EXPERIENCE-MEDIATED PLASTICITY IN MATE PREFERENCES: MATING ASSURANCE IN A VARIABLE ENVIRONMENT

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An individual's prior experience of sexual signals can result in variation in mate preferences, with important consequences for the course of sexual selection. We test two hypotheses about the evolution of experience-mediated plasticity in mate preferences: mating assurance and mismating avoidance. We exposed female *Enchenopa binotata* treehoppers (Hemiptera: Membracidae) to treatments that varied their experience of signal frequency, the most divergent sexual signal trait in the *E. binotata* species complex. Treatments consisted of (1) signals matching the preferred frequency, (2–3) signals deviating either 100 Hz above or 100 Hz below the preferred frequency, and (4) no signals. Females experiencing preferred signals showed the greatest selectivity. However, experience had no effect on peak preference. These results support the hypothesis that selection has favored plasticity in mate preferences that ensures that mating takes place when preferred mates are rare or absent, while ensuring choice of preferred types when those are present. We consider how experience-mediated plasticity may influence selection on sexual advertisement signals, patterns of reproductive isolation, and the maintenance of genetic variation. We suggest that the plasticity we describe may increase the likelihood of successful colonization of a novel environment, where preferred mating types may be rare.

KEY WORDS: *Enchenopa*, form of selection, maintenance of variation, mate choice, strength of preference.

Adaptive plasticity evolves under selection favoring the expression of different phenotypes in different environments (Gomulkiewicz and Kirkpatrick 1992; Via et al. 1995; Schlichting and Pigliucci 1998; West-Eberhard 2003; Ghalambor et al. 2007). Existing patterns of adaptive plasticity should therefore reflect the environmental variables that have been important in shaping its expression (Jennions and Petrie 1997; Qvarnström 2001; Beckers and Schul 2010). Here, we apply this rationale to the evolution of plasticity in mate preferences—that is, we expect that observed patterns of plasticity in mate preferences will help identify the environmental variation that has influenced the cost-benefit balance of variable mate preferences across environments. Mate preferences have traditionally been viewed as largely invariant, and this typological perspective has influenced most of the theory re-

garding sexual selection (reviewed in Andersson 1994; Mead and Arnold 2004). However, recent work shows that mate preferences are highly variable both among and within individuals, and this variation may represent adaptive plasticity (Jennions and Petrie 1997; Qvarnström 2001; Chaine and Lyon 2008). Evolutionary biologists are thus challenged to include plastic variation in mate preferences into models of sexual selection and diversification (Qvarnström 2001; Verzijden et al. 2005; Dukas 2006; Verzijden and ten Cate 2007; Chaine and Lyon 2008; Servedio et al. 2009).

We focus on variation in the social environment—specifically variation in the types of potential mates available—as a source of selection favoring plasticity in mate preferences. The social environment is highly variable, and a prominent cause of variation in fitness (West-Eberhard 1983; Kingsolver et al. 2001; Kokko et al.

2006; Wolf et al. 2007). Across social contexts, the costs and benefits of mate choice will vary with the abundance of preferred and nonpreferred mates, the opportunity and cost of mating with nonpreferred mates, the level of sympatry with heterospecifics, and the degree of similarity with heterospecific signals (Jennions and Petrie 1997; McPeek and Gavrilets 2006). Because these factors are often dynamic, plasticity in mate preferences in response to variation in the social environment may be highly advantageous (Jennions and Petrie 1997; Qvarnström 2001; Dukas 2006). The sexual signals of other individuals in the social environment provide cues about the costs and benefits of mate choice, and animals in many taxa adjust mate preferences in response to their experience of sexual signals (Hebets and Sullivan-Beckers 2010). This experience-mediated plasticity can take a variety of forms: it can result in acquired or strengthened preferences (Dewinter and Rollenhagen 1993; Collins 1995; Wagner et al. 2001; Hebets and Vink 2007; Kozak and Boughman 2009), weakened preferences (Bailey and Zuk 2008), or a change in the preferred mate type (Miller and Fincke 1999; Hebets 2003; Walling et al. 2008; Bailey and Zuk 2009).

We test two hypotheses about selection on experience-mediated plasticity in mate preferences: the "mating assurance" hypothesis and the "mismating avoidance" hypothesis. Both hypotheses assume that selection has favored mating with a preferred mate type over a nonpreferred type (Andersson 1994; Kokko et al. 2006), and that selection has favored mating with a nonpreferred mate type over not mating at all. This means that when individuals encounter a nonpreferred mate type, they must trade off the cost of foregoing a mating opportunity against the costs of accepting a nonpreferred mate (e.g., reduced direct benefits, reduced offspring fitness). How the composition of the social environment has varied in the past will determine the balance between these costs. Each hypothesis describes a different balance, and makes specific predictions about patterns of variation in two basic features of mate preferences in response to variation in the social environment. These two features are peak preference (Fig. 1a), which is the most preferred signal value, and selectivity (Fig. 1b and d), which is how responses to signals decrease as the signals deviate from the preferred value.

The "mating assurance" hypothesis posits that the balance between the costs and benefits of mate choice has favored ensuring that mating occurs. This means that the relative cost of foregoing a mating opportunity has been greater than the relative cost of mating with a nonpreferred mate. Consequently, selection would have favored plasticity in mate preferences that ensures that mating takes place when preferred mates are rare or absent, while ensuring choice of preferred types when those are present. This hypothesis predicts that individuals will show higher selectivity when they experience preferred signals, compared to lower selectivity when they experience nonpreferred signals or have no
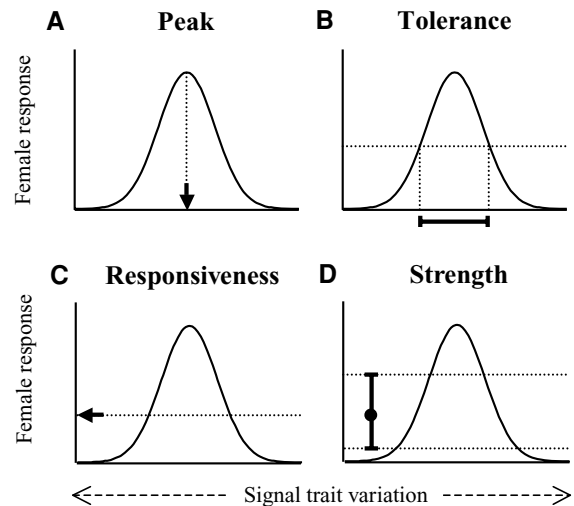

Figure 1. Description of traits used to describe variation in female preference functions. (A) Peak: the stimulus frequency eliciting the highest female response, indicated by the arrow on the *x*-axis. (B) Tolerance: width of the preference function at the 50% drop from the highest response, indicated on the *x*-axis with a bracketed bar. (C) Responsiveness: mean response across the range of stimuli, indicated by the bold arrow on the *y*-axis. (D) Strength: the square of the coefficient of variation in responses across the range of stimuli (Schluter 1988), represented by the length of bracketed bar.

experience with signals (Table 1). This hypothesis also predicts no effect of experience on peak preference (Table 1).

The "mismating avoidance" hypothesis posits that the balance between the costs and benefits of mate choice has favored the prevention of mismating with nonpreferred mates. Specifically, the relative cost of mating with a nonpreferred mate type has been greater than the relative cost of foregoing a given mating opportunity. Consequently, selection would have favored plasticity that minimizes the likelihood of mating with nonpreferred mate types when they are present. This hypothesis makes two independent predictions, either of which would decrease the likelihood of mating with a nonpreferred mate. The first prediction is that individuals will show higher selectivity when they experience nonpreferred signals, compared to lower selectivity when they experience preferred signals or have no experience with signals (Table 1). The second prediction is that individuals will shift their peak preference away from a nonpreferred signal that they have experienced (Table 1). Either of these predictions would support the mismating avoidance hypothesis.

We tested the predictions of "mating assurance" and "mismating avoidance" hypotheses using the *Enchenopa binotata* species complex of treehoppers (Hemiptera: Membracidae). We varied female experience of sexual signals by manipulating the frequency (Hz) of male advertisement signals of which a female had experience. The advertisement signals consist of a

Table 1. Hypotheses about selection shaping experience-mediated plasticity, and the predicted patterns of selectivity and peak preference according to variation in the experience of signals in the social environment.

Experience	Mating assurance hypothesis		Mismating avoidance hypothesis	
	Comparative selectivity	Change in peak preference	Comparative selectivity	Change in peak preference
Preferred signals	Higher	No difference	Lower	No difference
Nonpreferred signals	Lower	No difference	Higher	Shift away from nonpreferred
Silence	Lower	No difference	Lower	No difference

whine (a tone with a frequency sweep) followed by a series of pulses. Frequency is the signal trait that most differs among the members of the complex, and for which females have the strongest preferences (Rodríguez et al. 2004, 2006; Cocroft et al. 2010). We then evaluated the resulting patterns of plasticity in mate preferences. Female preferences for frequency are closed functions in the *E. binotata* complex and can therefore be characterized in terms of peak preference and selectivity. The peak preference corresponds to the population mean and falls away the further the signal deviates from this mean (Rodríguez et al. 2004, 2006; Sullivan-Beckers and Cocroft 2010).

The *E. binotata* complex provides a useful framework for studying the evolution of plasticity in mate preferences. Most members of the complex are widely and sympatrically distributed across eastern North America (Wood 1993; Cocroft et al. 2008, 2010). The life history of these insects provides a window during which females may experience variation in the signals of potential mates before mating—males begin to signal ca. two weeks after the adult molt, and females become receptive ca. two weeks afterwards (Rodríguez et al. 2004; Cocroft et al. 2008). As a result, variation in female experience with signals has the potential to influence variation in mate preferences. Females mate only once (Sullivan-Beckers and Cocroft 2010), and choose their mate on the basis of species-specific plant-borne vibrational advertisement signals (Rodríguez et al. 2004; Cocroft et al. 2008). The signal types experienced by a female prior to mating may vary as a result of several factors. Several species can occur at the same locality, with each specializing on a different host plant (Wood 1993; Cocroft et al. 2008, 2010). Dispersal among plants is not uncommon in adults—young adult males fly in search of females and can land and signal on neighboring plants, sometimes including nonhost plants (Sattman and Cocroft 2003; Cocroft et al. 2008). Also, development is regulated by host plant phenology (Wood and Guttman 1983; Cocroft et al. 2008) and broods on different plants may develop at different rates. Some aggregations may thus be visited by older conspecific and/or heterospecific males from neighboring plants.

The *E. binotata* complex also provides a framework for understanding how experience-mediated plasticity in mate prefer-

ences may influence the course of sexual selection and speciation. This complex offers a case study of evolutionary divergence initiated by the colonization of novel environments and accompanied by sexual signal divergence (Wood 1993; Cocroft et al. 2008). Reproductive isolation arises from the differential use of host plants and assortative mating due to sexual communication (Wood 1993; Cocroft et al. 2008). Changes in female preferences are the main cause of signal evolution in the complex (Rodríguez et al. 2006), and there is evidence of current strong sexual selection on signals (Sullivan-Beckers and Cocroft 2010). Because of the importance of mate choice in the *E. binotata* complex, understanding the causes of variation in plasticity of mate preferences is important for understanding the role of sexual selection in diversification.

Materials and Methods
GENERAL METHODS
We used two members of the *E. binotata* complex, allowing for two separate tests of the hypotheses. One species is from Missouri and lives on *Ptelea trifoliata* (Rutaceae) host plants, and one species is from Wisconsin and lives on *Viburnum lentago* (Caprifoliaceae) host plants. Members of the *E. binotata* complex are good biological species, due to ecological and behavioral causes of reproductive isolation (Wood and Guttman 1982; Lin and Wood 2002; Rodríguez et al. 2004; Cocroft et al. 2008; McNett and Cocroft 2008). However, only some members of the complex have been described (Hamilton and Cocroft 2009). We therefore refer to our study species using their host plant names: *E. binotata* "Ptelea" and *E. binotata* "Viburnum."

Enchenopa binotata "Ptelea" were collected as second- to third-instar nymphs in the field (Boone County, Missouri) in late May 2009, from several *P. trifoliata* plants over a span of several meters to ensure genetic diversity in the nymphs collected. Even on a single stem, aggregations are the result of several females laying eggs in the fall (Wood and Guttman 1983). *Enchenopa binotata* "Viburnum" insects were the offspring of adults we collected in the field (Milwaukee County, Wisconsin) in the fall of 2008. These adults were collected from several trees over a span

of several meters. They were placed on a potted host plant where they laid eggs that hatched the following spring. We reared the nymphs from both species on potted plants in the University of Wisconsin-Milwaukee greenhouse. The nymphs of both species grew up in groups of similar densities, and thus had similar early experience. *Enchenopa binotata* "Ptelea" molted to adults May 31-June 4 2009, and *E. binotata* "Viburnum" molted to adults between 29 June and 10 July 2009. We separated females from males within two to three days after the adult molt. Because males do not start signaling until ca. 2 weeks post adult molt, this time frame allowed us to control the signal experience of females. Individuals of each species were randomized among treatments and replicates to minimize the relatedness among individuals within the same treatment or replicate.

EXPERIMENTAL DESIGN

This study consisted of two phases: an experience phase during which we manipulated the signal experience of females, and a testing phase in which we described female preferences for signal frequency. We generated and controlled all stimuli using a custom-written program in MATLAB version 7.5.0. An iMac computer delivered stimuli, standardized to an amplitude of 0.3 mm/s, through a piezo-controller (Thorlabs, NJ, MDT694A) and a piezo-electric stack (Thorlabs, NJ, AE0505D16). The piezo-electric stack was coupled to the stem of a potted host plant with accelerometer wax. The stimuli were presented to females placed on the plants a few centimeters away from the piezo-electric stack. We isolated the setup from building vibrations by placing it on a ca 135-kg iron plank that rested on partially inflated bicycle inner tubes on a table. The plant was isolated from the iron plank by shock-absorbing sorbothane (Edmund Scientifics, Tonawanda, NY). We monitored the stimuli and female responses by focusing the beam of a laser vibrometer (Polytec CLV 2534; Polytec Inc., Auburn, MA) on a small (ca 2 mm) piece of reflective tape attached to the plant stem. Signals from the laser were sent to an iMac computer and were recorded with the sound analysis program AUDACITY (version 1.2.5; http://audacity.sourceforge.net/).

Experience phase

The experience phase started when females were two to three weeks old, during the beginning of the peak signaling period for males, but before females are sexually receptive. The experience phase continued for each individual female until she was tested—any female not tested on a given day of the testing phase continued to receive experience the following day. To control for the amount of experience across treatments, we randomized testing across treatments and replicates over the length of the testing phase. The experience phase lasted 2.5 weeks on average.

We randomly assigned females to one of four signal-experience treatments. Each treatment had two replicates, each consisting of 20 females residing on a potted plant stem enclosed in a mesh sleeve cage. Females experienced the stimuli for 50 min per day until final testing. Females may experience signaling males for up to several hours per day in nature (Wood and Guttman 1982; Sullivan-Beckers and Cocroft 2010). The duration of our treatment each day represents a balance between a realistic duration of experience and the need to expose all females during the hours males naturally call. The treatments were as follows: (1) Silence—no playbacks. This treatment corresponds to a low abundance of mates. (2) Preferred frequency—stimulus with the mean signal frequency for the population (mean ± standard error: 338 ± 2.2 Hz *E. binotata* "Ptelea" and 183 ± 1.2 Hz for *E. binotata* "Viburnum"; Rodriguez et al. 2006; K. D. Fowelr-Firn, and R. L. Rodríguez, unpubl. data) with all other signal values set at the population mean. (3) Low-frequency—nonpreferred stimulus with the frequency 100 Hz below the mean signal frequency (238 Hz *E. binotata* "Ptelea" and 83 Hz for *E. binotata* "Viburnum") with all other signal values set at the mean. (4) High-frequency—nonpreferred stimulus with the frequency 100 Hz above the mean signal frequency (438 Hz *E. binotata* "Ptelea" and 283 Hz for *E. binotata* "Viburnum") with all other signal values set at the mean. These shifts of 100 Hz correspond to seven standard deviations for *E. binotata* "Ptelea" and 14 standard deviations for *E. binotata* "Viburnum." The 200 Hz difference between low and high frequency stimuli corresponds to 50% of the range of variation in the *E. binotata* complex. Our nonpreferred treatments (3 and 4) thus overlap with heterospecific signals (Rodríguez et al. 2006; Cocroft et al. 2010). This range of frequencies results in strong mate discrimination in playback experiments (Rodríguez et al. 2004, 2006).

Testing phase

The testing phase started 4 weeks after the adult molt, one week after the onset of female receptivity (Rodríguez et al. 2004; Cocroft et al. 2008). The signal features were set to the mean of each species as described for the experience phase, but varied in frequency in steps of 20 Hz, 50 Hz, and 100 Hz in both directions away from the mean for each species. These steps were designed to allow the detection of small shifts in peak preference away from the mean while also spanning frequencies that should be outside of the normal range of tolerance for females (Rodríguez et al. 2006). We presented a random sequence of synthetic signals in signal bouts separated by 15 sec. We used the mean number of signals per bout for each species: six signals/bout for *E. binotata* "Ptelea," and three signals/bout for *E. binotata* "Viburnum" (Rodríguez et al. 2006).

Our assay of mate preferences is based on the duetting system of the *E. binotata* complex. Pair formation in the *E. binotata*

complex involves male–female signal exchanges, referred to as duets. Mate-searching males fly from plant to plant, producing advertisement signals. If a receptive female finds a male's signals attractive, she produces her own response signals in a species-specific interactive pattern, prompting the male to search locally for her (Rodríguez et al. 2006; Cocroft et al. 2008). Duetting ceases when mating begins. Thus, whether a female responds to a male's signals influences the likelihood of her mating with him, and female responses can be used as an assay of mate preferences. This technique has been used to demonstrate mate choice on the basis of between- and within-species signal variation (Rodríguez et al. 2004, 2006). We analyzed the recordings using AUDACITY (version 1.2.5) by scoring the number of female responses per stimulus (0–6 for *E. binotata* "Ptelea" and 0–3 for *E. binotata* "Viburnum"). In the *E. binotata* complex, the likelihood of responding to a male signaling bout correlates with the number of responses, and also correlates with the total response length (Rodríguez et al. 2004). In this study, the number of responses was strongly correlated with total response length (Pearson product-moment correlation, $r = 0.98$–1.00). We obtained the same results using response length as the dependent variable (not shown). Therefore, we use response number as a proxy for an underlying continuous response variable.

We started with 160 females for each species, and survivorship to the testing phase resulted in 141 *E. binotata* "Ptelea" females tested, and 58 *E. binotata* "Viburnum" females tested. We lost one replicate for the "low frequency" treatment group in *E. binotata* "Viburnum" due to low survival on that specific plant. Survivorship may have been affected by the health of the host plants or species-specific mortality rates because *Viburnum* plants may have been stressed whereas *Ptelea* plants were not. However, differences in mortality between species had little influence on the overall patterns of plasticity, which were similar between species. We also had sufficient power for detecting significant differences among treatments despite the lower sample for *E. binotata* "Viburnum," (see Results). We therefore retain both species in the analyses. Ninety-two of the 141 tested *E. binotata* "Ptelea," and 39 of the 58 tested *E. binotata* "Viburnum" were responsive and included in the analyses. This resulted in a final sample size of 92 *E. binotata* "Ptelea" females, and 39 *E. binotata* "Viburnum."

DESCRIPTION OF FEMALE PREFERENCE FUNCTIONS

We characterize variation in mate preferences across treatments using preference functions, which are curves that describe female responses according to variation in signals (Ritchie 1996; Blows et al. 2003; Brooks et al. 2005; Bentsen et al. 2006; Rodríguez et al. 2006). We visualized preference functions using cubic splines—nonparametric regressions that make no assumptions about the shape of the preferences other than some level of

smoothness (Schluter 1988; Ritchie 1996). We calculated splines for each replicate within treatment based on 1000 bootstraps in the program created by D. Schluter (www.zoology.ubc.ca/~schluter/lab.html), using a smoothness value of lambda = 2. Lambda determines the window width used in the running regression, and therefore the "stiffness" of the curves.

We used a function-valued approach to describe female preferences (Meyer and Kirkpatrick 2005; McGuigan et al. 2008). This approach considers the entire female preference function as the trait of interest, instead of single points along the curve. Using this approach, we tested for overall differences in preference function shape among treatment groups using a linear mixed model. Because we sampled each individual female along the range of stimuli, each female contributes a full preference function. We therefore included female identity nested within treatment as a random effect in the model. The model additionally included treatment, replicate nested within treatment, the linear and quadratic stimulus-frequency terms, the treatment × linear stimulus-frequency interaction, and the treatment × quadratic stimulus-frequency interaction. The linear stimulus-frequency term refers to female response as a function of increasing/decreasing signal frequency (e.g., open preference) whereas the quadratic stimulus-frequency term refers to female response as a curvilinear function of signal frequency (e.g., closed preference). Significant interaction terms indicate variation among treatments in the shape of female preference functions. The treatment × quadratic stimulus-frequency interaction was the critical component of the model, as female preference for signal frequency is a curvilinear function in the *E. binotata* complex (Rodríguez et al. 2004, 2006; Sullivan-Beckers and Cocroft 2010). A significant effect of this interaction (see Results) indicated an effect on the shape of the preference functions. This result warranted further investigation into the specific aspects of the preference functions that varied among treatments. We used four traits to describe the shape of the preference function and determine where the treatments had the most effect: peak, responsiveness, tolerance, and strength of preference (Fig. 1). The peak describes the preferred mate type. Responsiveness, tolerance, and strength represent female mate choice selectivity; females are considered more selective as their responsiveness decreases, their tolerance decreases, and the strength of their preference increases. We described correlations between these four traits by calculating the correlations between each pair of traits for each experimental treatment, and then averaging across the four treatments. However, we analyzed each trait separately because of their different consequences for selection on male signal frequency. We obtained one value for each trait per female. We analyzed each trait with a nested ANOVA including the trait of interest as the response variable, treatment as an independent variable, and replicate nested within treatment as an additional variable. We
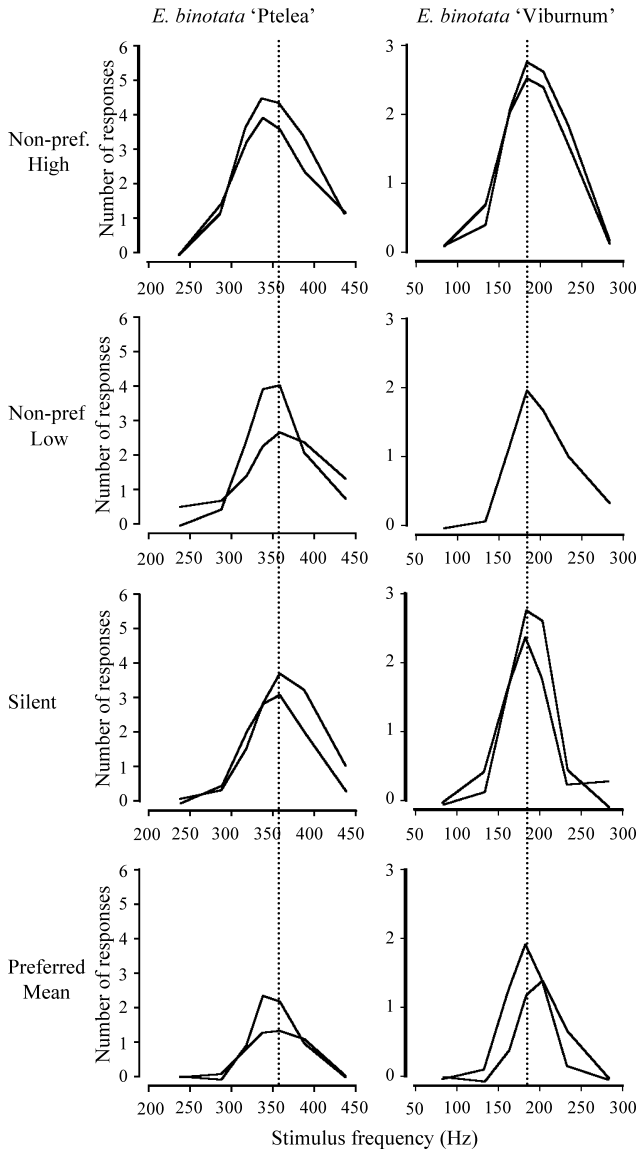
Figure 2. Tests for the overall shape of female preference functions show variation in preferences according to variation in the experience of signals. Replicate-level preference functions show that female *E. binotata* differ across treatments in traits that pertain to the selectivity of preference functions, but not in peak preference. The dashed line marks the average male signal frequency for the populations tested. Each curve corresponds to a replicate within a treatment, with two replicates per treatment. Treatment is indicated left of the *y*-axis.

performed all statistical analyses in JMP (Version 6.0). Analyses were performed separately for *E. binotata* "Ptelea" and *E. binotata* "Viburnum."

Results

We found experience-mediated plasticity in female preferences for *E. binotata* "Ptelea" and *E. binotata* "Viburnum." Patterns

Table 2. Variation in *E. binotata* female mate preferences, according to variation in experience of stimulus frequency (Hz). Significant *P*-values are highlighted in bold.

Factor	df	*F*-ratio	*P*
Enchenopa binotata "Ptelea"			
Whole model	97,546	5.82	**<0.0001**
Treatment	3,546	20.29	**<0.0001**
Replicate	4,546	0.94	0.4420
Linear	1,546	28.02	**<0.0001**
Treatment×linear	3,546	0.90	0.4427
Quadratic	1,546	149.75	**<0.0001**
Treatment×quadratic	3,546	4.97	**0.0021**
Individual (random effect)	82,546	3.18	**<0.0001**
Enchenopa binotata "Viburnum"			
Whole model	46,226	4.94	**<0.0001**
Treatment	3,226	12.76	**<0.0001**
Replicate	3,226	0.37	0.7758
Linear	1,226	3.64	0.0577
Treatment×linear	3,226	0.36	0.7799
Quadratic	1,226	104.18	**<0.0001**
Treatment×quadratic	3,226	3.07	**0.0287**
Individual (random effect)	32,226	1.54	**0.0393**

were similar for both species, but stronger in *E. binotata* "Ptelea." Inspection of the preference functions suggests that the treatments mostly affected selectivity (i.e., responsiveness, tolerance, and strength) rather than peak preference (Fig. 2). The significant treatment effect indicates variation in responsiveness across treatments (Table 2). The significant treatment × quadratic stimulus-frequency term indicates variation in the shape of preference functions across treatments (Table 2). The significant individual term indicates individual differences in responsiveness that may provide material for selection on preferences, and that may also influence selection on male traits.

To determine which aspects of the shape of preference functions varied among treatments, we analyzed four preference function traits: preference peak, responsiveness, tolerance, and strength. The three selectivity traits were highly correlated with each other, but they were each uncorrelated or weakly correlated with peak preference (Table 3). Females experiencing the preferred signal stimuli showed the greatest selectivity for preferred signals: this included lower responsiveness, lower tolerance, and greater strength of preference (Fig. 3; Table 4). Experience treatments did not affect peak preference (Fig. 3 and Table 4).

Discussion

We tested two hypotheses about selection on experience-mediated plasticity in mate preferences: "mating assurance" and "mismating avoidance." Each hypothesis describes a difference

Table 3. Correlations between peak preference, responsiveness, tolerance, and strength. The strongest correlations are those between the three aspects of selectivity.

	Responsiveness	Tolerance	Strength
E. binotata "Ptelea"			
Peak	0.24	0.33	0.01
Resonsiveness		0.80	−0.68
Tolerance			−0.67
E. binotata "Viburnum"			
Peak	0.35	0.41	−0.31
Responsiveness		0.91	−0.81
Tolerance			−0.74

balance of the costs and benefits of mate choice resulting from the evolutionary history of variation in preferred and nonpreferred mates. We tested the predictions of each hypothesis by characterizing patterns of plasticity across different perceived social environments. Our results support the mating assurance hypothesis: we found that females that experienced nonpreferred signals or silence were less selective than those that experienced preferred signals. Also, females did not change peak preference according to experience. Further, our results reject the mismating avoidance hypothesis: females that experienced nonpreferred signals showed lower, instead of higher, mate preference selectivity. Also, females did not shift peak preference away from the nonpreferred signals they experienced. We therefore infer that selection has favored experience-mediated plasticity in mate preferences that ensures mating when preferred mate types are rare or absent, while also ensuring the choice of preferred types when those are present. Consequently, females may be less likely to forgo mating by being too selective when preferred mate types are not available. At the same time, they remain likely to mate with preferred mate types when they are available. Therefore, females can maximally mate with preferred types without being so selective that reproductive success is sacrificed. That this is a real risk is suggested by a study of mating success in field-like conditions that found that some females had not mated during a six to eight week span of the mating season (Sullivan-Beckers and Cocroft 2010).

Our framework predicts that the nature of variation in the composition of the social environment will influence selection on experience-mediated plasticity. In support of this notion, we find some studies that are consistent with the mating assurance hypothesis and other studies that are consistent with the mismating avoidance hypotheses. For example, in some cases documenting experience-mediated plasticity, experience of preferred signals results in greater selectivity relative to no experience of signals (planthoppers, Dewinter and Rollenhagen 1993; Drosophila, Dukas 2005; field crickets, Bailey and Zuk 2008),
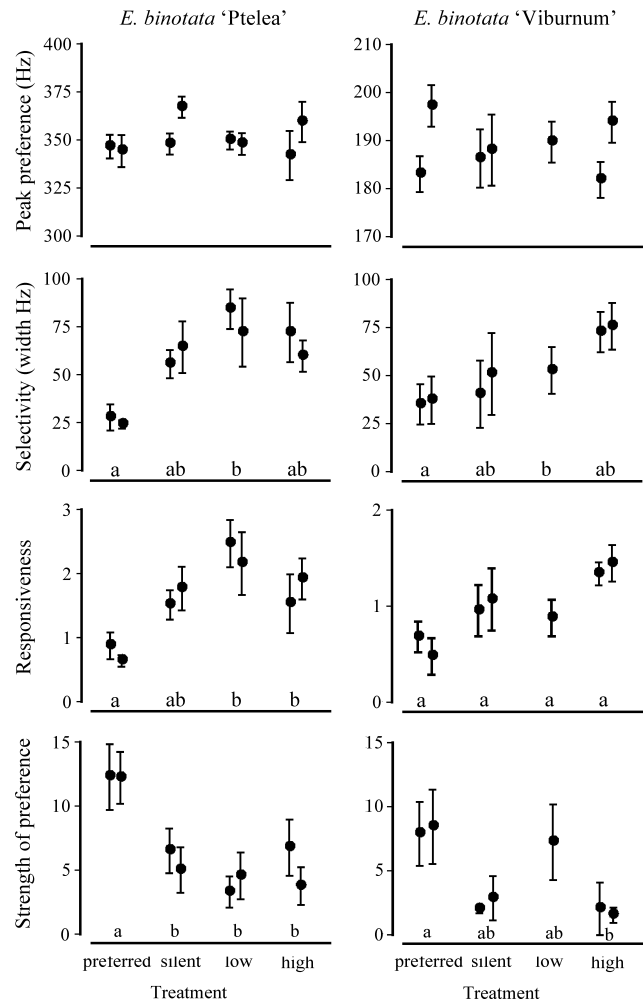

Figure 3. Tests for variation in each of four preference function traits showing variation in some aspects of preference function shape according to variation in the experience of signals. Peak preference did not vary among treatments. Females that experienced nonpreferred signals or had no experience with signals (silence) show greater responsiveness, greater tolerance, and weaker strength of preference. Variation among treatments in the four aspects of preference function shape are illustrated with means ± standard error corresponding to the circle symbols. Two values per treatment correspond to the two replicates in each treatment, with treatment indicated on the x-axis. Significant differences in responsiveness, tolerance, and strength of preference (B–D) between treatments that were identified by Tukey's HSD posthoc tests are indicated by a's and b's in the figure.

as predicted by the mating assurance hypothesis. In other cases, experience of nonpreferred signals results in greater selectivity (guppies, Magurran and Ramnarine 2004; stickleback, Kozak and Boughman 2009), as predicted by the mismating avoidance hypothesis. In one example, guppies vary among populations in the degree of sympatry with heterospecifics, which may result in corresponding differences in the strength and/or pattern of

Table 4. Effect of perceived social environment on variation in mate preferences. We consider variation in four aspects of preference function shape: peak preference, responsiveness, selectivity, and strength. Females show no significant change in preference peak across treatment groups, but show differences in responsiveness, selectivity, and strength of preference. Significant *P*-values highlighted in bold.

Variable	Factor	df	*F* ratio	*P*
Enchenopa binotata "Ptelea"				
Peak	Treatment	3,82	0.91	0.4417
	Replicate	4,82	1.15	0.2048
Responsiveness	Treatment	3, 82	5.09	**0.0028**
	Replicate	4,82	0.35	0.8439
Selectivity	Treatment	3,82	5.37	**0.0020**
	Replicate	4,82	0.37	0.8304
Strength	Treatment	3,82	6.16	**0.0008**
	Replicate	4,82	0.48	0.7491
Enchenopa binotata "Viburnum"				
Peak	Treatment	3,32	0.16	0.9207
	Replicate	3,32	3.60	**0.0238**
Responsiveness	Treatment	3,32	7.34	**0.0007**
	Replicate	3,32	0.32	0.8127
Selectivity	Treatment	3,32	3.79	**0.0197**
	Replicate	3,32	0.07	0.9849
Strength	Treatment	3,32	3.99	**0.0159**
	Replicate	3,32	0.03	0.9917

experience-mediated plasticity among populations (Magurran and Ramnarine 2004). We also see evidence in the literature for selection favoring experience-mediated plasticity where females acquire a preference for the most commonly experienced conspecific mate type (e.g., Hebets 2003). Our data do not support this hypothesis, but we used heterospecific-like nonpreferred stimuli, which were well outside the range of conspecific signals. Therefore a shift in peak preference to match the experienced signal frequency would require a shift to preferring heterospecific-like signals.

The patterns of plasticity that we find have interesting implications for selection on sexual displays, patterns of reproductive isolation, the maintenance of genetic variation, and the colonization of novel environments. Experience-mediated plasticity will vary selection on signals as a function of females' experience of the signals in the environment. In this study, peak preference did not vary with experience, and matched the mean signal frequency of males in the two respective species (Rodríguez et al. 2006). Thus, selection on signal frequency will remain stabilizing. However, preference selectivity will vary depending upon the signals that females experience, and therefore so will the strength of selection on sexual signals. This variation in the strength of selection may provide a mechanism that contributes to the maintenance

of genetic variation in sexual signals (Chaine and Lyon 2008; Morris et al. 2010). There may also be a positive feedback loop that will further contribute to the maintenance of genetic variation. In this feedback look, a high abundance of nonpreferred mates results in lower selectivity, which will weaken selection and increase the likelihood of mating with nonpreferred males. If male signals are heritable (e.g., *E. binotata* "Ptelea' shows a low but significant heritability in signal frequency; Rodríguez et al. 2008), the next generation will be composed of a greater number of nonpreferred males, again resulting in lower selectivity. Thus, populations that are less dense or that experience a greater number of nonpreferred signals should have a higher potential to maintain phenotypic and/or genetic variation in male signals. For example, in a population of field crickets where the majority of males do not produce advertisement signals, females that do not experience advertisement signals are less selective, and sexual selection on the male advertisement signal is likely relaxed (Zuk et al. 2006; Bailey and Zuk 2008).

Variation in selectivity resulting from experience-mediated plasticity should affect the level of reproductive isolation among species or populations. The pattern of experience-mediated plasticity that we find predicts that reproductive isolation should be weakened when females experience nonpreferred mates (e.g., males from another population or heterospecifics), or when females experience few or no preferred mates. Such weakened reproductive isolation may counter sources of divergent selection. However, it may, under some conditions, act in concert with divergent selection. For example, experience-mediated plasticity may facilitate the colonization of novel environments, which can often be an important step in the process of speciation; when new populations are established, differential ecological and sexual selection across variable environments can result in divergence and potentially speciation (Schluter 2001; Drès and Mallet 2002; Coyne and Orr 2004; Rundle and Nosil 2005; Nosil and Crespi 2006; Nosil 2007; Cocroft et al. 2008, 2010; Schluter 2009). However, fixed, strong female preferences may inhibit initial successful establishment in a novel environment, where signals may show a higher likelihood of deviating from the source population (Kaneshiro 1976, 1980). Therefore initial successful establishment in a novel environment may require females to mate with males with nonpreferred signals. The pattern of experience-mediated plasticity that we find predicts an increased likelihood of mating for individual females. This decreased selectivity may facilitate the successful establishment in a novel environment.

CONCLUSION
Experience-mediated plasticity in mate preferences may often be favored by selection to modulate the trade-off between the benefits of mate choice and the risk of losing mating opportunities. In turn, it may have important consequences for the course of

sexual selection and divergence. It may contribute to the maintenance of genetic variation, or weaken reproductive isolation between populations. Finally, it may set the stage for ecological speciation by facilitating the successful colonization of novel environments.

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