Physiological Entomology

Physiological Entomology (2011) **36**, 321–326 DOI: 10.1111/j.1365-3032.2011.00799.x

Response of *Brontispa longissima* to coconut palm (*Cocos nucifera*) leaf volatiles

YULING FANG, JIANGHUA SUN and ZHONGNING ZHANG

State Key Laboratory of Integrated Management of Pest Insects and Rodents, Institute of Zoology, Chinese Academy of Sciences, Beijing, China

Abstract. *Brontispa longissima* (Gestro) (Coleoptera: Hispidae) is a new invasive pest in China that has caused severe economic damage to palm trees (Arecaceae, Palmae). The response of this beetle to coconut palm (*Cocos nucifera*) leaf volatiles is investigated in laboratory bioassays. Both sexes are attracted to a mixture of β-myrcene, (−)-limonene and *E*-2-hexen-1-ol (1 : 6 : 1), which are key components of coconut palm leaf volatiles. A blend of β-myrcene and (−)-limonene (0.7 : 1–1 : 0.7) in low amounts (100 ng) elicits aggregation and oviposition in females. Chemical analyses of food-deprived, gravid female *B. longissima* show high concentrations of β-myrcene and (−)-limonene in their accessory glands, suggesting that female beetles sequester both compounds and release them during oviposition.

Key words. Attraction, behaviour, *Brontispa longissima*, *Cocos nucifera*.

Introduction

The coconut leaf beetle *Brontispa longissima* (Gestro) (Coleoptera: Hispidae) is one of the most notorious insect pests of palm trees (Arecaceae). Native to Indonesia, *B. longissima* is currently distributed throughout Southeast Asia, including the Maldives, Solomon Islands, Indonesia, Thailand, Vietnam, Australia and China. It was first reported as a new pest in China in Hainan Island in 2002, where it has four to five overlapping generations per year (Zhong *et al*., 2003). *Brontispa longissima* attacks more than ten species of coconut palms (*Cocos nucifera* L.), which are its preferred host plants (Zhong *et al*., 2003). Larvae and adults spend most of the time hiding in unopened leaf buds and feed on its tissue, although mature leaves can also be eaten. Attacks by beetles can affect the growth of coconut trees and reduce production of coconuts. If left uncontrolled, *B. longissima* infestations have the potential to cause enormous damage. Estimates of losses in Vietnam alone are approximately US$40 million annually (Satoshi *et al*., 2006).

Most studies on *B. longissima* focus on damage assessment and control tactics to mitigate its impact (Voegele, 1989; Liu *et al*., 1989; Chang, 1991). Despite the obvious aggregation phenomena of *B. longissima* in the wild, little is known about its chemical communication system. Many species of herbivorous pests rely on host emitted odours for locating and selecting suitable hosts (Bolter *et al*., 1997). In particular, chiral compounds are known to play important roles in modulating behaviour in the Coleoptera. For example, the use of two enantiomers together yields maximum aggregation effects in *Gnathotrichus sulcatus* (LeConte) (Coleoptera: Curculionidae: Scolytinae) (Borden *et al*., 1976) and, in *Dendroctonus brevicomis* LeConte (Scolytidae), opposite enantiomers (antipodes) can have antagonistic effects on behavioural responses, such as aggregation behaviour (Wood *et al*., 1976).

The present study aims to: (i) extract and identify volatiles produced by tender coconut leaves and by *B. longissima* adults and (ii) identify which of these volatiles are attractive to *B. longissima*.

Materials and methods

Insects

Adult *B. longissima* beetles and leaves from unopened leaf buds from coconut trees (*C. ucifera*) were obtained from Xionghai, Hainan Province, China, in 2007. The sex ratio of the beetles was 1 : 1. The beetles were maintained under an LD 12 : 12 h photocycle at 25 ± 1 °C and 80 ± 5% relative humidity and mass-reared on fresh coconut leaves in a plastic box (24 × 17 × 9 cm). Each box contained approximately 500 beetles. Two experimental groups were maintained. The first group was fed 30 g of fresh coconut leaves three times per

Correspondence: Zhongning Zhang, Institute of Zoology, Chinese Academy of Sciences, 1 Beichen West Road, Chaoyang District, Beijing 100101, China. Tel.: +86 10 6480 7071; e-mail: zhangzn@ioz.ac.cn

week, whereas the second group was fed 30 g of fresh coconut leaves only once every 10 days (food-deprived group). Beetle sex was determined by examination of the ventral side of the abdominal tip under the microscope.

Volatile collection

Volatile compounds produced by coconut leaves and beetles were collected by solvent extraction. Tender coconut leaves (100 g) were cut into small pieces and added to a 1 : 1 mixture of pentane/dichloromethane (200 mL) (high-performance liquid chromatography grade) for 24 h. The solution phase was removed, saturated with Na_2SO_4, then concentrated by fractional-distillation with an erect distillation column (30 cm), filled with tiny glass heads (outer diameter 3 mm), at a rate of 1 mL min^{-1} to a final volume of 5 mL. The sample was concentrated to 200 μL under N_2 and stored at −20 °C before analysis by gas chromatography-mass spectrometry (GC-MS) and laboratory bioassays. This collection method was performed seven times.

Solvent extracts of *B. longissima* were prepared by soaking different parts of the beetles (heads, wings, legs, abdomen, hindgut, ovaries, accessory glands, spermathecae, testes and ejaculatory ducts) in a 1 : 1 mixture of pentane/dichloromethane (1 mL) for 30 min at room temperature. Each sample contained a 50-beetle equivalent that was concentrated to 200 μL under N_2 and maintained at −20 °C for GC-MS analysis. All sample collections were repeated five times.

Chemical analysis

GC-MS analyses of coconut leaf extracts were performed on a HP 6890 Gas Chromatograph interfaced with a HP 5973 Mass Spectrometer (electron impact ionization, 70 eV) through a HP-5MS column (inner diameter 30 m × 0.25 mm) (J&W Scientific, Folsom, California). The temperature programme comprised: 30 °C for 1 min, increased to 120 °C at 5 °C min^{-1}, increased to 180 °C at 20 °C min^{-1} and was then maintained at 180 °C for 10 min. Camphene was added to all volatiles collections (include the plants and beetle body parts) as an internal standard (10 ng) for quantification. The mass spectrometry data library was NIST 08 and MS SEARCH 2.0 software (NIST, Gaithersburg, Maryland) was used in the analyses.

The absolute configuration of the chiral compounds in the extracts was analyzed by GC-MS using a Cyclosil-B column (inner diameter 30 m × 0.25 mm) (J & W Scientific). The temperature programme comprised: 50 °C for 5 min, increased to 90 °C at 5 °C min^{-1}, maintained at 90 °C for 2 min, then increased to 96 °C at 2 °C min^{-1}, maintained at 96 °C for 10 min, then increased to 200 °C at 10 °C min^{-1}, and then maintained at 200 °C for 10 min. The absolute configuration of chiral compounds was determined by comparison of retention times with those of authentic standards. All volatile collections received furfural as an internal standard (10 ng).

Sources of compounds

Synthetic volatile compounds (Tables 1 and 2) were supplied by Acros Organics (Somerville, New Jersey), whereas solvents (hexane, pentane and dichloromethane) were obtained from Fisher Chemicals (Fairlawn, New Jersey).

Olfactometer bioassays

Tests were performed using a Y-tube glass olfactometer (outer diameter 12 mm). The vertical segment was 28 cm long and the arms were 9 cm long, spaced at an angle of 120°. During the test, baits (filter paper with different dose of chemicals) were placed at the end of one arm of the Y-tube and a solvent control (1 μL of 1 : 1 mixture of pentane/dichloromethane) was placed in the other arm. This sequence was interchanged randomly in subsequent replicates. Activated charcoal filtered air was passed through the olfactometer at a rate of 50 mL min^{-1} through both arms. An insect container with five test beetles was placed at the open end of the vertical segment. The number of beetles that chose each arm was counted after 5 min. The Y-tube was replaced with a clean one after every test. Any individual beetle was tested only once. The experiment was repeated 20 times. To identify an active blend, all the constituents (including the chiral forms) identified in the plant and insect extracts were tested using their natural ratios in a series of subtractive assays (with each constituent).

Effect of host plant volatiles on oviposition

To test the effect of the identified volatile compounds on oviposition, females were given a choice to oviposit between

Table 1. Chemical profile of coconut (*Cocos nucifera*) leaf extracts.

Chemical compound	Mean ± SE percentage relative to quantity of α-pinene[a] ($n = 7$)
Green leaf volatiles	
Hexanal	0.6 ± 0.2
3-Hexanol	1.0 ± 0.1
E-2-Hexenal	2.2 ± 0.3
2-Hexanol	1.5 ± 0.3
1-Hexanol	2.2 ± 0.3
Z-3-Hexen-1-ol	7.6 ± 0.5
E-2-Hexen-1-ol	5.7 ± 0.2
Terpenoid compounds	
α-Pinene	100
β-Pinene	28.0 ± 2.4
Myrcene	7.2 ± 1.0
3-Carene	12.7 ± 3.1
Limonene	45.8 ± 4.3
Terpinolene	20.7 ± 3.8
Other compounds	
Heptanal	0.2 ± 0.2
Nonanal	0.2 ± 0.1

[a]The content of α-pinene from tender leaves of *C. nucifera* varied in the range 3.1–7.2 ng g^{-1}.

Table 2. Chiral compounds present in coconut (*Cocos nucifera*) leaf extracts and accessory glands of food-deprived *Brontispa longissima* females.

Retention time (min)	Compound	Mean ± SE percentage relative to quantity of (+)-α-pinene ($n = 5$)	
		Leaf[a]	Accessory glands[b]
13.07	(−)-α-Pinene	32.2 ± 1.4	35.8 ± 3.7
13.25	(+)-α-Pinene	100	100
14.20	β-Myrcene[c]	11.7 ± 1.7	91.0 ± 4.6
15.02	β-Pinene	44.6 ± 2.4	43.1 ± 2.6
14.41	3-Carene	38.9 ± 3.2	36.8 ± 2.8
16.54	(−)-Limonene[c]	32.6 ± 5.2	76.0 ± 6.0
17.05	(+)-Limonene	9.7 ± 2.1	8.5 ± 1.8
19.19	Terpinolene	22.0 ± 3.1	19.6 ± 2.8

[a]Extract of unopened bud's leaves of *C. nucifera.* The content of (+)-α-pinene varied in the range 2.1–5.5 ng g^{-1}.
[b]Extract from the accessory glands of food-deprived gravid female *Brontispa longissima*. The content of (+)-α-pinene varied varied in the range 0.2–0.7 ng per beetle.
[c]Indicates a significant difference between leaf and accessory gland (*t*-test, $P < 0.05$).

fresh coconut leaves treated with the volatile compounds and control leaves treated with hexane. Samples of volatiles were diluted in analytical grade hexane (10 or 200 ng μL^{-1}). For each assay, a piece of fresh coconut leaf (2 × 15 cm) treated with 10 μL of the volatile was used. The control leaf was identical and was treated with 10 μL of hexane. The test was performed in a glass tube (5 × 20 cm) during 24 h, after which the number of eggs was recorded. The test was repeated 30 times. Twenty females were used in each test. Subtractive assays were used in this experiment.

Statistical analysis

Data were analyzed by using a paired-sample Student's *t*-test. For comparisons between several groups (subtractive blends series), one-way analysis of variance with Bonferonni analysis was used in SPSS, version 13.0 (SPSS Inc., Chicago, Illinois). $P < 0.05$ was considered statistically significant.

Results

Chemical analysis

The chemical profile of coconut leaf extracts is shown in Table 1. Terpenoids were the main components in the crude extracts and six-carbon compounds were in low abundance. The results are expressed by the relative percentage to α-pinene (5.3 ± 0.51 ng g^{-1}). Ratios of compositions were similar in the volatile composition of the gas phase and in the corresponding leaf-extract (Y. Fang, unpublished data).

Chiral chromatography revealed eight types of terpenoids, including (−)- and (+)- enantiomers of both α-pinene and limonene. The ratio of the terpenoids was similar in coconut leaves and in most female beetle extracts, with the exception of the accessory glands (Fig. 1 and Table 2). Higher amounts of β-myrcene (0.45 ± 0.07 ng/beetle) and (−)-limonene (0.45 ± 0.07 ng/beetle) were detected in accessory gland extracts of food-deprived gravid females, whereas the other parts of the beetles contained low amounts of myrcene (0.07 ± 0.04 ng/beetle) and (−)-limonene (0.17 ± 0.06 ng/beetle).

Olfactometer bioassays

Solvent extracts from coconut leaves elicited responses in both sexes. In subtractive assays, the results showed significant differences between controls and a mixture of β-myrcene, (−)-limonene and *E*-2-hexen-1-ol (1 : 6 : 1) (Bonferoni, $P = 0.004$). No significant difference was found between the coconut leaf extract and the synthetic mixture β-myrcene, (−)-limonene and *E*-2-hexen-1-ol (1 : 6 : 1) (Bonferoni, $P = 0.297$). Compared with the control, the blend comprising β-myrcene, (−)-limonene and *E*-2-hexen-1-ol (1 : 6 : 1) elicited significant attraction in female (*t*-test, $P = 0.001$) and

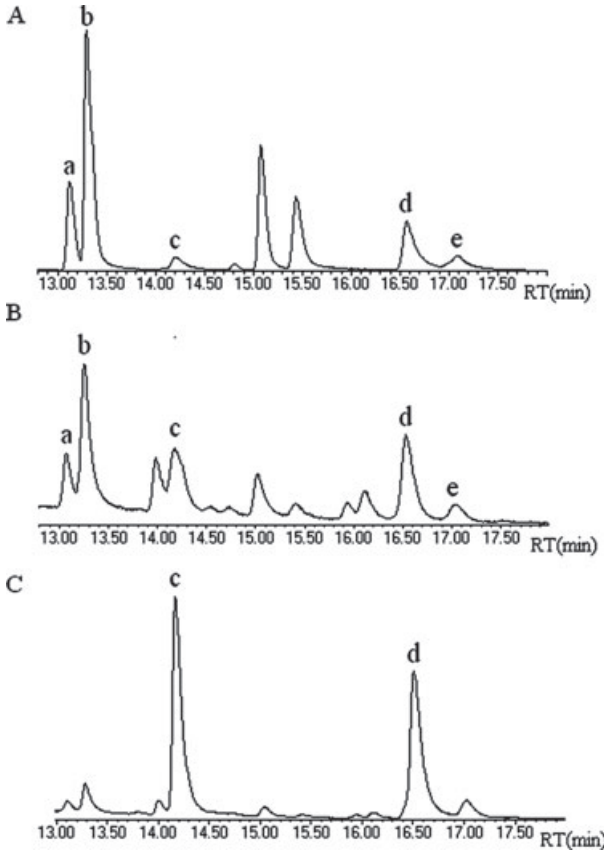

Fig. 1. Gas chromatography (cycloSil-B column) analysis of (A) coconut leaves, (B) accessory glands of food-deprived *Brontispa longissima* females, and (C) co-injections [β-myrcene + (−)-limonene] with accessory glands of food-deprived *B. longissima* females. a, (+)-α-Pinene; b, (−)-α-pinene; c, myrcene; d, (−)-limonene; e, (+)-limonene.

male (*t*-test, $P = 0.002$) beetles. There were no significant differences in the response of males versus female beetles (*t*-test, $P = 0.398$) (Fig. 2).

A mixture of β-myrcene and (−)-limonene (1 : 1) at a low concentration (100 ng) induced attraction in female beetles. A blend of β-myrcene and (−)-limonene (at the ratio of 0.7 : 1 to 1 : 0.7) was detected by female beetles at a dose of 100 ng. Activity was lost at higher concentrations (2 μg). No activity was elicited by a mixture of β-myrcene and (+)-limonene at a 1 : 1 ratio (Table 3). No significant differences were found in bioassays between the different subtractive blends compared with the final blend of β-myrcene and (−)-limonene (1 : 1) (Bonferroni, $P < 0.05$). No synthetic compound alone was able to induce attraction in adult beetles.

Effect of selected chemicals on oviposition

In the subtractive bioassays, we found that a blend of β-myrcene and (−)-limonene (0.7 : 1 to 1 : 0.7) at a low concentration (100 ng) induced oviposition (Bonferroni, $P = 0.011$) (Table 4). The activity was lost at higher doses (2 μg).

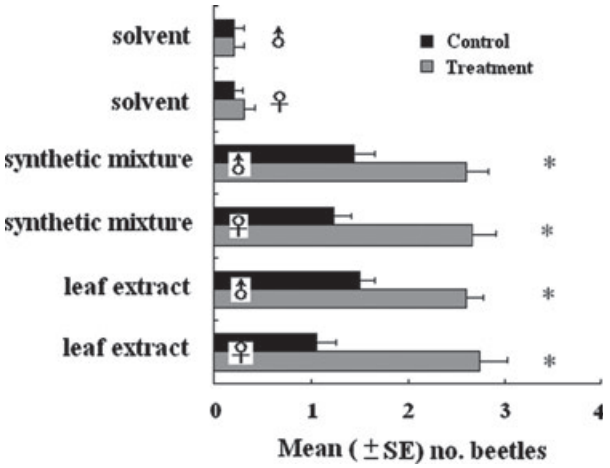

Fig. 2. Response of *Brontispa longissima* in a Y-tube olfactometer when given a choice between solvent control and coconut leaf solution extract or a mixture of [β-myrcene, (−)-limonene and *E*-2-hexen-1-ol in the ratio of 1 : 6 : 1] (synthetic mixture). *Statistically significant difference between treatment and control (*t*-test, $P < 0.05$).

A mixture of myrcene and (+)-limonene (1 : 1) did not elicit oviposition (*t*-test $P = 0.722$).

Discussion

As flavor-determining compounds, monoterpenes are important plant volatiles. Many studies demonstrate that monoterpenes present enantiomeric arrangements that can be utilized as a 'fingerprint' by insects for the detection of a host and/or mate (Silverstein, 1988; Seybold, 1993; Wilfried, 1998). The presence of both enantiomers of a single monoterpene is common for plant volatiles. As shown in Fig. 1, two monoterpenes, α-pinene and limonene, have both of their enantiomers present in the volatile blend of coconut leaves. Several studies suggest that enantiomeric ratios in many compounds are important for insects to discriminate between host and nonhost odors (Borden *et al*., 1976; Wood *et al*., 1976; Silverstein, 1988; Seybold, 1993; Atle *et al*., 1998). The data of the present study show that chirality was key to the behavioural activity of *B. longissima*. Even though a mixture of β-myrcene and (−)-limonene induces significant behavioural activity in olfactometer and oviposition bioassays, a mixture of β-myrcene and (+)-limonene does not. Similarly, the composition of volatile compounds is very important to beetle behaviour. A mixture of β-myrcene, (−)-limonene and *E*-2-hexen-1-ol (1 : 6 : 1) can attract both genders of beetles. On the other hand, a mixture of β-myrcene and (−)-limonene (1 : 1) induces attraction in female beetles only.

The volatile blend emitted by coconut leaves contains both β-myrcene and (−)-limonene. Remarkably, gravid females also contain β-myrcene and (−)-limonene when food-deprived. Only the accessory glands contain high amounts of β-myrcene and (−)-limonene, whereas other parts of the beetles contain low amounts of myrcene and (−)-limonene. This suggests that *B. longissima* sequesters β-myrcene and (−)-limonene and releases them when preparing for oviposition (probably triggered by food-deprivation). The data of the present study also show that *B. longissima* are very sensitive in behavioural bioassays to concentrations of β-myrcene and (−)-limonene, so it is likely that these volatiles have different functions at different concentrations. At low concentrations, they may act as an oviposition attractant, whereas, at higher concentrations, this behavioural effect could be lost. Perhaps at higher concentrations β-myrcene and (−)-limonene lose

Table 3. Response of *Brontispa longissima* to synthetic volatiles of coconut (*Cocos nucifera*) leaves in Y-tube olfactometer bioassays.

			Mean ± SE number of beetles ($n = 20$)	
Chemical	Dose	Gender	Treatment	Control
Myrcene + (−)-limonene (1 : 1)	100 ng	Male	2.33 ± 0.26	1.77 ± 0.21
Myrcene + (−)-limonene(1 : 1)[a]	100 ng	Female	2.56 ± 0.23	0.95 ± 0.18
Myrcene + (−)-limonene (0.7 : 1)[a]	100 ng	Female	2.50 ± 0.21	1.13 ± 0.20
Myrcene + (−)-limonene (1 : 0.7)[a]	100 ng	Female	2.47 ± 0.28	0.87 ± 0.15
Myrcene + (−)-limonene (1 : 1)	2 μg	Female	1.65 ± 0.27	1.91 ± 0.25
Myrcene + (+)-limonene (1 : 1)	100 ng	Female	1.57 ± 0.22	1.67 ± 0.19

[a]Indicates a significant difference between treatment and control (*t*-test, $P < 0.05$).

Table 4. Oviposition response of *Brontispa longissima* to coconut leaves in the presence of selected chemicals.

Chemical	Dose	Mean ± SE number of eggs ($n = 30$)	
		Treatment	Control
Myrcene + (−)-Limonene (1 : 1)[a]	100 ng	8.98 ± 0.86	4.89 ± 0.52
Myrcene + (−)-limonene (1 : 1)	2 µg	6.65 ± 0.79	7.00 ± 0.90
Myrcene + (−)-limonene (0.7 : 1)[a]	100 ng	6.07 ± 0.58	1.92 ± 0.34
Myrcene + (−)-limonene (1 : 0.7)[a]	100 ng	5.69 ± 0.56	1.84 ± 0.33
Myrcene + (+)-limonene (1 : 1)	100 ng	6.20 ± 0.77	6.14 ± 0.91
Myrcene	100 ng	6.22 ± 0.67	6.09 ± 0.79
(−)-Limonene	100 ng	6.12 ± 0.86	6.12 ± 1.04

[a] Indicates a significant difference between treatment and control (t-test $P < 0.05$).

their attractiveness for oviposition to prevent crowding and ensure that larvae have sufficient resources. Similar phenomena are reported for other insect species (Rothschild *et al*., 1977; Ronald & Juro, 1982; Poirier & Borden, 1991).

The classical view that the accessory gland simply yields material to provide protection from desiccation is challenged by the possibility of production of oviposition aggregation pheromones or oviposition-deterring pheromones in some species (Szopa, 1981; Schoonhoven *et al*., 1990; Rai *et al*., 1997). The accessory glands of many female insects produce secretions that are deposited on oviposited eggs. Gravid *B. longissima* females release high rates of β-myrcene and (−)-limonene under starved conditions, probably because gravid females wait until they are hungry before ovipositing. If females cease or decrease activity on the plant until they become hungry, they can then combine feeding and oviposition bouts into a single foraging expedition, cutting the predation risk considerably. Many studies report that natural enemies make very effective use of the volatiles emitted by plants when they are attacked, even when there is no plant damage (Nadel & van Alphen, 1987; Potting *et al*., 1995). Wallin & Ekbom (1994) show that only female beetles alter their behaviour with respect to both food deprivation and prey density. For *B. longissima*, there may be two advantages for gravid females when less food is consumed. First, they would induce a decreased reaction in the plant (less parasitoid attractants) and, second, changes of odour may change the chemical fingerprint, hence making females less prone to detection by natural enemies. Unexpectedly, the content of β-myrcene and (−)-limonene from the extract of female's accessory glands in the normal group (regularly fed) does not significantly increase compared with the food-deprived group. One explanation for this might be that hungry females are simply more motivated than fed ones for oviposition because food deprivation could quickly lead to death.

Only a few studies exist reporting attractants for species in the family Hispidae before the present study was conducted. Earlier studies on the natural history of *B. longissima* fail to report evidence of behavioural responses to volatile compounds produced by fresh, tender coconut leaves (Zeng *et al*., 2003; Zhou *et al*., 2004). *Brontispa longissima* lives in unopened leaf buds for most of its life, such that long distance communication may not be essential. Nevertheless, the identification of an active blend released by its host leave buds is a pioneer breakthrough for understanding the host-finding mechanisms in *B. longissima.* In addition, we raise the possibility that gravid females may sequester terpenoid compounds and release them when they prepare to oviposit. These results open new experimental perspectives for unravelling how *B. longissima* uses semiochemicals for communication.

Acknowledgements

We are grateful to Dr Dan Miller (USDA Forest Service) for his review of an earlier version of the manuscript and to Dr Christian Salcedo for editing the final version of the manuscript. The authors are also thankful to Hainan Province Forest Bureau for their assistance with beetle and coconut leaf collection. This work was supported by National Basic Research Program of China (2009 CB 119204).

References

Atle, W., Anna, K.B.K., Monika, P. *et al*. (1998) Enantiomeric composition of monoterpene hydrocarbons in some conifers and receptor neuron discrimination of α-pinene and limonene enantiomers in the pine weevil, *Hylobius abietis*. *Journal of Chemical Ecology*, **24**, 273–287.

Bolter, C.J., Dicke, M., Van Loon, J.J.A. *et al*. (1997) Attraction of Colorado potato beetle to herbivore-damaged plant during berbivory and after its termination. *Journal of Chemical Ecology*, **23**, 1003–1023.

Borden, J.H., Chong, L., Mclean, J.A. *et al*. (1976) Gnathotrichus sulcatus: synergistic response to enantiomers of the aggregation pheromone sulcatol. *Science*, **192**, 894–896.

Chang, Y.H. (1991) Integrated pest management of several forest defoliators in Taiwan. *Forest Ecology and Management*, **39**, 65–72.

Liu, S.D., Lin, S.C. & Shiau, J.F. (1989) Microbial control of coconut leaf beetle (*Brontispa longissima*) with green muscardine fungus, *Metarhizium anisopliae* var. *anisopliae*. *Journal of Invertebrate Pathology*, **53**, 307–314.

Nadel, H. & van Alphen, J.J.M. (1987) The role of host- and host-plant odours in the attraction of a parasitoid, *Epidinocarsis Iopezi*, to the habitat of its host. The cassava mealybug, *Phenacoccu manihoti*. *Entomologia Experimentalis et Applicata*, **45**, 181–186.

Poirier, L.M. & Borden, J.H. (1991) Recognition and avoidance of previously laid egg masses by the oblique-banded leafrooler (Lepidoptera: Tortricidae). *Journal of Insect Behavior*, **4**, 501–508.

Potting, R.P.J., Vet, L.E.M. & Dicke, M. (1995) Host microhabitat location by the stemborer parasitoid *Cotesia flavipes*: The role of locally and systemically induced plant volatiles. *Journal of Chemical Ecology*, **21**, 525–539.

Rai, M.M., Hassanali, A., Saini, R.K. *et al*. (1997) Identification of components of the oviposition aggregation pheromone of the gregarious desert locust *Schistocerca gregaria* (Forskal). *Journal of Insect Physiology*, **43**, 83–87.

Ronald, J.P. & Juro, K. (1982) Oviposition site partitioning in *Dacus cucurbitae*. *Entomologia Experimentalis et Applicata*, **31**, 428–432.

Rothschild, M. & Schoonhoven, L.M. (1977) Assessment of egg-load by *Pieris brassicae* (Lepidoptera: Pieridae). *Nature*, **266**, 352–55.

Satoshi, N., Kazuhiko, K. & Keiji, T. (2006) *Invasion of the Coconut Hispine Beetle,* Brontispa longissima*: Current Situation and Control Measures in Southeast Asia* [WWW document]. URL http://www.agnet.org/activities/sw/2006/589543823/paper-899851121.pdf [accessed 7 December 2008].

Schoonhoven, L.M., Beerling, E.A.M., Braaksma, R. & Van Vugt, Y. (1990) Dose the imported cabbageworm, *Pieris rapae*, use an oviposition deterring pheromone? *Journal of Chemical Ecology*, **16**, 1649–1656.

Seybold, S.J. (1993) Role of chirality in olfactory-directed behavior: aggregation of pine engraver beetles in the genus *Ips* (Coleoptera: Scolytidae). *Journal of Chemical Ecology*, **19**, 1809–1831.

Silverstein, R.M. (1988) Chirality in insect communication. *Journal of Chemical Ecology*, **14**, 1981–2004.

Szopa, T.M. (1981) The role of the accessory reproductive glands and genital ducts in egg pod formation in female *Schistocerca gregaria*. *Journal of Insect Physiology*, **27**, 23–29.

Voegele, J.M. (1989) Biological control of *Brontispa longissima* in Western Samoa: an ecological evaluation. *Ecosystems and Environment*, **27**, 315–329.

Wallin, H. & Ekbom, B. (1994) Influence of hunger level and prey densities on movement pattern in three species of *Pterostichus* beetles (Coleoptera: Carabidae). *Environment Entomology*, **23**, 1171–1181.

Wilfried, A.K. (1998) Enantioselective capillary gas chromatography in the investigation of stereochemical correlations of terpenoids. *Chirality*, **10**, 499–504.

Wood, D.L., Browne, L.E., Ewing, B. *et al*. (1976) Western pine beetles: specificity among enantiomers of male and female components of an attractant pheromone. *Science*, **192**, 896–898.

Zeng, L., Zhou, R., Cui, Z.X. *et al*. (2003) Effect of host plant on development and survival of *Brontispa longissima* (Gestro). *Journal of South China Agricultural University (Natural Science Edition)*, **24**, 37–39.

Zhong, Y.L., Wu, X.Y. & Liu, K. (2003) Brontispa longissima – a highly dangerous pest. *Chinese Journal of Tropical Agriculture*, **23**, 67–72.

Zhou, R., Zeng, L., Liang, G.W. *et al*. (2004) Life history of the laboratory population of the palm beetle *Brontispa longissima*. *Entomological Knowledge*, **41**, 336–339.

Accepted 29 June 2011
First published online 13 September 2011