Attractant for Vinegar Fly, *Drosophila busckii*, and Cluster Fly, *Pollenia rudis* (Diptera: Drosophilidae et Calliphoridae)

Vincas Būda^{a,*}, Sandra Radžiutė^b, and Erikas Lutovinovas^b

- ^a Vilnius University, Faculty of Natural Sciences, M. K. Čiurlionio 21/27, LT-03101 Vilnius, Lithuania. Fax: +3 70 5 2 72 93 52. E-mail: vinbuda@ekoi.lt
- ^b Institute of Ecology, Vilnius University, Akademijos 2, LT-08412 Vilnius-21, Lithuania
- * Author for correspondence and reprint requests
- Z. Naturforsch. **64c**, 267–270 (2009); received September 4/October 24, 2008

A field test carried out in an industrial greenhouse in Lithuania revealed the attractiveness of synthetic methyl salicylate (MeSa) to two dipteran species: the vinegar fly, $Drosophila\ busckii$ (Drosophilidae), and the cluster fly, $Pollenia\ rudis$ (Calliphoridae). The attractant for the former fly species was especially effective, as sticky traps containing 0.25 ml of MeSa captured (814 \pm 55) D. busckii flies/trap on average compared to (12 \pm 4) flies/trap in control traps. The mean capture of P. rudis [(42 \pm 4) flies/trap] was significantly higher in MeSa-baited traps compared to the control traps [(13 \pm 4) flies/trap]. The presence of MeSa in emissions of many fruits suitable for P. P0. P1. P2. P3. P3. P4. P4. P5. P5. P6. P8. P8. P8. P8. P9. P

Key words: Attractiveness, Field Trapping, Methyl Salicylate

Introduction

Chemical attractants are a useful tool for insect monitoring. Therefore, extension of our knowledge on both attractive compounds and species responding to the chemicals is of great importance.

The order Diptera is among the species-richest ones. It includes many species of economic importance, *i.e.* both useful insects (*e.g.*, pollinators) and pests (transmitters of human, animal and plant diseases). However, chemical attractants are known for relatively few species in this order, especially when compared to lepidopterans (El-Sayed, 2008), thus dipterans remain to be investigated much more thoroughly in this aspect.

The Drosophilidae family is one of the beststudied families within the order Diptera in different aspects. About 3000 species are known in this family (e.g. Nartshuk, 2003). However, available data on chemical attractants (including pheromones) are scarce. Sex and/or aggregation pheromones have been identified for 34 species and attractants for 12 species only (El-Sayed, 2008). The known attractants include methyl eugenol (attractive for fruit flies of 10 species) and two three-component blends (each attractive for a single species). Two of the chemicals in the three-component attractive blends are the same: ethanol and acetic acid, one component being different: either 2-phenylethanol or ethyl acetate (Casana-Giner *et al.*, 1999; Zhu *et al.*, 2003).

Within the family Calliphoridae, which includes more than 1000 species (Nartshuk, 2003), a pheromone of a single species was identified and attractants for only 8 species were revealed (El-Sayed, 2008). It should be noted that all 5 compounds known up to now as attractants for fruit flies (Drosophilidae) attract some species from the Calliphoridae family as well. Other attractants include phenols (phenol, *p*-cresol), organic acids (butyric, valeric, benzoic), aliphatic alcohols (2-methylpropan-1-ol, butan-2-ol, delta-1-octen-3-ol), hydrogen sulfide compounds (1,2-dimethyldisulfane and 1,3-dimethyltrisulfane), an ester (sec-butyl acetate) and a ketone (propan-2-one) (El-Sayed, 2008).

Taking into consideration the richness of species within the two dipteran families briefly reviewed, one can conclude, that the knowledge of attractant composition still remains poorly investigated.

Many chemicals of plant origin are potential attractants for insects including methyl salicylate

(MeSa). The aim of the present study was to test the activity of MeSa for some species within the order Diptera.

Material and Methods

Behavioural test

The behavioural test was conducted in a 0.5-ha greenhouse with green onions growing on compost-enriched ground substrate in Sakalai, Šalčininkai district, Lithuania in September 2006.

Sticky yellow cards [Biobest N.V., Westerlo, Belgium (Bug Scan, 20 × 10 cm in size)] were used. In the central part of the card a 2-ml plastic vial was fixed in a hole which has been cut at a height of ~2 cm above the centre of the card. The vials were plugged with cotton wool. Such sticky traps were fixed vertically to wire poles at a height of ~2 m above the ground. They were either baited with 0.25 ml of MeSa (Fig. 1) (purity ≥99%; Carl Roth GmbH, Karlsruhe, Germany) or unbaited (control). The traps were placed randomly in three rows with 15 m distance in between. Four replicates were conducted. The traps were collected after a 14-d period, and trapped insects were identified and counted.

Species identification

The trapped specimens were identified following the key of Shtakelberg (1970) for Drosophilidae and Grunin (1970) for Calliphoridae.

Statistical analysis

Catches were analyzed by the Kruskal-Wallis ANOVA test by ranks using the software Statistica 5.0 (Sokal and Rohlf, 1995).

Results and Discussion

Numerous catches of the dipteran species were recorded in yellow sticky MeSa-baited traps in the greenhouse. A total of 3255 *D. busckii* and

Fig. 1. Chemical structure of methyl salicylate (MeSa).

166 *P. rudis* specimens were captured. In control traps there were 46 and 51 flies of the two dipteran species, respectively. The traps contained some other Diptera species as well, however, the catches were low and no significant differences compared to the control were recorded.

Vinegar fly, D. busckii Coquillett (Drosophilidae)

The mean catch [(814 \pm 55) flies/trap] of vinegar flies in MeSa-baited traps significantly differed from that in control traps [(12 \pm 4) flies/trap], p = 0.02. The yellow colour itself attracted the flies as proved by catches in the control traps. However, the application of MeSa enhanced the attractiveness of the traps significantly (Fig. 2).

There is very little known on chemical compounds affecting the *D. busckii* behaviour. Two compounds [(S)-2-pentadecyl acetate and 2-pentadecanone] are reported to trigger an aggregation reaction of the flies. These compounds were extracted from the ejaculatory bulb of males of this species and were attributed to aggregation pheromones (Schaner *et al.*, 1989).

D. busckii like most vinegar flies, is attracted to various rotting vegetable materials (vegetables and fruit in particular), abundant in bacteria and yeasts. When flying from one rotting object to another, adults transmit rot-causing microorganisms. Thus, although vinegar flies are saprophagous, they can cause considerable damage by spreading the rot. It was demonstrated, that D. busckii transmits, to high extent, fruit rot caused by the fungus Aspergillus niger thus non-insecticide measures are searched for the control of the

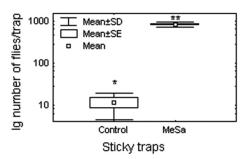


Fig. 2. Mean captures of *D. busckii* in yellow sticky traps baited with and without (control) methyl salicylate (MeSa). Values marked by different numbers of asterisks differ significantly.

flies with the aim to reduce a spread of the rot (Saxena and Saxena, 1990). It was demonstrated that D. busckii can be a vector of the bacterium Erwinia carotovora subsp. carotovora (Boers, 1997, after van der Sommen et al., 2000) which induces soft rot in chicory. Therefore D. busckii is considered a pest in commercial plantations of chicory, and insecticides are used to control it. It is known that D. busckii colonizes rotting oranges later than other species of Drosophila, larvae of which feed on the same resources and form a guild coexisting on the same food source (Nunney, 1996). This means that vinegar flies of this species are potentially more dangerous transmitters of pathogenic microorganisms than other Drosophila species.

Based on data on the *D. busckii* behaviour, one can conclude that the flies are likely to be attracted to compounds released by bacteria, yeasts, fungi or rotting plant substrate. It is known that MeSa is an important flavour constituent of many fruits (*e.g.* Goff and Klee, 2006) and some plants when they are damaged by bacteria or fungi (Cardoza *et al.*, 2002; Cardoza and Tumlinson, 2006). Thus, we assume that MeSa can be attributed to kairomone attractants for this species.

In our opinion, MeSa can be tested as a tool increasing the effectiveness of yellow sticky traps for monitoring *D. busckii* flies. The effectiveness of this attractant in yellow sticky traps for mass trapping of *D. busckii* in closed areas (such as greenhouses, fruit warehouses) should be evaluated in the future.

MeSa is the first attractant for the vinegar fly *Drosophila busckii*.

Cluster fly, Pollenia rudis (Fabricius) (Calliphoridae)

In yellow MeSa-baited traps the mean catch of cluster flies was (42 ± 4) flies/trap and differed significantly (p = 0.02) from the control $[(13 \pm 4)$ flies/trap on average]. The yellow trap itself is attractive to some extent to the flies as was proved by catches in control traps. However, the application of MeSa enhanced the attractiveness for this species significantly (Fig. 3). Thus, MeSa is an attractant for *P. rudis*. This is the second attractant for the cluster fly, *P. rudis*. The only attractant known so far for this species was methyl eugenol (Kido *et al.*, 1996).

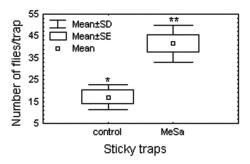


Fig. 3. Mean captures of *P. rudis* in yellow sticky traps baited with and without (control) methyl salicylate (MeSa). Values marked by different numbers of asterisks differ significantly.

As the lifespan of adults is quite long, they undoubtedly need some food. P. rudis has been often recorded on flowers of Asclepias syriaca trapped by pollinium (Frost, 1965). Also it is established that *P. rudis* is an important pollinator of the Mediterranean weed Vincetoxicum nigrum (Asclepiadaceae) (Lumer and Yost, 1995). Flowers of many plants emit MeSa as the common compound of floral scent (Knudsen et al., 2006). Since MeSa is a widespread flowers' compound, it could become an attractant for P. rudis indicating a suitable food source for adult flies. Thus, MeSa should be classified as attractant and synomone (compound beneficial for both receiver and sender) due to the pollination activity of flowersvisiting *P. rudis* adult flies.

However, the adult cluster fly is attributed to household nuisances because of its habitude of clustering in buildings during late summer and autumn for the purpose of overwintering. When cluster flies enter domestic or industrial premises, and particularly hospitals, the species is considered a pest due to the possible transmission of bacterial pathogens, and wherever it is possible, either biological, e.g. entomophilic nematodes (Barson et al., 1986), or chemical insecticides (Faulde et al., 2001) as control measures are applied against it. Besides, larvae of *P. rudis* are known as parasites of earthworms from several genera, Allolobophora, Eisenia, Lumbricus (Heath et al., 2004). However, it is not clear whether the flies of this species are agricultural pests or not (Szpila, 2003).

In our opinion, the combined application of the available biological pest control measures and attractants can markedly enhance the effectiveness of the control.

Acknowledgements

We thank Dr. Henrikas Ostrauskas from the Lithuanian State Plant Protection Service for guidance on a suitable trapping site, and Mr. Evaldas Masevičius for the kind permission to run field experiments in the greenhouse in Sakalai, Šalčininkai district, Lithuania. The research was supported by the Lithuanian State Science and Study Foundation (grant no. T-06079).

- Barson G., Renn N., and Richardson P. N. (1986), Controlling insects with entomophilic nematodes. GB Patent #GB2174907.
- Cardoza Y. J. and Tumlinson J. H. (2006), Compatible and incompatible *Xanthomonas* infections differentially affect herbivore-induced volatile emission by pepper plants. J. Chem. Ecol. 32, 1755–1768.
- Cardoza Y. J., Alborn H. T., and Tumlinson J. H. (2002), *In vivo* volatile emissions from peanut plants induced by simultaneous fungal infection and insect damage. J. Chem. Ecol. **28**, 161–174.
- Casana-Giner V., Gandia-Balaguer A., and Primo-Yufera E. (1999), Field trial of an attractant mixture for dipterans, including the pest *Ceratitis capitata* (Wiedemann) (Dipt., Tephritidae), in Valencia, Spain. J. Appl. Entomol. **123**, 47–48.
- El-Sayed A. M. (2008), The Pherobase: Database of Insect Pheromones and Semiochemicals. URL http://www.pherobase.com
- Faulde M., Sobe D., Burghardt H., and Wermter R. (2001), Hospital infestation by the cluster fly, *Pollenia rudis sensu stricto* Fabricius 1794 (Diptera: Calliphoridae), and its possible role in transmission of bacterial pathogens in Germany. Int. J. Hyg. Environ. Heal. **203**, 201–204.
- Frost W. (1965), Insects and pollinia. Ecology 46, 556–558.
- Goff S. A. and Klee H. J. (2006), Plant volatile compounds: sensory cues for health and nutritional value? Science 311, 815–819.
- Grunin K. Ya. (1970), Family Calliphoridae. In: Key to the Insects of the European Part of the USSR, Vol. 5(2) (Bei-Bienko G. Ya., ed.). Nauka, Leningrad, pp. 607–624 (in Russian).
 Heath A. C. G., Marris J. W. M., and Harris A. C. (2004),
- Heath A. C. G., Marris J. W. M., and Harris A. C. (2004), A cluster fly, *Pollenia pseudorudis* Rognes, 1985 (Diptera: Calliphoridae): its history and pest status in New Zealand. New Zeal. J. Zool. **31**, 313–318.
- Kido M. H., Asquith A., and Vargas R. I. (1996), Nontarget insect attraction to methyl eugenol traps used in male annihilation of the oriental fruit fly (Diptera: Tephritidae) in riparian Hawaiian stream habitat. Environ. Entomol. **25**, 1279–1289.

- Knudsen J. T., Eriksson R., Gershenzon J., and Stahl B. (2006), Diversity and distribution of floral scent. Bot. Rev. **72**, 1–120.
- Lumer C. and Yost S. E. (1995), The reproductive biology of *Vincetoxicum nigrum* (L.) Moench (Asclepiadaceae), a Mediterranean weed in New York State. B. Torrey Bot. Club 122, 15–23.
- Nartshuk E. P. (2003), Key to Families of Diptera (Insecta) of the Fauna of Russian and Adjacent Countries. Russian Academy of Sciences, St. Petersburg, pp. 158–183 (in Russian).
- Nunney L. (1996), The colonization of oranges by the cosmopolitian *Drosophila*. Oecologia 108, 552–561.
- Saxena P. and Saxena S. K. (1990), Effect of treating tomatoes with leaf extract of *Argemone mexicana* on the development of fruit rot caused by *Aspergillus niger* in the presence of *Drosophila busckii*. Natl. Acad. Sci. Lett. (India) **13**, 333–335.
- Schaner A. M., Tanico Hogan L. D., and Jackson L. L. (1989), (S)-2-Pentadecyl acetate and 2-pentadecanone, components of aggregation pheromone of *Dro*sophila busckii. J. Chem. Ecol. 15, 2577–2588.
- Shtakelberg A. A. (1970), Family Drosophilidae. In: Key to the Insects of the European Part of the USSR, Vol. 5(2) (Bei-Bienko G. Ya., ed.). Nauka, Leningrad, pp. 390–399 (in Russian).
- Sokal R. R. and Rohlf F. J. (1995), Biometry: The Principles and Practice of Statistics in Biological Research, 3rd ed. W. H. Freeman & Co., New York, USA.
- Szpila K. (2003), Actual impact of the blowfly-parasitoids (Diptera: Calliphoridae) on earthworms (Oligochaeta: Lumbricidae) of cultivated fields, against the literature. Wiadomości Entomol. 22, 211–220.
- van der Sommen A. T. C., Wertheim B., Vet L. E. M., and Dicke M. (2000), The attraction of *Drosophila busckii* by substrate and aggregation pheromone under laboratory conditions. P. Neth. Entomol. Soc. Exp. Appl. Entomol. (N.E.V.) 11, 189–194.
- Zhu J. W., Park K. C., and Baker T. C. J. (2003), Identification of odors from overripe mango that attract vinegar flies, *Drosophila melanogaster*. J. Chem. Ecol. **29**, 899–909.