

SHORT COMMUNICATION

Ecological correlates of daily mating frequency in a wild population of ambush bugs

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Abstract. 1. Ecological variables can exert considerable influence on the dynamics of mating systems, determining the population mean and variance in mating success.
2. Sex-specific daily estimates of mating frequency in a wild population of *Phymata americana* Melin were determined and the way in which these covaried with three ecological variables was evaluated.
3. Average daily temperature had strong influence on the mating frequency of both sexes, with evidence of a maximum at intermediate values.
4. Female mating frequency covaried with sex ratio but density did not predict mating frequency of either sex.

Key words. Mating rate, opportunity for selection, *Phymata*, temperature, temporal variation.

Introduction

Ecological factors, both biotic and abiotic, can play a crucial role in determining the dynamics of mating interactions, which in turn mediate selection on male and female phenotypes. For example, the relative abundance of prospective mates is expected to exert strong effects on mating patterns, because these determine the access to prospective mates as well as the relative costs and benefits of sexual interactions (Emlen & Oring, 1977; Thornhill & Alcock, 1983; Rowe *et al.*, 1994). The influence of such ecological factors is, therefore, the ultimate cause of selection and evolution (Wade & Kalisz, 1990; MacColl, 2011).

The ambush bug *Phymata americana* Melin is characterised by sexual colour dimorphism attributable to sexual selection (Punzalan *et al.*, 2008b), apparently due to the thermal advantages conferred by male secondary sex (dark) coloration (Punzalan *et al.*, 2008a). Previous work has shown that the strength and direction of sexual selection covary with population density and sex ratio (Punzalan *et al.*, 2010). These studies suggest the importance of these ecological variables on mating frequency, but a formal examination has been previously lacking. In the present study, we estimated population mean mating frequency of male and female *P. americana*, multiple times over a single season (generation) in the wild. We investigated the joint roles of ambient

temperature, density and sex ratio in determining the mating frequency of both sexes. Predicting the effects of multiple (sometimes correlated and potentially interacting) variables is difficult, but we illustrate an approach to disentangling the causes of natural variation in mating frequency and how this relates to selection on secondary sex traits.

Materials and methods

A population of ambush bugs *P. americana* Melin (Heteroptera: Reduviidae) was studied at the Koffler Scientific Reserve, King, Ontario, Canada (44°03'N, 79°29'W). This species is univoltine and mates multiply. On 27 days between 29 July and 26 September in 2003, we observed individuals during 2–6 h walks along 15 parallel transects (within a 2700 m² plot), noting sex and mating status. Observations were pooled across transects and based on a total of 1276 observations from 476 uniquely marked individuals (described in Punzalan *et al.*, 2010). Many individuals were observed repeatedly on the same as well as on different days. We acknowledge the non-independent nature of our observations but emphasise that our focus is on cross-sectional ‘snapshots’ of a population-level parameter (i.e. mating frequency) across varying environments, an approach advocated as having great utility for inferring agents of selection (Wade & Kalisz, 1990; MacColl, 2011). For each date, we calculated the sex-specific (population-level) daily mating frequency, q , as the number of unique individuals found to have mated over the total number of unique individuals at the site. From these data, we also

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calculated the sex ratio (males : females) and density (i.e. total number of individuals encountered, irrespective of sex). Average daily temperature data were obtained from a data logger installed on the (shaded) ground. Mating typically occurs in open, sunny areas, so the values obtained from the logger are almost certainly underestimates of the actual (e.g. ambient and body) temperatures experienced by the bugs. We excluded data for 5 days in which q was zero, probably owing to the non-reproductive (teneral) period following adult emergence as well as extremely low density at the beginning and end of season (in four cases, the density was less than five individuals of both sexes combined). In separate (by sex) evaluations of the proportional mating success, we evaluated effects of daily sex ratio, (\log_e) density and temperature using multiple linear regression, including the linear terms as well as the non-linear (quadratic and cross-product) effects. All analyses were performed in JMP v. 5.0.1a © (SAS Institute Inc., Cary, North Carolina).

Results and Discussion

Overall, the analyses suggested that the temperature, population density and sex ratio explained a large proportion of the variation in mating frequency of *P. americana* and indicated no significant interactions among them (Table 1). We found that temperature was a strong predictor of male and female mating

Table 1. Results of separate (by sex) multiple regressions of linear and quadratic effects of ecological variables on male and female mating proportional mating success, q .

| | b | SE | t | P -value |
|--------------------------------|--------|-------|-------|---------------|
| Male* | | | | |
| Intercept | 0.396 | 0.412 | 0.96 | 0.3550 |
| Density | 0.107 | 0.073 | 1.46 | 0.1694 |
| Sex ratio | 0.164 | 0.181 | 0.91 | 0.3823 |
| Mean temperature | -0.031 | 0.011 | -2.77 | 0.0171 |
| Density ² | 0.097 | 0.093 | 1.05 | 0.3158 |
| Density \times sex ratio | 0.040 | 0.666 | 0.06 | 0.9526 |
| Density \times temperature | -0.022 | 0.026 | -0.87 | 0.3989 |
| Sex ratio ² | 0.087 | 1.310 | 0.07 | 0.9483 |
| Sex ratio \times temperature | 0.098 | 0.111 | 0.89 | 0.3931 |
| Mean temperature ² | -0.011 | 0.006 | -1.81 | 0.0949 |
| Female* | | | | |
| Intercept | 0.136 | 0.267 | 0.51 | 0.6188 |
| Density | 0.072 | 0.047 | 1.53 | 0.1516 |
| Sex ratio | 0.491 | 0.117 | 4.18 | 0.0013 |
| Mean temperature | -0.026 | 0.007 | -3.63 | 0.0035 |
| Density ² | -0.021 | 0.060 | -0.36 | 0.7285 |
| Density \times sex ratio | 0.738 | 0.432 | 1.71 | 0.1130 |
| Density \times temperature | -0.012 | 0.017 | -0.70 | 0.4960 |
| Sex ratio ² | -0.798 | 0.849 | -0.94 | 0.3657 |
| Sex ratio \times temperature | 0.040 | 0.072 | 0.56 | 0.5858 |
| Mean temperature ² | -0.012 | 0.004 | -2.93 | 0.0126 |

SE, standard error of the partial regression coefficient, b . P -values in bold indicate significance at $\alpha = 0.10$.

*Whole model fit – male: $F_{9,12} = 4.65$, $P = 0.0079$, multiple $R^2 = 0.78$; female: $F_{9,12} = 6.2$, $P < 0.0001$, multiple $R^2 = 0.92$.

frequencies. Evidence of such temperature effects on the mating rates of ectotherms is widespread and, in *P. americana*, temperature is believed to be particularly important because dark male coloration appears to increase body temperature and enhance male mate search performance (Punzalan *et al.*, 2008a). Although Punzalan *et al.* (2010) did not observe any correlation between the strength of (phenotypic) sexual selection and temperature, their sample size was relatively small ($n = 10$), especially for detecting non-linear relationships. The larger sample size in the present study afforded improved resolution. We found a significant negative linear relationship (i.e. partial regressions) between temperature and q in both sexes, as well as a significant negative quadratic relationship for females (and a quadratic trend for male mating frequency), whereby the highest q was observed at intermediate temperatures, declining sharply with temperatures over 17 °C (Table 1, Fig. 1). This suggests that, for both sexes, there is an optimal temperature range beyond which dark colour is detrimental to mating success. This might explain episodes of negative selection on (i.e. disfavoured) dark male coloration reported in the wild (Punzalan *et al.*, 2010), and a trend towards negative selection under warmer ambient conditions in an experimental setting (Punzalan *et al.*, 2008a). The mechanistic causes are unclear but excessively high temperature is known to hinder various aspects of insect physiology and fitness (e.g. Hoffmann *et al.*, 2003; Sørensen *et al.*, 2003).

Based on the partial regression coefficients (Table 1), density did not predict mating frequency of either sex. For females, but not males, q declined as a function of sex ratio. No quadratic effects were detected for either variable in either sex (Fig. 1). The importance of demographic variables for mating success is generally established in insects (e.g. Thornhill & Alcock, 1983; Rowe *et al.*, 1994) and, with respect to *P. americana*, previous work showed that the strength of sexual selection on male colour covaried positively with density and negatively with sex ratio (Punzalan *et al.*, 2010). Our findings suggest that fluctuating selection on male secondary sex coloration is tied to the effects of sex ratio on female mating frequency. Reanalysis of data in Punzalan *et al.* (2010) showed a negative but non-significant relationship between sexual selection on male (lateral) coloration and female q ($r = -0.412$, $P = 0.4448$, $n = 12$). This highlights the non-independent nature of sexual reproduction and its complications for disentangling the causes of variation in mating patterns. Mating rate is a trait shared between the sexes and the outcome of both inter- and intra-sexual interactions. All else being equal, ecological conditions that promote/inhibit the mean and variance in the mating rate of one sex will often have similar effects on the other sex, for both deterministic and stochastic reasons (Sutherland, 1985; Downhower *et al.*, 1987; Shuster & Wade, 2003; Jennions *et al.*, 2012). The prominence of demographic factors differed between the present study and the results of Punzalan *et al.* (2010), possibly reflecting the influence of additional, unmeasured factors, which are always a concern with correlational approaches (e.g. Lande & Arnold, 1983; Mitchell-Olds & Shaw, 1987).

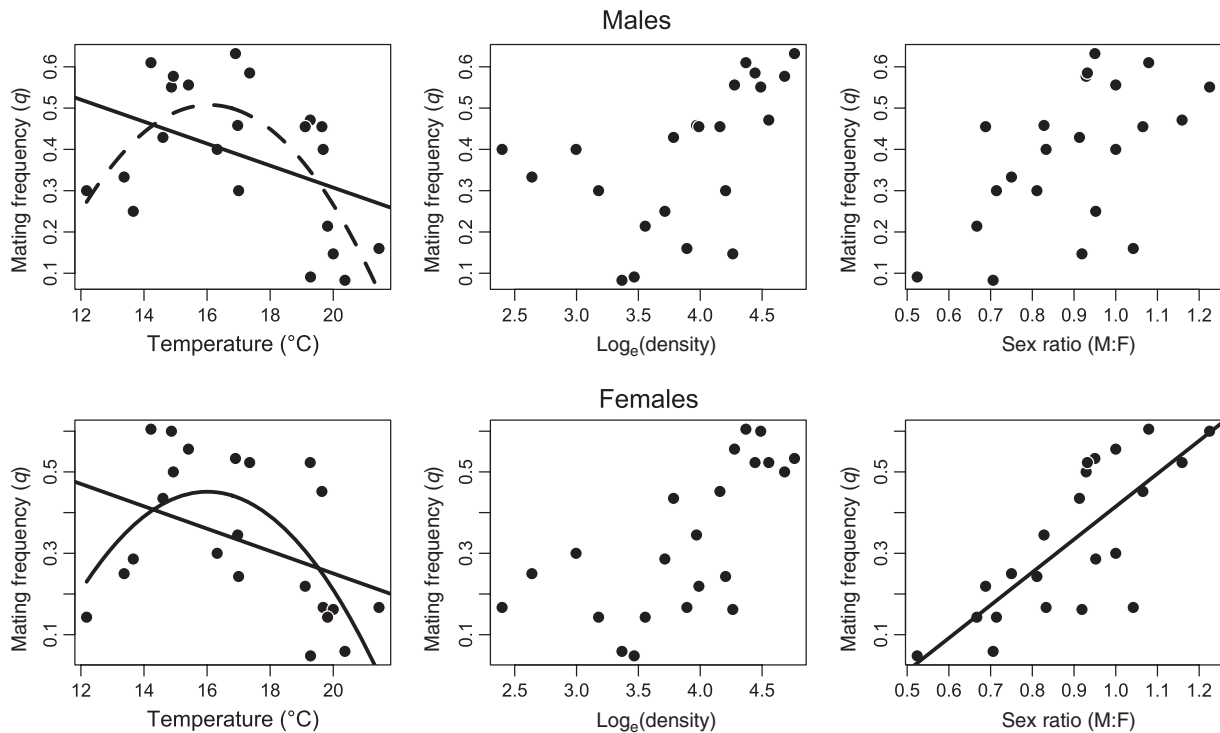


Fig. 1. Ecological correlates of daily estimates of mean mating frequency (i.e. proportion of individuals mating, q). Bivariate plots depict male (upper panels) and female (lower panels) mean mating success (y-axis) against the three measured ecological variables (x-axis). For simplicity, linear and quadratic ordinary least-squares regression lines are shown to illustrate the instances where partial regression coefficients from multiple regression were statistically significant at $\alpha = 0.05$ (solid lines) or $\alpha = 0.10$ (dashed line).

We should point out that mating frequency, q , is also closely tied to the evolutionary parameter I , or the opportunity for selection. I defines the upper limit for the strength of phenotypic selection and evolutionary response (Crow, 1958; Wade, 1979; Arnold & Wade, 1984). When dealing with binary measures of fitness (e.g. mated versus unmated) in cross-sectional samples, q is equivalent to mean absolute fitness (mating success) and $I = (1 - q)/q$ (Brodie & Janzen, 1996). Thus, ecologically driven fluctuations in q (and concomitant inflation/contraction of I) might contribute to fluctuating sexual selection observed for this population (Punzalan *et al.*, 2008b, 2010).

Our repeated cross-sectional measures of q illustrate the potential impact of ecological variables on mating systems and how these may relate to observed patterns of phenotypic selection. Collecting adequate data to identify ecological determinants of spatiotemporal variation in selection is often logistically demanding. However, the present study demonstrates that even coarse, but more readily obtained, measures (e.g. mean and variance in mating frequency) can provide a complementary means for identifying potential agents of sexual selection when repeated across a varying environment. Such an approach provided results that partially mirror previous observations of demography-mediated selection as well as new insights into the role of temperature in determining mating success in *P. americana*.

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REFERENCES

- Arnold, S.J. & Wade, M.J. (1984) On the measurement of natural and sexual selection: theory. *Evolution*, **38**, 709–719.
- Brodie, E.B. III & Janzen, F.J. (1996) On the assignment of fitness values in statistical analyses of selection. *Evolution*, **50**, 437–442.
- Crow, J.F. (1958) Some possibilities for measuring selection intensities in man. *Human Biology*, **30**, 1–13.
- Downhower, J.F., Blumer, L.S. & Brown, L. (1987) Opportunity for selection: an appropriate measure for evaluating variation in the potential for selection? *Evolution*, **41**, 1395–1400.
- Emlen, S.T. & Oring, L.W. (1977) Ecology, sexual selection, and the evolution of mating systems. *Science*, **197**, 215–222.
- Hoffmann, A.A., Sørensen, J.G. & Loeschke, V. (2003) Adaptation of *Drosophila* to temperature extremes: bringing together quantitative and molecular approaches. *Journal of Thermal Biology*, **28**, 175–216.
- Jennions, M.D., Kokko, H. & Klug, H. (2012) The opportunity to be misled in studies of sexual selection. *Journal of Evolutionary Biology*, **25**, 591–598.

- Lande, R. & Arnold, S.J. (1983) The measurement of selection on correlated characters. *Evolution*, **37**, 1210–1226.
- MacColl, A.D.C. (2011) The ecological causes of evolution. *Trends in Ecology & Evolution*, **26**, 514–522.
- Mitchell-Olds, T. & Shaw, R.G. (1987) Regression analysis of natural selection: statistical inference and biological interpretation. *Evolution*, **41**, 1149–1161.
- Punzalan, D., Rodd, F.H. & Rowe, L. (2008a) Sexual selection mediated by the thermoregulatory effects of male color pattern in the ambush bug *Phymata americana*. *Proceedings of the Royal Society of London Series B*, **275**, 483–492.
- Punzalan, D., Rodd, F.H. & Rowe, L. (2008b) Sexual selection on sexually dimorphic traits in the ambush bug *Phymata americana*. *Behavioral Ecology*, **19**, 860–870.
- Punzalan, D., Rodd, F.H. & Rowe, L. (2010) Temporal variation in patterns of multivariate sexual selection in a wild insect population. *American Naturalist*, **175**, 401–414.
- Rowe, L., Arnqvist, G., Sih, A. & Krupa, J.J. (1994) Sexual conflict and the evolutionary ecology of mating patterns: water striders as a model system. *Trends in Ecology & Evolution*, **8**, 289–293.
- Shuster, S.M. & Wade, M.J. (2003) *Mating Systems and Strategies*. Princeton University Press, Princeton, New Jersey.
- Sørensen, J.G., Kristensen, T.N. & Loeschke, V. (2003) The evolutionary and ecological role of heat shock proteins. *Ecology Letters*, **6**, 1025–1037.
- Sutherland, W.J. (1985) Chance can produce a sex difference in mating success and explain Bateman's data. *Animal Behaviour*, **33**, 1349–1352.
- Thornhill, R. & Alcock, J. (1983) *The Evolution of Insect Mating Systems*. Harvard University Press, Cambridge, Massachusetts.
- Wade, M.J. (1979) Sexual selection and variance in mating success. *American Naturalist*, **114**, 742–747.
- Wade, M.J. & Kalisz, S. (1990) The causes of natural selection. *Evolution*, **44**, 1947–1955.

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