



Male mate choice by chemical cues leads to higher reproductive success in a bark beetle

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Darwin's sexual selection theory predicts that males should compete to gain access to females, while females should choose the best possible male. However, males can also be choosy, especially in monogamous mating systems with female-biased sex ratios and biparental care. A few studies show that in systems in which males are highly selective, chemical cues can lead to mating of individuals with high reproductive success. Using *Dendroctonus valens*, a monogamous bark beetle with typical biparental care, and Y-tube olfactometers, we provided males with a choice between two females and found strong evidence of odour-based male mate preference. We then tested whether this preference led to successful matings and if it had consequences for courtship displays and offspring fitness. Male and preferred female pairs had less aggressive courtship interactions, longer copulation times, constructed galleries faster, and had cleaner galleries. Preferred females had faster oviposition rates and larval development. Our study shows that, in *D. valens*, males choose females based on chemical cues and that the resulting male–female pairs have superior reproductive success.

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Since Darwin (1871), sexual selection has been divided into intersexual selection (mate choice by one sex) and intrasexual selection (competition by the same sex for access to mates). In intersexual selection, the sex with higher investment in reproduction must be choosy (Andersson 1994), and in many cases females fulfil this role (i.e. conventional sex roles) (Berglund et al. 2005). Female mate choice is more likely to evolve because the number of offspring a female produces increases either minimally or not at all with the number of matings she achieves (Bonduriansky 2001). Male mate choice, nevertheless, does not evolve as easily as female mate choice (Bonduriansky 2001; Clutton-Brock 2007). In male mate choice, the number of offspring a male produces increases in proportion to the number of matings he achieves (Bonduriansky 2001). Under certain conditions, males are expected to discriminate carefully among alternative potential mates. These include limited opportunities to obtain additional matings (e.g. monogamy), high parental investment (e.g. parental care), low effort required to find mates (e.g. several potential mates are encountered simultaneously), and high variation in female quality (Bonduriansky 2001; Barry & Kokko 2010).

Successful mate selection requires efficient sensorial and behavioural mechanisms, including highly evolved vision systems for detection of specific colour patterns, elaborate sexual rituals and chemical cues (Andersson 1994). Chemical cues play an important role in mate choice in many taxa (Johansson & Jones 2007). In insects, the use of pheromones is almost ubiquitous and researchers have typically emphasized the role of pheromones as sex attractants (Lewis 1984; Roitberg & Isman 1992; Morgan 2004). It is often assumed that the use of chemical cues only leads to simple discriminations, for example, to determine whether another individual is a conspecific or a heterospecific (species recognition), or either a male or a female (mate recognition). However, a growing body of evidence indicates that chemical cues may be used by males or females to derive complex information about potential mates (Johansson & Jones 2007; Coleman 2009). Insect chemical cues have been shown to provide information regarding size, body condition, parasite load, immunocompetence, familiarity, reproductive status and degree of relatedness (Johansson & Jones 2007). Despite this, there is little evidence showing that, in systems in which males are highly selective, interindividual variation in chemical cues allows assessment of potential mates with potential consequences for reproductive success.

Chemical communication has long been recognized in bark beetles (Sauvard 2004; Blomquist et al. 2010). Some evidence suggests that recognition of chemical signals is probably the first

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cue to bring males and females together (Wood 1982; Blomquist et al. 2010) but there is no experimental evidence on the effects of chemical cues on male reproductive decisions in bark beetles.

The red turpentine beetle, *Dendroctonus valens*, native to North America and introduced to China in the early 1980s, is one of China's most destructive invasive pests of pine forests killing over 500 000 km² of Chinese oil pines, *Pinus tabulaeformis* (Yan et al. 2005). To overcome pine's natural defence mechanisms, *D. valens* attacks in large numbers (Smith 1971). Although the sex ratio of *D. valens* in natural populations is nearly 1:1, a skewed operational sex ratio (i.e. ratio of sexually active females to males; Emlen & Oring 1977) occurs during a mass attack, which varies from being female biased in the initial attack phase, to unbiased with an occasional male bias at the end of the attack phase (H. F. Chen & J. H. Sun, unpublished data). *Dendroctonus valens* is monogamous (i.e. the male pairs with only one female) and is characterized by a high level of cooperative biparental care (Smith 1971; Kirkendall 1983; Kirkendall et al. 1997; Liu et al. 2006). Females initiate boring through the outer bark of pines and excavate a small, circular nuptial chamber in the phloem (H. F. Chen & J. H. Sun, personal observation). Pheromones released by the female in the nuptial chamber typically attract a male within 3–5 days of colonization (Smith 1971; Liu et al. 2008). After excavating at least 2–3 cm of gallery, the pair begins to excavate a wider gallery in which eggs are oviposited (egg gallery). Egg gallery length has a positive correlation with offspring fitness because, as shown in other bark beetles, longer egg galleries give each newly hatched larva a larger area of fresh phloem on which to feed (Robins & Reid 1997; Pureswaran et al. 2006). Larvae of each family feed gregariously, that is, side by side in an irregular line, steadily moving forwards into fresh phloem (Smith 1971). Within a tree, multiple galleries occur and, often, neighbouring larval tunnels from different families meet. As a result, larval competition for phloem is fierce; in fact, this has been documented as an important fitness factor in bark beetles, including *D. valens* (Beaver 1974; Anderbrant et al. 1985; Anderbrant & Schlyter 1989; Kirkendall 1989).

An additional factor that plays a major role in all the aforementioned ecological interactions in *D. valens* is chemical communication. Recent studies reported that the hindguts of *D. valens* males and females contain five compounds (cis- and trans-verbenol, myrtenal, myrtenol and verbenone) and documented their effects on the beetle's colonization behaviour (Zhang & Sun 2006; Zhang et al. 2006, 2009; Liu et al. 2008). Furthermore, there is evidence suggesting that *D. valens* males may use chemical cues for mate recognition (Liu et al. 2006).

Given the importance of chemical cues in the mating system in *D. valens*, the occurrence of male mate choice, modulated via female chemical cues, would not be surprising. If evolution has favoured this type of sexual selection in *D. valens*, male–female pairs formed through this mechanism are likely to have fitness benefits. To test this hypothesis, we performed laboratory experiments to determine (1) whether males select females using chemical cues, (2) whether males' odour-based preference for females leads to successful matings, including consequences for courtship displays, and (3) whether the offspring of females selected by their odour have higher fitness.

METHODS

Study Subject

Laboratory experiments were conducted from 8 May to 30 June 2010 in a natural forest of *P. tabulaeformis* at the Beishe Mountain near the foot of the Luliang Mountains (37°48'N, 111°44'E; average elevation 1400 m). The onset of experiments coincided with the

spring dispersal flight of newly emerged adult beetles (i.e. virgin females and males). In this period, which lasts 15 days, newly emerged virgin females fly to find a suitable tree to make galleries. To evaluate the condition of females, we dissected 132 field-collected females to determine presence of sperm in the spermathecal sac (details on this procedure are reported by Lissemore 1997). Only 3.8% (5/132) of females were mated. Beetles were collected daily from traps baited with *D. valens* kairomone lure (+)-(3)-carene. Sexes were separated based on the stridulation of males (Lyon 1958). Beetles were used within 2 days after capture. Prior to experiments, body length (anterior tip of head to posterior end of elytra) of each beetle was measured with an accuracy of 0.02 mm using a Vernier calliper. To factor out potential size effects, male mean body length was estimated (6.72 ± 0.06 mm, $N = 427$) and males whose size fell within the mean \pm SE were used.

Experiment 1: Mate Choice with Live Females and Frass

Females collected from field traps were individually placed in labelled transparent plastic centrifuge tubes (12.5 cm height \times 2.2 cm diameter). Five pieces of fresh phloem (each piece 11 \times 0.5 \times 0.3 cm) were placed inside each tube for 36 h to simulate natural conditions of attack. This also allowed us to obtain female frass (i.e. faeces + phloem bore-dust) for our experiments. Female frass contains female-produced male volatile attractants (Liu et al. 2006; Shi & Sun 2010). To test male mate choice based on chemical cues alone, we carried out an experiment with two phases: phase 1 (tests with live females) and phase 2 (tests with frass produced by females from phase 1).

In phase 1, male preference for females was assessed using a glass Y-tube olfactometer, consisting of two female chambers, each being at the end of one arm, and one male start area at the beginning of the stem (Fig. 1). Wire-mesh walls (mesh size 2 mm) were placed in the female chambers to restrain the female to that area of the olfactometer. Unidirectional airflow was passed through the female chambers, at 150 ml/min, so male beetles had to walk upwind; air was filtered with activated carbon before entering the apparatus (Fig. 1). To factor out visual cues, the whole set-up was covered with a black blanket. Prior to each trial, females were first placed in the female chambers for 30 min for acclimatization before we placed the male in the male start area (Fig. 1). Males were given 10 min to make a decision after being released. We deemed that a choice was made when a male walked at least halfway into the chosen arm. Chosen females were labelled as P (preferred) and nonchosen females were labelled as NP (nonpreferred). Trials in which a male stayed in the male start area or did not walk into an arm further than halfway were discarded. Females were placed in either chamber (i.e. left or right) of the Y-tube randomly. Y-tubes were cleaned with acetone and dried for 5 h, to remove all possible chemical cues, before each trial. The whole set-up was maintained at 27 ± 3 °C and $60 \pm 12\%$ relative humidity during trials. A total of 181 trials were performed in this experiment. After phase 1, each individual was placed separately in a labelled vial with fresh phloem pieces for 48 h before phase 2.

To verify that male mate choice was based on chemical cues alone, we carried out phase 2 using the same set of beetles (one male and two females) in which the male had successfully made a choice in phase 1. Phase 2 was performed with the same Y-tube olfactometers used in phase 1, except that the females' frass was used instead of the females. In phase 2, frass was placed in opposite orientation (i.e. left or right) compared to the position of the female donor in phase 1. We used 100 ± 5 mg of frass from each female. We only used frass collected no more than 1 h prior to each trial. A total of 148 trials were conducted in phase 2.

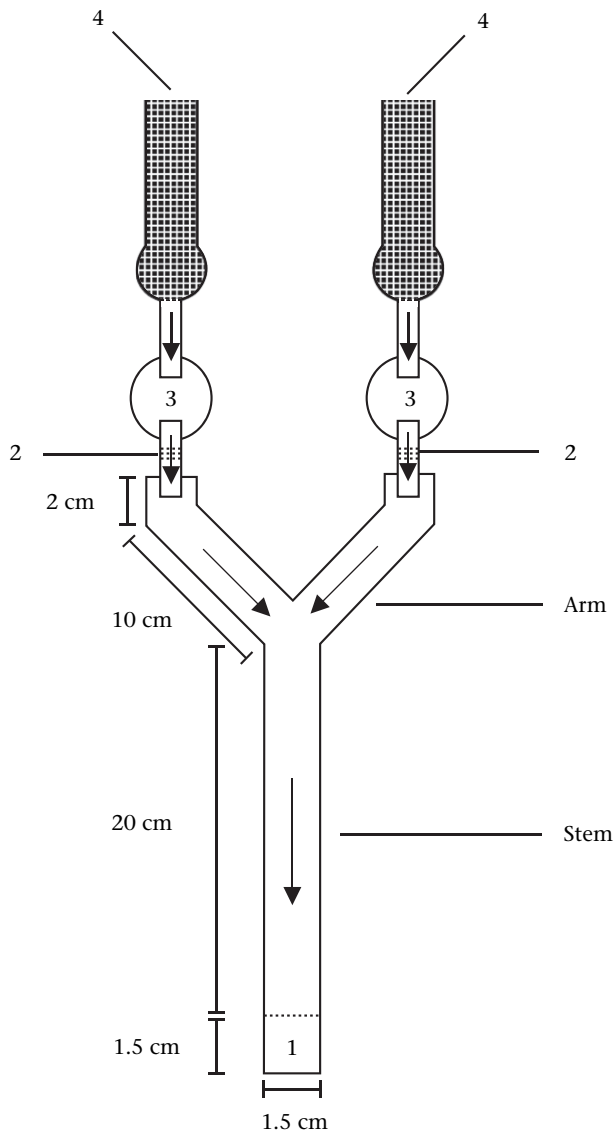


Figure 1. Diagram of the Y-tube olfactometer. 1: male start area; 2: wire-mesh barrier; 3: test chambers for odour stimulus presentation (live female or frass); 4: activated carbon filters. Arrows show airflow direction.

Experiment 2: Effects of Mate Choice on Mating Interactions

To determine whether female odour-based attractiveness would lead to mating success, we randomly selected either a P or an NP female from experiment 1 and placed it with its matching male in a mating chamber that allowed monitoring of the beetles while simulating natural conditions. The mating chamber consisted of a 'phloem sandwich' (a 25 cm square piece of fresh pine phloem pressed between two 25 cm square glass plates). Sandwich edges were covered with polyvinylidene chloride wrap to prevent water loss. To allow ventilation, six holes were pierced with a pin on each side of the sandwich. In each trial, one female was placed in a hole drilled in the centre of each phloem square. After the female had bored a gallery (24 h later), the matching male was placed in the gallery entry hole and allowed to walk in. For 30 min, we recorded (1) occurrence of mating, (2) duration of copulation and (3) number of 'walk away' behaviours (i.e. when a male faced the posterior part of the abdomen of the female and then walked away from her). A total of 21 P and 20 NP females were used. Females that did not

build a gallery were not included in the data analysis. Beetle interactions were recorded using a digital video camera (Sony DCR-HC40E) to identify courtship behavioural traits.

Experiment 3: Reproductive Success

We also tested whether there were any differences in oviposition rate and offspring development when males were paired with P or NP females. We used bolts as nesting arenas to simulate natural conditions. Thick bolts can increase the success of nesting, so the bolts we chose were 29–35 cm diameter, 70 cm long, and cut from the bottom part of live Chinese oil pines. Bolt cut ends were sealed with paraffin wax to reduce desiccation. To establish the breeding galleries, six or eight spaced holes (30 cm apart), 1.9 cm in diameter, were drilled in a uniform distribution into the phloem layer of the bark in each bolt. The holes had enough space between them to allow feeding of larvae without accidentally connecting galleries. One female was placed per hole, allowed to begin gallery construction, and 24 h later a male was added for pairing. The day a male was placed in a hole was defined as the first day of pairing. In total, 49 P and 48 NP females were placed in the bolts. All holes were covered with iron wire mesh (mesh size, 2 mm) to ensure that the beetles remained in the gallery. All bolts were kept upright in a rearing room and maintained at temperatures of $27 \pm 3^\circ\text{C}$ and $60 \pm 12\%$ relative humidity with a 12:12 h photoperiod.

All the females that constructed a gallery were monitored daily throughout the experiment. We dissected a sample of tunnels at 5, 10, 20 and 30 days after gallery initiation. To do so, the outer bark was thinned by shaving or chipping, the inner bark on each side of the notches, and the eggs or larvae were removed with a fine moist brush. To determine differences in reproductive outputs (fitness) of females, we made observations of the galleries and recorded the following: (1) main gallery and egg gallery length at day 5, 10 and 20; (2) number of eggs at day 5 and 10; (3) number of eggs and larvae at day 20 and 30; (4) proportion of 'frass-covered gallery length' to 'total gallery length' at day 5, 10 and 20; and (5) number of larvae and their weight at day 30. We then quantified gallery lengths by measuring a pencil-traced profile (on tracing paper) with a Vernier calliper. Main gallery length was defined as the length of the main tunnel from the entrance to the division with the egg gallery. Egg gallery was defined as the part of the gallery that contained the eggs. We refer to eggs and larvae collectively as 'offspring' but distinguish them when appropriate. In (5) each larva was weighed using an electronic analytical balance with an accuracy of 0.01 mg.

Data Analyses

All statistical tests were based on Zar (1996) and were carried out with the statistical software SPSS 12 (SPSS Inc., Chicago, IL, U.S.A.). We used *F* tests (one- and two-way ANOVA and independent two-sample *t* tests) after verifying that the assumptions of the tests were met. Normality of data was tested with a Shapiro–Wilk test, and homogeneity of variance was tested using a Levene's test. Nonparametric statistics were used when transformations were unsuccessful to meet the assumptions of parametric analysis. Reported *P* values are from two-tailed tests, with $\alpha = 0.05$. Values are presented as mean \pm SE.

RESULTS

Experiment 1: Mate Choice with Live Females and Frass

In phase 1, males were attracted to a particular female (P) in 148 of the 181 trials (binomial test: $P < 0.001$). For the remaining 33

males that did not make a choice, seven stayed in the male start area, 12 entered one of the Y-tube arms but did not walk further than halfway, and 14 walked further from the start area but remained in the stem. Within pairs of females, body length of P females (6.86 ± 0.04 mg, $N = 148$) and NP females (6.89 ± 0.04 mg, $N = 148$) had no effect on mate choice (paired t test: $t_{147} = -1.512$, $P = 0.133$). In phase 2, 139 males confirmed the decision made in phase 1 by being attracted to the corresponding female frass; nine males failed to make a choice (binomial test: $P < 0.001$).

Experiment 2: Effects of Mate Choice on Mating Interactions

Courtship interactions before mating proceeded with little variation (Fig. 2). Following the initial encounter, where usually the male moved towards the female, the male oriented its head to the posterior part of the female abdomen. A brief pause usually followed, after which the male either 'walked away' or mounted the female and attempted to effect intromission. 'Walk away' events were often quick and occurred after no more than a brief physical contact. Encounters usually led to mounting and intromission attempts but not necessarily to copulation. In several cases, females attempted to block male intromission while being mounted (six of 23 P and 12 of 21 NP). Females did this by first moving outwards while pushing the male away, and then moving back. After this, the female rendered her genitalia inaccessible to the male. If the genital insertion attempt was not successful, the male dismounted and entered the courtship phase once again.

Courtship of males paired with P females led to copulation in 17 of 19 trials (Fig. 2a). For males paired with NP females, only nine of 17 trials led to copulation (Fig. 2b; Fisher's exact test: $P = 0.025$). Encounters between males and NP females resulted in significantly more 'walk away' behaviours (54 'walk away' for males–NP females and six for males–P females; Mann–Whitney U test: $U = 63.5$, $N_1 = 19$, $N_2 = 17$, $P = 0.002$; Fig. 2). Duration of copulation was significantly higher for P females (P females: 3.31 ± 0.21 min, $N = 17$; NP females: 2.47 ± 0.17 min, $N = 9$; independent-samples t test: $t_{24} = 2.697$, $P = 0.013$).

Experiment 3: Reproductive Success

In the bolt inoculation trials, 92.1% ($N = 101$) of the females placed in bolts for 24 h began gallery construction (eight females failed to build a gallery, two P, six NP). The main gallery and egg

gallery of P females were significantly longer than NP female galleries by day 5 but not after day 20 (Table 1). By day 5, 66.7% ($N = 12$) of P females and only 20% ($N = 10$) of NP females had built an egg gallery (Fisher's exact test: $P = 0.043$). Gallery length in different treatments across all of the bolts was independent of trees ($\chi^2 < 2.35$, all NS) and bolts from the same tree ($\chi^2 = 0$, all NS).

Egg gallery cleaning was substantially different between treatments. The percentage of egg galleries clear of frass was 78.8% for P ($N = 33$) and 53.6% for NP ($N = 28$; chi-square test: $\chi^2_1 = 4.371$, $P = 0.037$). Among the galleries that were blocked with frass, twice as much belonged to NP females ($23.86 \pm 2.55\%$ for P, $N = 7$; $46.62 \pm 5.97\%$ for NP, $N = 13$; Mann–Whitney U test: $U = 13.5$, $N_1 = 7$, $N_2 = 13$, $P = 0.011$).

By day 5 and 10, P females had produced more eggs than NP females (day 5: Mann–Whitney U test: $U = 28$, $N_1 = 12$, $N_2 = 10$, $P = 0.027$; day 10: independent-samples t test: $t_{25} = 2.603$, $P = 0.015$; Fig. 3). There were no significant differences in number of offspring at day 20 (Fig. 4) and day 30 (independent-samples t test: $t_{15} = 0.682$, $P = 0.506$). However, at day 20, the proportion of larvae to eggs was significantly higher for P females (independent-samples t test: $t_{25} = 4.864$, $P < 0.001$; Fig. 4), indicating rapid larval development. Females laid most of their eggs within 20 days of gallery initiation (Fig. 3). However, unhatched eggs were present in only one (NP) of the 17 galleries dissected after 30 days. At day 30, there was a significant effect of treatment on the weight of the larvae (independent-samples t test: $t_{15} = -6.188$, $P < 0.001$; Fig. 5), indicating larvae produced by P females were significantly heavier than larvae produced by NP females. Offspring number and weight were not affected by family (all $F < 1.536$, all NS) or bolt (all $F < 5.752$, all NS) in each treatment. NP parents had higher mortality compared to P parents over the 30-day period (P: 16.67%, $N = 96$; NP: 32.22%, $N = 90$; chi-square test: $\chi^2_1 = 6.129$, $P = 0.013$).

DISCUSSION

Our study reveals that male *D. valens* are able to assess and choose mating females using odour cues alone and that the resulting male–female pairs have enhanced reproductive success.

Sexual selection in *D. valens* is a complex process that is strongly influenced by several factors. The operational sex ratio in *D. valens* changes from being female biased during the initial attack phase (when mating occurs) to unbiased with an occasional male bias at the end of the attack phase (H. F. Chen & J. H. Sun, unpublished

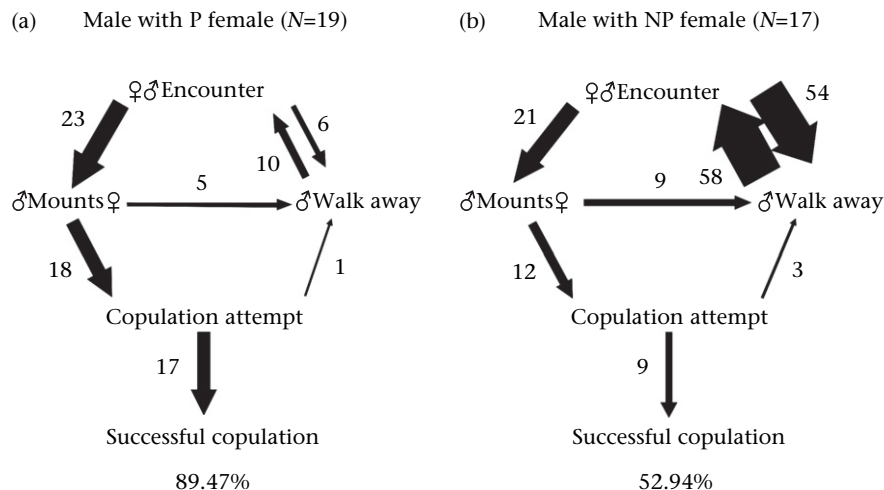


Figure 2. Kinematic diagrams showing transitions between common action patterns in male courtship attempts with (a) P females and (b) NP females. The width of the arrow represents the frequency of a particular transition. Values at the bottom of (a) and (b) show the percentages of successful copulations.

Table 1
Mean ± SE main gallery length and egg gallery length (cm) constructed by male–female pairs at day 5, 10 and 20

Type of pair	5 days		10 days		20 days	
	Main gallery	Egg gallery	Main gallery	Egg gallery	Main gallery	Egg gallery
♂ + P ♀	6.49±0.77	0.96±0.37	9.66±1.09	5.68±0.51	9.92±0.89	6.13±0.37
♂ + NP ♀	4.18±0.58	0.38±0.26	6.81±0.68	3.58±0.56	9.01±0.75	6.20±0.51
<i>t/U</i>	2.308	3.627	2.256	2.768	0.750	−0.111
<i>P</i>	0.032	0.041	0.033	0.010	0.460	0.912

For 'egg gallery' at 5 days statistical analysis was based on a Mann–Whitney *U* test; other comparisons were tested with an independent-samples *t* test. The *t/U* values in each column represent tests for differences between the mean gallery lengths for the two types of male–female pairs. P ♀ = preferred female; NP ♀ = nonpreferred female; *N* = number of pairs.

data). The female-biased sex ratio at the mass attack stage leads to increased female competition and, as a result, mate-searching males have access to large numbers of females. In addition, *D. valens* is a monogamous species with biparental care. Males must be careful when selecting a female because there is only one chance at pairing with the best possible female and because of their investment in parental care. Sexual asymmetries may then evolve as a by-product of high selectivity in *D. valens* males and variation in the operational sex ratio. These asymmetries may involve evolution of effective signals to attract males. Chemical cues are a likely signal to be used in male attraction because males are not able to see the females before they enter the chosen gallery. Our results clearly show that males are attracted to pheromones emitted by particular females and their frass. Pheromone variation among individuals can be used for mate assessment (Johansson & Jones 2007). Variation may be achieved through quantitative or qualitative differences in the pheromone blend or via differential release rates (Johansson & Jones 2007). In bark beetles, there is evidence for high individual variation (i.e. males and females) in pheromone production and in frass scent (Birgersson et al. 1988; Birgersson & Bergström 1989; Schlyter & Birgersson 1989; Pureswaran & Borden 2003; Pureswaran et al. 2006, 2008). Furthermore, bark beetle pheromones are generally assumed to have a fixed number of active components (Wood 1982; Symonds & Elgar 2004; Blomquist et al. 2010) but the role of a large number of 'minor' components, which are often considered residuals or precursors (Zhang & Sun 2006; Johansson & Jones 2007; Shi & Sun 2010), may be important in sexual selection.

Although we did not record qualitative or quantitative data on the pheromone cues emitted by *D. valens* females, we detected

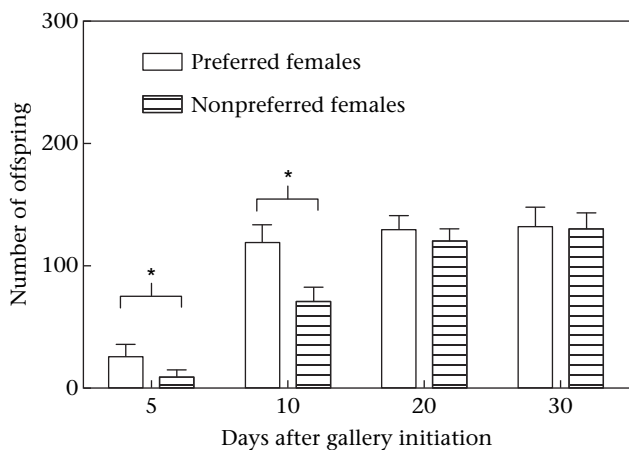


Figure 3. Numbers of offspring (eggs + larvae) after 5, 10, 20 and 30 days of gallery initiation. Values are mean + SE. **P* ≤ 0.05.

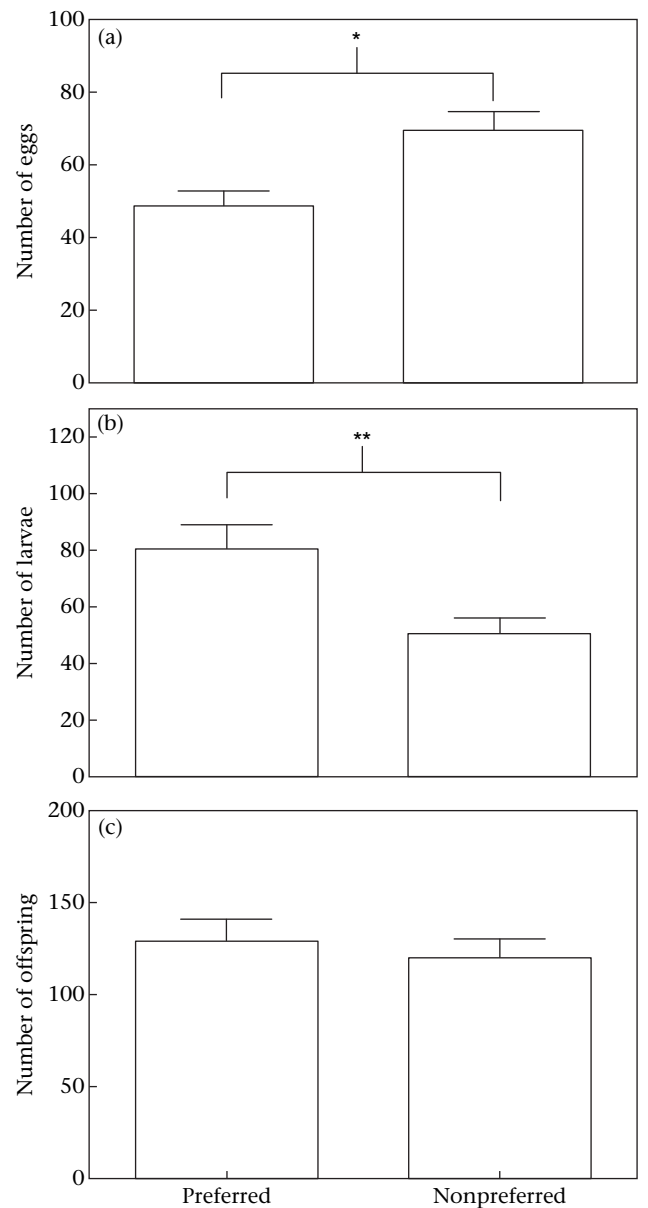


Figure 4. Number of (a) eggs, (b) larvae and (c) offspring (larvae + eggs) produced by P or NP females 20 days after gallery initiation. (a) *t*₂₅ = −2.391, *P* = 0.025, (b) *t*₂₅ = 3.612, *P* = 0.001, (c) *t*₂₅ = 1.051, *P* = 0.303. Values are mean + SE. **P* ≤ 0.05; ***P* ≤ 0.01.

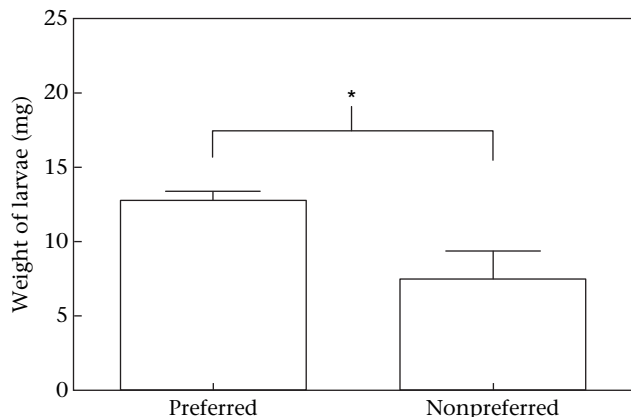


Figure 5. Weight of larvae produced by P or NP females 30 days after gallery initiation. Values are mean + SE. * $P \leq 0.05$.

important behavioural and reproductive differences between selected and nonselected females and their mates. When males were paired with P females, males and females constructed larger galleries during the first 10 days after gallery initiation. Preferred females also had an overall higher proportion of larvae to eggs than the other treatments by day 20, and P female larvae had higher weights by day 30. Galleries are normally constructed within 20 days (personal observation). Our results suggest that the first 10 days are crucial and have important consequences. By constructing larger galleries, during the first 10 days, P females and their mates created optimal conditions for egg laying and larval development. Larger galleries were beneficial to the offspring because they had access to more feeding resources (i.e. phloem surface area) and more space, thus increasing developmental speed, reducing competition between larvae and enhancing survival (see also Robins & Reid 1997). These conditions are key for invasive 'r' strategy insects such as *D. valens* and may prepare larvae for fierce interfamily competition within the tree, which is typical in successful mass attacks (Faccoli 2009). Low-performing larvae imply detrimental effects in future adult size, weight, fat content, pheromone production, survival and dispersal ability (Botterweg 1982, 1983; Anderbrant et al. 1985; Anderbrant & Schlyter 1989; Faccoli 2009).

We did not determine the exact contribution of males and P females in gallery construction and parental care because these behaviours occur inside the gallery (i.e. under the bark of the host tree) and are difficult to observe. However, the observed enhanced reproductive success could be the result of a combination of factors. Chemical cues emitted by *D. valens* P females in premating interactions may bear information about their ability to tunnel and clean galleries, and to detect optimal conditions to increase egg laying during the first 10 days of gallery establishment. Preferred females produced more eggs and larvae during the first 10 days after gallery initiation compared to NP females. Nevertheless, this enhanced reproductive success could also have been influenced by a significant male contribution (i.e. gallery cleaning and tunnelling), which would allow females to invest more metabolic energy in vitellogenesis, oviposition and brood rearing. Furthermore, offspring chemical cues may play a role in regulating male or female contribution in the gallery (e.g. Kölliker et al. 2005; Mas et al. 2009).

Systems in which males are highly selective in mate choice and where this choice is mediated by chemical cues are poorly understood (Edward & Chapman 2011). Our work shows a unique case, in which red turpentine beetle males choose females using female-generated chemical cues. Remarkably, the resulting pairs have superior reproductive success. Further work is needed to determine

the exact contribution of *D. valens* males and females in gallery construction and parental care as well as possible behavioural influences of offspring chemical cues. In addition, if mate choice mediated by chemical cues is an essential component in *D. valens* sexual selection, disrupting this process may be an ideal control method for *D. valens*, which is a notorious pest in China. Whether this phenomenon is widely spread among other bark beetles with parental care, or even other taxa, remains to be answered.

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