ANIMAL BEHAVIOUR, 2007, **74**, 143–152
doi:10.1016/j.anbehav.2006.12.004

Male postcopulatory reproductive success in the beetle, *Diaprepes abbreviatus*

LAURA K. SIROT*†, H. JANE BROCKMANN* & STEPHEN L. LAPOINTE‡
*Department of Zoology, University of Florida
†Department of Molecular Biology and Genetics, Cornell University
‡United States Department of Agriculture, Agricultural Research Services, United States Horticultural Research Laboratory

*(Received 28 April 2006; initial acceptance 8 June 2006;
final acceptance 4 December 2006; published online 20 June 2007; MS. number: A10433R)*

For most species that have been studied, considerable variation exists in male postcopulatory reproductive success, including female sperm use, reproductive investment and remating patterns. We investigated postcopulatory sexual selection in the beetle *Diaprepes abbreviatus* (L.) (Coleoptera: Curculionidae) by examining the correlation of two measures of a male's success (proportion of eggs fertilized (P_2) and number of eggs that a female laid after mating with him) with male body size, copulatory behaviour and copula duration. We allowed each female to mate with an irradiated sterile male and a normal male. Relative duration of intromission (i.e. the difference in the intromission durations of the two males) and relative stroking rate, but not the absolute duration and stroking rate, were positively correlated with P_2. The number of eggs that a female laid was negatively related to the thrusting rate of her mate. Irradiated males mated for a shorter duration and thrusted at a lower rate than normal males. Irradiated males also fertilized a lower proportion of eggs than normal males, and females were more likely to remate after mating with an irradiated male than after mating with a normal male. These findings suggest that behavioural traits may influence male postcopulatory reproductive success. Furthermore, since variation in male stroking rate and intromission duration are associated with variation in male fertilization success, these traits may have evolved through postcopulatory sexual selection.

Keywords: Coleoptera; copulatory courtship; *Diaprepes abbreviatus*; postcopulatory reproductive success; root weevil; sexual selection

Competition among males for reproductive success does not end with the onset of copulation. Rather, in many species, there are processes that occur within the female during and after copulation (postcopulatory) that influence the quantity and quality of offspring sired by particular males (Parker 1970; Eberhard 1996). In most species in which male postcopulatory reproductive success (MPCRS) has been measured, substantial unexplained variation occurs both in the likelihood that a particular male will fertilize his mate's eggs (Birkhead & Møller 1998) and in the reproductive investment by the female after mating with a particular male (e.g. differential allocation; Arnqvist & Danielsson 1999a; Sheldon 2000). In this study, we investigated the correlation of MPCRS with male body size, copulatory behaviour and copula duration in the beetle *Diaprepes abbreviatus* (L.) (Coleoptera: Curculionidae).

In insects, the best-documented pattern of MPCRS is that, on average, the first or last male to mate fertilizes the majority of eggs (Simmons 2001a). However, in many cases, mating order actually explains very little variation in sperm use patterns, with male fertilization success ranging from 0 to 100% for both first and last males to mate (Lewis & Austad 1990; Simmons 2001a). Investigations into other correlates of sperm use patterns have resulted in important insights regarding the strength and direction

Correspondence: L. Sirot, Department of Molecular Biology and Genetics, 421 Biotechnology Building, Cornell University, Ithaca, NY 14853, U.S.A. (email: ls286@cornell.edu). H. J. Brockmann is at the Department of Zoology, 223 Bartram Hall, University of Florida, Gainesville, FL 32611, U.S.A. S. Lapointe is at the USDA, ARS, USHRL, 2001 South Rock Road, Ft Pierce, FL 34945, U.S.A.

0003–3472/07/$30.00/0

of sexual selection on male traits (Danielsson 2001) and the mechanisms underlying sperm transfer, storage and use (Simmons & Parker 1992; Otronen 1997a, b; Andrés & Cordero 2000; Schäfer & Uhl 2002; Tallamy et al. 2002; Bloch Qazi 2003). Yet, for most species of insects, the study of MPCRS has not gone beyond the effect of mating order on sperm use patterns. There is a need for a more comprehensive understanding of the causes of variation in MPCRS both to determine whether the currently known patterns can be generalized and to gain a better understanding of mechanisms of postcopulatory sexual selection, circumstances under which it occurs, and how it affects the evolution of traits.

Like many insect species, *D. abbreviatus* is characterized by (1) promiscuous mating, (2) female storage of sperm from multiple males and (3) substantial variation in MPCRS (Harari et al. 2003). *Diaprepes abbreviatus* provides an appropriate system in which to investigate correlates of MPCRS because body size, copula duration and male copulatory behaviour vary greatly within populations (Sirot 2004). In this study, we investigated influences on MPCRS by studying the correlation of two measures of a male's success (the proportion of eggs that he fertilized and the number of eggs that a female laid after mating with him) with male body size, copulatory behaviour and copula duration. Based on the results of previous studies of postcopulatory sexual selection, we made the following predictions.

(1) Male body size should be positively correlated with MPCRS. This prediction is based on previous studies that have shown that a female's sons and daughters will benefit from larger size through increased competitive ability and increased fecundity (Harari et al. 1999). Male size also may be positively related to MPCRS if larger males contribute more to egg production than smaller males. Although male contribution to female fecundity has not been demonstrated in *D. abbreviatus*, male-derived molecules are found in the ovaries and eggs of mated females (Sirot et al. 2006).

(2) Duration of intromission (Table 1) should be positively related to MPCRS since it may (1) be related to the number of sperm that a male transfers or to the number of rival sperm that a male removes (Simmons 2001a) and/or (2) indicate male competitive ability and endurance.

(3) Male stroking rate (Table 1) during copulation should be positively related to MPCRS, as it may be an indicator of male endurance (Watson & Lighton 1994; Kotiaho et al. 2001; Tallamy et al. 2002). In other insect species, the rate of similar behavioural patterns is positively related to male fertilization success (Edvardsson & Arnqvist 2000; Tallamy et al. 2002; Bloch Qazi 2003).

(4) The number or rate of genitalic thrusts (Table 1) by males should be positively related to male fertilization success because thrusting could (1) be a method of sperm transfer or sperm positioning within the female reproductive tract (Eberhard 1993) or (2) stimulate the female to (a) release stored sperm from previous matings (Córdoba-Aguilar 1999) or (b) relax muscles that prevent full penetration by and expansion of the male genitalia (Eberhard 1993; Eberhard & Kariko 1996; Tallamy et al. 2002).

Taken together, our study provides evidence consistent with postcopulatory sexual selection acting on male behavioural traits in *D. abbreviatus* and provides insights into the mechanisms underlying variation in MPCRS.

METHODS

Study Subjects

Diaprepes abbreviatus is a weevil found in Florida, Texas and California, U.S.A. (accidentally introduced into these areas), and much of the Caribbean (native to this region), where it feeds on a variety of plants including citrus. Females used in our experiment were reared in 30-ml plastic cups containing an artificial insect diet (product no. F1675, Bio-Serv, Inc., Frenchtown, New Jersey, U.S.A.) mixed with water, agar and preservatives (one female per cup; 26°C, >95% RH; Lapointe & Shapiro 1999; Lapointe 2000). After adult emergence, we maintained females in all-female cages (45 × 45 × 45 cm; wire mesh; 30 females per cage) and provisioned them with new citrus leaves and oviposition substrate every 3–4 days. Females were 2–4 weeks postemergence at the time of the experiment and had not mated. Females had a narrow range of body size (coefficient of variation = 5%), which minimized the effect of female size on the outcome of the experiment. Males were collected in the wild from a plant nursery in Homestead, Florida (Dade County) 2–5 days before the beginning of the experiment and were maintained in all-male cages under the same conditions as females. Wild males were used because their body sizes are more variable than laboratory-reared males. The length of the right elytron (Table 1) of males used in our study ranged between 7.4 and 11.9 mm ($\overline{X} \pm \mathrm{SE} = 9.5 \pm 0.1$; $N = 150$). We used each male for only one mating in the experiment.

Experimental Design

We used males sterilized by gamma radiation to measure paternity (Boorman & Parker 1976). We allowed virgin laboratory-reared females to mate with two males on consecutive days; one of her mates was sterilized. We assigned females to either the IN (Irradiated male first, Normal male second) or NI (Normal male first, Irradiated male second) treatment. This allowed us to determine the paternity of eggs laid after the two matings. We then tested for correlations of the two measures of MPCRS (proportion of eggs fertilized by the second male to mate or P_2 and the number of eggs females laid after mating) with body size, copulatory behaviour and copula duration (Table 1). The experiment was conducted in June and July 2003 at the U.S. Horticultural Research Laboratory (USHRL), Ft Pierce, Florida (FL).

The day before the experiment began (Rugman-Jones & Eady 2001), we exposed a subset of males to 10 kR of irradiation (dosed intermittently: 1 min of irradiation at 1020 R/min and 4 min without irradiation; Cesium-137 source: Florida Department of Agriculture and Consumer Services, Gainesville, FL). Matings were conducted in an