Prionic Acid: An Effective Sex Attractant for an Important Pest of Sugarcane, *Dorysthenes granulosus* (Coleoptera: Cerambycidae: Prioninae)

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Abstract

Male *Dorysthenes granulosus* (Thomson, 1860) (Coleoptera: Cerambycidae: Prioninae) were caught in traps baited with racemic 3,5-dimethyldodecanoic acid (prionic acid) during field screening trials in China that tested known cerambycid pheromones. This species is an important pest of sugarcane (*Saccharum officinarum* L.). In follow-up dose-response trials, plastic sachets loaded with 1 or 0.1 mg of prionic acid were equally attractive to male beetles, whereas lower doses were no better than controls. Two commercial prionic acid lures also were attractive, suggesting that traps baited with prionic acid can be rapidly incorporated into integrated pest management programs targeting this major pest. It is likely that this compound is a major component of the female-produced sex pheromone of *D. granulosus* because this species is in the same subfamily as *Prionus californicus* Motschulsky, 1845, the species from which prionic acid was originally identified.

Key words: longhorned beetle, sugarcane borer, pheromone, pest management

The cerambycid beetle, *Dorysthenes granulosus* (Thomson, 1860) (Prioninae, Prionini), is distributed throughout southern China, India, Myanmar, and Thailand (Drumont and Komiya 2010), where it is an important pest of sugarcane (Yu et al. 2006). Damage is caused by the larvae, which feed and develop in the sugarcane roots. In recent decades, this species has spread rapidly in the Chinese provinces of Hainan, Guangxi, Guangdong, and Guizhou, where sugarcane is now grown on ~1.5 million hectares (Gong et al. 2008). In these regions, sugarcane is cultivated using the ratoon method, in which only the upper part of the plant is harvested, leaving the stump and roots to resprout for several seasons before replanting. The beetle larvae take 2 yr to complete their life cycle, so ratooning strongly favors their development. The nonfeeding adults live for ~3 wk (Zeng and Huang 1981) and begin emerging in April in Hainan Province (Wu et al. 2013), and as late as June in Guangxi Province and other more northern parts of its range (Yu et al. 2006). Current control measures include intensive cultural control (plowing, harrowing, and furrowing before planting), manual collection of adults, sanitation (complete removal of plants at harvest; Gong et al. 2008), biological control with entomopathogenic fungi (Yu et al. 2010), and use of various insecticides (Gong et al. 2008, Feng 2009). Attract-and-kill light-trapping techniques have also been employed because both sexes are attracted to ultraviolet light (Yu et al. 2008).

A growing body of evidence has shown that volatile sex pheromones of many species of cerambycid beetles are highly conserved, with closely related species within the same genus, more distantly related species in different subfamilies, and even species on different continents using the same pheromones or close analogs (Mitchell et al. 2011, 2015; Ray et al. 2012; Imrei et al. 2013). One such example is (3R,5S)-3,5-dimethyldodecanoic acid, which was first identified as the female-produced sex pheromone of the root-feeding cerambycid *Prionus californicus* Motschulsky, 1845 (Rodstein et al. 2009, 2011). This species is in the same subfamily and tribe as *D. granulosus*. Further field trials determined that racemic 3,5-dimethyldodecanoic acid (prionic acid) was highly attractive to...
males of a number of other North American Prionus species, as well as the European species Prionus coriarius L. (Barbour et al. 2011).

As part of a larger project involving field testing known cerambycid pheromones in different regions of the world, we deployed traps baited with various pheromones in Guangxi Province in southern China in June 2012. During those trials, four male D. granulosus were caught in pitfall traps baited with prionic acid, whereas no beetles were caught in unbaited control traps (J.D.W., unpublished data). We report here the results from follow-up field trials that confirmed that prionic acid is highly attractive to male D. granulosus and likely a component of its female-produced sex pheromone.

Materials and Methods

A dose-response trial that targeted D. granulosus was carried out in a sugarcane plantation near Danzhou, Hainan Province (19° 31’ N, 109° 34’ E). Racemic prionic acid (synthesized as described in Rodstein et al. 2009) was diluted with ethanol to concentrations of 0.001, 0.01, 0.1, and 1 mg per ml, and 1 ml of each solution was then pipetted into resealable polyethylene sachets (Cat. #018161A, 5.1 by 7.6 cm, Thermo-Fisher, Waltham, MA). Sachets containing only ethanol were used as controls. Treatments were randomly assigned to pitfall traps (30 cm wide by 60 cm deep holes with vertical sides) that were ~10 m apart in linear transects. Each lure was tied to a stick, which was then laid over the pitfall trap so that the sachet hung centered above the trap. Transects of traps were deployed at the edges of each of four sugarcane fields that were at least 100 m apart. The traps were left uncovered because preliminary observations confirmed that captured beetles could not escape. The experiment was deployed on 25 April 2013 (N = 10 replicates per treatment), with beetles collected and treatments rotated one position within transects on 29 April 2013. Beetles were collected again on 3 May 2013 when the experiment was terminated.

A second set of experiments carried out in Chongzuo city, Guangxi Province (22° 33’ N, 107° 55’ E), tested two commercial lures that had been designed for use in mating disruption and mass trapping trials with P. californicus in the United States: plastic twist-tie lures (Shin-Etsu Chemical Co., Tokyo, Japan) and amber-colored plastic sachets (Contech, Victoria, British Columbia, Canada). Both types of lures were loaded with 50 mg of racemic prionic acid and were hung above pitfall traps, as described above, in transects that included one lure treatment of each type and an unbaited control trap (traps ~10 m apart). Five transects of traps (at least 300 m apart) were deployed on 9 June 2013, and serviced on 14, 19, and 26 June 2013, when the experiment was terminated. On each service date, treatments were rotated one position within transects.

Differences among means of treatments were tested using the nonparametric Friedman’s test (PROC FREQ, option CMH; SAS Institute 2011). Differences between pairs of means were tested with the REGWQ means-separation test, which controls for maximum experiment-wise error rates (PROC GLM, SAS Institute 2011).

In an effort to verify that female beetles actually produced prionic acid, female beetles were collected from field sites and shipped to Beijing for collection and analysis of volatiles. However, no females survived long enough to obtain collections of headspace odors for analysis.

Results and Discussion

During the dose-response trial, 107 male D. granulosus were captured, with doses of 1 and 0.1 mg of prionic acid being equally attractive and lower doses and controls capturing few beetles, if any (Fig. 1; overall Friedman’s Q_{3,36} = 29.4, P < 0.0001). A single female was captured in a trap baited with the 1 mg dose, likely as a random catch. A total of 284 males were captured during the trial that compared attraction to commercial lures of different design, with no females captured. Twist-tie and amber plastic sachet lures were equally attractive, with means of 9.47 ± 2.45, 2.45, 2.2, and 9.0 ± 2.0 males per trap, respectively, and both were significantly different from controls (mean 0.47 ± 0.3 beetles per trap; Friedman’s Q_{2,45} = 26.6, P < 0.0001).

Our results suggest that traps baited with these commercially available lures can be immediately adopted into integrated pest management programs for detecting this important pest of sugarcane and monitoring the seasonal activity of adults. Within a population,

![Fig. 1. Mean (±SE) number of adult male D. granulosus captured by pitfall traps baited with varying doses of racemic prionic acid. Traps were deployed in a sugarcane plantation near Danzhou, Hainan Province (five sets of traps, two sample dates). Treatment means with different letters are significantly different (REGWQ means separation test, P < 0.05).](http://journals.oxfordjournals.org/)

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the flight period of adult *D. granulosus* is typically less than one month (Yu et al. 2006). Thus, the commercial lures for *P. californicus*, which are designed to last season long, should be entirely adequate for monitoring *D. granulosus* throughout its entire flight season.

Our experiments also showed that pitfall traps are simple and effective traps for these large and clumsy beetles, as they are for other large prionine species (Barbour et al. 2011). During our experiments, we marked a subset of the trapped beetles and returned them to the traps. Over the course of several days, none of the marked beetles could climb the vertical sides of the traps or fly out of the traps. The beetles were clearly capable of flight because they are attracted to light traps, but they apparently could not fly vertically upwards and out of the pitfall traps.

Although there already is considerable evidence for the conservation of pheromone structures among related cerambycid taxa, as discussed in the Introduction, the results described here represent the first evidence for likely use of prionic acid as a pheromone by a cerambycid species outside the genus *Prionus*. Although we have yet to verify that female *D. granulosus* do indeed produce prionic acid as their sex pheromone, the strong and specific attraction of only male beetles certainly suggests that this is the case. Furthermore, several other *Dorysthenes* species are significant pests of sugarcane as well as tree fruit crops in tropical and subtropical areas of southeast Asia, particularly *Dorysthenes buqueti* Guerin-Meneville, 1844 and *Dorysthenes buegli* Redtenbacher, 1848 (Hill 2008). Thus, it is possible and even likely that prionic acid could be rapidly developed for detection and monitoring of these species as well. In addition, given that the racemic pheromone is relatively inexpensive to synthesize and that it is highly attractive to male beetles in low doses (Barbour et al. 2011), it may be possible to use the pheromone to control these beetles through mating disruption or mass trapping, analogous to the highly successful pheromone-based mass trapping of other long-lived tropical beetles (Faleiro 2006).

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