



Context-dependent preferences vary by multicomponent signals in a swordtail



Luke Reding*, Molly E. Cummings

Department of Integrative Biology, University of Texas at Austin, Austin, TX, U.S.A.

ARTICLE INFO

Article history:

Received 4 January 2017
Initial acceptance 1 March 2017
Final acceptance 1 March 2017
Available online 24 June 2017
MS. number: A17-00024R

Keywords:

complex signal
decision making
mating preference
multicomponent
swordtail

Female mating preferences can be both context dependent and based on the assessment of multicomponent male signals. Here, we assess the social context dependence of female mating preferences for two components of a male's multicomponent signal. We dissected the visual signal of male *Xiphophorus nigrensis* swordtails, a species in which males vary by both size and degree of courtship, to test (1) how the identity of males in a given choice influences female mating preferences and (2) how females perceptually integrate a male's multicomponent signal. We used validated male animations that generate repeatable female responses to test mating preferences for size and courtship vigour, separately and together, using dichotomous choice tests. When keeping courtship vigour constant, females discriminated between males only when there was a large size difference between them. When keeping size constant, the identity of males in a choice reversed a preference for a vigorously courting male. We found no evidence that females perceptually bind the separate components of a male's signal additively. However, females were faster to approach males when the males varied in both size and courtship than when the males only varied in size, perhaps favouring the evolution of multicomponent signals in males. © 2017 The Association for the Study of Animal Behaviour. Published by Elsevier Ltd. All rights reserved.

Although Darwin and Wallace agreed on many things, the relative importance of male ornamentation to a female's mate choice decision was not one of them. While Darwin contended that ornamentation was more important to females (Darwin, 1859), Wallace maintained that courtship vigour was the main interest (Wallace, 1889) and that ornamentation simply amplified the intensity of the male's courtship. Although the surge of interest in the past three decades on female mating preferences has mostly focused on the importance of ornamentation, there is evidence to suggest Wallace might have been right as well (Byers, Hebets, & Podos, 2010; Cornuau, Rat, Schmeller, & Loyau, 2012).

Male mating signals of the sort that Darwin and Wallace disagreed about are 'multicomponent signals' if females perceive the parts of the signal—e.g. ornamentation and courtship vigour—in a single sensory modality (Rowe, 1999). Multimodal signals span modalities, like vision and audition (Hebets & Papaj, 2005). Both types of signals, which we call 'complex signals' (following Hebets & Papaj, 2005), are ubiquitous in mating interactions (Candolin, 2003) in part because they are thought to be more detectable, eliciting faster reactions from females (Rowe, 1999). The study of

complex signalling is indebted to the descriptive categorizations of Johnstone (1996) and Møller and Pomiankowski (1993) that were later expanded by Partan and Marler (1999, 2005). These categorizations are useful in understanding how different components of a complex signal interact to affect some response in a receiver. A common assumption is that females can independently assess the different components of a signal and integrate them by additively combining them (Hebets, 2005; Hebets & Papaj, 2005; Stange, Page, Ryan, & Taylor, in press), although this need not be the case.

One of the challenges still facing the study of complex signal processing and design is understanding how females process multiple signal components that vary continuously. As noted by Wagner (1998), presentation of two extremes of a signal component (i.e. presence and absence) is best suited for testing for directional preferences. In many species, the components of a signal vary continuously, and work in treehoppers (Fowler-Finn & Rodríguez, 2012; Rodríguez, Hallett, Kilmer, & Fowler-Finn, 2013), guppies (Blows, Brooks, Kraft, & Phillips, 2003; Cole & Endler, 2015) and crickets (Bentsen, Hunt, Jennions, & Brooks, 2006; Gray, Gabel, Blankers, & Hennig, 2016; Reichert, Finck, & Ronacher, 2017) shows that female preferences for components of male traits are often not directional. Smith and Evans (2013) recognized this limitation and encouraged researchers to consider continuous variation in signal components when studying multimodal communication. Ronald,

* Correspondence: L. Reding, Department of Integrative Biology, University of Texas at Austin, 1 University Station C0990, Austin, TX 78712, U.S.A.

E-mail address: lukereding@utexas.edu (L. Reding).

Zeng, White, Fernández-Juricic, and Lucas (in press) further expanded on this work and created a theoretical framework for understanding how continuously varying signal components influence the overall response to a complex signal.

At the same time, evolutionary biologists interested in the patterns of multivariate selection that females impose on males have studied complex signalling for decades. Female guppies, for example, impose complex linear and nonlinear selection on male coloration (Blows et al., 2003; Brooks & Endler, 2001; Cole & Endler, 2015). These studies used statistics to understand patterns of female preference for the separate components of a male's coloration. But by relying on naturally occurring variation in male traits, it becomes difficult to fully understand the individual contributions of signal components on the receiver's response. Use of live males also makes it difficult to partition a female's response to the male's coloration from other components of his visual signal, such as his behaviour. Correlations between signal components make teasing apart these contributions even more difficult. A more powerful approach would be to synthetically engineer signals (Bentsen et al., 2006; Gray et al., 2016; McClintock & Uetz, 1996; Reichert et al., 2017; Rosenthal & Evans, 1998), giving the researcher precise control over the signal and the ability to decouple signal components that might typically be tightly correlated, although this has rarely been done for visual signals.

While most of the ideas about how complex signals are perceived by receivers assume that signals will be perceived the same way regardless of context, female mating preferences can also be influenced by the environment (Jennions & Petrie, 1997). For example, female mating preferences for colourful males can be abolished in the presence of a predator (Forsgren, 1992; Godin & Briggs, 1996). Although context-dependent or plastic preferences are often studied by examining how mating preferences change in different physical (e.g. lighting) environments (Fuller & Noa, 2010; Maan, Seehausen, & Van Alphen, 2010), the social environment is increasingly recognized as playing a large role in shaping a female's choice of mate (Rodríguez, Rebar, & Fowler-Finn, 2013). Mate choice copying is one example where the presence of a conspecific interacting with a potential partner can increase that individual's perceived attractiveness by an observer (Auld & Godin, 2015; Schlupp, Marler, & Ryan, 1994; Witte & Ryan, 2002). Furthermore, multiple experiments have demonstrated that exposure to specific male types at developmental (Fowler-Finn & Rodríguez, 2012; Hebets, 2003; Verzijden & Rosenthal, 2011) or adult (Tudor & Morris, 2011) life stages can alter female responses towards males during subsequent encounters.

Here we explore how the immediate social environment, specifically the available set of males that a female is deciding among (the 'choice set' of males), influences her choice behaviour. This relatively unexplored mechanism of how the social environment could affect mating preference is particularly important with multicomponent signals that show continuous variation. How will a female respond when confronted with two males, each more attractive for a different component of his multicomponent signal? In humans (Huber, Payne, & Puto, 1982) and other animals (Lea & Ryan, 2015; Locatello, Poli, & Rasotto, 2015; Royle, Lindström, & Metcalfe, 2008), the options immediately available to a female chooser can alter her choice behaviour (Bateson & Healy, 2005). In túngara frogs, *Engystomops pustulosus*, for example, an initial preference for one of two male calls in a dichotomous choice test can be reversed by adding a third call to the 'choice set' of available calls (Lea & Ryan, 2015). This mechanism is distinct from the social exposure experiments outlined above because all females have the same exposure to male signals: the difference is in the males that are immediately available for comparison (Patricelli & Hebets, 2016). Many studies

that measure female mating preferences for different male signals use no-choice tests (Fowler-Finn & Rodríguez, 2012; Girard, Elias, & Kasumovic, 2015; Hebets, Vink, Sullivan-Beckers, & Rosenthal, 2013; Wagner, Smeds, & Wiegmann, 2001). These are tests in which a female is presented with a single type of male or male signal and her response to that signal alone is recorded. Thus the role that the choice set of males has on female preferences for different males has often been neglected in studies of mate choice behaviour, and even more so in studies of complex signalling.

Here, we study context dependence of female mating preference for two continuously varying components of a male multicomponent signal in the El Abra pygmy swordtail *Xiphophorus nigrensis*. Males of this species have genetically based alternative mating phenotypes: large males court females, small males attempt coercive matings with females, and intermediate males can do both (Ryan & Causey, 1989; Zimmerer & Kallman, 1989). Females prefer large males over small males in dichotomous tests with live males (Cummings & Mollaghan, 2006; Wong, So, & Cummings, 2011) and respond to synthetic animations of males (Rosenthal, Wagner, & Ryan, 2002), allowing the decomposition of a male's visual signal. *Xiphophorus nigrensis* is therefore an attractive species in which to test both perceptual integration of signals and context dependence.

Our goals in this study were three-fold. First, we wanted to test whether females perceptually bind two components of a male's multicomponent signal in an additive or nonadditive way. To do this, we presented female swordtails with a series of animated males that differed (1) only in size, (2) only in courtship vigour or (3) in both size and courtship vigour. This dissection of a male's visual signal allowed us to measure female mating preferences for each component separately and together. To test for additive preferences, we compared female preference responses in each of the single component trials to the combined multicomponent trials. If females bind separate components of a male's visual signal in a strictly additive way, then their preference scores for the multicomponent trials should be predicted by the summed quantity of their responses for each of the individual component trials.

Second, we tested whether female preferences for specific male components (his size or courtship rate) were absolute or varied depending on the expression of these components between the available males present (the 'choice set' of males). To test the influence of a female's choice set, for each signal component we created three types of males that varied in component properties (small, intermediate and large males that displayed either no, low or high levels of courtship). We tested female mating preferences in dichotomous choice tests for each pairwise combination of males. This allowed us to assess female mating preferences for a given type of male when paired against two other types of males. If female preferences for specific components are absolute, then preferences for that male component should be constant across different choice sets. If, however, a female's preference for male signal components is dependent on how those signals are perceived in the presence of other males with different component states, then we predicted that female preferences would vary across choice sets.

Third, to test a possible adaptive function of multicomponent signalling, we tested whether multicomponent signals evoke faster reaction times than individual stimuli (as predicted by Rowe, 1999). To test this hypothesis, we examined whether latency to approach males in our experiments was shorter when males varied in both courtship intensity and size relative to the single component experiments. We predicted that females would approach males more quickly when males varied in size and courtship compared to when males varied in only one dimension (size or courtship).

METHODS

Fish and Rearing

In total, we tested 81 mature adult *X. nigrensis* females (22 females for validation experiments, 59 females for preference experiments). All fish were descendants of wild-caught fish from the Nacimiento de Río Choy in Mexico kept outside in seminatural 800-gallon (3028.3-litre) tanks at Brackenridge Field Laboratory at the University of Texas at Austin. Fish were housed under a 14:10 h light:dark cycle and fed flakes daily, supplemented once a week with live brine shrimp larvae. Test females were not virgins, but unlike guppies, female *X. nigrensis* show stable preferences across their reproductive cycle (Ramsey, Wong, & Cummings, 2011). To standardize female responsiveness to males, we sexually isolated females from males (but housed them with other females) for 2 weeks prior to the start of the experiment. All fish were fed prior to testing. All experimental procedures were approved by the Institutional Animal Care and Use Committee of the University of Texas at Austin (protocol number AUP-2013-00156). Fish were originally collected in 2009 under fishing permit number DGOPA.07311.130709.-2261 from the Mexican National Commission of Aquaculture and Fisheries and maintained for several generations in seminatural conditions. All fish were returned to communal breeding tanks in the laboratory after use in the experiment.

Animation Design

We created animations in Blender (<https://www.blender.org/>) based on lateral and dorsal photographs of male fish in our laboratory (see [Supplementary Material](#) for details). Briefly, we used these two photos to generate a three-dimensional representation of a large and a small *X. nigrensis* male. We then overlaid the photographs onto this model and associated 'bones' with different parts of the model to simulate movement. The behaviours in the animations were based on videos of large males courting females and of small males approaching females in the laboratory. For our validation experiment, we used animations of a large male (Fig. 1b) performing courtship displays compared against a screen that displayed the background image only (Fig. 1c) and the same large male compared against a small male (Fig. 1a and d).

In our single and multicomponent preference experiments, we used the model from our large male animations and varied his size and/or courtship intensity. We presented females with large, intermediate and small male animations that were 36 mm, 28.75 mm and 21.5 mm, respectively. These sizes were chosen because they are well within the ranges for each size class of *X. nigrensis* (Ryan, Pease, & Morris, 1992). The sizes of the large and small animations were the average sizes of large and small males used in dichotomous choice tests in our laboratory (Cummings & Mollaghan, 2006; Wong et al., 2011).

Each animation consisted of a fish swimming onto the screen, performing a series of courtship behaviours or not, then swimming off the screen. Based on videos of courting males, we defined a courtship sequence in our animation. The behaviour involved a male orienting himself laterally towards the female, fanning his pectoral fins, curving his body towards the female and flaring his dorsal fin. Males that lacked courtship never showed this behaviour. Males that showed low courtship behaviour swam placidly with little movement following each courtship behaviour. High courtship males constantly displayed towards females after swimming onto the screen. An example animation (the intermediate-sized, intensely courting stimulus male) can be found in the [Supplementary Material](#), Video S1; all animations are available from the authors upon request.

Supplementary data related to this article can be found online at <http://dx.doi.org/10.1016/j.anbehav.2017.05.017>.

Validation Experiment

To validate our animations as salient mating stimuli, we performed two tests with 22 females (following Künzler & Bakker, 1998). First, if females are sexually motivated, they should spend more time with an animated male than with the background against which the male is presented. We created an animated large courting male and tested whether females spent more time with the animation than with the background image against which the male was presented. Second, females should be able to discriminate between animated males as they do live males. In *X. nigrensis*, females generally show a preference for large courting males when compared to small coercive males (e.g. Wong et al., 2011). We used the same large male animation from the validation experiment and created a second animation of a small male that exhibited behaviours typical of his morph. We predicted that females would spend more time with the large male in this experiment if they are able to discriminate among animations as they do live males. Additional details about these tests are provided in the [Supplementary Material](#).

Preference Experiments

Housing

In each of the three experiments (manipulation of size only, courtship only, size and courtship), each female received three different binary (dichotomous) tests. The time between trials was at least 5 days, but no more than 14 days. We wanted a long period in between trials to ensure that females were not fatigued and to keep their receptivity to the animations high. Because the animations were not interactive, females tested repeatedly day after day could become nonresponsive to the same animation. Females were housed in pairs. We used the difference in size between females to identify each fish. We were therefore able to keep track of individual fish identities while allowing them social companionship throughout the course of the experiment.

Acclimation

We first acclimated the females to the test environment by placing groups of seven to eight females ($N = 8$ groups) in the tank for 2 h at a time for 2 days. The screens showed the background image against which the animations were presented. The purpose of these acclimation trials was to habituate the female to the tank and reduce her stress in the tank environment.

Animation presentation

We used the large male model from the validation experiment for all preference experiments. This allowed us to test the effect of size and courtship vigour independently from all other aspects of a male's phenotype (see [Supplementary Material](#) for details regarding animation design). The animations were presented on two 7-inch 1024 × 600 LCD screens (<https://www.adafruit.com/products/2300>) that were directly attached to opposite ends of an aquarium (45 × 45 cm) and connected to a Mac mini computer. The small size and thinness of these screens allowed us to attach each screen directly to the side of the experimental tank, minimizing the distance light travelled through air before going through the Plexiglas of the tank and through the water to the fish. The test tank had a gravel substrate and an artificial plant in the middle, adjacent to one of the tank walls, and was filled to a depth of 10 cm with water at 25.6–26.7 °C. The tank was otherwise covered in blue felt to minimize reflections and other distractions. We used full-spectrum fluorescent lights with filters to mimic more natural

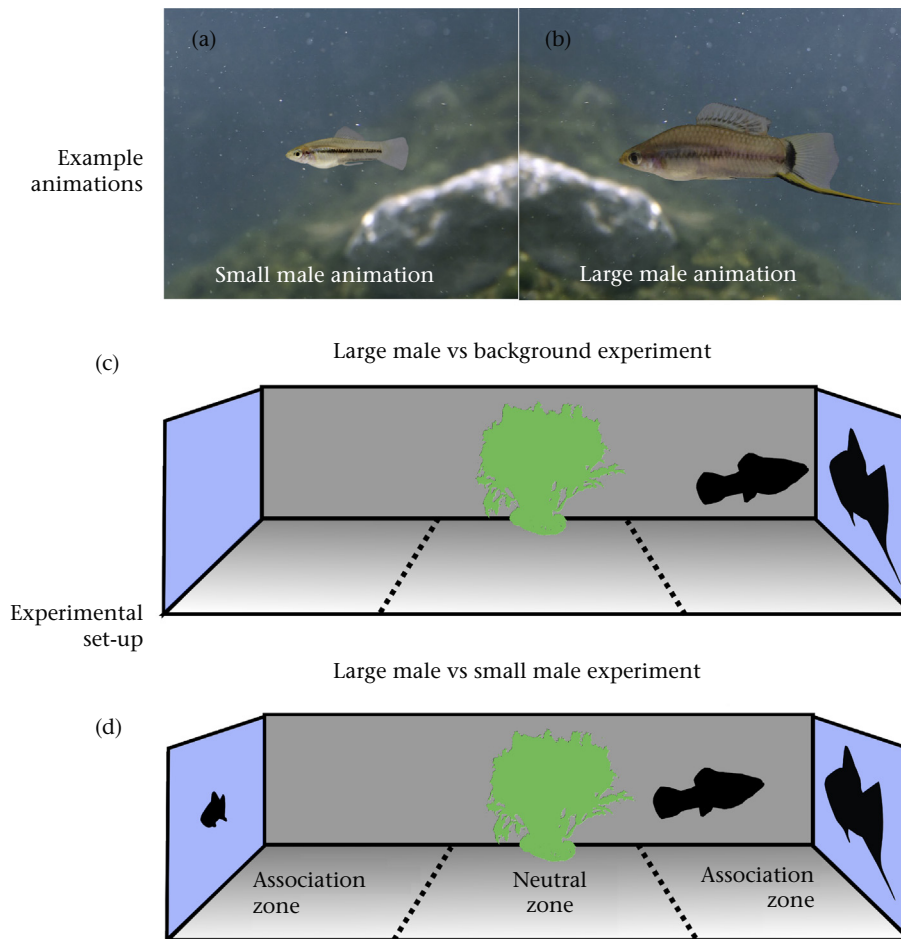


Figure 1. Validation experiment. Example screenshots from the (a) small male and (b) large male animations. Experimental set-up for (c) the large male versus background experiment and (d) the large versus small male experiment.

lighting conditions to illuminate the tank. A Microsoft LifeCam webcam mounted above the tank recorded each trial.

Testing

We used 20 females in the size-only experiment, 19 in the courtship-only experiment and 40 in the size and courtship experiment. This latter test consisted of 20 females that had not been tested, 10 females that were tested in the size-only experiment and 10 females that were tested in the courtship-only experiment. The order of the three dichotomous choice trials was determined by a random number generator, as was the side of the tank on which each male first appeared.

Each test lasted 21 min. The first 5 min showed only the background image against which the animations were presented. The next 5 min showed the animations, one animation on each side of the tank. The next minute was of the background images. The animations then appeared for another 5 min, appearing on the opposite site of the tank as they had originally. Each trial ended after an additional 5 min of the background image. The animations were shown using a custom Python script using psychopy (Peirce & Peirce, 2009) and the videos were recorded with ffmpeg (see [Supplementary Material](#) for further details).

Analysis and Statistics

We used an improvement of a tracking script written in Python using OpenCV (Bradski, 2000) to track the fish in each trial. This makes data collection more precise, repeatable and objective. The correlation

between measures output by the programme and by blinded observers is very high ($r = 0.97$, $P < 0.001$; Reding & Cummings, 2016). We used GNU parallel (Tange, 2011) to batch process the videos.

All analyses based on the tracking output were done in R v.3.3.3 using packages in the tidyverse (Wickham, 2016). We defined two equally sized regions extending 5 cm in front of each screen and 15 cm wide as the preference zones. We calculated how long the fish spent in front of each screen while the screens were showing animations of males. We used the proportion of time that a female spent with a given male relative to the time she spent with both males as our measure of preference. This metric is highly repeatable (see below) and is correlated with similar measures of preference (e.g. difference in time spent between the two males; see [Supplementary Material](#)). We calculated a female's latency to approach either screen as the amount of time elapsed between when the animations began playing and when she entered one of the preference zones in front of either screen, excluding females that were in a preference zone when the screens began to show animations of males. To account for side bias, we excluded females that, on average (across the three trials), spent more than 90% of their time with male animations presented on one side of the tank ($N = 1$ in the size-only experiment, $N = 2$ in the courtship-only experiment, $N = 11$ in the size and courtship experiment).

Our main statistical question was whether the population-level preference for a given male in each dichotomous choice trial would differ from chance, or from our null expectation of 0.5 (equal time spent with each male). We assessed the statistical significance of all preference measures with the permutation equivalent of one-

sample t tests. We used permutation tests because they make no assumptions about the distribution from which that data are drawn. We used 9999 permutations and calculated P values as recommended by Ruxton and Neuhäuser (2013).

To test our hypothesis that the experiment in which males varied in two components would elicit faster responses from females, we compared the latency of females to enter the male preference zones between the experiments. We used the `lsmmeans` package (Lenth, 2016) in R to test for a difference between the two unicomponent experiments and the multicomponent experiment. The input to the `lsmmeans` function was a linear mixed effects model created using the `nlme` package in R (Pinheiro, Bates, DebRoy, Sarkar, & R Core Team, 2017). Latency was the response variable, experiment was a fixed effect and female was a random effect to control for repeated testing of females. Tukey post hoc tests were implemented by the `multcomp` package (Hothorn, Bretz, & Westfall, 2008). To test whether trial order affected female preferences, we used a linear mixed effect model with preference as the response variable and experiment, trial and order as fixed explanatory variables. Female was a random effect to control for repeated testing. We assessed significance of the main effects with type II sums of squares using the `car` package (Fox & Weisberg, 2011). In the size + courtship experiment, some of the females had previously been tested in an earlier experiment. To test whether prior use in an experience influenced a female's preferences, we used a linear mixed effects model with preference as the response variable and whether the female was experienced or not was the explanatory variable. Female was again included as a random effect. All statistical tests were two tailed.

RESULTS

To validate that females respond to the animations of males as they do live males, we first tested whether females spent more time

near an animated male compared with the background against which the male was presented (Fig. 1c), as in Künzler and Bakker (1998). Females indeed spent more time with the large male than with the background image ($P < 0.001$; Fig. 2a). We next tested whether the preference for large relative to small male animations was similar to that found using live *X. nigrensis* males (average preference score of 0.685 in Wong et al., 2011; 0.7 in Cummings & Mollaghan, 2006). We found that females spent more time with the large male than with the small male ($P = 0.013$, average preference = 0.61; Fig. 2b), similar to their preference for live males.

Additionally, we tested whether a female's preference for the large male was repeatable across the two validation experiments. We found that female preference for the large male in the large male versus background image test significantly predicted her behaviour in the large male versus small male experiment (Pearson correlation test: $r_{17} = 0.74$, $P < 0.001$; Fig. 3a). This high repeatability is unusual for mating preferences: Fig. 3b shows the distribution of mating preference repeatability scores from a meta-analysis (Bell, Hankison, & Laskowski, 2009) compared with our estimate. Our estimate is substantially higher than previous preference assays with this species using live males ($r_{13} = 0.32$ in Cummings & Mollaghan, 2006). Thus, our male animations allow us to generate highly repeatable mating preferences in female swordtails.

To test for both perceptual integration of a male's multimodal signal and context dependence of preferences, we tested female swordtails in a series of dichotomous choice tests using animations of males. An example testing sequence is shown in Fig. 4a. In the first experiment, male courtship vigour was fixed at an intermediate level, but male sizes were typical of small, intermediate or large males (Fig. 4b). All other visual features of the male were kept constant. Across all three size pairings, we only found a preference for size when the large male was paired with the small male ($P = 0.0431$; Fig. 4e). There was no overall preference for the large

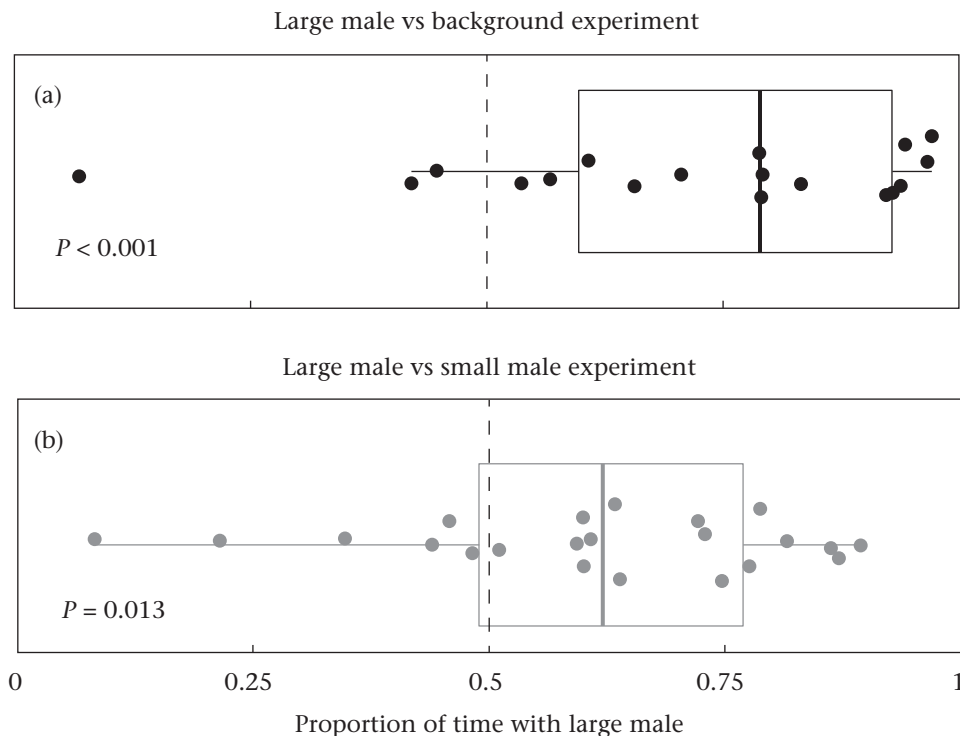


Figure 2. Results for (a) the large male versus background experiment and (b) the large versus small male experiment. Box plots show the first and third quartiles (boxes), median (line inside each box) and range excluding outliers (the whiskers that extend away from each box). Points represent the raw data.

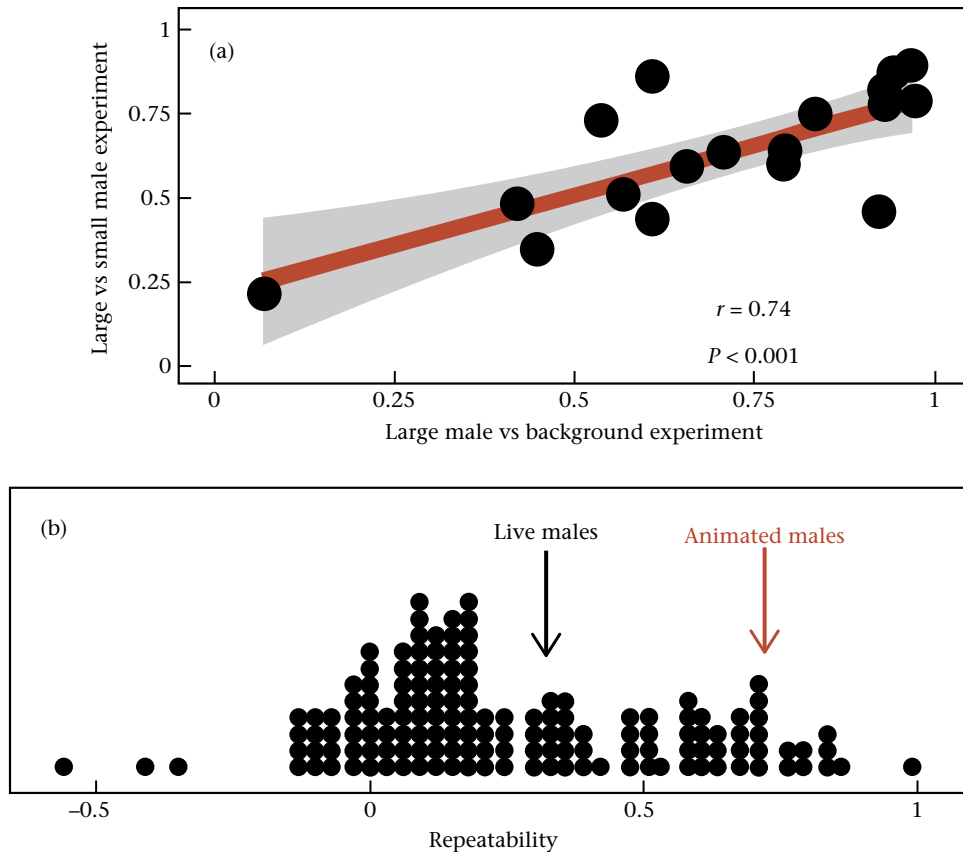


Figure 3. (a) Correlation between a female's preference for the large male in the large male versus background experiment (X axis) and the large male versus small male experiment (Y axis). (b) Repeatability estimates from this study compared with estimates from the literature. Each point represents an estimate of female mating preferences taken from Bell et al. (2009). The two arrows represent repeatability estimates using animated males and live males from Cummings and Mollaghan (2006).

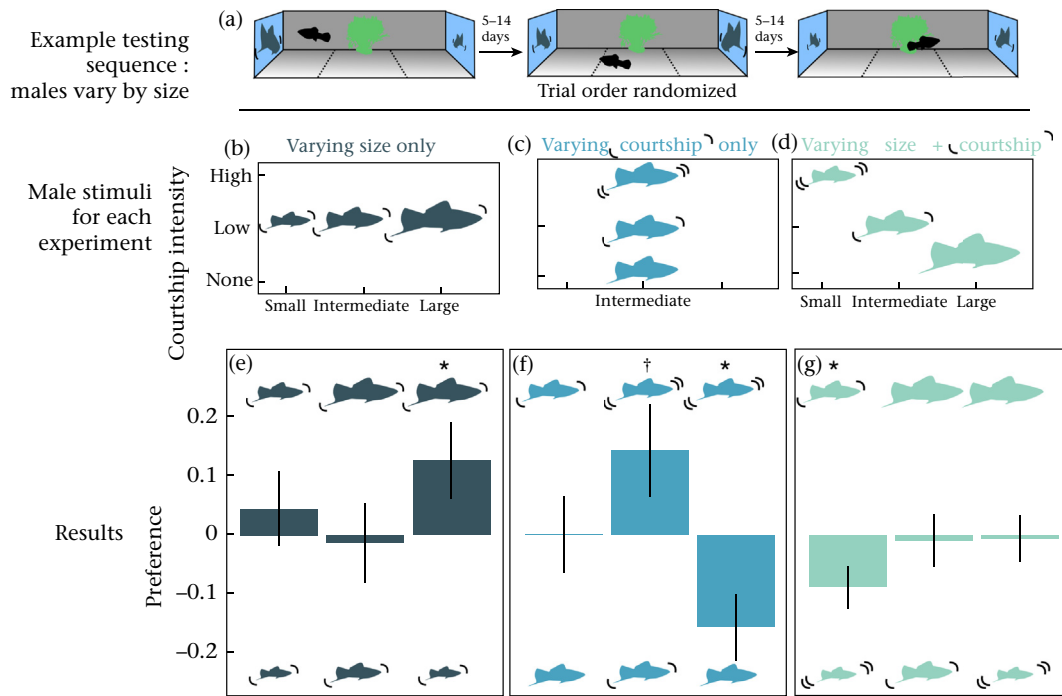


Figure 4. (a) Example testing sequence of females when males varied only in size. The order of the trials and of the initial location of each male was randomized. Size and courtship intensity of male animations in the (b) size-only, (c) courtship-only and (d) size and courtship experiments. Brackets denote level of courtship: none; (low); ((high)). Results for the (e) size-only, (f) courtship-only and (g) size and courtship experiments. The Y axis is the difference in the proportion of time spent with the male shown at the top of each bar plot and the bottom of each bar plot. Each bar tends towards the more preferred male in each trial. † $P < 0.10$; * $P < 0.05$. Values are means \pm SE.

male when paired with an intermediate male, nor for an intermediate male when paired with the small male.

We next varied courtship vigour while keeping the male animation at the intermediate size (Fig. 4c). Females showed no preference between a male that lacked courtship and one that courted at a low rate (Fig. 4f). However, patterns of female preference for the vigorously courting male were highly unexpected and context dependent. When paired with a male having low courtship intensity, there was a borderline statistically significant preference for the male with high courtship intensity ($P = 0.0538$). When a noncourting male was paired with a male of high courtship intensity, there was a significant preference for the noncourting male ($P = 0.0036$).

To test how swordtail females perceptually bind the components of a male's visual signal, we varied both size and courtship intensity (Fig. 4d). Females only showed a significant preference for a small highly courting male when paired with an intermediate-sized male displaying an intermediate amount of courtship (Fig. 4g). A female's prior exposure to animated males did not explain a significant proportion of variation in mating preferences in this experiment (linear mixed effects model: $\chi^2_1 = 0.177$, $P = 0.674$). Order did not have a statistically significant effect on preferences in any of the experiments ($\chi^2_2 = 2.767$, $P = 0.251$).

Finally, we were interested in whether females responded more quickly to animations in which males varied in both size and courtship compared to animations where males varied only in one way. We compared the average latency to approach the male animations when the males varied in both size and courtship and when the males varied only in one way. Female latency to approach animated males varied by experiment ($\chi^2_2 = 13.086$, $P = 0.001$; Fig. 5a). The females in the size and courtship experiment showed a significantly reduced latency compared with females in the other two experiments (lsmeans comparison: $t_{82} = -3.567$, $P = 0.001$). Tukey post hoc tests showed a significant difference between the size + courtship experiments and each of the unicomponent experiments (difference with the size-only experiment: $P = 0.003$;

difference with courtship-only experiment: $P = 0.024$). When we excluded females from the size and courtship experiment that had previously experienced the experimental set-up, there was no overall effect of experiment on latency (linear mixed effects model: $\chi^2_2 = 4.374$, $P = 0.1122$), although this difference was due to reduced power, not to a change in the average latency of females in the size and courtship experiment: the average latency in this experiment declined by less than 1 s when we removed these experienced females. In this reduced data set, the difference in average latency between the size + courtship experiment and the other two experiments was nearly statistically significant (lsmeans comparison: $t_{45} = -1.97$, $P = 0.055$).

DISCUSSION

Multicomponent signals are ubiquitous in mating interactions, evoke myriad responses from receivers and are thought to benefit signallers. Here we explored how social context dependence (the presence of certain types of males) affects mating preferences for two components of a swordtail's multicomponent visual signal and possible benefits of the males producing them. We found that female preferences for both size and courtship vigour depended on the males in a female's 'choice set'. When varying male size, preference was only found when the size difference between males was substantial, not when it was incremental. For courtship vigour, we found that the choice set of males can reverse female preferences for a vigorously courting male. When we varied both the size and courtship intensity of males in opposite directions, we found no evidence for preference functions resulting from additive processes across the signal components. However, females responded fastest to males in experiments where males varied in multiple dimensions (size and courtship) compared with experiments where males only varied in a single dimension, suggesting that a possible mechanism driving the evolution of multicomponent signalling might be ease of receiver processing.

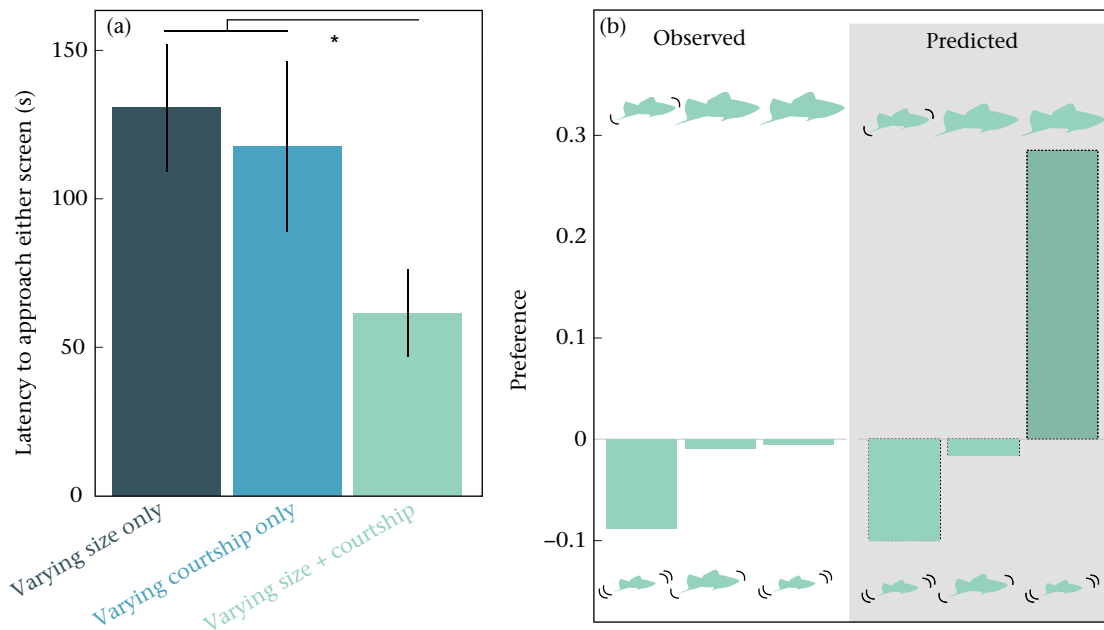


Figure 5. (a) Average latency (\pm SE) to approach either screen in each experiment for all females tested. The asterisk represents a significant difference between the pooled average of the two unicomponent experiments and the size + courtship experiment. (b) Observed responses of females in the size + courtship experiment and their predicted response if females additively bind components of a male's multicomponent signal. Their predicted responses were generated from adding relative preferences for different types of males from Fig. 4e and f. Observed responses are taken from Fig. 4g. See text for additional details.

Female preference for animations of large males was highly repeatable, much more so than estimates of mating preference repeatability using live males in this species (Fig. 3; Cummings & Mollaghan, 2006). Animations have previously been shown to generate more repeatable mating preferences than assays using live males. Morris, Nicoletto, and Hesselman (2003), for example, found in a series of choice tests using live and animated *Xiphophorus cortezi* male stimuli that preferences were highly repeatable when using animations but often unrepeatable when using live males. Because live males will differ in their behaviour, these unrepeatable female preferences for live males might not be capricious so much as reflect differing male behaviour in different trials. Animations also eliminate the possibility of male–female interaction, which in some cases may contribute additional noise to estimates of preference.

Animations might prove a valuable tool for understanding female mating preferences for specific features of male signals in a more repeatable, consistent way. For instance, our validation experiments allowed us to determine whether previously observed preferences for large males are due to actual preference for the large male or aversion to the small male. If female choice behaviour in this species is driven by aversion to the small male, we would have expected to see heightened preference for the large male in the large versus small male experiment (Fig. 2b) compared with the large male versus background experiment (Fig. 2a). Instead, we found comparable levels of preference for the large male in the two experiments, making it clear that females show preference towards large males even in the absence of small males.

We determined female mating preferences for size alone, courtship alone and when both size and courtship intensity varied. When we varied male size, keeping ornamentation (same large male image) and courtship (low rate) constant, females only expressed a preference for the large male when paired with the small male. This result largely parallels previous studies with live males (Ryan, Hews, & Wagner, 1990) and suggests that size is an important mediator of mating preferences in this fish as in other swordtails (Morris, Wagner, & Ryan, 1996; Rosenthal et al., 2002; Ryan & Wagner, 1987). To isolate a male's size from other features, we created some male stimuli that would appear unnatural to a female (e.g. a small male with the body shape and coloration of a large male). While unnatural, these stimuli allowed us to independently assess how different aspects of a male's multicomponent signal affect a female's response. This is especially important in species like *X. nigrensis* where a male's size covaries with his behaviour and ornamentation. Importantly, the use of these unnatural stimuli allowed us to disentangle a male's size from other features that covary with size to show that size alone matters in female mate choice decisions (Rosenthal & Evans, 1998). The sizes of males were within the ranges of the small, intermediate and large alternative male morphs in this species. There was no preference for the large male when paired with the intermediate male, suggesting that the 'choice set' of males affects female preferences for the large male. It also suggests that the female's ability to discriminate males of different sizes may suffer from an inability to perceive relatively small differences. Differences in stimuli that vary along a perceptual gradient are easier to detect when there is a large relative difference between them than a small one (i.e. Weber's law; Akre, Farris, Lea, Page, & Ryan, 2011). In our experiments, the large male was 67% larger than the small male and 25% larger than the intermediate male, which was 33% larger than the small male.

The pattern of preferences was much more complex when courtship intensity was varied independently of size. Intriguingly, the vigorously courting male was either preferred or unpreferred depending on the male that he was presented with. It is surprising

that this type of preference reversal could occur based only on the type of male that the highly courting male was presented with. It also generates a testable prediction about how highly courting males should spatially arrange themselves when courting females: high courting males should be more successful at attracting females when they place themselves near low courting males, but they should avoid being near males that lack courtship altogether. Something similar occurs in guppies, where males choose to surround themselves with unattractive competitors, making themselves look better by comparison (Gasparini, Serena, & Pilastro, 2013; but see Auge, Auld, Sherratt, & Godin, 2016). Neighbouring males also influence a male's attractiveness in finches (Oh, Badyaev, Irwin, & McPeck, 2010) and fiddler crabs (Callander, Hayes, Jennions, & Backwell, 2013; Callander, Jennions, & Backwell, 2011).

Why might females reverse their preferences for an intensely courting male? Perhaps when two males are courting a female, she is forced to make some comparison between the two and prefers the male that shows more intense courtship. But when paired with a noncourting male, the intensely courting male may appear too aggressive and challenging to a female. Something similar occurs with bowerbirds. Male bowerbird courtship behaviour resembles aggressive behaviours (as it does in *X. nigrensis*), and while males that court more intensely are preferred by females, females can be frightened by males that court too intensely (Borgia & Presgraves, 1998; Patricelli, Uy, Walsh, & Borgia, 2002). Males adjust their courtship intensity according to a female's perceived comfort (Patricelli et al., 2002), something that might occur with our swordtails but would be impossible to test using noninteractive animations. Additionally, female bowerbirds allow more intense displays by males that have attractive decorations at their bower (Patricelli, Uy, & Borgia, 2003). Perhaps in swordtails, females are more likely to tolerate intense courtship when it comes from a large, ornamented (sworded) male than from a smaller male that is less likely to be ornamented.

When we presented females with males that varied in both size and courtship, females only showed a preference for the small, high courting male when it was paired with an intermediate-sized male displaying an intermediate amount of courtship. What does this experiment tell us about how female swordtails combine these two components of a male's visual signal? One common assumption is that a female simply adds her individual responses to each component of a male's multicomponent trait. Fig. 5b shows the pattern of preferences we would expect if females perceptually bind size and courtship additively (based on the size-only and courtship-only experiments) and compares these expectations to what we observed in the experiment where male size and courtship were both varied. For example, if females simply add their relative preference for each component independently, referred to as 'atomism' by Rowe (1999), we would predict a very high preference for the large noncourting male in the final comparison in Fig. 4g. This prediction was based on our findings that large males were preferred over small males (Fig. 4e) and that noncourting males were preferred over high courting males (Fig. 4f). Instead, we found essentially no preference for either male (Fig. 4g). This suggests that females are binding the components of a male's signal in more complex ways, giving more credence to emergent properties (Partan & Marler, 2005) or 'configural' theories that hold that the whole is not simply the sum of its parts (Rowe, 1999). Female crickets appear to do something similar: when the pulse rate and loudness of a male's call was varied so that each call was more attractive for a different component of his chirp, females failed to express preferences, and the correspondence between their actual and predicted preferences (assuming an atomistic model) was poor (Gabel & Hennig, 2016).

Although many studies have quantified how independent aspects of a male's complex signal contribute to a female's mating response, few have manipulated more than one component of the signal at the same time, and even fewer have done so in a way that would allow a test of whether the male choice sets affect a female's preference. There are two notable exceptions. [Kodric-Brown and Nicoletto \(2001\)](#) created animations of male guppies that varied in colour (presence or absence of an orange spot) and in courtship rate (high or low). A series of dichotomous choice tests among these different males revealed an interaction between the two male traits: females preferred orange males only when both males courted at a low rate. When both males courted the female more intensely, no preference for colour was observed. Similarly, females only expressed a preference for more intensely courting males when males had an orange spot and not when they lacked the spot. These results suggest that female perception of individual components of a complex signal is highly dependent on the other components of the signal. This indicates that female guppies do not have simple preferences for each component that they sum to reach some overall preference for a given male and suggests an 'intersignal interaction' ([Hebets & Papaj, 2005](#)). [Künzler and Bakker \(2001\)](#) performed a similar study in threespine sticklebacks, *Gasterosteus aculeatus*, varying the throat colour of the male (dull or bright) and the type of courtship the male performed (zigzag or straight-line courtship). Males with redder throats were always preferred when the courtship intensity of the two males in the dichotomous choice was held constant. There was no overall preference for a male's display type when throat colour was kept constant. These results again suggest a more complex perceptual binding of a male's visual signal than would be predicted by additive or configural theories of signal integration ([Rowe, 1999](#)).

The above studies are also unique in that they allow a comparison of how the choice set of males influences a female's choice of mate. Both studies show clear evidence that the males available for comparison influence a female's choice. Female guppies, for example, prefer orange-spotted, high courting males when paired with a male that shows a low courtship rate, but they fail to show a preference when the formerly attractive male is paired with a noncoloured, highly courting male. Similarly, a colourful male that displays at a low rate is preferred when paired with a low courtship noncoloured male but not when paired with a high courtship noncoloured male ([Kodric-Brown & Nicoletto, 2001](#)). In sticklebacks, red-throated zigzagging males were usually preferred, except when paired with a red-throated straight-line courting male, where females failed to show a preference ([Künzler & Bakker, 2001](#)).

If female swordtails exhibit complex responses to male multicomponent signals, why and how might these signals evolve? One possibility is that males might benefit from evolving complex signals if these signals decrease decision-making time by females. Multicomponent signals are predicted to elicit faster reaction times than unicomponent signals ([Rowe, 1999](#)), thus potentially decreasing decision-making time by females. There are two non-mutually exclusive mechanisms that have been proposed to account for faster decisions during multicomponent signal assessment relative to unicomponent ones. The first is that complex signals may be more salient than unicomponent signals, which allows for faster assessment ([Rowe, 1999](#)). The other is that multicomponent signals may carry 'multiple messages', resulting in higher information content per unit time than with unicomponent signals, and allow females to reach decisions sooner ([Partan & Marler, 2005](#)). Our experimental results provide limited support for this intriguing hypothesis. Although all the animations we presented to females were multicomponent—males both courted

and had a size—we found limited evidence that females showed a shorter latency to approach males when the males varied in two ways than when the males varied in a single way ([Fig. 5a](#)). Thus, when potential mates vary in more ways, females may be more responsive to them, potentially driving the evolution of these multicomponent signals. Our results are similar to those with bumblebees, where bees trained to discriminate between flowers that differed in both colour and odour made better, faster foraging decisions than bees trained on flowers that differed only in colour or only in scent ([Kulahci, Dornhaus, & Papaj, 2008](#)). Jumping spiders show a quicker latency to approach potential prey when given visual and seismic cues as opposed to either cue in isolation ([Roberts, Taylor, & Uetz, 2007](#)).

In contrast with most studies of complex (multicomponent or multimodal) signalling that test behavioural responses in the presence or absence of signal components, we varied our two signal components continuously. Why is this important? By only testing two levels of a given component, the shape of the female preference function (linear, disruptive, stabilizing) cannot be known. Additionally, classic frameworks for classifying complex signals ([Partan & Marler, 2005](#)), while valuable, are not general enough to include complex signals in which the components vary continuously ([Smith & Evans, 2013](#)). Our study, as well as others that have continuously varied male traits ([Gabel & Hennig, 2016](#); [Gray et al., 2016](#); [Stange et al., in press](#)), reveal more complex communication dynamics.

Context dependence of the type considered here is closely related to the idea of rationality: that options can be absolutely ranked on some one-dimensional scale. Rationality is implicitly assumed in nearly all models of behaviour but is often violated in humans and other animals ([Huber et al., 1982](#); [Lea & Ryan, 2015](#); [Locatello et al., 2015](#)). Our findings show that the 'value' of a male to a female depends on the other males with which a male is presented: thus, a male's value changes as his competitors change. It is therefore impossible to construct the scale needed to fulfil the basic tenants of rational decision making. Our results are thus incompatible with the idea that swordtails are strictly rational in their mating preferences.

Our results also shed some light on the relative importance of courtship vigour and size to overall female mating preferences in *X. nigrensis*. Male size was clearly important to females; however, male courtship was also important and produced plastic responses. Courtship, a more dynamic component of a male's visual signal than size, appeared to elicit stronger bias in female preference than did size and was also more prone to context dependence. For *X. nigrensis* females, it appears that Darwin and Wallace were both right.

Acknowledgments

We thank Kelly Wallace and Sarah Price for feedback on an early draft. We thank Brackenridge Field Laboratories for use of their facilities. Critiques from two anonymous referees and the Editor substantially improved the manuscript. This research was funded by a student research award through the Animal Behavior Society, the DDIG-like award from the Ecology, Evolution, and Behavior programme at the University of Texas at Austin, and a National Science Foundation Graduate Research Fellowship (DGE-1110007) to L.R.

Supplementary Material

Supplementary data related to this article can be found at <http://dx.doi.org/10.1016/j.anbehav.2017.05.017>.

References

- Akre, K. L., Farris, H. E., Lea, A. M., Page, R. A., & Ryan, M. J. (2011). Signal perception in frogs and bats and the evolution of mating signals. *Science*, 333(6043), 751–752. <http://dx.doi.org/10.1126/science.1205623>.
- Auge, A.-C., Auld, H. L., Sherratt, T. N., & Godin, J.-G. J. (2016). Do males form social associations based on sexual attractiveness in a fission–fusion fish society? *PLoS One*, 11(3), e0151243. <http://dx.doi.org/10.1371/journal.pone.0151243>.
- Auld, H. L., & Godin, J.-G. J. (2015). Sexual voyeurs and copiers: Social copying and the audience effect on male mate choice in the guppy. *Behavioral Ecology and Sociobiology*, 69(11), 1795–1807. <http://dx.doi.org/10.1007/s00265-015-1992-z>.
- Bateson, M., & Healy, S. D. (2005). Comparative evaluation and its implications for mate choice. *Trends in Ecology & Evolution*, 20(12), 659–664. <http://dx.doi.org/10.1016/j.tree.2005.08.013>.
- Bell, A. M., Hankison, S. J., & Laskowski, K. L. (2009). The repeatability of behaviour: A meta-analysis. *Animal Behaviour*, 77, 771–783. <http://dx.doi.org/10.1016/j.anbehav.2008.12.022>.
- Bentsen, C. L., Hunt, J., Jennions, M. D., & Brooks, R. (2006). Complex multivariate sexual selection on male acoustic signaling in a wild population of *Teleogryllus commodus*. *American Naturalist*, 167(4), E102–E116. <http://dx.doi.org/10.1086/501376>.
- Blows, M. W., Brooks, R., Kraft, P. G., & Phillips, P. (2003). Exploring complex fitness surfaces: Multiple ornamentation and polymorphism in male guppies. *Evolution*, 57(7), 1622–1630. <http://dx.doi.org/10.1554/02-507>.
- Borgia, G., & Presgraves, D. C. (1998). Coevolution of elaborated male display traits in the spotted bowerbird: An experimental test of the threat reduction hypothesis. *Animal Behaviour*, 56, 1121–1128. <http://dx.doi.org/10.1006/anbe.1998.0908>.
- Bradski, G. (2000). The opencv library. *Doctor Dobbs Journal*, 25(11), 120–126.
- Brooks, R., & Endler, J. A. (2001). Female guppies agree to differ: Phenotypic and genetic variation in mate-choice behavior and the consequences for sexual selection. *Evolution*, 55(8), 1644–1655. [http://dx.doi.org/10.1554/0014-3820\(2001\)055\[1644:FGATDP\]2.0.CO;2](http://dx.doi.org/10.1554/0014-3820(2001)055[1644:FGATDP]2.0.CO;2).
- Byers, J., Hebets, E., & Podos, J. (2010). Female mate choice based upon male motor performance. *Animal Behaviour*, 79, 771–778. <http://dx.doi.org/10.1016/j.anbehav.2010.01.009>.
- Callander, S., Hayes, C. L., Jennions, M. D., & Backwell, P. R. Y. (2013). Experimental evidence that immediate neighbors affect male attractiveness. *Behavioral Ecology*, 24(3), 730–733. <http://dx.doi.org/10.1093/beheco/ars208>.
- Callander, S., Jennions, M. D., & Backwell, P. R. Y. (2011). Female choice over short and long distances: Neighbour effects. *Behavioral Ecology and Sociobiology*, 65(11), 2071–2078. <http://dx.doi.org/10.1007/s00265-011-1216-0>.
- Candolin, U. (2003). The use of multiple cues in mate choice. *Biological Reviews*, 78(4), 575–595. <http://dx.doi.org/10.1017/S1464793103006158>.
- Cole, G. L., & Endler, J. A. (2015). Variable environmental effects on a multicomponent sexually selected trait. *American Naturalist*, 185(4), 452–468. <http://dx.doi.org/10.1086/680022>.
- Cornuau, J. H., Rat, M., Schmeller, D. S., & Loyau, A. (2012). Multiple signals in the palmate newt: Ornaments help when courting. *Behavioral Ecology and Sociobiology*, 66(7), 1045–1055. <http://dx.doi.org/10.1007/s00265-012-1355-y>.
- Cummings, M., & Mollaghan, D. (2006). Repeatability and consistency of female preference behaviours in a northern swordtail, *Xiphophorus nigricans*. *Animal Behaviour*, 72, 217–224. <http://dx.doi.org/10.1016/j.anbehav.2006.01.009>.
- Darwin, C. (1859). *On the origin of species*. London, U.K.: J. Murray.
- Forsgren, E. (1992). Predation risk affects mate choice in a gobioid fish. *American Naturalist*, 140(6), 1041–1049.
- Fowler-Finn, K. D., & Rodríguez, R. L. (2012). The evolution of experience-mediated plasticity in mate preferences. *Journal of Evolutionary Biology*, 25(9), 1855–1863. <http://dx.doi.org/10.1111/j.1420-9101.2012.02573.x>.
- Fox, J., & Weisberg, S. (2011). *An R companion to applied regression* (2nd ed.). Thousand Oaks, CA: Sage. Retrieved from: <http://socserv.socsci.mcmaster.ca/jfox/Books/Companion>.
- Fuller, R. C., & Noa, L. A. (2010). Female mating preferences, lighting environment, and a test of the sensory bias hypothesis in the bluefin killifish. *Animal Behaviour*, 80, 23–35. <http://dx.doi.org/10.1016/j.anbehav.2010.03.017>.
- Gabel, E., & Hennig, R. M. (2016). Evidence for comparative decision making in female crickets. *Behavioral Ecology*, 27(4), 1216–1222. <http://dx.doi.org/10.1093/beheco/arw030>.
- Gasparini, C., Serena, G., & Pilaastro, A. (2013). Do unattractive friends make you look better? Context-dependent male mating preferences in the guppy. *Proceedings of the Royal Society B: Biological Sciences*, 280(1756), 20123072. <http://dx.doi.org/10.1098/rspb.2012.3072>.
- Girard, M. B., Elias, D. O., & Kasumovic, M. M. (2015). Female preference for multimodal courtship: Multiple signals are important for male mating success in peacock spiders. *Proceedings of the Royal Society B: Biological Sciences*, 282(1820), 20152222. <http://dx.doi.org/10.1098/rspb.2015.2222>.
- Godin, J.-G. J., & Briggs, S. E. (1996). Female mate choice under predation risk in the guppy. *Animal Behaviour*, 51, 117–130. <http://dx.doi.org/10.1006/anbe.1996.0010>.
- Gray, D. A., Gabel, E., Blankers, T., & Hennig, R. M. (2016). Multivariate female preference tests reveal latent perceptual biases. *Proceedings of the Royal Society B: Biological Sciences*, 283(1842), 20161972. <http://dx.doi.org/10.1098/rspb.2016.1972>.
- Hebets, E. A. (2003). Subadult experience influences adult mate choice in an arthropod: Exposed female wolf spiders prefer males of a familiar phenotype. *Proceedings of the National Academy of Sciences of the United States of America*, 100(23), 13390–13395. <http://dx.doi.org/10.1073/pnas.2333262100>.
- Hebets, E. A. (2005). Attention-altering signal interactions in the multimodal courtship display of the wolf spider *Schizocosa uetzi*. *Behavioral Ecology*, 16(1), 75–82. <http://dx.doi.org/10.1093/beheco/arl133>.
- Hebets, E. A., & Papaj, D. R. (2005). Complex signal function: Developing a framework of testable hypotheses. *Behavioral Ecology and Sociobiology*, 57(3), 197–214. <http://dx.doi.org/10.1007/s00265-004-0865-7>.
- Hebets, E. A., Vink, C. J., Sullivan-Beckers, L., & Rosenthal, M. F. (2013). The dominance of seismic signaling and selection for signal complexity in *Schizocosa* multimodal courtship displays. *Behavioral Ecology and Sociobiology*, 67(9), 1483–1498. <http://dx.doi.org/10.1007/s00265-013-1519-4>.
- Hothorn, T., Bretz, F., & Westfall, P. (2008). Simultaneous inference in general parametric models. *Biometrical Journal*, 50(3), 346–363.
- Huber, J., Payne, J. W., & Puto, C. (1982). Adding Asymmetrically dominated alternatives: Violations of regularity and the similarity hypothesis. *Journal of Consumer Research*, 9(1), 90–98.
- Jennions, M. D., & Petrie, M. (1997). Variation in mate choice and mating preference: A review of causes and consequences. *Biological Reviews*, 72(2), 283–327. <http://dx.doi.org/10.1111/j.1469-185X.1997.tb00015.x>.
- Johnstone, R. A. (1996). Multiple displays in animal communication: 'Backup signals' and 'multiple messages'. *Philosophical Transactions of the Royal Society B: Biological Sciences*, 351(1337), 329–338. <http://dx.doi.org/10.1098/rstb.1996.0026>.
- Kodric-Brown, A., & Nicoletto, P. F. (2001). Female choice in the guppy (*Poecilia reticulata*): The interaction between male color and display. *Behavioral Ecology and Sociobiology*, 50(4), 346–351. <http://dx.doi.org/10.1007/s002650100374>.
- Kulahci, I. G., Dornhaus, A., & Papaj, D. R. (2008). Multimodal signals enhance decision making in foraging bumble-bees. *Proceedings of the Royal Society B: Biological Sciences*, 275(1636), 797–802. <http://dx.doi.org/10.1098/rspb.2007.1176>.
- Künzler, R., & Bakker, T. C. M. (1998). Computer animations as a tool in the study of mating preferences. *Behaviour*, 135(8/9), 1137–1159.
- Künzler, R., & Bakker, T. C. M. (2001). Female preferences for single and combined traits in computer animated stickleback males. *Behavioral Ecology*, 12(6), 681–685. <http://dx.doi.org/10.1093/beheco/12.6.681>.
- Lea, A. M., & Ryan, M. J. (2015). Irrationality in mate choice revealed by túngara frogs. *Science*, 349(6251), 964–966. <http://dx.doi.org/10.1126/science.aab2012>.
- Lenth, R. V. (2016). Least-squares means: The R package lsmeans. *Journal of Statistical Software*, 69(1), 1–33. <http://dx.doi.org/10.18637/jss.v069.i01>.
- Locatello, L., Poli, F., & Rasotto, M. B. (2015). Context-dependent evaluation of prospective mates in a fish. *Behavioral Ecology and Sociobiology*, 69(7), 1119–1126. <http://dx.doi.org/10.1007/s00265-015-1924-y>.
- Maan, M. E., Seehausen, O., & Van Alphen, J. J. M. (2010). Female mating preferences and male coloration covary with water transparency in a Lake Victoria cichlid fish. *Biological Journal of the Linnean Society*, 99(2), 398–406. <http://dx.doi.org/10.1111/j.1095-8312.2009.01368.x>.
- McClintock, W. J., & Uetz, G. W. (1996). Female choice and pre-existing bias: Visual cues during courtship in two *Schizocosa* wolf spiders (Araneae: Lycosidae). *Animal Behaviour*, 52, 167–181. <http://dx.doi.org/10.1006/anbe.1996.0162>.
- Møller, A. P., & Pomiankowski, A. (1993). Why have birds got multiple sexual ornaments? *Behavioral Ecology and Sociobiology*, 32(3). <http://dx.doi.org/10.1007/BF00173774>.
- Morris, M. R., Nicoletto, P. F., & Hesselman, E. (2003). A polymorphism in female preference for a polymorphic male trait in the swordtail fish *Xiphophorus cortezi*. *Animal Behaviour*, 65, 45–52. <http://dx.doi.org/10.1006/anbe.2002.2042>.
- Morris, M. R., Wagner, W. E., & Ryan, M. J. (1996). A negative correlation between trait and mate preference in *Xiphophorus pygmaeus*. *Animal Behaviour*, 52, 1193–1203. <http://dx.doi.org/10.1006/anbe.1996.0267>.
- Oh, K. P., Badyaev, A. V., Irwin, A. E. D., & McPeck, E. M. A. (2010). Structure of social networks in a passerine bird: Consequences for sexual selection and the evolution of mating strategies. *American Naturalist*, 176(3), E80–E89. <http://dx.doi.org/10.1086/655216>.
- Partan, S., & Marler, P. (1999). Communication goes multimodal. *Science*, 283(5406), 1272–1273. <http://dx.doi.org/10.1126/science.283.5406.1272>.
- Partan, S. R., & Marler, P. (2005). Issues in the classification of multimodal communication signals. *American Naturalist*, 166(2), 231–245. <http://dx.doi.org/10.1016/j.cobeha.2016.09.011>.
- Patricelli, G. L., Uy, J. A. C., & Borgia, G. (2003). Multiple male traits interact: Attractive bower decorations facilitate attractive behavioural displays in satin bowerbirds. *Proceedings of the Royal Society B: Biological Sciences*, 270(1531), 2389–2395. <http://dx.doi.org/10.1098/rspb.2003.2530>.
- Patricelli, G. L., Uy, J. A. C., Walsh, G., & Borgia, G. (2002). Sexual selection: Male displays adjusted to female's response. *Nature*, 415(6869), 279–280. <http://dx.doi.org/10.1038/415279a>.
- Peirce, J. W., & Peirce, J. W. (2009). Generating stimuli for neuroscience using PsychoPy. *Frontiers in Neuroinformatics*, 2(10). <http://dx.doi.org/10.3389/fninf.2010.2008>.
- Pinheiro, J., Bates, D., DebRoy, S., Sarkar, D., & Core Team, R. (2017). *nlme: Linear and nonlinear mixed effects models*. Retrieved from <https://CRAN.R-project.org/package=nlme>.
- Ramsey, M. E., Wong, R. Y., & Cummings, M. E. (2011). Estradiol, reproductive cycle and preference behavior in a northern swordtail. *General and Comparative Endocrinology*, 170(2), 381–390. <http://dx.doi.org/10.1016/j.ygcen.2010.10.012>.

- Reding, L., & Cummings, M. E. (2016). Does sensory expansion benefit asexual species? An olfactory discrimination test in Amazon mollies. *Behavioral Ecology*, 27(2), 411–418. <http://dx.doi.org/10.1093/beheco/arv168>.
- Reichert, M. S., Finck, J., & Ronacher, B. (2017). Exploring the hidden landscape of female preferences for complex signals. *Evolution*, 71(4), 1009–1024. <http://dx.doi.org/10.1111/evo.13202>.
- Roberts, J. A., Taylor, P. W., & Uetz, G. W. (2007). Consequences of complex signaling: Predator detection of multimodal cues. *Behavioral Ecology*, 18(1), 236–240. <http://dx.doi.org/10.1093/beheco/arl079>.
- Rodríguez, R. L., Hallett, A. C., Kilmer, J. T., & Fowler-Finn, K. D. (2013). Curves as traits: Genetic and environmental variation in mate preference functions. *Journal of Evolutionary Biology*, 26(2), 434–442. <http://dx.doi.org/10.1111/jeb.12061>.
- Rodríguez, R. L., Rebar, D., & Fowler-Finn, K. D. (2013). The evolution and evolutionary consequences of social plasticity in mate preferences. *Animal Behaviour*, 85, 1041–1047. <http://dx.doi.org/10.1016/j.anbehav.2013.01.006>.
- Ronald, K. L., Zeng, R., White, D. J., Fernández-Juricic, E., & Lucas, J. R. (2017). What makes a multimodal signal attractive? A preference function approach. *Behavioral Ecology*, 28(3), 677–687. <http://dx.doi.org/10.1093/beheco/arn015>.
- Rosenthal, G. G., & Evans, C. S. (1998). Female preference for swords in *Xiphophorus helleri* reflects a bias for large apparent size. *Proceedings of the National Academy of Sciences of the United States of America*, 95(8), 4431–4436.
- Rosenthal, G. G., Wagner, W. E., Jr., & Ryan, M. J. (2002). Secondary reduction of preference for the sword ornament in the pygmy swordtail *Xiphophorus nigrensis* (Pisces: Poeciliidae). *Animal Behaviour*, 63, 37–45. <http://dx.doi.org/10.1006/anbe.2001.1887>.
- Rowe, C. (1999). Receiver psychology and the evolution of multicomponent signals. *Animal Behaviour*, 58, 921–931. <http://dx.doi.org/10.1006/anbe.1999.1242>.
- Royle, N. J., Lindström, J., & Metcalfe, N. B. (2008). Context-dependent mate choice in relation to social composition in green swordtails *Xiphophorus helleri*. *Behavioral Ecology*, 19(5), 998–1005. <http://dx.doi.org/10.1093/beheco/arn059>.
- Ruxton, G. D., & Neuhäuser, M. (2013). Improving the reporting of *P*-values generated by randomization methods. *Methods in Ecology and Evolution*, 4(11), 1033–1036. <http://dx.doi.org/10.1111/2041-210X.12102>.
- Ryan, M. J., & Causey, B. A. (1989). 'Alternative' mating behavior in the swordtails *Xiphophorus nigrensis* and *Xiphophorus pygmaeus* (Pisces: Poeciliidae). *Behavioral Ecology and Sociobiology*, 24(6), 341–348. <http://dx.doi.org/10.1007/BF00293262>.
- Ryan, M. J., Hews, D. K., & Wagner, W. W. (1990). Sexual selection on alleles that determine body size in the swordtail *Xiphophorus nigrensis*. *Behavioral Ecology and Sociobiology*, 26(4), 231–237. <http://dx.doi.org/10.1007/BF00178316>.
- Ryan, M. J., Pease, C. M., & Morris, M. R. (1992). A genetic polymorphism in the swordtail *Xiphophorus nigrensis*: Testing the prediction of equal fitnesses. *American Naturalist*, 139(1), 21–31.
- Ryan, M. J., & Wagner, W. E. (1987). Asymmetries in mating preferences between species: Female swordtails prefer heterospecific males. *Science*, 236(4801), 595–597. <http://dx.doi.org/10.1126/science.236.4801.595>.
- Schlupp, I., Marler, C., & Ryan, M. J. (1994). Benefit to male sailfin mollies of mating with heterospecific females. *Science*, 263(5145), 373–374. <http://dx.doi.org/10.1126/science.8278809>.
- Smith, C. L., & Evans, C. S. (2013). A new heuristic for capturing the complexity of multimodal signals. *Behavioral Ecology and Sociobiology*, 67(9), 1389–1398. <http://dx.doi.org/10.1007/s00265-013-1490-0>.
- Stange, N., Page, R. A., Ryan, M. J., & Taylor, R. C. (2017). Interactions between complex multisensory signal components result in unexpected mate choice responses. *Animal Behaviour*. <http://dx.doi.org/10.1016/j.anbehav.2016.07.005> (in press).
- Tange, O. (2011). GNU Parallel: The command-line power tool. *login: The USENIX Magazine*, 36(1), 42–47. <http://dx.doi.org/10.5281/zenodo.16303>.
- Tudor, M. S., & Morris, M. R. (2011). Frequencies of alternative mating strategies influence female mate preference in the swordtail *Xiphophorus multilineatus*. *Animal Behaviour*, 82, 1313–1318. <http://dx.doi.org/10.1016/j.anbehav.2011.09.014>.
- Verzijden, M. N., & Rosenthal, G. G. (2011). Effects of sensory modality on learned mate preferences in female swordtails. *Animal Behaviour*, 82, 557–562. <http://dx.doi.org/10.1016/j.anbehav.2011.06.010>.
- Wagner, W. E. (1998). Measuring female mating preferences. *Animal Behaviour*, 55, 1029–1042. <http://dx.doi.org/10.1006/anbe.1997.0635>.
- Wagner, W. E., Smeds, M. R., & Wiegmann, D. D. (2001). Experience affects female responses to male song in the variable field cricket *Gryllus lineaticeps* (Orthoptera, Gryllidae). *Ethology*, 107(9), 769–776. <http://dx.doi.org/10.1046/j.1439-0310.2001.00700.x>.
- Wallace, A. R. (1889). *Darwinism: An exposition of the theory of natural selection with some of its applications*. London, U.K.: Macmillan.
- Wickham, H. (2016). *tidyverse: Easily install and load 'tidyverse' packages*. Retrieved from: <https://CRAN.R-project.org/package=tidyverse>.
- Witte, K., & Ryan, M. J. (2002). Mate choice copying in the sailfin molly, *Poecilia latipinna*, in the wild. *Animal Behaviour*, 63, 943–949. <http://dx.doi.org/10.1006/anbe.2001.1982>.
- Wong, R. Y., So, P., & Cummings, M. E. (2011). How female size and male displays influence mate preference in a swordtail. *Animal Behaviour*, 82, 691–697. <http://dx.doi.org/10.1016/j.anbehav.2011.06.024>.
- Zimmerer, E. J., & Kallman, K. D. (1989). Genetic basis for alternative reproductive tactics in the pygmy swordtail, *Xiphophorus nigrensis*. *Evolution*, 43(6), 1298–1307. <http://dx.doi.org/10.2307/2409364>.