

Optimization of a Phenylacetaldehyde-Based Attractant for Common Green Lacewings (*Chrysoperla carnea* s.l.)

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Abstract In field trapping tests, the catch of *Chrysoperla carnea sensu lato* (Neuroptera: Chrysopidae) increased when acetic acid was added to lures with phenylacetaldehyde. The addition of methyl salicylate to the binary mixture of phenylacetaldehyde plus acetic acid increased catches even further. The ternary blend proved to be more attractive than β -caryophyllene, 2-phenylethanol, or 3-methyl eugenol (compounds previously described as attractants for chrysopids) on their own, and no influence on catches was recorded when these compounds were added as fourth components to the ternary blend. There were minimal changes in activity when (*E*)-cinnamaldehyde or methyl anthranilate (both evoking large responses from female or male antennae of *C. carnea* in this study) were

added, although both compounds showed significant attraction on their own when compared to unbaited traps. In subtractive field bioassays with the ternary mixture, it appeared that the presence of either phenylacetaldehyde or methyl salicylate was important, whereas acetic acid was less so in the ternary mixture. The ternary blend attracted both female and male lacewings at sites in southern, central, and northern Europe. Possible applications of a synthetic attractant for lacewings are discussed.

Keywords Acetic acid · Attractant · *Chrysoperla* · Chrysopidae · Green lacewings · Methyl salicylate · Neuroptera · Phenylacetaldehyde

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Introduction

The common green lacewing, *Chrysoperla carnea sensu lato* (Neuroptera: Chrysopidae), is one of the most important lacewing species used in biological control. Adults of some genera and all lacewing larvae feed on pest aphids, scales, caterpillars, and other pests of many crops (McEwen et al. 2001). A synthetic attractant for green lacewings might be useful for monitoring lacewing abundance or manipulating lacewing population densities (for a review, see Szentkirályi 2002).

Lacewings of different genera (*Chrysopa* sp. and *Peyerimhoffina* sp.) are highly attracted to semiochemicals that lacewings produce themselves (Hooper et al. 2002). In *Chrysopa oculata* Say, a semiochemical apparently secreted by elliptical epidermal glands found only on the male abdominal sternites (Zhang et al. 2004) is a powerful aggregation pheromone (Chauhan et al. 2007). Although specific pheromones have yet to be found for lacewings in the genus *Chrysoperla*, several chemicals occurring in the

scent of several flowers attract *Chrysoperla* and other lacewings. 3-Methyl eugenol (Suda and Cunningham 1970; Umeya and Hirao 1975), methyl salicylate (Molleman et al. 1997; James 2003, 2006; James and Price 2004), β -caryophyllene (Flint et al. 1979), and 2-phenylethanol (Zhu et al. 1999, 2005) have all been reported to attract *C. carnea* or some other species of Chrysopidae in the field. Recently, we also discovered that phenylacetaldehyde was attractive to *C. carnea* in field tests in Hungary and Italy (Tóth et al. 2006). The objective of the present study was to optimize multicomponent attractant blends with phenylacetaldehyde as the core component. We investigated new, optimized attractant combinations and searched for synergists by using electroantennographic and field screening techniques and by utilizing data from other synthetic lacewing attractants known from literature. In addition, the most attractive combination of volatiles was tested for its attractivity in several areas in Europe.

Methods and Materials

Electroantennograms Insects for electroantennogram (EAG) analyses were collected from the edge of an oak forest at Julianna major (Budapest, Hungary). To present the stimuli to the antenna during EAG studies, a stainless steel tube (Teflon-coated inside) with a constant humidified airflow of ca. 0.7 l/min was set up. An antenna was freshly amputated at the base from a live lacewing and mounted between two glass capillaries containing 0.1 N KCl solution. The mounted antenna was placed at ca. 3 mm distance from the out-coming airflow. One of the electrodes was grounded, while the other was connected to a high impedance DC amplifier (IDAC-232, Syntech, Hilversum, The Netherlands). Test compounds (10 μ g each) were administered in hexane solution onto a 10 \times 10 mm piece of filter paper inside a Pasteur pipette. Stimuli consisted of pushing 1 ml of air through the Pasteur pipette into the airstream flowing towards the antenna. Response amplitudes were normalized against the means of responses to phenethyl alcohol (eliciting medium high responses from antennae), which was tested before and after the test compounds. Stimuli were administered at ca. 20–30 s intervals.

Trap Types For field studies conducted in Hungary, we used standard CSALOMON[®] VARL+ funnel traps (www.julia-nki.hu/traps) produced by the Plant Protection Institute, HAS (Budapest, Hungary). This trap type was developed originally for capturing moths (Tóth et al. 2000; Subchev et al. 2003) but later proved to be suitable for the green lacewing (Tóth et al. 2006). The trap consists of an opaque plastic funnel (top opening outer diameter, 13 cm; funnel hole

diameter, 3 cm; height of funnel, 16 cm), with a 20 \times 20 cm flat plastic roof. The trap also has a round transparent plastic collection container (ca. 1 l capacity) attached to the bottom funnel by a rubber band. The bait was suspended from the middle of the roof and positioned slightly above the level of the upper edge of the large funnel opening. A small piece (1 \times 1 cm) of household anti-moth strip (Chemotox[®], Sara Lee, Temana Intl. Ltd, Slough, UK; active ingredient 15% dichlorvos) was placed in the collection container to kill the captured insects.

For trials conducted in Italy, we used sticky delta traps (the standard CSALOMON[®] RAG traps produced by Plant Protection Institute, HAS, Budapest, Hungary; Szöcs 1993; Tóth and Szöcs 1993; www.julia-nki.hu/traps). In Norway, we used delta traps with sticky bottoms produced by PheroNet AB, Lund, Sweden.

Baits All synthetic compounds (>95% chemical purity as per the manufacturer) were obtained from Sigma-Aldrich Kft. (Budapest, Hungary). For preparing the baits, compounds were loaded onto a 1-cm piece of dental roll (Celluron[®], Paul Hartmann AG, Heidenheim, Germany), which was put into a polyethylene bag (ca. 1.0 \times 1.5 cm) made of 0.02 mm linear polyethylene foil (FS471-072, Phoenixplast BT, Pécs, Hungary). Loaded dose of single compounds was kept at 100 mg each, unless otherwise stated in the respective experiments. The dispensers were heat sealed and attached to 8 \times 1 cm plastic handles for easy handling when assembling the traps. Dispensers were wrapped singly in pieces of aluminum foil and stored at -18°C until used. In the field, baits were changed at 2 to 3-week intervals, as previous experience showed that they do not lose their attractiveness during this period (Tóth et al. 2006)

Field Tests Field comparison tests were conducted routinely by using the methods applied in similar studies on pheromones and attractants (e.g., Roelofs and Cardé 1977; Arn et al. 1986). Trapping experiments were conducted in fruit orchards. Green lacewings are frequently seen flying at 1.5–2.0 m in the crown of trees, thus traps were suspended in the trees at a height of 1.5–1.7 m above ground. One trap of each treatment was incorporated into a block so that individual treatments were 5–8 m apart (according to the distance between trees at the respective sites). Blocks were situated 15–20 m apart. Unless otherwise stated, five blocks of traps were applied in an experiment, and traps were inspected twice weekly. In Hungary, field tests were conducted at Halásztelek, in a sour cherry orchard, and at Érd-Elviramajor, in a cherry orchard (both sites situated in Pest county). In Italy, field tests were conducted in an apricot orchard near Rotondella (southern Italy), and in Norway, traps were placed in sweet cherry orchards in two

different areas in Ås (southeastern Norway) and Ullensvang (western Norway).

Experimental Details

Experiments 1A and 1B The objective of these preliminary tests was to study the influence of acetic acid added to phenylacetaldehyde. These tests were originally aimed at catching noctuid moths (Lepidoptera: Noctuidae). Treatments included traps baited with phenylacetaldehyde on its own [a well-known moth attractant (e.g., Creighton et al. 1973; Cantelo and Jacobson 1979)], or phenylacetaldehyde plus acetic acid (1:1). In the treatment containing both compounds in experiment 1A, acetic acid and phenylacetaldehyde were loaded into separate dispensers (so in this case, traps with this treatment contained two dispensers), whereas in experiment 1B, the two compounds were loaded into a single dispenser. Both tests were conducted at Halásztelek; experiment 1A was carried out from 9 May to 10 June, 2003, whereas experiment 1B was carried out from 10 June to 23 September, 2003. There were five blocks in the former and six blocks in the latter.

Experiment 2 The objective was to study the optimal ratio of acetic acid added to phenylacetaldehyde. Treatments included traps baited with phenylacetaldehyde or acetic acid alone, and their binary mixtures in 10:1, 1:1, and 1:10 ratios. The test was conducted at Halásztelek from 11 August to 24 August, 2003.

Experiment 3 In this test, we studied the activity of methyl salicylate or β -caryophyllene alone or in addition with the optimal blend of phenylacetaldehyde and acetic acid. Treatments included methyl salicylate or β -caryophyllene alone, the binary mixture of phenylacetaldehyde and acetic acid (1:1), ternary mixtures of phenylacetaldehyde, acetic acid, and β -caryophyllene or methyl salicylate, and unbaited traps. The test was conducted at Érd-Elviramajor from 14 June to 23 July, 2004.

Experiment 4 This test was designed to study the influence of varying doses of methyl salicylate added to the optimal phenylacetaldehyde + acetic acid blend. Traps baited with methyl salicylate on its own, the binary phenylacetaldehyde + acetic acid blend, and ternary mixtures of phenylacetaldehyde + acetic acid + methyl salicylate (1:1:1, 10:10:1, 1:1:10) and unbaited controls were set out. An extra treatment which contained the phenylacetaldehyde + acetic acid blend plus methylsalicylate in separate dispensers was also tested. The test was conducted at Érd-Elviramajor from 23 July to 13 October, 2004.

Experiment 5 This test was aimed at comparing efficacy of the phenylacetaldehyde + acetic acid + methyl salicylate ternary blend with that of the known lacewing attractants 2-phenylethanol and 3-methyl eugenol. The test consisted of the treatments 2-phenylethanol and 3-methyl eugenol on their own, the ternary phenylacetaldehyde + acetic acid + methyl salicylate blend, and unbaited traps. The test was conducted at Érd-Elviramajor from 4 to 19 July, 2005.

Experiment 6 The objective was to study the influence of 2-phenylethanol and 3-methyl eugenol when added to the ternary phenylacetaldehyde + acetic acid + methyl salicylate blend. Treatments included the ternary phenylacetaldehyde + acetic acid + methyl salicylate mixture (1:1:1), quaternary blends of these compounds with 2-phenylethanol or 3-methyl eugenol added (1:1:1:1), a blend of all five compounds together (1:1:1:1:1), and unbaited traps. The test was conducted at Érd-Elviramajor from 19 July to 16 August, 2005.

Experiment 7 In this test, we studied the activity of cinnamaldehyde and methyl anthranilate, both compounds with high EAG activity. Traps with (*E*)-cinnamaldehyde or methyl anthranilate on their own were compared to traps with the ternary phenylacetaldehyde + acetic acid + methyl salicylate mixture, the quaternary blends containing (*E*)-cinnamaldehyde or methyl anthranilate as fourth components, and unbaited traps. The test was conducted at Halásztelek from 1 June to 13 July, 2007.

Experiments 8 and 9 The objective of these tests was to compare the relative importance of the single components in the ternary blend of phenylacetaldehyde, acetic acid, and methyl salicylate. In both experiments, treatments included phenylacetaldehyde on its own, its binary combinations with acetic acid or methyl salicylate (1:1), and the ternary combination (1:1:1). Experiment 9 also included unbaited traps. The tests were conducted at Érd-Elviramajor from 16 August to 12 September, 2005 (experiment 8) and at Halásztelek from 13 July to 21 August, 2007 (experiment 9).

Activity of the Optimized Attractant in Different European Regions Apart from the comparison tests performed in Hungary (central Europe), the optimized ternary attractant of phenylacetaldehyde + acetic acid + methyl salicylate (1:1:1) also was tested at sites in southern Europe (Italy) and in northern Europe (Norway). The purpose of these tests was to confirm attraction of *C. carnea* in these regions.

In Italy, five traps with bait dispensers and five unbaited control traps were checked for adult *C. carnea* weekly between 14th April and 27th July.

In Ullensvang (western Norway), ten traps with bait dispensers and ten unbaited control traps were checked for

adult *C. carnea* four times between 16th May and 15th June. From 16th June until 12th September 3, traps with bait dispensers and five control traps were checked for adults 12 times. Further, these traps were checked for *C. carnea* eggs twice between 16th May and 15th June (ten baited traps and ten control traps) and 12 times between 16th June and 12th September (five baited traps and five control traps). In Søråsjordet, Ås (southeastern Norway), five traps with bait dispensers and one unbaited control trap were checked ten times for both adults and eggs between 29th May and 8th August. A second field in this area, Norderåshagen, had seven traps and one control trap and was checked five times from 29th May until 13th June.

Identification of Species Within the *C. carnea* Group The widespread and abundant *C. carnea s.l.* is composed of several sibling, cryptic, and sympatric species (Henry et al. 2001). For their identification, we used several morphological traits of adults (Thierry et al. 1992, 1998; Henry et al. 1996, 2002, 2003). As we were aware of the debates about a valid nomenclature within the *C. carnea* group (Canard and Thierry 2005), we accepted the species names given by Henry et al. (1996, 2002, 2003).

Statistical Analyses The data units for the field experiments were number of insects caught/inspection. The catches from field trapping tests were transformed by using $(x+0.5)^{1/2}$ as suggested by Roelofs and Cardé (1977) for similar experiments. In many field trapping tests, trap catches are below ten or even zero, so this or a similar transformation is desirable (Tukey 1949, 1955). Data were analyzed by a Student's *t* test (two treatments only) or analysis of variance (ANOVA; more than two treatments). Treatment means were separated by the Games–Howell Test (Games and Howell 1976; Jaccard et al. 1984). Where one of the treatments caught no insects, the Bonferroni–Dunn test (Dunn 1961) was used to check that mean catches in other treatments were not significantly different from zero catch (see also table and figure legends). To analyze EAG responses, we used ANOVA followed by Fisher's protected least significant difference (LSD) for significance levels (Fisher 1966). All statistical procedures were conducted by using the software packages StatView® v4.01 and SuperANOVA® v1.11 (Abacus Concepts, Berkeley, CA, USA).

Results

EAG Response Spectra Both female and male antennae showed similar response intensities to the range of compounds tested (Fig. 1). The largest responses were evoked by (*E*)-cinnamaldehyde, and these responses were signifi-

cantly different from all other compounds. This was followed in tendency by responses to methyl anthranilate, methyl salicylate, β -ionone, benzyl alcohol, and 2-phenylethanol, but responses to these compounds were not significantly higher than those to the phenethyl alcohol standard. Phenylacetaldehyde evoked medium large responses similar to those of the standard. The rest of the compounds evoked rather uniform, low to medium EAG responses.

Field Comparison Tests, Hungary In experiment 1A, traps baited with the binary combination of phenylacetaldehyde and acetic acid caught significantly more green lacewings than traps with only phenylacetaldehyde throughout the test period. Mean catches/trap/inspection (\pm SE) were 3.31 ± 0.60 vs. 1.56 ± 0.37 , respectively ($P=0.011$ by Student's *t* test). Similar results were recorded in experiment 1B, the respective mean catches were 6.69 ± 0.41 vs. 4.70 ± 0.34 specimens ($P=0.002$ by Student's *t* test). Unbaited control traps caught a mean (\pm SE) of 0.00 ± 0.00 (experiment 1A) or 0.02 ± 0.01 (experiment 1B) lacewings, respectively.

In the ratio test (experiment 2), traps with the 1:1 blend of phenylacetaldehyde + acetic acid caught the highest number of *C. carnea*, and mean catch in these traps was significantly higher compared to traps with phenylacetaldehyde alone (Fig. 2). The 10:1 blend produced intermediate catches. Very little was captured with the 1:10 blend, and zero catches were recorded in traps with acetic acid only.

The known lacewing attractant β -caryophyllene (Flint et al. 1979) did not catch more than unbaited controls and did not influence catches when added to the binary phenylacetaldehyde + acetic acid combination (Table 1, experiment 3). Methyl salicylate (Molleman et al. 1997; James 2003) was also inactive on its own; however, it increased catches significantly when added to the binary phenylacetaldehyde + acetic acid blend (Table 1, experiment 3).

The synergistic action of methyl salicylate was confirmed in experiment 4 (Fig. 3). Addition of methyl salicylate in equal or ten times lower amounts compared to the other two compounds in the ternary blend produced higher catches than the binary phenylacetaldehyde + acetic acid mixture. Similarly high catches were observed when methyl salicylate was added in a separate dispenser or loaded together in the same dispenser. When the amount of phenylacetaldehyde + acetic acid was decreased in the ternary blend, catches decreased. Methyl salicylate on its own again showed no activity (Fig. 3).

When comparing efficacy of the 1:1:1 ternary blend with that of the described lacewing attractants 2-phenylethanol (Zhu et al. 1999, 2005) and 3-methyl eugenol (Suda and Cunningham 1970; Umeya and Hirao 1975), the ternary blend caught approximately one magnitude more than the single compounds, although both 2-phenylethanol and 3-

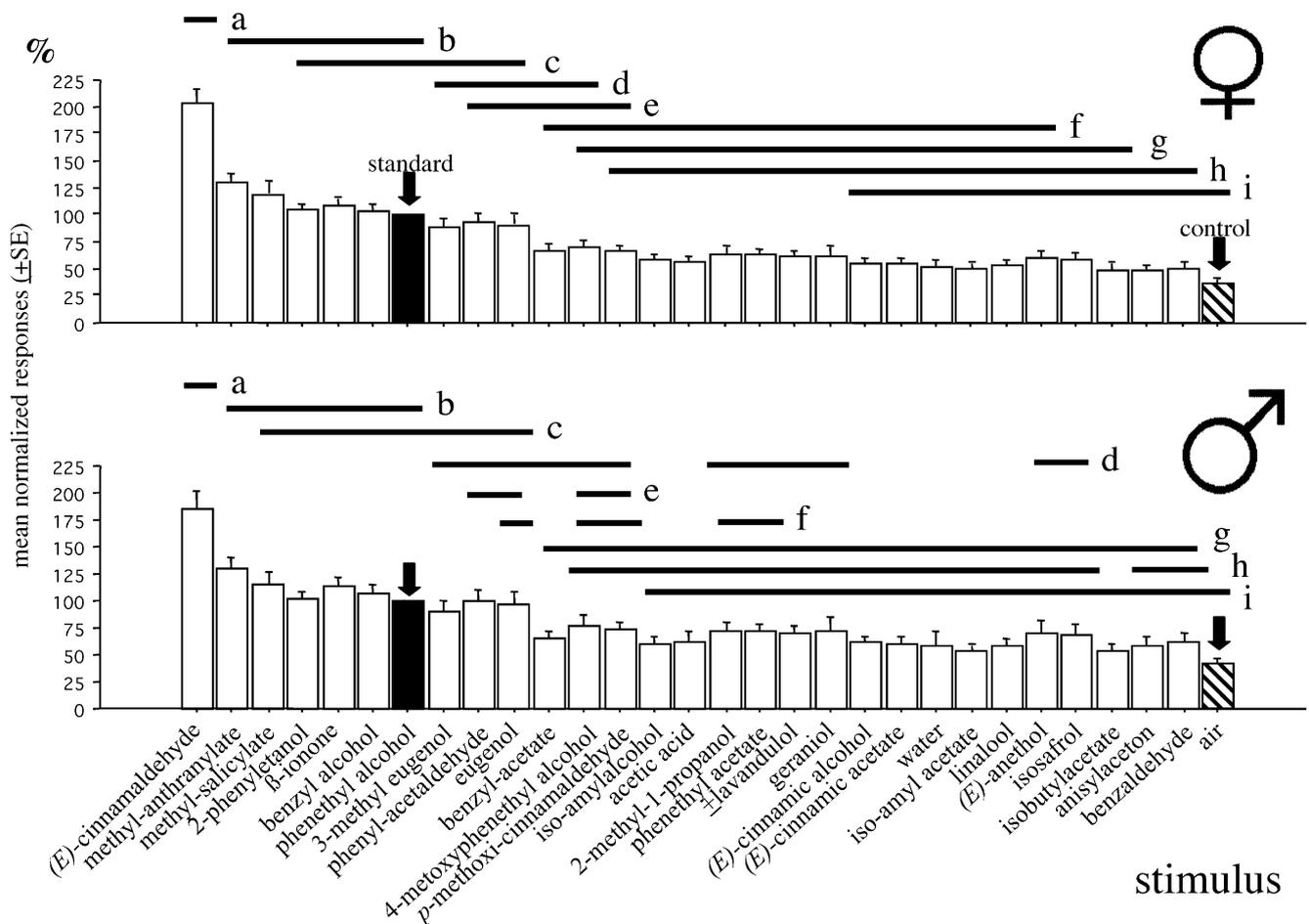


Fig. 1 Normalized EAG responses of female or male green lacewing antennae to a range of attractant candidate compounds. Columns show mean responses of antennae from nine female or 11 male insects tested. Responses were normalized to the response evoked by the

standard phenethyl alcohol. Columns with horizontal bars not significantly different ($\alpha=0.05$) by ANOVA, Fisher's Protected LSD test

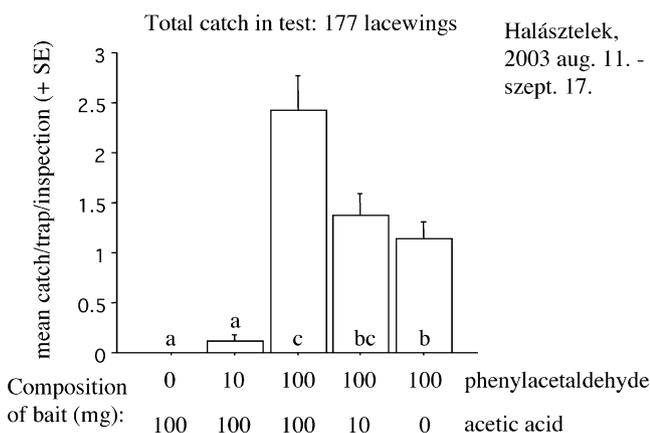


Fig. 2 Mean catches of green lacewings in traps baited with blends in different ratios of phenylacetaldehyde and acetic acid or with the single compounds (experiment 2; total caught in test, 177 lacewings). Columns with same letter not significantly different ($\alpha=0.05$) by ANOVA, Games–Howell test. Significant difference from zero catch (acetic acid on its own) was tested by Bonferroni–Dunn test

methyl eugenol showed significantly increased catches as compared to unbaited controls (Table 1, experiment 5). However, when these compounds were added to the ternary blend, no influence on catches was observed (Table 1, experiment 6).

Finally, when the activity of (*E*)-cinnamaldehyde and methyl anthranilate, both evoking high EAG responses from antennae of both sexes (Fig. 1) were studied, the single compounds showed significant attraction as compared to the unbaited traps (Table 1, experiment 7). However, their addition to the ternary blend did not increase catches. In fact, the quaternary mixture containing methyl anthranilate caught less than the ternary blend.

Subtraction tests aimed to study the relative importance of the components of the ternary phenylacetaldehyde + acetic acid + methyl salicylate blend yielded similar trends of catches in both experiment 8 and 9 (Fig. 4). Lowest catches were recorded with the acetic acid + methyl salicylate binary combination; however, catches in these traps were higher than in unbaited traps (in experiment 9, where unbaited traps

Table 1 Catches of green lacewings, *Chrysoperla* spp., in traps baited with synthetic attractant candidates and their mixtures or in unbaited traps at various sites in Europe

Bait composition (mg)		Mean catch/trap/inspection									
Phenylacet- aldehyde	Acetic acid	Methyl salicylate	β -Caryo- phyllene	2-Phenyl- ethanol	3-Methyl eugenol	Cinnam- aldehyde	Methyl anthranilate	Experiment 3	Experiment 5	Experiment 6	Experiment 7
100	100	–	–	–	–	–	–	4.90b	n.t.	n.t.	n.t.
100	100	100	–	–	–	–	–	7.21c	8.95a	4.38a	10.02d
–	–	100	–	–	–	–	–	0.32a	n.t.	n.t.	n.t.
100	100	–	100	–	–	–	–	3.39b	n.t.	n.t.	n.t.
–	–	–	100	–	–	–	–	0.03a	n.t.	n.t.	n.t.
–	–	–	–	100	–	–	–	n.t.	1.15b	n.t.	n.t.
–	–	–	–	–	100	–	–	n.t.	1.05b	n.t.	n.t.
100	100	100	–	100	–	–	–	n.t.	n.t.	5.43a	n.t.
100	100	100	–	–	100	–	–	n.t.	n.t.	4.91a	n.t.
100	100	100	–	100	100	–	–	n.t.	n.t.	4.66a	n.t.
–	–	–	–	–	–	100	–	n.t.	n.t.	n.t.	4.78c
100	100	100	–	–	–	100	–	n.t.	n.t.	n.t.	11.08d
–	–	–	–	–	–	–	100	n.t.	n.t.	n.t.	2.22b
100	100	100	–	–	–	–	100	n.t.	n.t.	n.t.	2.93b
–	–	–	–	–	–	–	–	0.09a	0.10c	0.03b	0.06a
Total number of lacewings caught in test:								1822	225	675	1864

Means with same letter within one column are not significantly different ($\alpha=0.05$) by ANOVA, Games–Howell

n.t. not tested

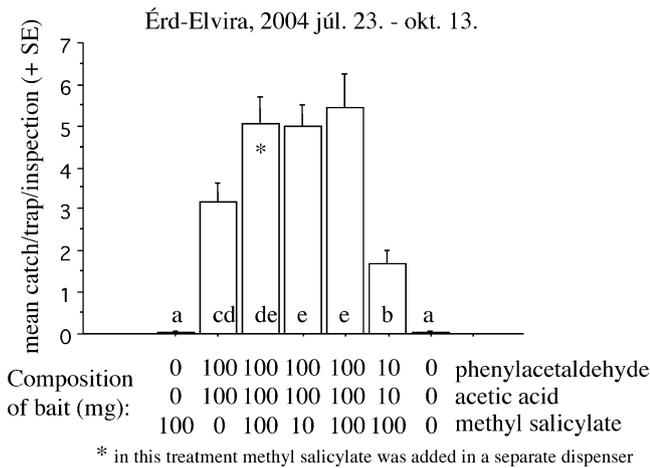
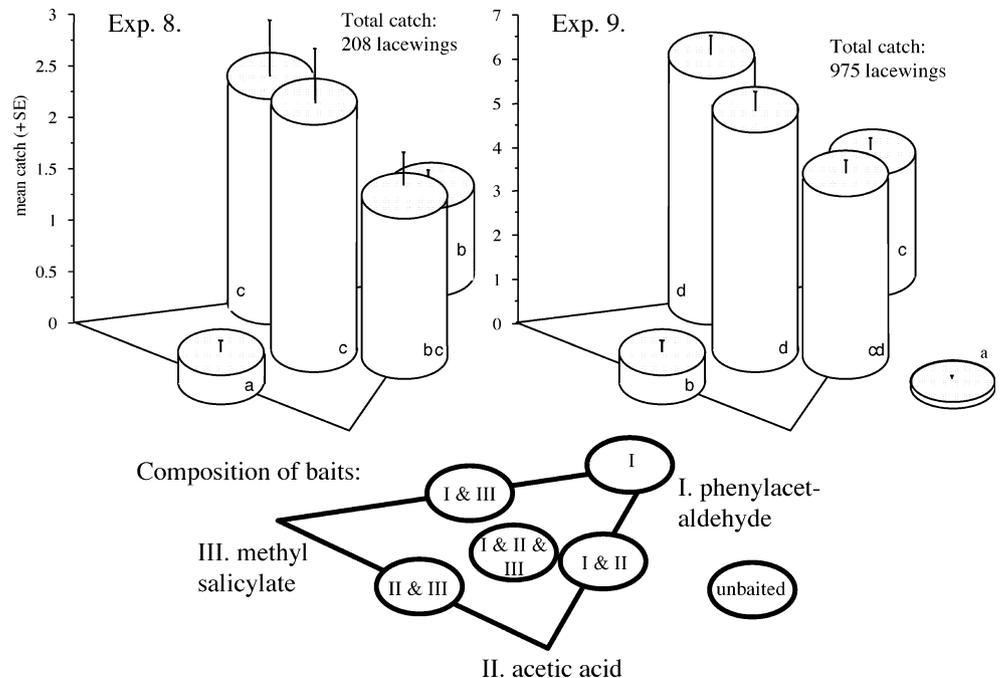


Fig. 3 Mean catches of green lacewings in traps baited with blends of phenylacetaldehyde, acetic acid, and methyl salicylate in different ratios or with the single compounds (experiment 4; total caught in test, 1,210 lacewings). Columns with same letter not significantly different ($\alpha=0.05$) by ANOVA, Games–Howell test

were also operated). Phenylacetaldehyde on its own produced medium high catches. The binary phenylacetaldehyde + acetic acid blend showed a tendency of catching more than phenylacetaldehyde on its own. Largest catches were observed in the binary phenylacetaldehyde + methyl salicylate mixture or the ternary blend, and no significant difference was found between these blends.

Among randomly selected specimens caught in experiments 8 and 9 (138 and 941, respectively), 30.0% and 62.2% were females. There was no apparent difference in ratio of sexes caught among the different treatments.

Fig. 4 Mean catches of green lacewings in traps baited with binary and ternary blends of phenylacetaldehyde, acetic acid, and methyl salicylate or with phenylacetaldehyde by itself (experiments 8 and 9; total caught in tests, 208 and 975 lacewings, respectively). Columns with same letter not significantly different ($\alpha=0.05$) by ANOVA, Games–Howell test



A determination effort of a random sample of 205 (experiment 8, 92% out of a total of 222 captured) and 559 (experiment 9, 50.4% out of a total of 1,109 captured) lacewing specimens resulted in 84.1% and 49.1% of *C. carnea* s. str. (Stephens), 11.6% and 39.1% of *Chrysoperla lucasina* Lacroix, 4.3% and 9.9% of *Chrysoperla pallida* (Henry et al. 2002), respectively. The remaining 3.5% of specimens from experiment 9 were morphologically similar to *Chrysoperla agilis* (Henry et al. 2002); however, the occurrence of this species in the Hungarian fauna has not been reported previously. To sum it all up, all captured specimens investigated belonged to the *C. carnea* group.

Activity of the Optimized Attractant in Different European Regions In Rotondella, a total of 128 adult *C. carnea* were caught (83 females, 16 males, and 29 unsexed specimens) in traps with the ternary lure between 14th April and 27th July (16 inspections on five traps) resulting in a mean (\pm SE) of 1.60 ± 0.18 adults/trap/inspection. Only five adults (four females and one male) were found in unbaited control traps.

In Ullensvang, Western Norway, a total of 18 adult *C. carnea* were caught in traps with the ternary lure between 16th May and 15th June (four inspections on ten traps) resulting in a mean (\pm SE) of 0.45 ± 0.19 adults/trap/inspection. Between 16th June and 12th September, a total of 14 adults were caught (12 inspections on three traps) resulting in a mean (\pm SE) of 0.23 ± 0.10 . Further, a total of 287 eggs were found inside ten delta traps with baits between 16th May and 15th June, and a total of 26 eggs were found inside five delta traps with baits between 16th June and 12th September. No adults or eggs were found in

unbaited control traps. In Søråsjordet, Ås (southeastern Norway), a total of 63 adults were caught in delta traps with the ternary lures between 29th May and 8th October (10 inspections on 5 traps) resulting in a mean (\pm SE) of 1.26 ± 0.22 adults/trap/inspection. In Norderåshagen, Ås (five inspections on seven traps), a total of 137 adults were caught between 29th May and 13th June (3.91 ± 0.45 adults/trap/inspection). A total number of 99 and 156 eggs were found in the two fields, respectively, inside the traps during the corresponding periods. In the unbaited control traps, a total of one adult and two eggs were found.

Discussion

In this study, the addition of acetic acid clearly enhanced the behavioral activity of the previously known synthetic attractant phenylacetaldehyde. Acetic acid is known to be weakly attractive to several insect species (i.e., Yothers 1927; Keiser et al. 1976; Casana-Giner et al. 1999). Attractant combinations of acetic acid and other food- or host-derived volatile compounds have been reported to show increased activity in yellowjacket wasps (Landolt 1998), in noctuid or pyralid moths (Landolt and Alfaro 2001; Tóth et al. 2002; Landolt 2005), and, more recently, in the codling moth (Landolt et al. 2007). The presence of acetic acid usually is thought to indicate fermentation products, and attraction to acetic acid-emitting sources has been explained as orientation towards food sources by insects that feed on fermenting materials (Utrio and Eriksson 1977; Landolt et al. 2007). A similar hypothesis can explain the increased response of green lacewings to blends containing acetic acid.

Phenylacetaldehyde will produce (*E*)-2,4-diphenylbut-2-enal in the presence of acetic acid (Goetz et al. 1990). This reaction can be expected to take place when both compounds are loaded into the same dispenser. Because the addition of acetic acid resulted in a significant increase in catches both when the two compounds were presented in two different dispensers (experiment 1A) or when they were loaded into the same dispenser (experiment 1B), it suggests that the possible formation of (*E*)-2,4-diphenylbut-2-enal had no dramatic effect on behavioral activity. In this study, no attempt was made to examine in detail the possible influence of (*E*)-2,4-diphenylbut-2-enal on the activity of the phenylacetaldehyde + acetic acid blend, but this interaction should be clarified in future experiments.

We demonstrated in this work that trap catches to the binary mixture of phenylacetaldehyde and acetic acid can be further increased by the addition of methyl salicylate. This compound attracted *C. carnea* green lacewings in a test originally aimed at predators of the pear psylla

(Molleman et al. 1997) and was also attractive to *Chrysopa nigricornis* Burmeister (James 2003). We did not observe any significant attraction to methyl salicylate on its own (Table 1, Fig. 3).

The compound β -caryophyllene has earlier been described as an attractant for *C. carnea* (Flint et al. 1979), but we did not confirm these findings. Caryophyllene has also been found to be behaviorally inactive in other studies (Zhu et al. 2005; Tóth et al. 2006). The reason for this discrepancy in attraction is not clear.

3-Methyl eugenol, which has been reported as attractive for some chrysopid spp. (Suda and Cunningham 1970; Umeya and Hirao 1975), showed some activity with *C. carnea* when presented alone in our tests. The attraction of *C. carnea* to 2-phenylethanol presented alone (e.g., Zhu et al. 2005) was also confirmed in our study. However, none of the above described compounds increased catches of *C. carnea* when added to the ternary blend optimized in this study.

We screened synthetic compounds by EAG to look for attractant candidates, as suggested by Dodds and McEwen (1998). Both compounds that evoked the highest responses, (*E*)-cinnamaldehyde and methyl anthranilate, were attractive in the field when presented alone. To our knowledge, neither has been reported to be attractive to lacewings. However, when (*E*)-cinnamaldehyde was added to the ternary blend optimized in this study, we observed no impact on the trap catches. Addition of methyl anthranilate possibly reduced catches (although this phenomenon should be confirmed through further testing).

There are other potential semiochemicals that might bear further study. For example, in a laboratory olfactometer bioassay, Van Emden and Hagen (1976) reported that tethered female green lacewings spent ca. 70% of the total time flying towards reaction mixtures thought to produce indole acetaldehyde, which led the authors to suggest that indole acetaldehyde was a lacewing attractant. However, Van Emden and Hagen (1976) never tested the synthesized pure compound in the laboratory or in the field.

Based on our results, it appears that the ternary blend described in this study is the most powerful synthetic attractant reported to date for *C. carnea*. Green lacewing populations from southern to northern and central Europe responded equally well in this study, and there was no indication of geographical differences in response, so this ternary attractant may prove to have widespread applicability. It remains to be seen whether *C. carnea* populations outside of Europe will also be as responsive.

From the subtraction tests of the ternary blend (experiments 8 and 9), it appears that the removal of phenylacetaldehyde had the most impact on the activity, whereas the removal of methyl salicylate resulted in a quantitative but not significantly lower reduction in trap catch. Removal of acetic acid did not lower trap catch. The combination of

methyl salicylate and phenylacetaldehyde was two to three times more attractive than phenylacetaldehyde alone was. More detailed studies should be carried out to evaluate whether acetic acid can be omitted altogether without seriously affecting efficiency.

Most insect predators show omnivorous feeding behavior, i.e., by feeding directly or indirectly on plant materials such as pollen or nectar (Jervis and Kidd 1996; Villenave et al. 2005). Green lacewings are adapted to unpredictable food supplies and larval habitats. Adult females perform obligatory migration flights where they do not react to olfactory stimuli. After the initial migration flight, females start responding to plant odors by a combination of appetitive down- and upwind flights (Duelli 1980, 2001).

The three “cryptic” lacewing species, *C. lucasina*, *C. pallida*, and *C. carnea* s. str. captured in baited traps in this study all belong to the *C. carnea* species complex. No other chrysopids were caught in significant numbers. An explanation could be that floral food sources are more important for green lacewing adults of genera *Chrysoperla* (and probably also for *Dichochrysa*), whose diet regime is palynoglyphic, than for other chrysopids (Canard 2001).

The attractant described in this study appears to be highly attractive to females, which is advantageous from the point of view of applicability in biological control efforts. *C. carnea* females attracted by our ternary attractant may spend a long time in the vicinity of the bait dispenser, searching for a food source and oviposition sites. Preliminary observations of eggs deposited in traps indicate that the ternary attractant may give rise to major bouts of oviposition. If confirmed in more extensive experiments, this response may be advantageous in biological control. A large number of lacewing larvae will reduce aphid populations on that particular plant. The ternary attractant might also increase biological control by increasing lacewing numbers in overwintering boxes, a method widely used by organic growers.

In the closely related *C. oculata*, the synthetic main component of the aggregation pheromone, (1*R*,2*S*,5*R*,8*R*)-iridodial, attracted many males into traps (Zhang et al. 2004). More recently, Chauhan et al. (2007) reported that although few females entered traps baited with iridodial, they found many female insects in the vicinity of the traps. As the pheromonal communication and host attraction systems of the *C. carnea* complex become better known, combinations of pheromones with floral attractants may provide optimal management tools to exert better biological control by this group of beneficial insects.

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