MATING BEHAVIOUR OF GREYBACK AND GRATA CANE BEETLES, *DERMOLEPIDA ALBOHIRTUM* (WATERHOUSE) AND *LEPIDIOTA GRATA* BLACKBURN (COLEOPTERA: SCARABAEIDAE)

D.P. LOGAN[1] and M.S. SALLAM[2]

[1]*Bureau of Sugar Experiment Stations, PO Box 117, Ayr, Qld 4807*

(Current address: HortResearch, 412 No. 1 Road, RD2, Te Puke, New Zealand)

[2]*Bureau of Sugar Experiment Stations, PO Box 566, Tully, Qld 4854*

Abstract

Mating behaviour of greyback cane beetle, *Dermolepida albohirtum* (Waterhouse) and grata cane beetle, *Lepidiota grata* Blackburn, is described and compared. Mating pairs of *D. albohirtum* were observed within feeding aggregations in *Ficus opposita* and *Acacia* spp. during 4 hours from late afternoon to early evening. Copulation by *D. albohirtum* lasted on average 12 minutes and was preceded by the male resting piggy-back on the female and vibrating her body. Mating by *L. grata* occurred during a 30 minute period in trees at dusk. Females of *L. grata* called by repeatedly extending and retracting their ovipositor. Males appeared shortly after, flying upwind. Copulation followed immediately a male mounted a female, and lasted on average 18 minutes. Any sex pheromone produced by *D. albohirtum* may be active over a distance of less than 1 m, and may not be useful for monitoring numbers. By comparison, the mating behaviour of *L. grata* is consistent with a sex pheromone that acts over longer distances.

Introduction

Greyback canegrub, *Dermolepida albohirtum* (Waterhouse), is the most important insect pest of sugarcane in northern Queensland from Mossman (16°21'S, 145°15'E) to Sarina (21°22'S, 149°13'E). Since the mid-1990s, it has been in outbreak numbers in the Burdekin region, centred on Ayr (19°34'S, 147°24'E). It has an annual lifecycle, spent mainly as larvae in free-draining soils. In the Burdekin, beetles can be observed in favoured feeding trees (*Ficus* spp., *Corymbia tesselaris*, *Acacia* spp. and palms) between October and January. Beetles fly and feed during the evening and night and some beetles are also present during the day, resting in feeding trees.

Grata canegrub, *Lepidiota grata* Blackburn, is an occasional pest of sugarcane in northern and central Queensland from Ingham (18°43'S, 146°10'E) to Gin Gin (25°00'S, 151°58'E). Its life cycle is 1 or 2 years depending on field conditions (Chandler and Chapman 1989). Each year in the Burdekin, adults fly and feed in eucalypts, particularly *Eucalyptus tereticornis*, during the evening and night during late spring and summer.

Many scarabs use pheromones to facilitate mate location (Leal 1998). Allsopp (1993) showed that unmated females of the canegrubs *Antitrogus consanguineus* (Blackburn), *A. parvulus* Britton and *Lepidiota picticollis* Lea probably use pheromones to attract males. The identification and isolation of pheromones may provide the basis for monitoring and management tools for cane beetles. Collection, isolation and field-testing of putative pheromones of cane beetles are aided by an understanding of their mating behaviour. Knowledge of the mating behaviour of *D. albohirtum* and

L. grata is limited. Mating by *D. albohirtum* has been observed during a 1 hour period in the evening in trees up to 5-7 m above ground (Illingworth and Dodd 1921, Jarvis 1933). Illingworth and Dodd (1921) observed swarming by *L. grata* at dusk followed by mating on vegetation from near ground level up to 10 m above ground and copulation for one pair of *L. grata* took 35 minutes. Here we describe in more detail the mating behaviour of *L. grata* and *D. albohirtum*.

Materials and Methods

The daily pattern of mating for *D. albohirtum* was determined by counting mating pairs at four sites in the Burdekin for observation periods of 1.5 to 23.5 hours (Table 1). At site 1, pairs were counted at 15 minute intervals. A thunderstorm prematurely ended counting and beetles were collected to estimate the proportion of mated and non-mated beetles and males and females in the feeding aggregation. A count of all beetles was made at sites 2-4 during the evening. Beetles were collected at site 2 on the day following the observation of mating and were sexed. Two-by-two contingency tables were used to test whether the sex ratio of beetles in the aggregation was different to unity at sites 1 and 2. At site 4, the number of males resting on the back of females (piggy-back pairs), a stage preceding copulation, was counted every 30 minutes.

Table 1. Site description and period of observation for feeding aggregations of *D. albohirtum* in which mating was observed in our study. The proportion of *D. albohirtum* observed mating was based on an estimate of the number of beetles at each site after no further mating was observed.

Site	Site description	Date and period of observation	Pairs	Proportion of beetles mating in aggregation
1	Outer row of cane field, Burdekin Sugar Experiment Station	19 Nov 1999 1700 – 1830	32	0.06
2	1 tree (*Ficus opposita*), Burdekin River bank	17-18 Nov 2000 1515 – 0715	41	0.04
3	6 trees (1 *Acacia cincinnata*, 4 *A. mangium*, 1 *F. opposita*), Ayr golf course	1-2 Dec 2000 1500 – 0130	40	0.08
4	7 trees (*F. opposita*), Haughton River bank	8-9 Dec 2000 0900 – 0830	29	0.06

As adult *L. grata* are crepuscular and nocturnal, mating pairs and individuals were counted during the late afternoon and evening. Pairs and individuals were counted on a group of three small (< 3m tall) trees (*Eucalyptus tereticornis*, *Acacia cincinnata* and *Nauclea orientalis*) on the Ayr golf course from 1830 to 2100 hours on 22 January 2001. Counts were made for five consecutive 15 minute intervals and then after 30 minutes and one hour. Other occasional observations of *L. grata* behaviour were made between 2100 and 0100 hours and later in the morning from 0700 to 0800 hours.

Females of both species (n = 35) were collected in copula, frozen and dissected to determine ovarian status as an approximate estimate of age. Females of *D. albohirtum* emerge with no ovarian development and must feed for one to two weeks before oocytes are fully grown (D. Logan, unpubl. obs.). Ovarian status at emergence by *L. grata* is not known. Development of ovaries was scored as either with no fully grown oocytes or with some or all oocytes fully grown. As feeding may be necessary before females can produce pheromones, the hindgut of females was examined for evidence of ingested plant matter.

Duration of copulation was determined with stop-watches for 15 pairs of *D. albohirtum* and for 18 pairs of *L. grata*. Copulation times for *D. albohirtum* and *L. grata* was compared by two-sample t test at $P < 0.05$. Pre-copulatory behaviour was timed for seven pairs of *D. albohirtum*.

Results and Discussion

Mating behaviour of D. albohirtum

Mating by *D. albohirtum* occurred during 4 hours from the late afternoon to early evening (Fig. 1), much longer than the 1 hour period found by Illingworth and Dodd (1921). No swarming by males was observed. Instead, mating by *D. albohirtum* occurred amongst aggregations of feeding beetles. Mating pairs were observed hanging from leaves or branches from < 0.5 m to 4 m above the ground. We observed pre-copulatory behaviour that involved a male resting piggy-back on a female for up to two hours (Fig. 2a) and often vibrating his body and hers. Vibration occurred without leverage from adjacent branches or leaves. Between and during vibration events, the male was often perched forward on the female with antennal club segments fanned and may have been palpating the female's head. The male extended his aedeagus (Fig 2b) before successful intromission took place (Fig. 2c). The male remained in this position (Fig. 2c) relatively briefly (mean time in minutes $\pm$ SD = 2.51 $\pm$ 0.79, n = 7, range = 1.66 – 3.90) prior to leaning back and hanging upside down (Fig. 2d) often held only by the aedeagus. On average, males remained hanging upside down for 12.04 minutes (SD = 3.29, n = 15, range = 6.03 – 16.53). Some unsuccessful pairings were observed in which the piggy-back male vibrated the female and extended and retracted his aedeagus one or more times, but never achieved intromission. At site 4, the number of piggy-back pairs increased in the afternoon prior to the formation of mating pairs (Fig. 2d). Some pairs remained as piggy-back pairs for at least two hours before mating or moving apart. One piggy-back pair at site 4 mated at 1730 hours after being first observed at 1100 hours and marked with liquid paper. Within aggregations of *D. albohirtum*, there were few occasions when any movement to form pairs was seen. This was generally a flight of less than 1 m by a male to a female. Thus, any sex pheromones released by females of *D. albohirtum* may be most active over short distances.

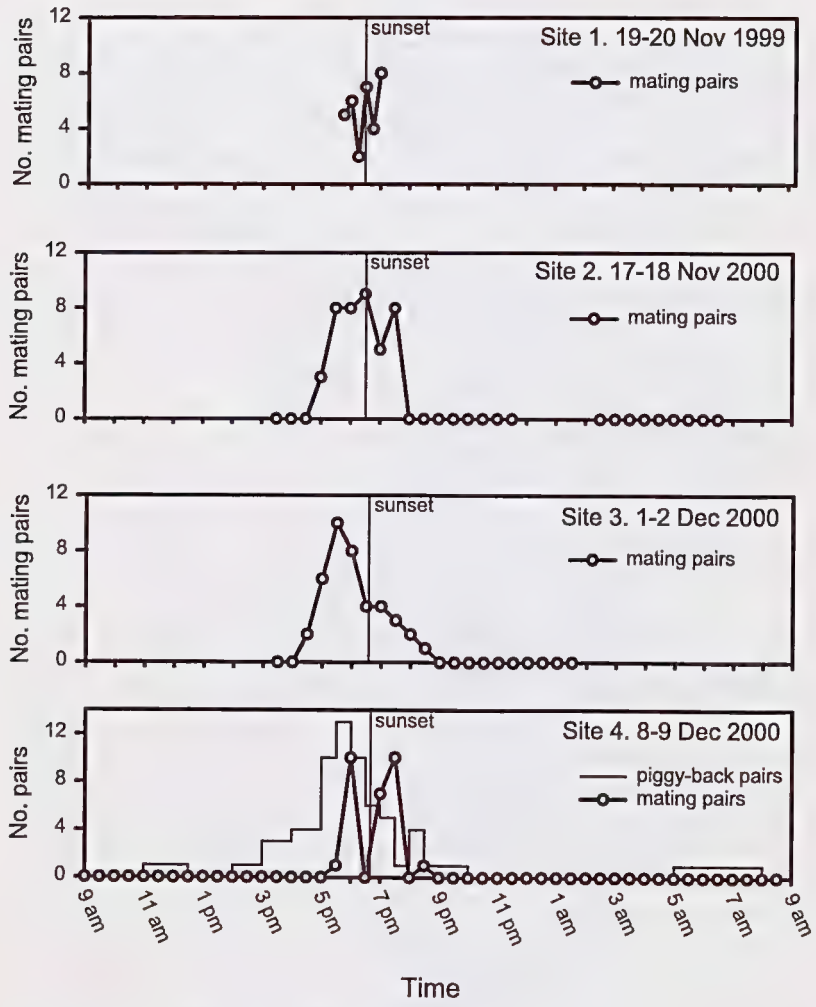

Fig. 1. Time of mating for *D. albohirtum* at four sites in the Burdekin.

Most females (80%) collected during mating had food in their hindguts, consistent with being part of a feeding aggregation. Most mated females (66%) had no fully developed oocytes indicating they may have recently emerged. The sex ratio of beetles in the aggregation at site 1 was unity (two by two contingency test, $\chi^2 = 0.11$, P = 0.74, n = 166) and at site 2 was female-biased ($\chi^2 = 4.87$, P = 0.03, n = 159, 62% females). A very small proportion of beetles in feeding aggregations mated at each site on any one night (Table 1).

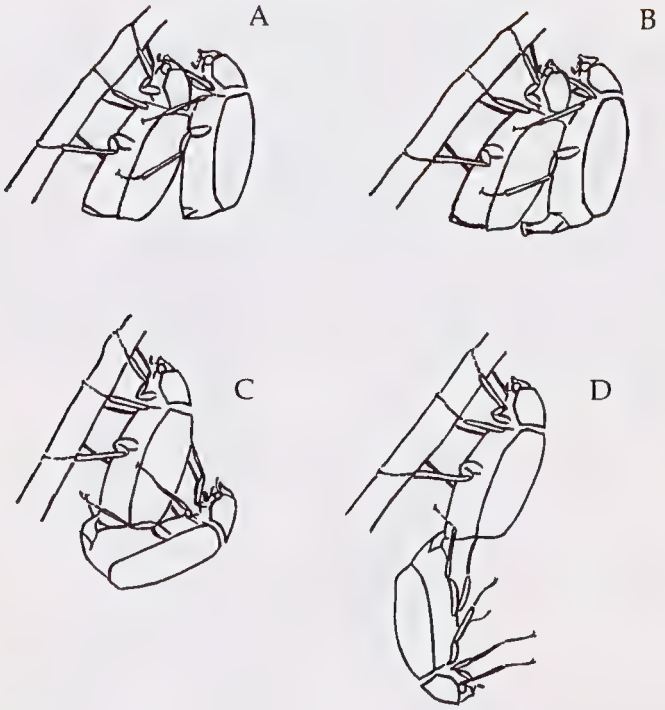

Fig. 2. Sequence of mating behaviour for *D. albohirtum*. (A) Male *D. albohirtum* crawls onto the back of female and vibrates female; (B) Male extends aedeagus and (C) intromits; (D) After several minutes, male hangs backwards and remains in this position for 6-16 minutes.

Mating behaviour of L. grata

Mate location by male *L. grata* was consistent with the release of pheromones by females. Beetles of *L. grata* appeared in flight shortly after sunset. Females landed usually at least 1 m above ground level on tree trunks, branches and leaves of a variety of trees and shrubs including *Eucalyptus* spp., *Acacia* spp., and *Melaleuca* spp. Females began 'calling' immediately by repeatedly extending and retracting their ovipositor (Fig. 3). One or more males often arrived within a minute, flying upwind, landed on the female or near her and crawled onto her back. The male immediately inserted his aedeagus and fell back to hang upside down, as with *D. albohirtum* (Fig. 2d). There was no pre-copulatory behaviour as with *D. albohirtum*. After insertion of the aedeagus, other males on and near the mating pair or circling nearby

quickly dispersed. Mating took on average 18.08 minutes (SD = 3.19, n = 18, range = 9.82 – 25.37), significantly longer than for *D. albohirtum* (T = 5.35, df = 31, P < 0.0001), but shorter than the 35 minutes recorded for one pair by Illingworth and Dodd (1921). If the male was not immediately successful in mating, other males attempted to copulate with either the female or the initial male, if he remained on her back. Many females called from the same tree or shrub and a swarm of males was present during the 10 to 15 minute period when mating pairs formed. No pairs remained 30 minutes after swarming started (Fig. 4). Males often flew away immediately, but females tended to remain longer. Mating pairs were only observed during 30 minutes at dusk (Fig. 4). After mating, beetles were seen feeding on trees (especially *Eucalyptus tereticornis*) (Fig. 4) until at least 0100 hours. No beetles were present after 0700 hours, probably having returned to the soil where they spend the day.

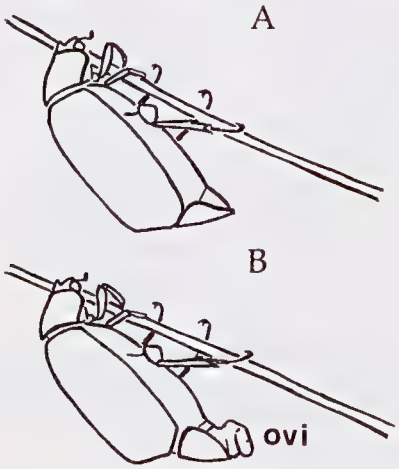

Fig. 3. Behaviour of female *L. grata* immediately prior to mating. (A) Female lands on branch and (B) starts to repeatedly extrude and retract her ovipositor (ovi).

In contrast to mated females of *D. albohirtum*, most females (91%) of *L. grata* collected while mating, had not fed. Most females (77%) had some or all of their oocytes fully grown. Feeding may not be necessary to develop oocytes or for the production of pheromones in *L. grata*. Females of *L. negatoria* Blackburn emerging from the soil for the first time have some of their oocytes fully grown and often mate on the same night (Logan 2000). Females of *L. grata* may have a similar behaviour, mating on their first night of emergence.

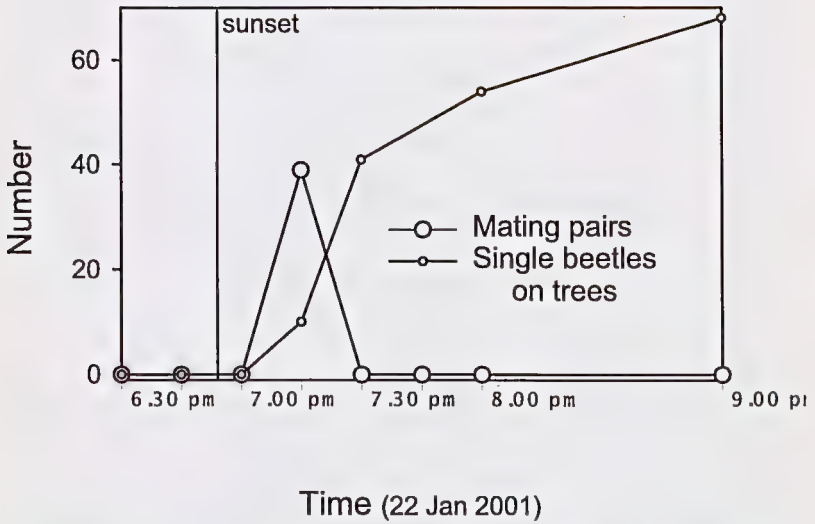

Fig. 4. Time of mating for *L. grata* and number of beetles feeding in a group of three trees at one site in the Burdekin.

Comparison of mating behaviour

Mating behaviour of *L. grata* is similar to that of the cane beetles *Lepidiota* spp. and *A. parvulus* (Illingworth and Dodd 1921, Logan 2000, K. Chandler, pers. comm.). Males of *Lepidiota* spp. and *A. parvulus* swarm around a female prior to mating and are probably attracted by a pheromone (Allsopp 1993). Swarming and mating occurs during 30 minutes at dusk.

Mating behaviour in *D. albohirtum* is unusual in comparison with other cane beetles. Copulation is more rapid than for *L. grata*, *L. negatoria* (about 20 minutes: Logan 2000) and *L. frenchi* Blackburn (20-29 minutes: Illingworth and Dodd 1921). Mating may occur sometime during a four hour period from the late afternoon to early evening, rather than during the brief period following the evening emergence of other cane beetles. Many females of *D. albohirtum* have fed prior to mating; as far as is known, other cane beetles mate before feeding (Logan 2000). The tendency for *D. albohirtum* to form piggy-back pairs is unknown for other cane beetles. However, piggy-back pairs occur in feeding aggregations of the rutelines *Anoplognathus porosus* (Dalman), *A. boisduvalii* Boisduval and *Popillia japonica* Newman, which remain as pairs for up to two hours after copulation (Barrows and Gordh 1978). Mounting in *D. albohirtum* precedes mating and may be prompted by a sex pheromone released by females.

Aggregation in feeding trees by *D. albohirtum* may be due to plant volatiles released from leaves during feeding. The forest cockchafer *Melolontha hippocastani* F. was attracted to mechanically damaged leaves of two favoured feeding trees and a non-host tree and a synthetic mix of green leaf volatiles (Ruther *et al.* 2000). Male *M. hippocastani* were also attracted to extracts washed from unmated females. Ruther *et al.* (2000) concluded that males used volatiles released from leaves damaged during feeding as well as volatiles released from females in order to facilitate mate location. Males of *D. albohirtum* may use a similar mechanism to locate females. Sex pheromones or other sex-specific volatiles from females may have a limited range and thus are unlikely to be useful for monitoring numbers of *D. albohirtum*. By comparison, females of *L. grata* release a pheromone that is active over a relatively wide area and may be useful for monitoring or trapping beetles.

Acknowledgements

Staff at the Queensland Herbarium identified the species of *Acacia* at the Ayr golf course on which *D. albohirtum* were feeding and mating. Sunset times for Ayr were from the website of the Australian Surveying and Land information group, Department of Industry, Science and Resources (www.auslig.gov.au). Peter Allsopp made some useful comments on the draft manuscript.

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