



How predatory mites learn to cope with variability in volatile plant signals in the environment of their herbivorous prey

BAS DRUKKER, JAN BRUIN, GERRIT JACOBS, ANNEMARIE KROON and MAURICE W. SABELIS*

IBED – Section Population Biology, University of Amsterdam, P.O. Box 94084, 1090 GB Amsterdam, The Netherlands

(Received 17 February 2000; accepted 17 November 2000)

Abstract. When the chemical cues co-occurring with prey vary in time and space, foraging predators profit from an ability to repeatedly associate chemical cues with the presence of their prey. We demonstrate the ability of a predatory arthropod (the plant-inhabiting mite, *Phytoseiulus persimilis*) to learn the association of a positive stimulus (herbivorous prey, *Tetranychus urticae*) or a negative stimulus (hunger) with a chemical cue (herbivore-induced plant volatiles or green leaf volatiles). It has been suggested that the rate at which the integration of information becomes manifest as a change in behaviour, differs between categories of natural enemies (parasitoids versus insect predators; specialist versus generalist predators). We argue that these differences do not necessarily reflect differential learning ability, but rather relate to the ecologically relevant time scale at which the biotic environment changes.

Key words: associative learning, sensitisation, innate response, herbivore-induced plant volatiles, *Phytoseiulus persimilis*, olfactometer

Introduction

Plants are able to exploit the cognitive abilities of their inhabitants for their own interests, especially when these overlap with the interests of the inhabitants. One of many examples of this is the recruitment of arthropod predators to remove herbivorous attackers. Plants betray the presence of herbivores to predators by emitting odours induced by herbivore feeding (e.g. Turlings *et al.*, 1995). These odours consist of blends which may vary with the species of host plant – even when attacked by the same herbivore, and with the species of herbivore – even when they attack the same species of host plant (Dicke *et al.*, 1998; De Moraes *et al.*, 1998). To cope with this bewildering variety

* Author for correspondence: Tel.: +31 20 525 7738; Fax: +31 20 525 7754; E-mail: sabelis@bio.uva.nl

of information, predators may either specialise on one cue (innately or by imprinting) or they may adjust their behavioural response to any relevant odour associated with prey. The ability to learn associations between host and host-related cues has been extensively studied in parasitoids (Papaj and Lewis, 1993), but its role with respect to induced or constitutively released plant volatiles is little studied (but see Geervliet *et al.*, 1998). For arthropods that are true predators, studies on associative learning and plant volatiles are in their infancy.

We studied whether and how associative learning plays a role in the orientation of the predatory mite, *Phytoseiulus persimilis* Athias-Henriot, to plant volatiles induced by feeding of their prey, the two-spotted spider mite *Tetranychus urticae* Koch. This prey mite is a phytophage with a vast array of host plants and the blends of herbivory-induced plant volatiles (HIPV) differ between hosts in qualitative and quantitative respects (e.g. Takabayashi *et al.*, 1991, 1994). Olfactory responses of *P. persimilis* to these HIPV have been assessed for many combinations of host plant species and spider mites (Dicke *et al.*, 1998), and using various types of experimental set-ups: Y-tube olfactometers (Sabelis and Van de Baan, 1983; Dicke *et al.*, 1990a), vertical airflow olfactometers (Sabelis *et al.*, 1984), wind tunnels (Sabelis and Van der Weel, 1993) and greenhouse releases (Janssen, 1999). These studies show that HIPV mediate arrestment on and attraction towards spider-mite infested plants. Among the many factors that influence these responses, the role of dietary history of the predatory mites is of particular relevance. Dicke *et al.* (1990b) found that *P. persimilis* reared on *T. urticae*-infested Lima bean prefer the odour from *T. urticae*-infested Lima bean over the odour from *T. urticae*-infested cucumber, and that this preference changes gradually to a preference for *T. urticae*-infested cucumber during a period of 7 days in which the predators were reared on *T. urticae*-infested cucumber (see also Krips *et al.*, 1999 for similar results with gerbera as a host plant). The authors suggested that the predators learned to respond to cucumber odours by sensitisation, that is, by prolonged exposure the predators got used to cucumber odours and responded to them in the same way as they previously responded to bean odours (see also Takabayashi and Dicke, 1992). Papaj and Prokopy (1989) defined sensitisation as the gradual increase in response to a stimulus with repeated exposure to that stimulus. What Dicke *et al.* (1990b) have not yet shown is a gradual increase to cucumber odours in absence of a (positive) reinforcing stimulus (food).

We suppose that *P. persimilis* would greatly benefit from an ability to learn the association between *T. urticae*, that is its prey, and HIPV, that is the odours in the immediate vicinity of the prey. This supposition is rooted in the predator's biology: it disperses passively on air currents, lands randomly

and then searches upwind until it either decides to become airborne again or encounters a prey item. It is not very likely that passive dispersal will bring the predator to a specimen of its original host plant. Hence, upon encounter with prey on a new host plant the predatory mite may experience the association with a new blend of HIPV. Because *T. urticae* produces colonies on leaves and the infested leaves usually occur in clusters, the predator benefits from continued search for prey using the new HIPV blend as a cue.

The ability to associatively learn combinations of prey and HIPV in unfamiliar environments is experimentally analysed in this study. Associative learning (used here in the narrow sense of operant conditioning; Thorpe 1963) requires that the conditioned (e.g. odour) and the unconditioned stimulus (e.g. food, hunger) are paired, resulting in context-dependent preference or aversion. We also analysed the existence of an innate (i.e., 'experience-free') response by rearing the predatory mites on a 'plant-free' and probably 'HIPV-poor' substrate, viz. by feeding the predators *T. urticae* rinsed off tomato plants onto a filter paper dish.

To test which mechanism alters the response of predators, we paired the odour of *T. urticae*-infested bean leaves to food abundance and to food absence. The predators were from the culture deprived of host plant odours (i.e. they were cultured on prey that were washed off plants). If sensitisation were the mechanism, the resulting response of the predators would in both cases be an increase in preference for the odour as only the duration of exposure counts, not the context. If the mechanism were associative learning, the expected response is preference if the odour is paired with food, and aversion if paired with food absence.

Materials and Methods

Predators and prey

The predatory mites, *P. persimilis*, were originally obtained from a commercial supplier in 1990 (Koppert BV, Berkel en Rodenrijs, The Netherlands). Since then they were reared on detached bean leaves with two-spotted spider mites, *T. urticae*, on water-soaked clay pots in a climate room.

In some experiments, experience with the full blend of spider-mite-infested bean volatiles was avoided. This was done by keeping the predators on moistened filter paper in a 10-cm Petri dish, and feeding them all stages of *T. urticae* washed from tomato leaves (leaves rinsed in a soap solution, subsequently poured over a series of metal sieves of decreasing mesh size, then rinsed with clean water). The Petri dish rested on water-soaked cotton

wool inside a 20-cm Petri dish, covered by a lid with gauze for ventilation and sealed with parafilm.

Two strains of spider mites were used: bean spider mites (green strain) kept on bean plants in the laboratory for more than 10 years, and tomato spider mites (red strain) kept on tomato since their collection in a commercial tomato greenhouse in 1993 (Houten, The Netherlands) (Drukker *et al.*, 1997). Culturing and experiments were carried out under constant climatic conditions (25°C, 80% RH, 16:8 LD).

Olfactory response of predators

The olfactory response of predatory mites towards various (combinations of) odour sources was investigated using a glass Y-tube olfactometer (*cf.* Sabelis and Van de Baan, 1983; Janssen *et al.*, 1997). An odour source consisted of two detached leaves, either with or without spider mites, on wet tissue cloth, in an 18 × 15 × 8 cm plastic box (see Janssen *et al.*, 1997 for a detailed description). When 'clean air' was the odour source, an identical plastic box was used, including wet tissue but without leaves. The boxes with odour sources were connected to the arms of the Y-tube.

In the centre of the Y-tube, a Y-shaped metal wire served to railroad the mites. The base of the tube was connected to a pump, pulling air through the set-up with a constant flow of 0.25–0.35 m/s in both arms, continuously monitored by a flow meter. In this set-up the odour plumes from the two odour sources form two neatly separated fields in the base tube of the olfactometer with the interface coinciding with the metal wire (Sabelis and Van de Baan, 1983).

Predators are released, one at a time, at the downwind end of the wire. Typically, a mite walks upwind towards the junction, where it must choose for odour from either source. A run is finished when the predator reaches the end of the wire in either arm or after 5 min since release. After each fifth run the odour sources are interchanged, to cancel out any unforeseen asymmetries in the set-up. About 20 adult female predators were tested per replicate experiment. Two to five replicates were done per experiment (each with a fresh pair of odour sources). After collection from the culture, predators were kept for 1 h on a clean Petri dish without food until they were tested, unless stated otherwise.

Three odour sources were used:

- (1) mite-infested Lima bean, the original host plant on which the predators were reared (two spider-mite infested Lima bean leaves, with 14–90 adult female spider mites per leaf, 80–200 juveniles and males, and numerous eggs);

- (2) uninfested tomato, a host plant novel to the predators (four to six tomato leaflets of uninfested plants); and
- (3) clean air from the ambience. Infested Lima bean represents the environment familiar to the parental strain, clean tomato an unfamiliar environment, and clean air is a control.

Assessment of innate response towards infested bean

To obtain naïve predators, eggs were collected randomly from the culture on bean and spider mites, and transferred to small glass vials (10-15 eggs per vial), where they were reared to adulthood on a diet of washed spider mites (all stages) on filter paper. It is assumed that after reaching adulthood on this diet, predators will have had no experience with (the complete blend of) plant volatiles. Their response was tested towards infested bean leaves versus clean air (familiar environment against control), and towards infested bean leaves versus uninfested tomato leaves (familiar against unfamiliar environment). An innate preference will appear as a deviation from a 50/50 distribution of predators over the two odour sources.

The responses of the naïve predators were compared to responses of experienced predators, reared from egg to adulthood on Lima bean plus spider mites. For clarity, we emphasize that the period over which the predators gained experience, did not exactly stop at adulthood, but included 0–2 days of adulthood (this period is on average equal to the pre-oviposition period).

Assessment of acquired response towards infested bean

Naïve predators, reared from egg to adulthood on filter paper and fed washed spider mites, were first tested for their response towards spider-mite-infested Lima bean versus clean air. Immediately after the test, they were transferred to Petri dishes with spider-mite-infested Lima bean leaves on soaked cotton wool, and allowed to feed for *ca.* 16 h. Subsequently their response towards spider-mite-infested Lima bean versus clean air was tested again. In this way it was tested how the response changed after experiencing odour combined with a positive stimulus, that is, odour from the (+) source in the olfactometer test combined with food.

In separate experiments it was tested how the mites' response changed after experiencing the same odour, but now combined with food *absence*, a negative stimulus. Naïve predators were first tested for their response towards air from spider-mite-infested Lima bean versus air from clean tomato. Immediately after this test they were transferred into a glass tube with nylon gauze at both ends to allow free air exchange. The mites were deprived of food, but water was supplied on filter paper. Humidified air carrying odour from

a container with spider-mite-infested Lima bean leaves was pulled through the tube for 16–24 h. Then the response towards spider-mite-infested Lima bean versus clean tomato was tested again. After this test, the mites were put back in the glass tube, but now they were in *presence* of food (spider mite eggs washed off tomato leaves onto filter paper), while being exposed for 24 h to odour. In one experiment, this odour came from clean tomato, in another experiment it came from spider-mite-infested Lima bean. Subsequently, the response of the predators towards spider-mite-infested Lima bean leaves versus clean tomato leaves was tested again, in both experiments. In this way it was tested whether two sequentially offered pairs of stimuli had an additive effect on the olfactory response of *P. persimilis*.

In all experiments where groups of mites were given serial experiences, random samples of *ca.* 25 individuals were taken for olfactometer tests. After each test the mites were returned to the group for a subsequent experience (when applicable), regardless of the choice they had made in the olfactometer. In other words, the groups of mites tested consisted of mites that had been tested before, and mites that were tested for the first time. Thus, the treatment prior to the olfactometer experiment could in no way alter the genetic composition of the predator population; any change in response is unlikely to be the consequence of directional selection, but rather to be due to learning.

Statistical analysis

Each replicate experiment was subject to a binomial test against the null hypothesis that the distribution of predators reaching the end of either arm is equal. Based on common sense, replicate experiments were pooled or not. Pooled data for different treatments were tested against each other by means of 2×2 contingency table analysis, where appropriate.

Results

Innate response towards infested bean?

Well-fed predators reared on spider-mite infested bean leaves and hence exposed to HIPV throughout their entire life, showed a significant preference for odour from infested bean leaves (familiar environment), both over odour from uninfested tomato leaves (unfamiliar environment; Table 1) and over clean air (Table 2). They did not prefer odour from uninfested tomato leaves to clean air (Table 1).

Naïve predators raised on washed tomato-reared spider mites (eggs and emerging larvae) on filter paper, showed a very weak preference for odour from mite-infested bean leaves over clean air. Two sets of replicates, each

Table 1. Olfactory response of *Phytoseiulus persimilis* females reared on bean leaves infested with *Tetranychus urticae*

Odour source		<i>n</i> (+)	<i>n</i> (-)	<i>n</i> (0)	% (+)*	<i>P</i> (2-sided)
+	-					
TUB	CT	19	1	1	95	0.00004
		17	3	1	85	0.0026
		15	5	1	75	0.041
		15	5	2	75	0.041
		15	5	0	75	0.041
Total		81	19	5	81	<10 ⁻⁶
CT	CA	13	7	2	65	0.26
		9	11	0	45	0.82
Total		22	18	2	55	0.64

*% (+) = $n(+)/\{n(+)+n(-)\} \times 100$.

Pairs of odour sources were: *T. urticae*-infested bean leaves (TUB) versus clean tomato leaves (CT), and clean tomato leaves (CT) versus clean air (CA). Rows indicate independent replicate trials.

being part of a separate experiment, were carried out: three replicates (Table 2) and four replicates (Table 3). None of the seven replicates gave a significant deviation from the 50:50 distribution expected under the null hypothesis. Pooling within a set gave a significant effect only in Table 3. In a separate experiment predators were raised on washed bean-reared spider mites on filter paper, and tested as adults for their response towards mite-infested bean leaves versus clean tomato leaves. In none of the four replicates a preference was found, nor when the results were pooled (Table 2). These results support our initial assumption that predatory mites will have had no significant experience with (the whole blend of) plant volatiles after reaching adulthood on a diet of washed spider mites on filter paper.

Naïve predators showed no significant preference for odour from infested bean leaves over odour from uninfested tomato leaves (Table 4). These negative results are unlikely to be due to inferior quality of the odour sources, because the very same odour sources elicited a significant response in predators obtained from spider-mite-infested bean leaves (not shown in Table; with odour sources from trial A–E: (A) 19 predators went towards infested bean leaves/1 predator towards uninfested tomato leaves, (B) 17/3, (C) 15/5, (D) 15/5, (E) 15/5; in total 81% of 100 females chose for odour from infested bean leaves, all five replicates significant (two-sided binomial test)). Thus, the slight preference of the ensemble of naïve predators to spider mite-infested

Table 2. Effect of pre-adult experience on the olfactory response of *Phytoseiulus persimilis* females towards odour from *Tetranychus urticae*-infested bean leaves (TUB) versus clean air (CA) or clean tomato (CT)

Experience	<i>n</i> (TUB)	<i>n</i> (CA)	<i>n</i> (CT)	<i>n</i> (0)	% (TUB)*	<i>P</i> (2-sided)
Spider mites on bean leaves						
	15	1		2	94	0.00052
	18	6		0	75	0.023
	20	2		0	91	0.00012
	19	6		0	76	0.015
Total	72	15		2	83	<10 ⁻⁶
Tomato spider mites on filter paper						
	12	7		6	63	0.36
	12	12		3	50	1.00
	11	9		0	55	0.82
Total	35	28		9	56	0.45
Bean spider mites on filter paper						
	6		4	1	60	0.75
	6		4	1	60	0.75
	11		5	0	69	0.21
	8		8	0	50	1.00
Total	31		21	2	60	0.21

*see Table 1.

Predators were reared from egg to adulthood either on *T. urticae*-infested bean leaves, on filter paper with spider mites washed off tomato leaves, or on filter paper with spider mites washed off bean leaves. Rows indicate independent replicate trials.

bean leaves over clean air suggests that an innate response, if present at all, is weak.

Acquired response: sensitisation or operant conditioning?

Naïve predators were first tested for their response to spider-mite infested bean leaves versus clean air. As pointed out before, their responses were not significant in any of the four replicate experiments (Table 3). Subsequently, these predators were given a 16 h experience with mite-infested bean leaves, and then tested again with fresh, but similar, odour sources. The results showed a strong preference for odour from infested bean leaves over clean air, significant in all four replicates (Table 3). The change in response due to experience with mite-infested bean leaves was highly significant ($\chi^2 = 18.9$, d.f. = 1, $p < 0.001$). This can be interpreted as strong evidence for an acquired

Table 3. Effect of conditioning on olfactory response of *Phytoseiulus persimilis* females towards odour from *Tetranychus urticae*-infested bean leaves (TUB) versus clean air (CA)

Condition	<i>n</i> (TUB)	<i>n</i> (CA)	<i>n</i> (0)	% (TUB)*	<i>P</i> (2-sided)
Inexperienced	15	11	0	58	0.56
	15	8	5	65	0.21
	14	5	1	74	0.064
	12	7	3	63	0.36
Total	56	31	9	64	0.0097
Experienced	16	1	2	94	0.00027
	19	0	1	100	0.000004
	18	1	1	95	0.00008
	20	3	1	87	0.00049
Total	73	5	5	94	<10 ⁻⁶

*see Table 1.

Predators were reared from egg to adulthood on filter paper with spider mites washed off tomato leaves as food. Subsequently, these females, inexperienced with odour from infested leaves, were given experience for 16 h with HIPV, spider mites and bean leaves. Groups of females were tested in the olfactometer before and after this experience. Rows indicate independent replicate trials.

response to HIPV when paired to a positive unconditioned stimulus (i.e. prey presence).

The acquired response may result from two underlying mechanisms: sensitisation or associative learning. For sensitisation the increase in responsiveness to the odour should be gradual and independent of the stimulus with which it is paired. To investigate independence of the conditioned and unconditioned stimuli, HIPV was paired to a contrasting stimulus, viz. food absence. Naïve predators were exposed to HIPV for 16 or 24 h in absence of prey. Before exposure to the paired stimuli the naïve predators had no preference for odour from spider-mite infested bean leaves over odour from uninfested tomato leaves (Table 4). After 24 h the response had dropped from 54% to 22% (via 50%, after 16 h; Table 4). This points at a switch to the opposite response: the predators acquired a significant aversion to the odour of spider-mite-infested bean leaves. The change in response due to the 24 h experience with HIPV in absence of food was significant ($\chi^2 = 15.3$, d.f. = 1, $p < 0.001$). Together with the results in Table 3, this is evidence to reject sensitisation as the underlying mechanism, because a change of context (from positive to negative) in which the odour is presented, also changes the quality

of the response (from positive to negative). Rather, it suggests associative learning.

Associative learning was further tested in two ways. After 24 h experience with hunger in presence of the odour of spider-mite-infested bean leaves, and

Table 4. Effect of odour during starvation and subsequent satiation on the olfactory response of *Phytoseiulus persimilis* females towards odour from *Tetranychus urticae*-infested bean (TUB) versus clean tomato (CT)

Experience		Trial	n(TUB)	n(CT)	n(0)	% (TUB)*	P (2-sided)
Feeding state	Odour given						
Fed	None	A	10	10	2	50	1.00
		B	11	9	0	55	0.82
		C	11	9	0	55	0.82
		D	9	11	0	45	0.82
		E	13	7	0	65	0.26
Total			54	46	2	54	0.48
16 h starved	TUB	A	13	7	1	65	0.26
		B	7	13	3	35	0.26
Total			20	20	4	50	1.00
24 h starved	TUB	C	8	16	1	33	0.15
		D	3	17	0	15	0.0026
		E	3	17	0	15	0.0026
Total			14	50	9	22	0.000007
Fed	CT	B	6	17	8	26	0.035
		C	3	17	1	15	0.0026
		D	4	16	0	20	0.012
Total			13	50	9	21	0.000003
Fed	TUB	F	7	2	1	78	0.18
		G	11	4	1	73	0.12
		H	12	4	1	75	0.08
		I	4	1	0	80	0.38
		J	9	5	1	64	0.42
Total			43	16	4	73	0.0006

*see Table 1.

Predators were first reared from egg to adulthood on filter paper with spider mites washed off tomato leaves as food. These females, inexperienced with odour from infested leaves, were first tested in the olfactometer, then starved for 16 h or 24 h while exposed to TUB, and tested. Subsequently during 24 h they were fed *ad libitum* washed spider mites while either exposed to CT, or while exposed to TUB, and then tested again. Rows indicate independent replicate trials; trial codes identify groups of mites.

after a choice test displaying aversion to the odour of spider-mite-infested bean leaves, predators were given 24 h experience either with food in presence of the odour of clean tomato leaves, or with food in presence of the odour of spider-mite-infested bean leaves. When prey presence (unconditioned stimulus) was paired to odour from the unfamiliar environment, that is, uninfested tomato (conditioned stimulus), a preference for odour from clean tomato leaves over odour from spider-mite-infested Lima bean leaves was found (79% of mites towards tomato leaves; Table 4) – similar to the previous response found after the negative experience with odour of spider-mite-infested bean leaves (78%). Thus ironically, by association of prey and odour from clean tomato, the predators now respond positively to the odour from the unfamiliar environment that in actual fact does not harbour the prey!

When prey presence (unconditioned stimulus) was paired to odour from the familiar environment, that is spider-mite-infested bean leaves (conditioned stimulus) – after the mites had been given 24 h experience with food absence combined with volatiles from mite-infested bean leaves – a change of preference was found. Now predators preferred odour from spider-mite-infested Lima bean leaves over odour from clean tomato leaves (73% of mites towards infested bean leaves; Table 4). Although due to small sample sizes none of the five replicates gave a significant deviation from the 50:50 distribution, the pooled data showed a highly significant effect (Table 4). Thus, by imposing a subsequent, qualitatively different experience on predatory mites, a qualitative change in response was found, which was highly significant ($\chi^2 = 30.1$, d.f. = 1, $p < 0.001$). This is strong evidence for context-dependent adjustment of behaviour, in other words: associative learning in predatory mites.

Discussion

Evidence for associative learning

Our results are consistent with the hypothesis that naïve, inexperienced predators have no, or at best a very faint, preference for volatile chemicals until they first perceive odours in association with a rewarding or unrewarding experience. Inspired by Papaj *et al.* (1994), we tested this hypothesis by offering stimuli that are relevant in the context of foraging predatory mites, both in realistic combinations (odours from spider-mite-infested bean leaves in association with prey presence, and clean air in association with prey absence) and unrealistic combinations (clean air or odour from uninfested tomato plants associated with prey, and odour from spider-mite-infested bean

leaves associated with prey absence). In both cases the predators responded according to the context and not according to how familiar their parental strains were with the odour. Thus, predatory mites can interpret the coincidence of two independent events or states, as a relation between these events, which is defined as associative learning. Sensitisation cannot be the mechanism because a change of context (from positive to negative) in which the odour is presented, also changes the quality of the response (from positive to negative).

Taking a devil's advocate position, one could argue that the population of predatory mites used for the experiments exhibits a genetic polymorphism of olfactory responses to different stimuli. So, each combination of stimuli offered would elicit an innate, fixed response of a different subset of individuals in the population, whereas, for the sake of argument, the remaining individuals respond indifferently in two-choice tests. This hypothesis, however, is highly unlikely for two reasons. First, the predatory mites can be fooled to make non-adaptive choices by offering unrealistic combinations of stimuli. Second, the response switches are too intense to be explained by a polymorphic population response. Recent experiments on prey choice of another predatory mite, *Hypoaspis aculeifer* (Mesostigmata: Laelapidae), showed that local populations exhibit between-individual variability in prey choice and that prey choice has a simple genetic basis (Lesna and Sabelis, 1999). However, isofemale lines of this predator selected for choosing a particular prey (bulb or copra mites), retain their ability to learn the association between prey and (novel) odour, and switch their choice accordingly (Lesna and Sabelis, unpublished data).

To our best knowledge, this is the first evidence for associative learning in predatory mites, and it is almost the first case for associative learning in predatory arthropods in general. Similar evidence was obtained in recent studies on olfactory responses of heteropteran bugs (Drukker *et al.*, 2000) and laelapid predatory mites (Lesna and Sabelis, unpublished data). Although new for predators, associative learning has been demonstrated for parasitic insects (Lewis and Tumlinson, 1988; Lewis and Takasu, 1990; Vet and Groenewold 1990; Vet *et al.*, 1995) and honey bees (Gould, 1993; Menzel, 1993).

Some authors have argued that experience changes behaviour of parasitoids and bees much faster (say in seconds), than that of predatory mites (say in days) (Krips *et al.*, 1999). This generalization, however, should be treated with caution, as becomes clear when considering a functional, rather than a causal, perspective: why should a forager change its behaviour fast when its natural environment changes relatively slow? In other words, learning in parasitoids should be scaled to the rate at which hosts in a patch become

parasitized, whereas learning in predators should be tuned to the time scale at which prey densities change. Thus, slow or fast changes in behaviour do not necessarily reflect interspecific differences in learning capacity.

Search phases and the relevant time scale of learning

One may wonder whether predatory mites learn fast when the biotic environment changes rapidly. To explore this possibility it is useful to divide the searching process into five phases (Sabelis and Dicke, 1985):

- (1) take-off,
- (2) passive aerial dispersal,
- (3) searching for prey patches (clusters of prey colonies on leaves),
- (4) searching for prey colonies within a patch,
- (5) searching for prey within a colony.

These phases differ in the characteristic time scale of change in the prey environment. During phase 4 and 5, prey density (number of prey per occupied leaf area) will certainly not change in seconds or minutes, but rather in days or even slower. When local prey populations are wiped out, the vast majority of predatory mites take off for aerial dispersal (phase 1 and 2). After landing, these hungry predators may find themselves on a specimen of the same or a new host plant species, harbouring the same or other potential prey, or no prey at all. From the predator's perspective this is where the biotic environment changes fast and the predatory mites do better by learning fast accordingly. This will involve learning to associate herbivore-induced volatiles (HIPV) from the new host plant with the herbivore infesting it. Since the predatory mites are hungry in phase 1–3 and well fed during most of phase 4 and 5, we expect starved predators to learn fast and fed predators to learn slowly.

We also argue that fast or slow learning does not relate to the degree of specialisation of the predator (Poolman Simons *et al.*, 1992; Vet *et al.*, 1995; Potting *et al.*, 1997; Geervliet *et al.*, 1998), but rather to the ecologically relevant time scale of change in the prey environment. Thus, before taking position in the debate over whether some species (should) learn fast and others slow, whether generalist predators require learning and specialist predators can do without, it seems wiser to investigate at which time scale the predator should integrate information before deciding to alter its behaviour.

Acknowledgements

B. Drukker, J. Bruin and G. Jacobs were supported by the Dutch Technology Foundation (NWO/STW). Martijn Egas, Arne Janssen, Marcel Dicke, Jetske

de Boer and two anonymous referees kindly provided thoughtful comments on the manuscript.

References

- De Moraes, C.M., Lewis, W.J., Paré, P.W., Alborn, H.T. and Tumlinson, J.H. 1998. Herbivore-infested plants selectively attract parasitoids. *Nature* 393: 570–573.
- Dicke, M., Van Beek, T.A., Posthumus, M.A., Ben Dom, N., Van Bokhoven, H. and De Groot, Æ. 1990a. Isolation and identification of volatile kairomone that affect acarine predator-prey interactions. *J. Chem. Ecol.* 16: 381–396.
- Dicke, M., Van der Maas, K.J., Takabayashi, J. and Vet, L.E.M. 1990b. Learning affects response to volatile allelochemicals by predatory mites. *Proc. Exper. Appl. Entomol., N.E.V. Amsterdam* 1: 31–36.
- Dicke, M., Takabayashi, J., Posthumus, M.A., Schütte, C. and Krips, O.E. 1998. Plant-phytoseiid interactions mediated by herbivore-induced plant volatiles: variation in production of cues and in response of predatory mites. *Exp. Appl. Acarol.* 22: 311–334.
- Drukker, B., Janssen, A., Ravensberg, W. and Sabelis, M.W. 1997. Improved control capacity of the mite predator *Phytoseiulus persimilis* (Acari: Phytoseiidae) on tomato. *Exp. Appl. Acarol.* 21: 507–518.
- Drukker, B., Bruin, J. and Sabelis, M.W. 2000. Anthocorid predators learn to associate herbivore-induced plant volatiles with presence or absence of prey. *Physiol. Entomol.* 25: 260–265.
- Geervliet, J.B.F., Vreugdenhil, A.I., Dicke, M. and Vet, L.E.M. 1998. Learning to discriminate between infochemicals from different plant-host complexes by the parasitoids *Cotesia glomerata* and *C. rubecula* (Hymenoptera: Braconidae). *Entomol. Exp. Appl.* 86: 241–252.
- Gould, J.L. 1993. Ethological and comparative perspectives on honey bee learning. In: *Insect Learning: Ecological and Evolutionary Perspectives*, D. R. Papaj and A. C. Lewis (eds), pp. 18–50. Chapman and Hall, New York.
- Janssen, A. 1999. Plants with spider-mite prey attract more predatory mites than clean plants under greenhouse conditions. *Entomol. Exp. Appl.* 90: 191–198.
- Janssen, A., Bruin, J., Jacobs, G., Schraag, R. and Sabelis, M.W. 1997. Predators use volatiles to avoid prey patches with conspecifics. *J. Anim. Ecol.* 66: 223–232.
- Krips, O.E., Willems, P.E.L., Gols, R., Posthumus, M.A. and Dicke, M. 1999. The response of *Phytoseiulus persimilis* to spider-mite induced volatiles from gerbera: influence of starvation and experience. *J. Chem. Ecol.* 25: 2623–2641.
- Lesna, I. and Sabelis, M.W. 1999. Diet-dependent female choice for males with ‘good genes’. *Nature* 401: 581–584.
- Lewis, W.J. and Tumlinson, J.H. 1988. Host detection by chemically mediated associative learning in a parasitic wasp. *Nature* 331: 257–259.
- Lewis, W.J. and Takasu, K. 1990. Use of learned odours by a parasitic wasp in accordance with host and food needs. *Nature* 348: 635–636.
- Menzel, R. 1993. Associative learning in honey bees. *Apidologie* 24: 157–168.
- Papaj, D.R. and Prokopy, R.J. 1989. Ecological and evolutionary aspects of learning in phytophagous insects. *Annu. Rev. Entomol.* 34: 315–350.
- Papaj, D.R. and Lewis, A.C. (eds.) 1993. *Insect Learning: Ecological and Evolutionary Perspectives*. Chapman and Hall, New York.

- Papaj, D.R., Snellen, H., Swaans, K. and Vet, L.E.M. 1994. Unrewarding experieexinces and their effect on foraging in the parasitic wasp *Leptopilina heterotoma* (Hymenoptera: Eucoilidae). *J. Ins. Behav.* 7: 465–481.
- Poolman Simons, M.T.T., Suverkropp, B.P., Vet, L.E.M. and de Moed, G. 1992. Comparison of learning in related generalist and specialist eucoilid parasitoids. *Entomol. Exp. Appl.* 64: 117–124.
- Potting, R.P.J., Otten, H. and Vet, L.E.M. 1997. Absence of odour learning in the stemborer parasitoid *Cotesia flavipes*. *Anim. Behav.* 53: 1211–1223.
- Sabelis, M.W. and Van de Baan, H.E. 1983. Location of distant spider mite colonies by phyto-seiid predators: demonstration of specific kairomones emitted by *Tetranychus urticae* and *Panonychus ulmi*. *Entomol. Exp. Appl.* 33: 303–314.
- Sabelis, M.W. and Dicke, M. 1985. Long-range dispersal and searching behaviour. In: *Spider Mites: Their Biology, Natural Enemies and Control*, W. Helle and M.W. Sabelis (eds.), pp. 141–160. Elsevier, Amsterdam.
- Sabelis, M.W. and Van der Weel, J.J. 1993. Anemotactic responses of the predatory mite, *Phytoseiulus persimilis* Athias-Henriot, and their role in prey-finding. *Exp. Appl. Acarol.* 17: 1–9.
- Sabelis, M.W., Vermaat, J.E. and Groeneveld, A. 1984. Arrestment responses of the predatory mite, *Phytoseiulus persimilis*, to steep odour gradients of a kairomone. *Physiol. Entomol.* 9: 437–446.
- Takabayashi, J. and Dicke, M. 1992. Responses of predatory mites with different rearing histories to volatiles of uninfested plants. *Entomol. Exp. Appl.* 64: 187–193.
- Takabayashi, J., Dicke, M. and Posthumus, M.A. 1991. Variation in composition of predator attracting allelochemicals emitted by herbivore-infested plants: Relative influence of plant and herbivore. *Chemoecology* 2: 1–6.
- Takabayashi, J., Dicke, M. and Posthumus, M.A. 1994. Volatile herbivore-induced plant–mite interactions: variation caused by biotic and abiotic factors *J. Chem. Ecol.* 20: 1329–1354.
- Thorpe, W.H. 1963. *Learning and Instinct in Animals*. Methuen, London.
- Turlings, T.C.J., Loughrin, J.H., McCall, P.J., Röse, U.S.R., Lewis, W.J. and Tumlinson, J.H. 1995. How caterpillar-damaged plants protect themselves by attracting parasitic wasps. *Proc. Natl. Acad. Sci. USA* 92: 4169–4174.
- Vet, L.E.M. and Groenewold, A.M. 1990. Semiochemicals and learning in parasitoids. *J. Chem. Ecol.* 16: 3119–3135.
- Vet, L.E.M., Lewis, W.J. and Cardé, R.T. 1995. Parasitoid foraging and learning. In: *Chemical Ecology of Insects – 2*, R. T. Cardé (ed.), pp. 65–101. Chapman and Hall, New York.