Female guppies use orange as a mate choice cue: a manipulative test

R. Brooks and N. Caithness

Communication Biology Research Group, Department of Zoology, University of the Witwatersrand, Private Bag 3, Wits 2050, Republic of South Africa
e-mail: robb @ gecko.biol.wits.ac.za

Received 6 February 1995; accepted 30 August 1995

Female guppies from a feral South African population respond sexually to more orange males in correlative trials. We impaired the female's ability to use orange elements of male colour patterns by conducting choice trials under orange light. Under orange light, there was no relationship between male colour pattern and female sexual response, but under white and blue light treatments there was such a relationship. Other male ornaments were not preferred by females either under normal lighting conditions or when their ability to use orange as a cue was impaired. The implications for sexual selection theory and the evolution of multiple male ornaments are discussed.

Evidence of female mate choice using visual cues is often correlative, failing to show that the character studied is the object of preference rather than a correlate of the real cue. Distinguishing between correlation and causation is especially important when males have several secondary sexual characters, each of which may be sexually selected. One solution is to experimentally manipulate the ornament in question (eg. Andersson 1982), but colour patterns are often not as easily manipulated as, for example, the tails of birds. Long & Houde (1989) devised an ingenious test of whether the correlation observed between orange (carotenoid) area and female preference in a Trinidadian guppy population (Houde 1987) indicated a real preference for orange males. Instead of altering the colour patterns of males, they varied the colour of incident light, thus altering female perception of these patterns.

Previously, we found a significant positive correlation between orange areas of males and both female preference (Brooks & Caithness 1995) and mating success (Brooks & Caithness, unpubl.) in a feral guppy population from Durban, South Africa. We designed the following experiment both as a manipulative test of this trend and an examination of the effect of other secondary sexual characters when the cue of choice is factored out.

Furthermore, the importance of courtship and female choice as a determinant of male mating success changes with lighting conditions (Endler 1987; Reynolds, Gross & Coombs 1993), and so we examined the effects of light colour on general female responsiveness.

We used wild-caught or first generation laboratory-bred guppies. The colour patterns (under white light) of each male were drawn onto standard guppy outlines (after Houde 1987) and relative area of each colour spot was estimated using a grid of squares. This method of quantifying colour patterns is sufficient for discriminating which male of a pair has a greater relative area of a pigment. Also, it provides a good measure of iridescent colour areas which do not show up well on photographic film. We measured pattern complexity (after Nicoletto 1993) by ranking each male (relative to others in the sample) for the number of spots of each colour (orange, blue, white, black and yellow) and then adding the five ranks to give a measure of male pattern complexity relative to the other males in the sample.

Two males (paired to vary dramatically in the percentage of their surface covered by orange spots) and two virgin females were placed in a tank (900 mm × 315 mm filled to 270 mm, with brown paper on three sides to standardise background colour) at noon, and behavioural trials took place on the next three mornings. Ten hours of light from fluorescent tube lighting was provided beginning at 12h00 each day, preceded by 2 h of either orange, blue or white light such that each colour was presented on one morning of the trial. The order in which the lighting colours were presented was random in each of twenty replicates.

After a 15-min adjustment period, we watched each male for two 10-min periods, 15 min apart. Gonopodial thrusts, nipping a females' gonopore, chasing the female, and chasing, nipping and displaying to the other male were all noted. We scored the response of a female to a displaying male as either: no response, 'orient' towards male, 'glide' towards male or 'mate' (described by Brooks & Caithness 1995). The proportion of displays that received an 'orient response' and the proportion 'glide' response all significantly predict mating success (Brooks & Caithness submitted; Houde 1988). Only results for glide response are presented here as it is the best predictor of mating success (Brooks & Caithness, unpubl.) and other measures did not provide substantially different results.

Non-parametric statistical tests (Siegel & Castellan 1988) were used because behavioural measures could not be transformed to normality. We examined the three treatments (orange, blue and white light) for systematic variation in the frequency of the various behaviours studied using a Kruskal-Wallis one-way analysis. Wilcoxon rank-sum tests were used to check if the male with a greater relative area of a particular colour was preferred over the other within treatments.

Females preferred the more orange male of a pair under white and to a lesser extent under blue light, but not under orange light (Figure 1). Female response did not differ significantly between males with greater and lesser expression of any other secondary sexual character, including 'pattern complexity', measured under orange light as follows (Wilcoxon rank-sum tests, for all tests $n = 40$): black area ($z = 0{,}447$; n.s.), white area ($z = 1{,}069$; n.s.), iridescent blue ($z = 0{,}000$; n.s.), yellow ($z = -1{,}000$; n.s.) and complexity ($z = 0{,}0535$; n.s.). The only variable that may have influenced attractiveness is black area under blue light ($z = 1{,}820$, one-tailed $p = 0{,}035$).

There was significant systematic variation in female responsiveness (proportion 'glide' response to all male displays) between light colour treatments (Kruskal-Wallis test statistic 7,38; $p = 0{,}025$). Responsiveness was lowest under orange light (0,024 ± 0,06) and highest under white light (0,170 ± 0,19).

Our finding that females prefer males with a greater relative area of orange under white light, but not under orange

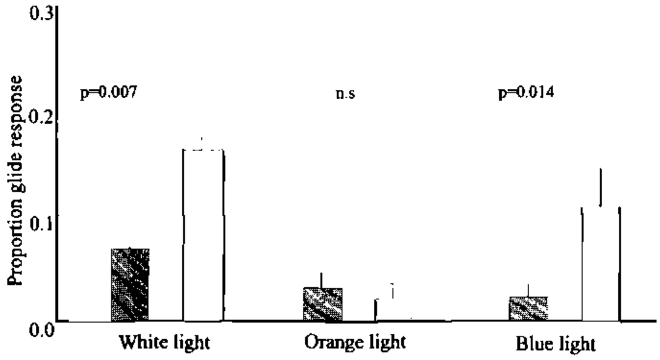

Figure 1 Female response (± 1 *SD*) to males with smaller (shaded) and greater (unshaded) relative area of orange. Probabilities are one-tailed probabilities according to Wilcoxon's Rank sum test. All comparisons are based on a sample of 20 trials (40 males).

light (when males look uniformly orange) supports the notion that carotenoid patches are the cue females choose. Paradoxically, under blue light, which contains little or none of the 'orange' wavelengths reflected by orange spots (causing them to appear black), females show significant preference both for orange (as Long & Houde also found) and for black areas. It is possible that females from this population use the complexity (Nicoletto 1993) or contrast (Endler & Houde 1995) of males' colour patterns, rather than orangeness, in mate choice. There are indications, from an earlier mating success experiment (Brooks & Caithness, submitted) that pattern complexity is important to choice in this population, despite not being significantly related to choice in the present study. Therefore, our results may indicate that females are choosing males with more complex patterns.

The redundant-signal hypothesis (Møller & Pomiankowski 1993) for the presence of multiple secondary sexual characters postulates several cues which advertise phenotypic quality to females. As Zuk, Ligon & Thornhill (1992) manipulated the combs of red jungle fowl cocks, hens switched to other secondary sexual characters to choose mates. The low female receptiveness despite sustained male courtship intensity under orange light may account for our failure to detect a switch in the cue of choice. However, this is difficult to distinguish from two possible effects of orange light on female perceptions of male patterns. Firstly, because unpigmented areas and orange spots both reflect all wavelengths of incident light, causing males to appear uniformly orange, it should be impossible for females to choose using orange. Alternatively, males may appear uniformly lacking in orange owing to visual adaptation and bleaching (by overstimulation) of female's orange-red photoreceptors (see Long & Houde 1989 for a fuller discussion). This could lead to a failure to meet a threshold criterion of mate choice. If female guppies failed to choose on the basis of orange area for either of these reasons, the redundant-signal hypothesis predicts that one or more other characters will be used for choice (Møller & Pomiankowski 1993). At this point, only orange area is convincingly the object of a preference.

Acknowledgements

Wendy Wolhuter, Dan Polakow, Mike Jennions, Anne Houde, Pat Backwell and two anonymous referees provided stimulating discussion and helpful comments on the manuscript. Financial support in the form of bursaries (RB) from the University and the FRD as well as FRD and University CBRG grants to Robin Crewe are gratefully acknowledged. Animal ethics screening committee number 93-32-1.

References

ANDERSSON, M. 1982. Female choice selects for extreme tail length in a widowbird. *Nature*. 299:818–820.

BROOKS, R.C. & CAITHNESS, N. 1995. Female choice in a feral guppy population: are there multiple cues? *Anim. Behav.* 50: 301–307.

ENDLER, J.A. 1987. Predation, light intensity and courtship behaviour in *Poecilia reticulata* (Pisces: Poecilidae). *Anim. Behav.* 35: 1376–1385.

ENDLER, J.A. & HOUDE, A.E. 1995. Geographic variation in female preferences for male traits in *Poecilia reticulata. Evolution* 49: 456–468.

HOUDE, A.E. 1987. Mate choice based upon naturally occurring color-pattern variation in a guppy population. *Evolution* 41: 1–10.

HOUDE, A.E. 1988. The effects of female choice and male-male competition on the mating success of male guppies. *Anim. Behav.* 36: 888–896.

LONG, K.D. & HOUDE, A.E. 1989. Orange spots as a visual cue for female mate choice in the guppy (*Poecilia reticulata*). *Ethology* 82:316–324.

MOLLER, A.P. & POMIANKOWSKI, A. 1993. Why have birds got multiple sexual ornaments. *Behav. Ecol. Sociobiol.* 32: 167–176.

NICOLETTO 1993. Female sexual response to condition-dependent ornaments in the guppy, *Poecilia reticulata*. *Anim. Behav.* 46: 441–450.

REYNOLDS, J.D., GROSS, M.R. & COOMBS, M.J. 1993. Environmental conditions and male morphology determine alternative mating behaviour in Trinidadian guppies. *Anim. Behav.* 45: 145–152.

SIEGEL, S. & CASTELLAN, N.J. 1988. Nonparametric Statistics. McGraw-Hill, Singapore.

ZUK, M., LIGON, J.D. & THORNHILL, R. 1992. Effects of experimental manipulation of male secondary characters on female mate preference in red jungle fowl. *Anim. Behav.* 44: 999–1006.