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VISUAL AND OLFACTORY CUES FOR CATCHING PARASITIC WASPS (HYMENOPTERA: SCOLIIDAE)

VUTS, J.[1], RAZOV, J.[2], KAYDAN, M. B.[3] and TÓTH, M.[1]

[1]*Plant Protection Institute, Centre for Agriculture Research, Hungarian Academy of Sciences H-2462 Martonvásár, Hungary; E-mails: joci0617@gmail.com, h2371tot@ella.hu*
[2]*Department of Mediterranean Agriculture and Aquaculture, University of Zadar Mihovila Pavlinovica bb. Zadar, HR-23000 Croatia; E-mail: jrazov@unizd.hr*
[3]*Faculty of Agriculture, Department of Plant Protection, Yüzüncü Yıl University Van, TR-65080 Turkey; E-mail: bkaydan@gmail.com*

Scoliid wasps (*Scolia* spp.) are day-flying flower visitors. Earlier field observations indicated preference of *Scolia hirta*, *S. flavifrons* and *S. sexmaculata* for a combination of a light blue colour and a quaternary floral blend [(*E*)-anethol, 3-methyl eugenol, 1-phenylethanol and lavandulol], constituents of traps used for catching the scarabs *Cetonia aurata aurata* and *Potosia cuprea*. Subsequent field experiments for *Scolia hirta*, *S. flavifrons* and *S. sexmaculata* in Hungary, Croatia and Turkey, respectively, comparing different colours and floral blends previously found efficient for catching several scarab species, confirmed observations on the preference of scoliids for the *Cetonia/Potosia* trap. When investigating the relative importance of the colour and the quaternary floral blend released from the *Cetonia/Potosia* trap for *S. hirta*, joint application of the visual and chemical cues resulted in a significant increase of catches, compared to those in traps containing only one of the cues, or in unbaited traps with no colour. An explanation of the activity of the *Cetonia/Potosia* trap on scoliid wasps can be that, since the wasps feed on flowers and the bait of the *Cetonia/Potosia* trap comprises common floral compounds, it represents olfactory stimuli essential in host plant-finding. Role of the blue colour also seems to be important, however, more detailed comparative studies, including different colours and chemicals, are necessary to better understand interactions mediating host plant-finding. Possibilities of practical application of the *Cetonia/Potosia* trap for monitoring *S. hirta* and *S. flavifrons* are discussed.

Key words: floral compounds, colour, synergism, attractant, trapping, biological control, Scarabaeidae, Cetoniinae

INTRODUCTION

Many species of day-flying insects are flower visitors, using various sensory cues to locate the flowers (RAGUSO 2004, CHITTKA & RAINE 2006). Visual and olfactory cues from food sources for adults of numerous parasitic wasp species are well documented, although their relative importance is still unclear (e.g., GODFRAY 1994, HILKER & MCNEIL 2008). Solitary wasps in the family Scoliidae (Hymenoptera) (known as scoliid wasps or dagger wasps) are active during the day, and preferably feed on plants with composite flowers, umbels, panicles or other aggregated flower heads, including *Daucus* (Apiaceae), *Solidago, Cirsium* (Asteraceae), *Melilotus* (Fabaceae), *Thymus, Origanum* (Lamiaceae) and *Veronica* (Plantaginaceae) spp. (OSTEN 2000, LANDECK 2002, VEREECKEN & CARRIERE 2003). Nectar is particularly essential for females for their eggs to mature (BARRATT 2003).

Scoliid wasps are distributed worldwide, especially in the tropical and subtropical regions, and are parasitoids of different scarab and curculionid beetles (Coleoptera: Scarabaeidae; Curculionidae) (OSTEN 2005). In Europe, they are more common in drier, warmer areas with mostly sandy soil (e.g., the Mediterranean Basin) (N. BAJÁRI 1956, OSTEN 2000, FARD & ABINEH 2004, CHABROL 2007, SMIT 2007) where the soil-dwelling host larvae are more abundant and easier to access, thus their biotope preference is closely connected to that of the hosts (OSTEN 2000).

Trapping of day-flying insects with different combinations of colours and floral compounds is relatively well reported (e.g., IMAI *et al*. 2002, TÓTH *et al.* 2004, 2005). As for scoliids, field trials, investigating the visual and olfactory stimuli-mediated behaviour of several scarabs in the subfamily Cetoniinae with traps originally developed for the capture of *Epicometis hirta* PODA (TÓTH *et al*. 2004, SCHMERA *et al*. 2004), *Cetonia aurata aurata* L./*Potosia cuprea* F. (TÓTH *et al.* 2005, VUTS *et al*. 2010), and *Oxythyrea funesta* PODA (TÓTH *et al*. 2005, VUTS *et al*. 2008), indicated high numbers of the scoliid *Scolia hirta* SCHRANK in the *Cetonia/Potosia* traps (J. VUTS & M. TÓTH, unpubl. data). These traps comprise a light blue colour and the floral compounds (*E*)-anethol, 3-methyl eugenol, 1-phenylethanol and lavandulol (VUTS *et al.* 2010). Simultaneously, in Turkey and Croatia, two other species, *Scolia sexmaculata* var. *quadripunctata* F. and *Scolia flavifrons* var. *haemorrhoidalis* L., respectively, were also found in high numbers in the *Cetonia/Potosia* traps (J. VUTS and M. TÓTH, unpubl. data). In addition, captures of *S. hirta* were observed in Hungary in traps with a light blue colour and baited with a mixture of general floral compounds (phenethyl propionate, eugenol and geraniol) (M. TÓTH, pers. comm.).

Our aims in this study were twofold: to confirm preliminary observations on the activity of the visual and olfactory cues of the *Cetonia/Potosia* trap by carrying out comparative investigations on the colour and odour preference of *S. hirta*, *S. sexmaculata* and *S. flavifrons* in field trapping trials in Hungary, Turkey and Croatia, respectively, and to study the relative importance of the visual and olfactory cues of the *Cetonia/Potosia* trap for *S. hirta*. Development of a trap suitable for monitoring *Scolia* spp. could be useful in conservation and pest control practice.

MATERIALS AND METHODS

Baits. Preparation of the polythene bag dispensers used here has previously been described by SCHMERA *et al.* (2004). Dispensers were renewed at 3-week intervals as volatile activity was not reduced after several weeks of field exposure (M. TÓTH, G. SZŐCS and Z. IMREI, unpubl. data). Dispensers were wrapped singly in pieces of aluminum foil and were stored at –30 °C until use. Synthetic compounds were obtained from Sigma-Aldrich Kft. (Budapest, Hungary). All compounds were >95% pure.

Field tests. CSALOMON® modified funnel traps (codenamed as VARb3, and produced by MTA ATK Plant Protection Institute, Budapest, Hungary) were used and had proved efficient for the capture of different scarabs (i.e., IMREI *et al*. 2001, SCHMERA *et al*. 2004). Briefly, the trap is made of plastic and consists of two parts. The upper one is the capture surface providing chemical and visual stimuli, as well as a landing platform, whereas the lower part serves to hold captured insects.

Experiment 1–3: These tests were performed to study the importance of different colours and floral compounds on catches of *S. hirta, S. flavifrons* and *S. sexmaculata* in Hungary, Croatia and Turkey, respectively. Combinations of colour and floral volatile compounds, previously found to be suitable for the capture of scarab beetles, were tested, in order to confirm preliminary field observations on wasp occurrence in these traps. Period: Experiment 1, Mar 26–Sep 19 2008; Experiment 2, Apr 3–Aug 29 2008; Experiment 3, Apr 15–Aug 26 2008 (number of blocks: 4, number of traps/site: 12). Sites: Experiment 1, Telki, Hungary (HU); Experiment 2, Bascica, Croatia (HR); Experiment 3, Isparta, Turkey (TR). Biotope: Experiment 1, bushy edge of an oak forest; Experiments 2–3, edge of a mixed orchard. Upper funnels of traps developed for catching *C. aurata aurata/P. cuprea* (trap codenamed 'CE'; VUTS *et al*. 2010) and *E. hirta* (trap codenamed 'EP'; TÓTH *et al*. 2004, SCHMERA *et al*. 2004) were light blue (for reflectance spectrum, see SCHMERA *et al*. 2004) and differ with respect to the volatiles released (Table 1). Upper funnel of traps developed for catching *O. funesta* (trap codenamed 'OX'; TÓTH *et al*. 2005, VUTS *et al*. 2008) was fluorescent yellow (for reflectance spectrum, see JENSER *et al*. 2010). Combinations of colour and volatile compounds set up in experiment 1–3 are shown in Table 1.

Experiment 4: This preliminary test was carried out to investigate the effect on catches of elimination of the olfactory cue [the quaternary (*E*)-anethol, 3-methyl eugenol, 1-phenylethanol and lavandulol blend] from the CE trap, which caught more *S. hirta* in experiment 1. The treatments set up comprised: a combination of the colour plus the quaternary blend of the CE trap; colour without the quaternary blend. Period: Sept 10–22 2008 (number of blocks: 4, number of traps/site: 8). Site: Telki (HU). Biotope: bushy edge of an oak forest.

Experiment 5: The relative importance of the colour and the quaternary floral blend released from the CE trap was studied in detail for *S. hirta*. Period: Sept 7–23 2009 (number of blocks: 7, number of traps/site: 28). Site: Telki (HU). Biotope: bushy edge of an oak forest. The treatments set up comprised: a combination of the colour plus the quaternary blend of the CE trap; colour without the quaternary blend; the quaternary blend without colour (transparent); and transparent traps with no added colour and no volatiles.

Traps were set up in a randomized complete block design, with each block containing one replicate of every treatment. The distance between traps was 10–15 m, with a minimum distance of 25 m between blocks. Traps were set up in sunny places with no shade from trees or buildings, attached to poles at a 30–40 cm height above the ground. Traps were inspected twice weekly. Captured *Scolia* specimens were removed, their species identified and numbers recorded.

Statistics. The data units for the field experiments were number of insects caught/trap. As raw data did not fulfil requirements for parametric statistics, capture data were analysed by Kruskal-Wallis one-way analysis of variance, followed by pairwise comparisons by Mann-Whitney U test. All statistical procedures were conducted using the software GenStat 11th edition (VSN International Ltd).

Table 1. Treatments used in experiments 1–3

Trap code	Originally developed for the capture of	Bait composition (mg)						Trap colour
		(*E*)-Anethol	3-Methyl eugenol	1-Phenyl-ethanol	(*E*)-Cinnamyl alcohol	(±)-Lavandulol	2-Phenyl ethanol	
CE	*Cetonia aurata aurata* / *Potosia cuprea* (TÓTH *et al*. 2005, VUTS *et al*. 2010)	100	100	100	–	100	–	blue
OX	*Oxythyrea funesta* (TÓTH *et al*. 2005, VUTS *et al*. 2008)	–	–	–	–	200	200	fluorescent yellow
EP	*Epicometis hirta* (TÓTH *et al*. 2004, SCHMERA *et al*. 2004)	200	–	–	200	–	–	blue

RESULTS

Experiment 1–3. In Experiment 1 (Telki, HU), CE traps caught significantly more *S. hirta* than either the EP or OX traps, with the latter two not differing significantly from each other (Table 2). In Experiment 2 (Bascica, HR), greatest numbers of *S. flavifrons* were also observed in the CE trap, with catches in the other two treatments being significantly lower and not differing from each other (Table 2). In Experiment 3 (Isparta, TR), *S. sexmaculata* was caught only in low numbers, with catches showing a similar distribution to Experiments 1–2 (Table 2). The seasonal pattern of catches of *S. hirta*, *S. flavifrons* and *S. sexmaculata* at Telki (HU), Bascica (HR) and Isparta (TR), respectively, in the CE trap, is shown in Fig. 1.

Table 2. Catches of *Scolia* spp. in the CE, OX and EP traps in experiment 1-3. *P* values derive from paired comparisons by Mann-Whitney U test, preceded by Kruskal-Wallis one-way analysis of variance

Trap code	*S. hirta* [experiment 1, Telki (HU)]		*S. flavifrons* [experiment 2, Bascica (HR)]		*S. sexmaculata* [experiment 3, Isparta (TR)]	
	Total catch	*P* values from paired comparisons	Total catch	*P* values from paired comparisons	Total	*P* values from paired comparisons
CE	90	CE vs. EP: 0.046	20	CE vs. EP: 0.042	6	Due to low numbers caught, no *P* values were calculated
OX	1	EP vs. OX: 0.143	3	EP vs. OX: 0.229	0	
EP	20	OX vs. CE: 0.029	8	OX vs. CE: 0.029	3	
	$P < 0.008$ (by Kruskal-Wallis one-way analysis of variance)		$P < 0.001$ (by Kruskal-Wallis one-way analysis of variance)			

Table 3. Mean catches of *Scolia hirta* in experiment 5, testing the relative importance of the colour and the floral blend of the CE trap in different combinations. *P* values derive from paired comparisons by Mann-Whitney U test, preceded by Kruskal-Wallis one-way analysis of variance

Treatment	Total catch	*P* values from paired comparisons
1. colour + quaternary blend	25	Treatment 1 vs. 2: 0.048
2. colour, no quaternary blend	6	Treatment 1 vs. 3: 0.037 Treatment 1 vs. 4: 0.021
3. quaternary blend, no colour	1	Treatment 2 vs. 3: 0.315
4. no colour, no quaternary blend	0	Treatment 2 vs. 4: 0.192
$P < 0.014$ (by Kruskal-Wallis one-way analysis of variance)		Treatment 3 vs. 4: 1.000

Experiment 4. Elimination of the quaternary floral blend of the CE trap considerably reduced catches of *S. hirta*, although the difference was not significant (Kruskal-Wallis one-way analysis of variance, total catch: 75 wasps, $P = 0.141$; data not shown).

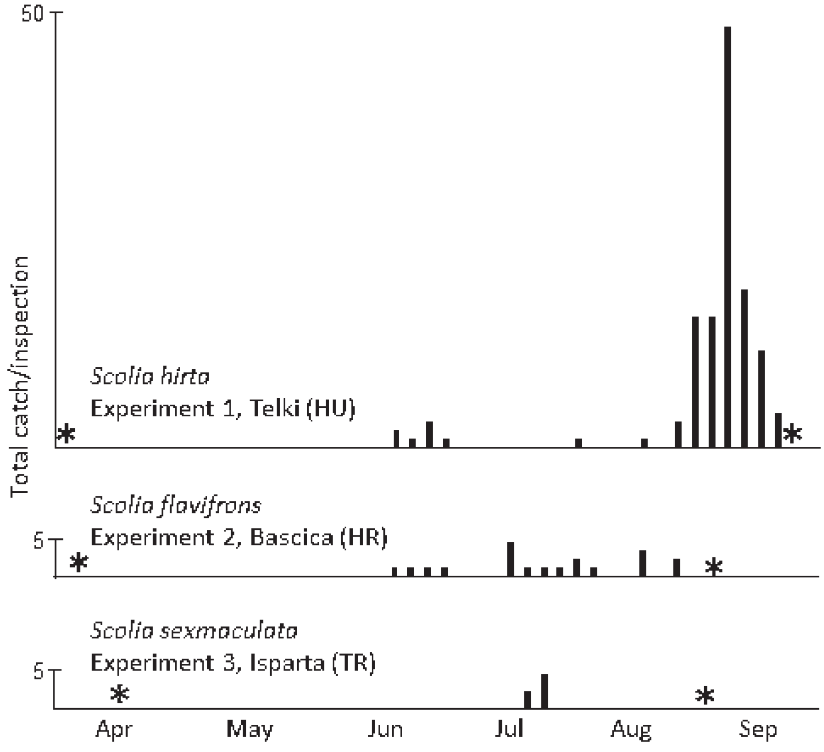

Fig. 1. Seasonal distribution of catches of *Scolia hirta*, *S. flavifrons* and *S. sexmaculata* at Telki (HU), Bascica (HR) and Isparta (TR), respectively, as observed in the CE trap. Asterisks within one diagram indicate the beginning and the end of the trapping period

Experiment 5. Numbers of *S. hirta* were significantly higher in traps with both a light blue colour and the quaternary blend, compared with traps comprising either the light blue colour or the quaternary blend, or unbaited transparent traps (Table 3).

DISCUSSION

An explanation for the similarly high numbers of *S. hirta, S. flavifrons* and *S. sexmaculata* in the CE trap can be that these wasps feed on flowers, and the bait of the CE trap comprises common floral compounds occurring in a number of plant families. The synthetic blend of the CE trap is assumed to represent olfactory stimuli essential in host plant-finding of flower-visiting scarabs (TÓTH *et al.* 2005, VUTS *et al.* 2010). A similar scenario could be outlined for scoliid wasps, since components of the blend can be found in many plant taxa (KNUDSEN *et al.* 1993, 2006), including families found to be attractive for different *Scolia* spp.

It has been reported that scoliid wasps prefer to visit flowers appearing blue to the human eye (OEHLKE 1974, WITT 1989, OSTEN 2000). In contrast, LANDECK (2002) found no colour preference. Although in experiment 1–3 of the present study, the three *Scolia* species were captured in high numbers in blue-coloured traps, these data do not allow for conclusions about colour preference yet. To address this question, experiments with different colours but the same lures will be necessary. At the sensory level, it can be assumed that scoliids, similar to many flower-visiting hymenopterans, i.e., members in the closely related Vespidae family (BROTHERS 1999), possess photoreceptors with a spectral sensitivity between 340 and 540 nm, which is the wavelength range of ultraviolet, blue and green (BRISCOE & CHITTKA 2001, LANDECK 2002).

When specifically investigating the importance of the visual (light blue colour) and olfactory (quaternary floral blend) cues of the CE trap for *S. hirta* in experiment 5, application of the floral blend together with the light blue colour synergistically increased catches, compared to those in traps with only one of the cues. Such synergism between colour and odour was reported, for example, for *E. hirta* (SCHMERA *et al.* 2004) or for *Rhagoletis cerasi* L. (Diptera: Tephritidae) (TÓTH *et al.* 2004). A comparative study testing a wider range of colours and plant volatile compounds would possibly lead to a greater understanding of the importance of different visual and olfactory cues for *S. hirta.* However, such an investigation would be limited by the fact that innate preferences of insects for certain key stimuli are typically modified or entirely overwritten by individual experience (e.g., GLINWOOD *et al*. 2011).

Seasonal distribution of *S. hirta* catches in the CE trap at Telki (HU) in 2008 shows a peak in the beginning of September, with some scarce numbers in the end of June, which is in agreement with the reported flight period of the species (June–September) (N. BAJÁRI 1956). *S. flavifrons* was found in the CE trap between June and August at Bascica (HR) in 2008, reflecting well literature data on the seasonal activity of the adults (N. BAJÁRI 1956). Finally, at Isparta (TR), the CE trap captured low numbers of *S. sexmaculata* in July. Although, according to literature data, occurrence of *S. sexmaculata* was detected accurately by the CE trap within its known flight period (June–August, N. BAJÁRI 1956), the low numbers caught do not allow for making explicit conclusions about the suitability of the CE trap for monitoring *S. sexmaculata* yet. To address this question, more detailed studies are needed to assess whether performance of the CE trap in 2008 for *S. sexmaculata* was connected to the lack of certain relevant visual and/or olfactory cues, or it was due to low abundance of local population in the experimental period.

Potential of the CE trap to catch *S. hirta* and *S. flavifrons* may make it suitable for various purposes: (i) monitoring distribution in areas where they are rare or endangered (see Red Book of Germany, for example), thereby contributing to enhanced conservation decisions; (ii) in the case of use as biological control agents against pest scarabs, monitoring with such trap could help evaluate population densities and distribution in treated areas to assess success of release, as reported for different *Scolia* spp. against pest dynastiins (Coleoptera: Scarabaeidae, Dynastinae) (SIMMONDS 1949, BEDFORD 1980), or related *Typhia* spp. against chafer grubs (ROGERS & POTTER 2002). The most common host species of *S. hirta*, *C. aurata aurata* and *P. cuprea*, are important pests (N. BAJÁRI 1956, HURPIN 1962, HOMONNAY & HOMONNAYNÉ-CSEHI 1990). *S. sexmaculata* parasites smaller cetoniins, mostly *Tropinota* and *Oxythyrea* spp. which can cause damage to fruit trees and ornamental plants (HURPIN 1962, VEREECKEN & CARRIERE 2003), while *S. flavifrons* is a parasitoid of large lamellicorn beetles, primarily *Oryctes nasicornis* Drury (Coleoptera: Scarabaeidae, Dynastinae) and *Lucanus cervus* L. (Coleoptera: Lucanidae, Lucaninae) (N. BAJÁRI 1956, NACHTIGALL 1992), but also of the pest *Polyphylla fullo* L. (Coleoptera: Scarabaeidae, Melolonthinae) (VEREECKEN & CARRIERE 2003). A better understanding of the factors mediating host-searching behaviour of these scoliid wasps, together with an effective monitoring tool, might facilitate the development of environmentally-sound pest control practices in areas where damage of their hosts occur, as well as might help to understand the possible evolutionary aspects and ecological relevance of these specific host-parasitoid interactions.

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