visible to prey. An alternative we cannot fully reject is that red females were distracted by the unnatural color of their own wings (Endler JA, personal communication). It seems unlikely, however, that distraction would lead to a quantitative match between the weight gain rates of males and red females. We did not test males and red females side-by-side, but in comparison to unmanipulated females, their weight gain rates were quite similar.

Why should red wing spots be conspicuous to red-blind prey? Some insects have photoreceptors tuned to long wavelengths (Bernard, 1979; Meinertzhagen et al., 1983; Peitsch et al., 1992; Yang and Osorio, 1991), but most do not (Stavenga and Schwemer, 1984). Red may in fact be the color to which rubyspot prey are the least sensitive. Nevertheless, to be cryptic, a color pattern must resemble a random sample of the visual background (Endler, 1984). To a completely red-blind animal, red would essentially appear black (Shepherd, 1988). The visual background, from the perspective of rubyspot prey, is the sky or vegetation on the bank of the creek facing the sun. Black wings would obviously be more conspicuous than clear unpigmented wings, when moving against a background of well-lighted vegetation or sky.

In general, since clear unpigmented wings transmit light from the background with little modification, no pigment or combination of pigments is likely to enhance the crypsis of insect wings in flight. The red pigment on the wings of male rubyspots and numerous other odonates (e.g., *Sympetrum* spp.) may represent a compromise between sexual selection and survival selection. Yellow or blue hues might be superior for intraspecific signaling but probably would be more conspicuous to odonate prey. Endler (1978, 1991) presented a parallel argument for male coloration and predation risk in guppies.

This study may be the first to suggest that sexual selection makes predators more conspicuous to their prey. Increased conspicuousness of predators to prey is probably less common than the reverse effect, since many sexually selected species are not predators, and not all predators display secondary sexual characters while hunting (e.g., frogs). Nevertheless, conspicuous-to-prey effects remain possible but unexamined in numerous taxa (e.g., piscivorous fish, insectivorous birds and lizards). Møller (1989) found that male barn swallows with experimentally elongated tails fed nestlings smaller prey, but he attributed the effect to reduced aerial maneuverability.

The results of this study suggest that a mutation producing male wing coloration on females would sharply reduce female hunting efficiency. Reduced efficiency could translate into decreased egg production, increased exposure to predators, or starvation when food is scarce. Interestingly, in many species of damselfly, a percentage of females (≤ 100%) develop andromorphic (male-like) wing or body coloration (De Marchi, 1990; Dunkle, 1990; Johnson, 1975; Robertson, 1985; Tillyard, 1917). In species with multiple female morphs, andromorphs are usually the rare morph, and rare morphs may benefit from reduced rates of superfluous male clasping attempts (Cordero, 1992; Forbes, 1991, 1994; Hinnekint, 1987; but see Fincke, 1994a,b). Thus far, no study has provided convincing evidence that andromorphic coloration is costly (Fincke, 1994a,b), as would be required to explain why andromorphs are usually rare. Our results suggest that it may be fruitful to examine morph differences in hunting efficiency.

The long-term weight changes of females in the wing color manipulation experiment raised questions that may warrant further study. Females lost weight for a few days after their wings were painted with red spots (Figure 5). This logically follows from their low rates of weight gain while hunting (Figure 3), but other interpretations are possible. We made focal observations only during the morning hunting period. Later in the day, when males compete for territories, red females might have suffered abnormally high rates of attack by males.

Eventually red females appeared to compensate for their handicap, gaining weight more rapidly than expected from the normal increase in weight with age (Figure 5). We found no evidence that they learned to hunt more efficiently, but two alternatives remain. The increase in weight may have been caused by an accumulation of unfertilized eggs. We saw several red females copulate and oviposit, however. Alternatively, red females may have spent a greater proportion of each day hunting.

Could males reduce the cost of conspicuousness by hunting for a greater proportion of the day? Males rarely hunt on territory, but males lacking territories frequently hunt at midday and defeated residents often hunt immediately after being ousted (Grether GF and Veldhuizen TC, in preparation). These observations suggest that conspicuous wing coloration may partly offset the advantages of territoriality.

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REFERENCES

Altmann J, 1974. Observational study of behavior: sampling methods. Behaviour 49:227–67.

Andersson M, 1994. Sexual Selection. Princeton: Princeton University Press.

Arnqvist G, 1989. Sexual selection in a water strider: the function, nature of selection and heritability of a male grasping apparatus. Oikos 56:344–350.

Arnqvist G, 1994. The cost of male secondary sexual traits: Developmental constraints during ontogeny in a sexually dimorphic water strider. Am Nat 144:119–132.

Balmford A, Thomas ALR, Jones IL, 1993. Aerodynamics and the evolution of long tails in birds. Nature 361:628–631

Bernard GD, 1979. Red-absorbing visual pigment of butterflies. Science 203:1125–1127.

Cade W, 1975. Acoustically orienting parasitoids: Fly phonotaxis to cricket songs. Science 190:1312–1313.

Cade WH. 1979. The evolution of alternative male reproductive strategies in field crickets. In: Sexual Selection and Reproductive Competition in Insects. (Blum M, Blum NA, eds). London: Academic Press; 343–379.

Clutton-Brock TH, Albon SD, Guinness FE, 1985. Parental investment and sex differences in juvenile mortality in birds and mammals. Nature 313:131–133.

Conner J, 1989. Density-dependent sexual selection in the fungus beetle, *Bolitotherus cornutus*. Evolution 43:1378–1386.

Cordero A, 1992. Density-dependent mating success and colour polymorphism in females of the damselfly *Ischnura graellsii* (Odonata: Coenagrionidae). J Anim Ecol 61:769–780.

Darwin C, 1871. The descent of man, and selection in relation to sex. London: J. Murray.

De Marchi G, 1990. Precopulatory reproductive isolation and wing colour dimorphism in *Calopteryx splendens* females in southern Italy (Zygoptera: Calopterygidae). Odonatologica 19:243–250.

Dunkle SW, 1990. Damselflies of Florida, Bermuda and the Bahamas. Gainesville: Scientific Publishers.

Eberhard WG, 1985. Sexual Selection and Animal Genitalia. Cambridge: Harvard University Press.

Endler JA, 1978. A predator's view of animal color patterns. Evol Biol 11:319–364.

Endler JA, 1980. Natural selection on color patterns in *Poecilia reticulata*. Evolution 34:76–91.

Endler JA, 1983. Natural and sexual selection on color patterns in poeciliid fishes. Environ Biol Fish 9:173–190.

Endler JA, 1984. Progressive background in moths, and a quantitative measure of crypsis. Biol J Linn Soc 22:187–231.

Endler JA, 1991. Variation in the appearance of guppy color patterns to guppies and their predators under different visual conditions. Vision Res 31:587–608.

Endler JA, 1992. Signals, signal conditions, and the direction of evolution. Am Nat 139:1–27.

Enquist M, Arak A, 1993. Selection of exaggerated male traits by female aesthetic senses. Nature 361:446–448.

Fincke OM, 1994a. Female colour polymorphism in damselflies: failure to reject the null hypothesis. Anim Behav 47:1249–1266.

Fincke OM, 1994b. On the difficulty of detecting density-dependent selection on polymorphic females of the damselfly *Ischnura graellsii*—failure to reject the null. Evol Ecol 8:328–329.

Fisher RA, 1930. The genetical theory of natural selection. Oxford: Clarendon Press.

Forbes MRL, 1994. Tests of hypotheses for female-limited polymorphism in the damselfly, *Enallagma boreale* Selys. Anim Behav 4:724–726.

Forbes MRL, 1991. Female morphs of the damselfly *Enallagma boreale* Selys (Odonata, Coenagrionidae)—a benefit for androchromatypes. Can J Zool 69:1969–1970.

Grafen A, 1990. Sexual selection unhandicapped by the Fisher process. J Theor Biol 144:473–516.

Grether GF, 1995. Natural and sexual selection on wing coloration in the Rubyspot Damselfly *Hetaerina americana.* (PhD dissertation), Davis: University of California.

Grether GF, in press. Intrasexual competition alone favors a sexually dimorphic ornament in the rubyspot damselfly *Hetaerina americana.* Evolution.

Grether GF, in press. Sexual selection and survival selection on wing coloration and body size in the rubyspot damselfly *Hetaerina americana.* Evolution.

Halliday TR. 1987. Physiological constraints on sexual selection. In: Sexual Selection: Testing the Alternatives (Bradbury JW, Andersson MB, eds). New York: Wiley; 247–264.

Harvey PH, Bradbury JW. 1991. Sexual selection. In: Behavioural ecology, An evolutionary approach (Krebs JR, Davies NB, eds). Oxford: Blackwell; 203–233.

Hasson O, 1991. Sexual displays as amplifiers: practical examples with an emphasis on feather decorations. Behav Ecol 2:189–197.

Hill GE, 1994. Geographic variation in male ornamentation and female mate preference in the house finch: a comparative test of models of sexual selection. Behav Ecol 5:64–73.

Hinnekint BON, 1987. Population dynamics of *Ischnura e. elegans* (Vander Linden) (Insecta: Odonata) with special reference to morphological colour changes, female polymorphism, multiannual cycles and their influence on behaviour. Hydrobiologica 146:3–31.

Johnson C, 1975. Polymorphism and natural selection in Ischnuran damselflies. Evol Theory 1:81–90.

Kirkpatrick M, 1982. Sexual selection and the evolution of female choice. Evolution 36:1–12.

Kodric-Brown A, Brown JH, 1984. Truth in advertising: the kinds of traits favored by sexual selection. Am Nat 124:309–323.

Lande R, 1981. Models of speciation by sexual selection on polygenic traits. Proc Natl Acad Sci USA 78:3721–3725.

Lloyd JE, Wing SR, 1983. Nocturnal aerial predation of fireflies by light-seeking flies. Science 222:634–635.

Meinertzhagen IA, Menzel R, Kahle G, 1983. The identification of spectral receptor types in the retina and lamina of the dragonfly *Sympetrum rubicundulum.* J Comp Physiol 151:295–310.

Meyer A, Morrissey JM, Schartl M, 1994. Recurrent origin of a sexually selected trait in *Xiphophorus* fishes inferred from a molecular phylogeny. Nature 368:539–542.

Møller AP, 1989. Viability costs of male tail ornaments in a swallow. Nature 339:132–135.

Neter J, Wasserman W, Kutner MH, 1985. Applied linear atatistical models: regression, analysis of variance, and experimental designs Homewood, Illinois: Irwin.

Partridge L, Endler JA. 1987. Life history constraints on sexual selection. In: Sexual selection: testing the alternatives (Bradbury JW, Andersson MB, eds). New York: Wiley & Sons; 265–277.

Peitsch D, Fietz A, Hertel H, de Souza J, Ventura DF, Menzel R, 1992. The spectral input systems of hymenopteran insects and their receptor-based colour vision. J Comp Physiol A 170:23–40.

Pomiankowski AN. 1988. The evolution of female mate preferences for male genetic quality. In: Oxford surveys in evolutionary biology (Harvey P, Partridge L, eds). Oxford: Oxford University Press; 136–184.

Robertson HM, 1985. Female dimorphism and mating behaviour in a damselfly, *Ishnura ramburi*, females mimicking males. Anim Behav 33:805–809.

Ryan MJ, 1985. The Tungara frog: A study in sexual selection and communication. Chicago: University of Chicago Press.

Ryan MJ. 1990. Sexual selection, sensory systems, and sensory exploitation. In: Oxford surveys in evolutionary biology (Futuyma D, Antonovics J, eds). Oxford: Oxford University Press; 157–195.

Ryan MJ, Tuttle MD, Rand AS, 1982. Bat predation and sexual advertisement in a neotropical anuran. Am Nat 119:136–139.

Seger J, 1985. Unifying models for the evolution of female choice. Evolution 39·1185–1193.

Shepherd GM, 1988. Neurobiology. New York: Oxford University Press.

Sokal RR, Rohlf FJ, 1981. Biometry. New York: W.H. Freeman and Co.

Stavenga DG, Schwemer J. 1984. Visual pigments of invertebrates. In: Photoreception and vision in invertebrates (Ali MA, ed). New York: Plenum Press; 11–62.

Tillyard RT, 1917. The biology of dragonflies. London: Cambridge University Press.

Vehrencamp SL, Bradbury JB, Gibson RM, 1989. The energetic cost of display in male sage grouse. Anim Behav 38:885–896.

Weichsel JI, 1987. The life history and behavior of Hetaerina americana (Fabricus) (Odonata: Calopterygidae). (PhD dissertation), Ann Arbor: University of Michigan.

Wingfield JC, Hegner RE, Dufty AMJ, Ball GF, 1990. The "challenge hypothesis": Theoretical implications for patterns of testosterone secretion, mating systems, and breeding strategies. Am Nat 136: 829–846.

Yang ENC, Osorio D, 1991. Spectral sensitivities of photoreceptors and lamina monopolar cells in the dragonfly, *Hemicordulia tau*. J Comp Physiol 169:663–669.

Zahavi A, 1978. Decorative patterns and the evolution of art. New Scientist 19:182–184.

Zahavi A, 1982. The pattern of vocal signals and the information they convey. Behaviour 80:1–8.