OIKOS

Research

Intraspecific variation in herbivore-induced plant volatiles influences the spatial range of plant–parasitoid interactions

Yavanna Aartsma, Benjamin Leroy, Wopke van der Werf, Marcel Dicke, Erik H. Poelman and Felix J. J. A. Bianchi

Y. Aartsma (http://orcid.org/0000-0002-2774-4611) (yavanna.aartsma@wur.nl), B. Leroy and F. J. J. A. Bianchi, Farming Systems Ecology, Wageningen Univ., Wageningen, the Netherlands. BL also at: Dept of Agroecology and Environment, ISARA Lyon, Lyon, France – YA, M. Dicke (http://orcid.org/0000-0001-8565-8896) and E. H. Poelman (http://orcid.org/0000-0003-3285-613X), Laboratory of Entomology, Wageningen Univ., Wageningen, the Netherlands. – YA and W. van der Werf, Centre for Crop Systems Analysis, Wageningen Univ., Wageningen, the Netherlands.

Oikos
00: 1–10, 2018
doi: 10.1111/oik.05151

Subject Editor: Marjo Saastamoinen
Editor-in-Chief: Dries Bonte
Accepted 9 July 2018

Chemical information influences the behaviour of many animals, thus affecting species interactions. Many animals forage for resources that are heterogeneously distributed in space and time, and have evolved foraging behaviour that utilizes information related to these resources. Herbivore-induced plant volatiles (HIPVs), emitted by plants upon herbivore attack, provide information on herbivory to various animal species, including parasitoids. Little is known about the spatial scale at which plants attract parasitoids via HIPVs under field conditions and how intraspecific variation in HIPV emission affects this spatial scale. Here, we investigated the spatial scale of parasitoid attraction to two cabbage accessions that differ in relative preference of the parasitoid *Cotesia glomerata* when plants were damaged by *Pieris brassicae* caterpillars. Parasitoids were released in a field experiment with plants at distances of up to 60 m from the release site using intervals between plants of 10 or 20 m to assess parasitism rates over time and distance. Additionally, we observed host-location behaviour of parasitoids in detail in a semi-field tent experiment with plant spacing up to 8 m. Plant accession strongly affected successful host location in field set-ups with 10 or 20 m intervals between plants. In the semi-field set-up, plant finding success by parasitoids decreased with increasing plant spacing, differed between plant accessions, and was higher for host-infested plants than for uninfested plants. We demonstrate that parasitoids can be attracted to herbivore-infested plants over large distances (10 m or 20 m) in the field, and that stronger plant attractiveness via HIPVs increases this distance (up to at least 20 m). Our study indicates that variation in plant traits can affect attraction distance, movement patterns of parasitoids, and ultimately spatial patterns of plant–insect interactions. It is therefore important to consider plant-trait variation in HIPVs when studying animal foraging behaviour and multi-trophic interactions in a spatial context.

Keywords: spatial ecology, animal behaviour, insect–plant interactions, plant odours

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Introduction

Organisms are usually confronted with resources that are scattered across space and time. The spatial scale at which organisms forage for resources depends – besides the spatial distribution of resources – on species traits, such as the capacity for movement and the ability to acquire information about the resource distribution from a distance (Nathan et al. 2008). Organisms use information (cues), including visual, acoustic and olfactory cues to make foraging decisions. In addition, foraging decisions can be influenced by the animal's internal state, such as hunger level, and learning (Bowler and Benton 2005, Clobert et al. 2009). Compared to large animals, smaller animals such as insects may experience spatial scales differentially. Although smaller animals need to gather information on the availability of resources just like large vertebrates, they sometimes have a limited capacity for directed movement and have a short lifespan to acquire and use information (Wajnberg 2006). For smaller animals, individual foraging behaviour is usually inferred from population patterns, leaving a knowledge gap on the possible use of information in individual foraging decisions (Holyoak et al. 2008). Yet, knowledge of how individual animals use information is important to understand at what spatial scale they interact with other individuals, both from the same and from other species.

A wide range of small animals have evolved foraging strategies based on olfactory cues (Raguso et al. 2015, Poelman and Kos 2016, Webster and Cardé 2016). Herbivore-induced plant volatiles (HIPVs), that are emitted by plants upon attack by herbivores, are remarkable olfactory cues in the sense that they provide indirect information on the presence of herbivores because they are produced by the plant under attack and not directly by the herbivores themselves (Vet and Dicke 1992, Hare 2011, Turlings and Erb 2018). HIPVs and other plant-derived volatiles are used during long-distance foraging by mammals (Finnerty et al. 2017), birds (Mäntylä et al. 2004, 2014, Amo et al. 2013), reptiles (Stork et al. 2011), and insects (Turlings and Erb 2018). Yet, HIPV use is particularly well studied for parasitoid wasps (Vet and Dicke 1992, Turlings and Erb 2018). Parasitoids comprise an important and highly species-rich group of carnivorous insects which may regulate herbivore populations (Forbes et al. 2018). They lay eggs and develop inside or on a host insect, and therefore their ability to find these hosts is closely linked to their fitness (Thiel and Hoffmeister 2009).

Because HIPVs provide information to parasitoids that in turn suppress herbivore attack, HIPVs can be considered a part of a plant's information-mediated indirect defences (Kessler and Heil 2011). Despite advances in the understanding of the role of HIPVs in parasitoid foraging behaviour, knowledge on the distance at which HIPVs are used by parasitoids is limited (Aartsma et al. 2017). Consequently, just like for chemical cues in general (Zimmer and Zimmer 2008), we have little understanding of the significance of plant volatiles in ecological processes at larger spatial scales under field conditions. With increasing distance from the source, a volatile blend will be diluted (Cardé and Willis 2008) and blend components differentially degrade under the influence of environmental factors, such as UV or ozone (Blande et al. 2014). Therefore, it is important to assess at what distances parasitoids interact with plants through HIPVs. While HIPVs are commonly referred to as long-distance cues, the spatial scale of 'long-distance' is under debate. Some studies report only very localized effects of HIPVs on insect distributions, less than 1.5 m away from the release point (Mallinger et al. 2011, Rodriguez-Saona et al. 2011). Others have recorded effects at larger spatial scales, up to 8 or 15 m (Bernasconi Ockroy et al. 2001, Braasch and Kaplan 2012). Insight into the distance at which HIPVs attract parasitoids is crucial for our understanding of movement patterns as a result of host-location behaviour of parasitoids, both in nature and in agroecosystems. Such insights will reveal the spatial scale at which herbivore-infested plants interact with the enemies of the herbivores, and will improve our understanding of the spatial processes underlying population and community ecology(Mills and Wajnberg 2008).

There is considerable variation within and between plant species in HIPV release (Degen et al. 2004, Gols et al. 2011). The identity of the herbivore, severity of attack, or presence of multiple herbivores may affect the strength and composition of HIPV blends, resulting in a dynamic and complex environment for parasitoids to locate their hosts (Dicke et al. 2009, de Rijk et al. 2013). It is well established that parasitoids prefer volatiles from damaged plants over those from non-damaged plants (Geervliet et al. 1996, Gols et al. 2012), but in a no-choice situation or combined with non-host plant species, parasitoids will also visit undamaged plants (Gohole et al. 2005, Pareja et al. 2007, Moraes et al. 2008). The effects of variation in plant-species traits have generally been studied in choice tests under controlled conditions. Preference for a certain plant accession can lead to higher parasitism rates in the field (Poelman et al. 2009), although differences in field parasitism on different varieties cannot always be explained by volatiles (Degen et al. 2012). It is unknown whether plants whose HIPVs are preferred by parasitoids can be detected from a larger distance than HIPVs from less preferred plants.

The objective of the present study was to assess the effect of plant-trait variation in terms of HIPV emission on the spatial scale of parasitoid attraction. We made use of our earlier findings on relative preference of the parasitoid *Cotesia glomerata* to HIPVs for white cabbage, *Brassica oleracea,* accessions (from here on: cabbage accessions) (Poelman et al. 2009). When damaged by *Pieris brassicae* caterpillars, the parasitoid's host, the cabbage accession 'Christmas drumhead' is preferred by *C. glomerata* parasitoids over the accession 'Badger shipper' in both wind tunnel assays (1 m distance) and field plots (Poelman et al. 2009). We hypothesized that parasitism rates decline with increasing distance between plants and that differential preference of parasitoids (here called attractiveness of plants), as recorded in the laboratory (Poelman et al. 2009),

results in differences in spatial ecology among the cabbage accessions under field conditions. We tested these hypotheses in a field experiment using different spatial arrangements (10 m or 20 m spacing) of cabbage plants that differed in attractiveness, and in a semi-field tent experiment (spacing up to 8 m) with the same system where we observed the behaviour of parasitoids in more detail and at a smaller spatial scale. These experiments provide novel information about the spatial scale at which plants interact with parasitoids that attack herbivores. We discuss the importance of plant genetic variation in volatile emission in the context of spatial scales, and the implications for foraging behaviour decisions based on HIPVs in the field.

Material and methods

Insects and plants

Seeds of two white cabbage accessions *Brassica oleracea* var. *alba*, the accession 'Badger shipper' and the accession 'Christmas drumhead', were obtained from the Centre for Genetic Resources (CGN-Wageningen, the Netherlands). Plants were sown in peat soil cubes in a greenhouse (L16:D8 photoperiod, at 18–26°C and 40–70% RH) and seedlings of three weeks old were placed outside in fine-meshed insect screen cages to acclimatize to field conditions. Five-week-old potted plants were used in the field experiments.

Caterpillars of *Pieris brassicae* (Lepidoptera: Pieridae) and adults of *Cotesia glomerata* (Hymenoptera: Braconidae) parasitoids were obtained from cultures at the laboratory of Entomology, Wageningen Univ. *Pieris brassicae* is a specialist herbivore of plants in the family Brassicaceae (Edger et al. 2015). Caterpillars were reared on Brussels sprouts plants *Brassica oleracea* var. *gemmifera* cultivar Cyrus, in a greenhouse compartment (L16:D8 photoperiod, at 21 ± 1°C and 50–70% RH). *Cotesia glomerata* is a small larval parasitoid specializing on *P. brassicae* and measures only 2–3 millimetres in size (Brodeur et al. 1996). Parasitoids were reared on *P. brassicae* caterpillars under similar greenhouse conditions. Cocoons were placed in a cage (30 × 30 × 30 cm) and the emerged adult parasitoids were allowed to mate and were fed with honey and water. The cage was kept in a climate cabinet at 24 ± 1°C, LD12:D12. We used 2–4 days-old adult female parasitoids in the experiments.

Wagon-wheel experiment

The effect of *Brassica* accession and spacing between plants on host plant finding of parasitoids was studied in the field, using two fields of perennial rye-grass *Lolium perenne* and a spatial set-up of caterpillar-infested cabbage plants in each field that had the shape of a wagon wheel (Fig. 1). The two fields were situated near Wageningen, the Netherlands, and about 200 m apart. No brassicaceous plants were present in the direct vicinity of the fields. Parasitoids were released in a 3 × 3 m square of 5 × 5 cabbage plants at 75 cm plant distance, in the centre of each wagon wheel that was surrounded by a wired metal fence to exclude rabbits. Five transects (i.e. 'spokes') were laid out, radiating from the release area at 72° angles. Each transect consisted of six single cabbage plants spaced 10 m apart, spanning a total length of 60 m from the centre. Each potted cabbage plant was placed in a hole in the ground and was also fenced with metal wire. During each replicate one 'Badger shipper' wagon wheel and one 'Christmas drumhead' wagon wheel were established, and accessions were alternated in each field in subsequent measurement sessions ('replicates') to prevent location bias.

To test whether the distance between infested plants affects parasitoid host finding, the plants in the five spokes

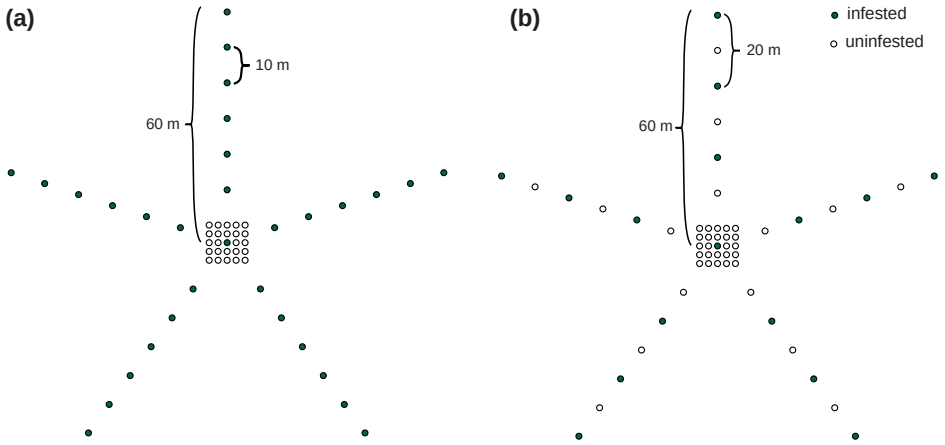


Figure 1. Spatial arrangement of wagon-wheel experiment. White cabbage plants *Brassica oleracea* var. alba of the attractive accession 'Christmas drumhead' or the less attractive 'Badger shipper' were placed in a homogeneous grassland. A central plot with 5 × 5 cabbage plants at 75 cm plant distance was created, from which five transects were laid out by placing six cabbage plants at 10-m intervals. Twenty-five mated *Cotesia glomerata* females were released from the central plant. In design a, each of the six plants on each transect was infested with 10 caterpillars of *Pieris brassicae*, the host of *C. glomerata*, while in design b, only every second plant (i.e. at 20, 40 and 60 m) was infested with caterpillars, while the other plants (at 10, 30 and 50 m) were not infested.

were either all infested (full infestation with 10 m distance between infested plants) or infested alternately within each spoke (half infestation with 20 m distance between infested plants). Replicates with distances of 10 or 20 m between infested plants on the spokes of the wagon wheels were alternated over time to realize a total of four replicate replicates with 10 m distance between infested plants and four replicate replicates with 20 m distance between infested plants. Replicates were done from May to September 2014. The grass fields were mown three times during this period in weeks that the fields were empty.

At the beginning of each replicate, potted cabbage plants were planted in the centre and the spokes of each wagon wheel. The middle plant of the central 5 × 5 plot was infested with 15 L1 *P. brassicae* caterpillars, while the rest of the plot remained uninfested. To verify that parasitism in our experiments was not caused by naturally occurring parasitoids or parasitoids released in previous experimental replicates, we assessed background levels of parasitism at the start of each replicate by introducing and recollecting the first batch of caterpillars on each plant in the spokes of the wheel 24 h before releasing the parasitoids. Subsequently, 25 female *C. glomerata* parasitoids were released from plastic tubes on the ground near the stem of the central plant. Infested plants in transects received 10 L1 *P. brassicae* caterpillars per plant. During four consecutive days, all caterpillars were recollected and replaced with new caterpillars each day. Caterpillars on the central plant, which were used to stimulate parasitoid host-searching behaviour, were removed 24 h after releasing the parasitoids and not replaced. In three earlier pilot trials, where 10 parasitoids were released and no caterpillars were present on the central plant, no parasitism was found. Recollected caterpillars were dissected under a stereomicroscope to record the presence of *C. glomerata* eggs. All cabbage plants were removed and the field was left empty for a week, after which the next replicate started.

Tent experiment

The influence of *B. oleracea* accession and spacing between infested plants on host finding by *C. glomerata* was studied in more detail and at a smaller spatial scale in a tent experiment. A large mesh tent (16 × 8 × 2.5 m) was set-up in a mown monoculture field of perennial rye grass near Wageningen. Infested or uninfested 'Badger shipper' or 'Christmas drumhead' plants were introduced at 1, 2, 4 or 8 m in the upwind direction from an uninfested plant where *C. glomerata* parasitoids were released. Infested plants were inoculated with 10 L1 *P. brassicae* caterpillars 24 h before the start of the experiment to induce HIPV production.

Female parasitoids were brought to the tent in a small mesh cage that included honey as food source and water. Parasitoids were stimulated to search for hosts by a short exposure to a Brussels sprouts leaf with *P. brassicae* caterpillars. Thereafter, we released five parasitoids near the stem of the release plant. To differentiate between parasitoids, four out of each five were marked a day prior to the observations with nail polish (Hema) of different colours. Parasitoids that were visibly wounded after marking were discarded. Unmarked parasitoids were used as a control to assess whether marking had an effect on foraging behaviour. Each parasitoid was only used once for the experiment. Twenty parasitoids were observed for each combination of accessions (Badger shipper or Christmas drumhead), distance between plants (1, 2, 4 or 8 m), and infestation with *P. brassicae* caterpillars (yes or no), for a total of 320 parasitoids.

Times until flight initiation and arrival at the (un)infested plant were recorded by visual examination of the release plant and target plant. Parasitoids that 1) did not initiate flight within 30 min, or 2) did not arrive within 30, 45, 60 or 70 min on infested plants at distances of 1, 2, 4 and 8 m, respectively, were considered non-responsive and were excluded from analyses. We used new sets of control and infested plants on each day, and accessions were tested in alternate weeks. Observations were conducted on dry days with temperatures ranging from 17–25°C in July and August 2014.

Statistical analysis

Wagon-wheel experiment

Background parasitism was found in one replicate of the wagon wheel experiment, which was excluded from the analysis. The final dataset included data from three full infestation replicates and four half infestation replicates for each cabbage accession.

Host-plant finding by *C. glomerata* was analysed as presence/absence data. If eggs of *C. glomerata* were found in at least one of the recollected caterpillars of a plant at a location on the spokes of the wagon wheel, parasitism at this location was recorded as 'present'. We used a generalized linear mixed model (GLMM) with binomial error distribution and plant accession (categorical, levels: 'Christmas drumhead' and 'Badger shipper'), spatial arrangement (categorical, levels: 10 and 20 m), distance from central plot (continuous) and day after release (continuous variable) as fixed effects. The factors 'week' (i.e. week in which the replicate was held) and 'plant ID' (as each plant was used for four consecutive days) were included as random effects. Three-way and higher term interactions were not fitted because the biological interpretation of such interactions is problematic. A full model containing all fixed variables with two-way interactions and random effects was the starting point for model selection using the dredge procedure in the R package MuMIN. This procedure results in a complete set of sub-models with combinations of the terms of the full model, and sorts the sub-models based on the corrected Akaike information criterion (AICc). Models with a ΔAICc of < 2 have a substantial support from the data. We present tables for the simplest model of the sub-models with substantial support, with the fewest degrees of freedom. The full list of sub-models and the selected variables is presented in the appendices.

To identify significant interactions, a Tukey post hoc test for the GLMM was used to compare combinations of factors. All analyses were conducted in R using the statistical packages

Table 1. Simplest generalized linear mixed model (<2 ΔAICc and lowest df) for host-plant finding success by *Cotesia glomerata* parasitoids in the wagon wheel with estimates, standard error (SE) of estimates, Wald's z-statistic and corresponding p value. Significant p-values are highlighted in bold (α = 0.05). The less attractive accession 'Badger shipper' and 10 m are reference treatments for accession and host spacing, respectively.

Fixed effect	Estimates	SE	Z value	p
Intercept	0.363	0.906	0.401	0.69
Accession 'Christmas drumhead' (categorical)	0.519	0.312	1.664	0.10
10 m host spacing (categorical)	–3.407	1.299	–2.623	**0.009**
Distance from release point (continuous)	–0.050	0.0080	–5.800	**<0.001**
Time since release (continuous)	–0.016	0.006	–2.688	**0.007**
Accession × host spacing (categorical)	2.815	0.798	3.530	**<0.001**

'lme4' (Bates et al. 2015), 'MuMIN' (Barton 2016), 'multcomp' (Hothorn et al. 2008) and 'car' (Fox and Weisberg 2011).

Tent experiment

Parasitoids that did not initiate flight were excluded from the analysis. Landing success was analysed as a binomial response variable. Parasitoids that landed on the infested plant within the time limit were recorded as a success, and other parasitoids as a failure. We used a generalized linear model with binomial error distribution and infestation treatment (categorical, levels: infestation and no infestation), distance from release point (continuous variable), plant accession (categorical, levels: 'Christmas drumhead' or 'Badger shipper'), and marking (categorical, levels: marked and non-marked) as fixed effects. The full model containing all these variables and two-way interactions was used for model selection with the dredge procedure as described above.

Search time of *C. glomerata* parasitoids was analysed for parasitoids that successfully found infested plants within the time limit. We used a general linear model with searching time as response variable and accession, distance, marking, and infestation treatment as fixed effects. Search time was square-root transformed to meet normality of residuals and equality of variance requirements. Model selection was conducted as described for the wagon-wheel experiment.

Data deposition

Data available from the Dryad Digital Repository: <http://dx.doi.org/10.5061/dryad.8n3d110> (Aartsma et al. 2018).

Results

Wagon-wheel experiment

A total of 12 000 *Pieris brassicae* caterpillars were placed in the field during the season and 7090 (59%) of these were recovered. Recovery rate was similar to previous studies using the same methodology (Poelman et al. 2009). Overall, *Cotesia glomerata* parasitism rate of recovered caterpillars was 9.7% (n = 790) and ranged from 0% to 30% of recovered caterpillars in the respective replicates (Supplementary material Appendix 1 Fig. A1–A2).

Model selection (Supplementary material Appendix 1 Table A1) indicated that host-plant-finding was influenced by accession, spacing (10 or 20 m between *P. brassicae*-infested plants), distance from release point, and days after release; there was a significant interaction between accession and spatial arrangement (Table 1). Host-plant finding was negatively associated with distance from the parasitoid release point and the number of days after release (Table 1).

In setups with 10 m spacing between infested plants, accession identity had no effect on host plant finding (Fig. 2, Tukey post hoc, z-value = 1.709, p = 0.28). There was no difference in host-plant finding between setups of 'Christmas drumhead' (attractive) with 10 or 20 m spacing (Tukey post hoc, z = -0.540, p = 0.94) and between setups of 'Badger shipper' (less attractive) with 10 m spacing and 'Christmas drumhead' with 20 m spacing (Tukey, z = –0.079, p = 1.0). In setups with 20 m spacing between infested plants, fewer plants with parasitized caterpillars were found in wagon wheels with the less attractive 'Badger shipper' accession than in those with the attractive 'Christmas drumhead' accession (Fig. 2; Tukey

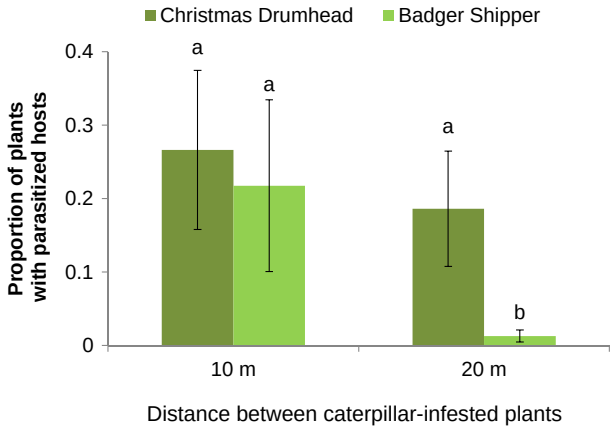


Figure 2. Proportion of *Pieris brassicae*-infested cabbage plants found by *Cotesia glomerata* parasitoids for the attractive accession 'Christmas drumhead' and less attractive accession 'Badger shipper' in 10 and 20 m plant-spacing wagon-wheel arrangements (Fig. 1). Bars and error bars represent averages across replicates and SEM, respectively (n = 3 for each accession in 10 m plant spacing, n = 4 for each accession in 20 m plant spacing). Different letters indicate significant differences between all bars ($p < 0.05$) based on pair-wise Tukey LSD post hoc tests.

Table 2. Simplest generalized linear model (<2 ΔAICc and lowest df) for host plant finding success by *Cotesia glomerata* parasitoids in a tent experiment with estimates, standard error (SE) of estimates, Wald's z-statistic and corresponding p value. Significant p-values are highlighted in bold (α = 0.05). The less attractive accession 'Badger shipper' and no infestation with caterpillars were the reference treatments for accession and infestation with caterpillars, respectively. n = 267 parasitoids.

Fixed effect	Estimates	SE	Z value	p
Intercept	–0.121	0.286	–0.421	0.674
Accession 'Christmas drumhead' (categorical)	0.728	0.260	2.806	**0.005**
Distance from release point (continuous)	–0.169	0.050	–3.416	**<0.001**
Infestation with caterpillars (categorical)	0.638	0.259	2.469	**0.014**

post hoc, z-value = 4.521, $p < 0.001$). Caterpillars were parasitized at a distance of 60 m after a time interval of 24 h after release (Fig. 2, Supplementary material Appendix 1 Fig. A1–A2).

Tent experiment

Out of the 320 parasitoids observed in the tent experiment, 127 parasitoids (39.7%) successfully landed on the experimental plant within the time limit, while 53 (16.5%) did not initiate flight within 30 minutes. The others (43.8%) initiated flight but did not arrive on the experimental plant within the time limit.

Host-plant finding success by *C. glomerata* was influenced by accession, distance from release point and whether the plant was infested with *P. brassicae* caterpillars or not (Supplementary material Appendix 1 Table A2). Marking of parasitoids had no effect on host-plant finding success (Table 2). Parasitoids were more often successful in finding plants near the release point than plants further away, and more parasitized hosts were found on plants of the attractive 'Christmas drumhead' accession than on plants of the less attractive 'Badger shipper' accession (Table 2, Fig. 3). The infestation of the focal plant with caterpillars also led to higher host-plant finding success on both accessions as compared to plants that did not contain caterpillars (Table 2, Fig. 3).

Search time of *C. glomerata* was influenced by distance from the release point and infestation with *P. brassicae* caterpillars. Parasitoids took more time to land on plants that were further away from the release point (Table 3, Fig 3): the shortest search time was 16 s to find a 'Christmas drumhead' plant at 1 m distance and the longest search time was 4531 s to find an uninfested 'Badger shipper' plant a 8 m distance. Search time of the parasitoids was lower when plants were infested with *P. brassicae* (Table 3, Fig. 3). The interaction between accession and infestation was also selected in some models, as was the main effect of marking of the parasitoids (Supplementary material Appendix 1 Table A3). Marking and accession did not influence search time.

Discussion

Animal behaviour is at the basis of many species interactions and, thus, at the basis of ecological dynamics (Sih et al. 2012) Animal behaviour is guided by information and chemical information is an important source of cues for foraging animals (Raguso et al. 2015). Chemical information as conveyed by volatile chemicals mediates interactions between organisms before they actually meet. Thus, volatile chemicals enlarge the interaction space. Intraspecific variation in the emission of volatile chemicals may result in intraspecific variation of species interactions with consequences for population dynamics and community ecology. Here, we investigated intraspecific variation in plant volatiles that are induced by insect herbivory and that attract the enemies of the inducing herbivores. Herbivore-induced plant volatiles consist of a complex blend of tens of chemical compounds produced by plants under herbivore attack, which provide

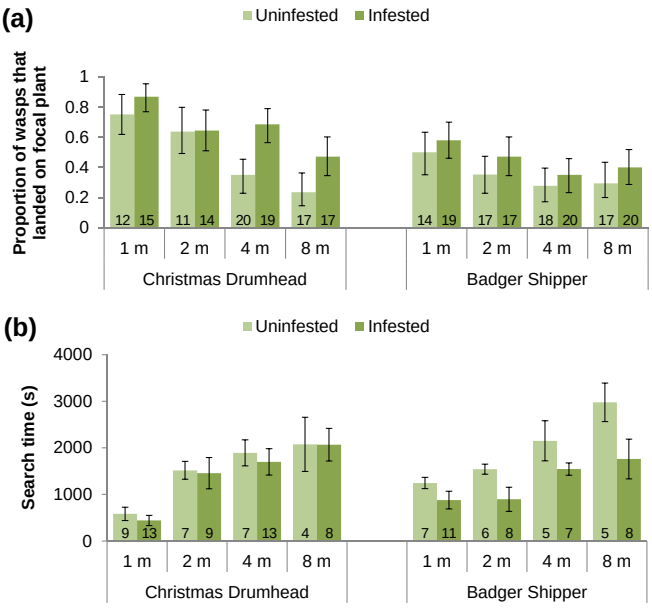


Figure 3. Proportion host location success (a) and search time of successful wasps (b) of *Cotesia glomerata* parasitoids when challenged to locate a focal white cabbage plant *Brassica oleracea* var. *alba* of either the attractive accession 'Christmas drumhead' or the less attractive accession 'Badger shipper' placed at different distances from the release plant (1, 2, 4 or 8 m). Colours indicate whether the plant was infested for 24 h with 10 L1 *Pieris brassicae* caterpillars or not. For (a), bars and error bars represent proportion of successful landings and binomial standard deviation, respectively. For (b), bars and error bars represent averages and SE, respectively. The number at the base of the bars indicates the number of parasitoids which initiated flight after release and the number of parasitoids which landed on the plant in (a) and (b) respectively.

Table 3. Simplest general linear model (<2 ΔAICc and lowest df) for square root transformed search time (s) by *Cotesia glomerata* parasitoids in a tent experiment with estimates, standard error (SE) of estimates, t-statistic and corresponding p value. Significant p-values are highlighted in bold (α = 0.05). No infestation with caterpillars was the reference treatment for infestation with caterpillars. n = 127 parasitoids.

Fixed effect	Estimates	SE	t value	p
Intercept	30.165	2.488	12.125	**<0.001**
Distance from release point (continuous)	2.637	0.398	6.625	**<0.001**
Infestation with caterpillars (categorical)	–6.325	2.075	–3.048	**0.003**

important information to both vertebrate and invertebrate natural enemies of herbivores that forage for insect herbivores (Poelman and Kos 2016).

Our field study shows that accession identity does not only influence the preference of parasitoids at a close range (Poelman et al. 2009), but also the distance from which parasitoids are recruited in field situations: the spatial scale of parasitoid attraction ranges from 10 to 20 m for the less attractive cabbage accession to 20 m and likely beyond for the more attractive accession over the time scale of the experiment. The hosts of *Cotesia glomerata* parasitoids, the lepidopteran *Pieris brassicae,* lays clutches of eggs and the emerging caterpillars feed as a group; to avoid competition, the butterflies distribute their egg batches patchily and distances between plants with hosts can easily be beyond 20 m, but within the range of the current field experiment. While accession identity did not influence the attraction of parasitoids at 10 m plant spacing in the field experiment, observations of parasitoid behaviour revealed that host finding success at distances smaller than 10 m was higher for the attractive accession than for the less attractive accession in a semi-field tent setting. Parasitoid searching behaviour was further influenced by the distance between the plant and the release point, and presence of hosts on the focal plant. Parasitism rates declined with time after release and increasing distance from the centre of the wagon-wheel setup.

While the movement ecology and perceptual range of larger animals, such as ungulates, have been relatively well studied (Zollner and Lima 2005, Fryxell et al. 2008), few studies have assessed the spatial scale of volatile emission and effects on the foraging behaviour or attraction of arthropods (Aartsma et al. 2017). Most field studies using HIPV blends assess the attraction of insects to a point source and quantify insect attraction at or close to the point source (reviewed by Aartsma et al. 2017). While this provides information on which HIPV compounds or blends attract certain insects, it does not tell us from which distance these insects are attracted. With increasing distance from an odour source, an odour plume will be diluted and change direction as a result of wind fluctuation (Cardé and Willis 2008). Moreover, individual components of the blend are differentially susceptible to environmental degradation (Blande et al. 2014). While parasitoids have navigational behavioural strategies to cope with these fluctuations in the strength of odour plumes (Kaiser et al. 1994), a higher probability of losing the plume during flight orientation might result in a lower host-plant finding success at larger distances, as observed in the tent experiment. The dynamics of odour-plume composition over time and space can compromise host-finding success under field conditions, and this is likely to influence the information value.

The spatial scale of arthropod attraction to infochemicals has predominantly been studied using synthetic volatiles. Increasing the number of pheromone point sources resulted in antennal responses of the moth *Plodia interpunctella* at larger distances (up to 60 m) downwind from these sources, suggesting that an increase in the number of volatile-emitting sources can lead to insect attraction from larger distances (Andersson et al. 2013). This may also apply to intraspecific variation in volatile-emission rates in plants, although we are not aware of any field electroantennogram (EAG) studies using plants as odour source. Applying vials with phenylethyl alcohol (as a synthetic volatile) led to both repellent and attractant effects on the abundance of insects from distances of at least 8 m (Braasch and Kaplan 2012). However, these studies did not assess whether a change in odour concentration influences the distance from which insects are recruited. Other field studies report attraction and aggregation of arthropods to methyl salicylate at only short distances (<1.5 m) from the point source (Mallinger et al. 2011, Rodriguez-Saona et al. 2011). Our study shows that, in field situations, parasitoids are able to find host-infested cabbage plants from distances up to 10 m and possibly beyond 20 m (ca 8000 parasitoid body lengths) within a day, depending on the HIPV profile of the accession. Future studies should identify the spatial scale of attraction in more complex settings, such as plant communities with host and non-host herbivore feeding, which are known to hamper parasitoid host location (de Rijk et al. 2013, Desurmont et al. 2014, Kostenko et al. 2015).

The attractive accession 'Christmas drumhead' attracted parasitoids from a larger distance than the less attractive accession 'Badger shipper'. Although the HIPV profiles of these accessions have been found to differ in quantitative and qualitative aspects under feeding damage by *Pieris* caterpillars (Poelman et al. 2009), it remains to be identified whether qualitative or quantitative components explain the distance of parasitoid attraction. Nevertheless, the present study provides the first empirical evidence that the differences in HIPV emission by the two accessions result in different attraction distances in the field, which has consequences for the overall spatial scale of attraction of parasitoids by HIPVs. Theoretically, a doubling of the attraction distance may lead to a 2- to-4-fold increase in catchment area of parasitoids depending on whether parasitoids are attracted from only the upwind direction or from all directions. This study shows

that parasitoids can spread within one day through the whole 60 m assemblage of cabbage plants when inter-plant distances are 10 m or less. This is particularly important for parasitoid species that specialize on particular host stages and therefore facing a limited window of opportunity to find a host in the right stage (van Nouhuys and Ehrnsten 2004).

Parasitism rates declined with time after release and increasing distance from the centre of the wagon-wheel setup. This may be explained by several non-exclusive mechanisms, including 1) mortality reducing the number of actively searching parasitoids in the field, 2) parasitoids leaving the experimental field because of a lack of floral food resources or simply due to random movement, 3) female parasitoids becoming egg limited, and 4) increasing distance between the spokes of the wagon wheel design at further distance from the release site. In the absence of HIPV information, parasitoids are expected to fly in cross-wind direction (Williams et al. 2007). However, in this case one would expect that with 20 m spacing between plants parasitoids may have encountered host-infested plants of the less-attractive accession when moving cross-wind without sensing cues and then tracing HIPV cues when they moved closer to the plants. Instead, we found that the parasitism rate in the wagon-wheel setups with the less attractive accession with 20 m spacing was negligible. It is known that parasitoids may switch from foraging to dispersing mode when information on hosts is not perceived or hosts are not found. Parasitoids may disperse by upward flight and the use of wind currents at higher altitudes (Kristensen et al. 2013, Schellhorn et al. 2014). However, the factors eliciting the switch from small-scale searching behaviour to passive dispersal are little understood and deserve further study. Future research on animal dispersal should take into account the availability of information, such as HIPVs or other cues, at different distances to better understand patterns of dispersal at the landscape scale.

The distance from which HIPVs attract natural enemies of herbivores is relevant in the context of information-mediated indirect defence by plants and biological control of insect pests. First, plant varieties with a larger attraction distance may have a higher chance of attracting the enemies of their attackers. Additionally, both natural enemy and host population dynamics might be influenced by the enemies' ability to detect host presence from a distance by increased attack rates. Second, in recent years there has been an increasing interest in manipulating plant traits, including indirect defence using HIPVs, to enhance biological control in agricultural fields (Cortesero et al. 2000, Kappers et al. 2011, Kaplan 2012, Penaflor and Bento 2013, Gish et al. 2015, Stenberg et al. 2015). There are, however, still many open questions on interactions of insects of different trophic levels with plants emitting HIPVs and the practical implementation of crops making use of increased indirect defences (Gish et al. 2015). In agricultural systems where monocultures cover large areas, it is important to consider from what distance HIPVs attract natural enemies and whether landscape features can supply these enemies and thus affect parasitism in the field. While our findings suggest that attractive accessions can attract larger parasitoid numbers because of an extended volatile 'catchment area', further work is needed to show how attraction by HIPVs can be exploited to increase parasitism rates in commercial agricultural field situations. Further studies are also needed to explore the implications of HIPV release for tritrophic interactions and effectiveness of biological control at a landscape level.

Insight in spatial ecology is important to understand population dynamics and community dynamics. Interactions between individuals, such as herbivore–plant, predator–prey, and pollinator–flower interactions, are at the basis of these processes. However, these interactions extend far beyond the physical interaction between the individual. Individuals emit cues that can influence the behaviour of others. Here, we have shown that a small insect of ca 2–3 mm small can interact with plants on which her hosts feed at a distance of 8000 body lengths. Thus, indirect cues produced by plants in response to herbivory have a large influence on the spatial scale at which parasitoids interact with herbivores. Moreover, this extension of the spatial interaction between two animal species is dependent on intraspecific variation among plant individuals. Investigating the consequences of the largely extended spatial scale of interactions between small animals for population dynamics and community dynamics will be an important challenge for future studies.

Acknowledgements – We thank Dine Volker and the staff of Unifarm for their help in the field work, and the rearing staff of the Laboratory of Entomology for the insects used in this experiment.
Funding – Our research is funded by the Netherlands Organisation for Scientific Research, NWO (grant 847.13.001) and the European Research Council (ERC) under the European Union's Horizon 2020 research and innovation programme (grant agreement n° 677139) to EHP.
Conflict of interest – The authors declare that they have no conflict of interest.
Author contributions – YA, WvdW, EHP, MD and FJJAB conceived the ideas and designed methodology; YA and BL collected the data; YA, EHP, WvdW and FJJAB analysed the data; YA, WvdW, EHP, MD and FJJAB led the writing of the manuscript. All authors contributed critically to the drafts and gave final approval for publication.

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Supplementary material (available online as Appendix oik-05151 at <www.oikosjournal.org/appendix/oik-05151>). Appendix 1.