Entomologia Experimentalis et Applicata **75:** 87–98, 1995.

Visual and olfactory location of biotopes, prey patches, and individual prey by the ladybeetle *Chilocorus nigritus*

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Accepted: September 8, 1994

Key words: foraging, behaviour, *Chilocorus*, Coccinellidae

Abstract

Foraging behaviour of the predator *Chilocorus nigritus* (Fabricius) (Coleoptera: Coccinellidae) at the three spatial levels of biotope, prey patch and individual prey, was studied in the laboratory, and related to behaviour in the field. Vertically oriented parallel lines were more attractive than the same shapes in a horizontal position. A simulated horizon with a tree line was preferred to a simulated flat horizon. They were attracted to a tree image for the first 2 h of exposure, but were less attracted after longer exposure, possibly due to habituation. Leaf shape was recognised, and simple ovate leaves were preferred to compound bipinnate leaves and to squares. These responses were associated with biotope selection for feeding and aggregation at aestivation sites. The location of prey patches by adults involved prey odour but the location of such sites by larvae did not. Adults detected individual prey visually and olfactorily over short distances but physical contact with prey was required for detection by larvae. Location of individual prey and prey patches by adults and larvae was facilitated by alternation between intensive and extensive search. The differences in the ability of larvae and adults to locate prey, stem from the adults being the active locators of biotope and patch, whereas the comparatively immobile larvae depend on their parents' ability for long-range location of prey. Two hypotheses concerning coccinellid foraging behaviour are proposed. Firstly, the duration of response to a visual cue is related to the distance over which such a cue may be perceived. It follows that habituation to closer range cues occurs more rapidly than to longer range cues. Secondly, visual cues used by adults at the different spatial levels of prey location, and the location of mates and aggregation sites, have the same or similar shape. These results also provide guidelines for orchard management to maximise the biocontrol value of this species.

Introduction

The foraging behaviour of many sensorily highly-developed insect predators and parasitoids has been well researched (Hassell & Southwood, 1978; Bell, 1990). An area of neglect has been the foraging behaviour of coccinellids, despite their biocontrol value. Hassell & Southwood (1978) describe three levels of predator foraging – single food items, patches or areas of aggregated food items and habitats (biotopes). There is, in particular, a dearth of information on coccinellid foraging at the level of patches, and especially of biotopes. Also, there is a need for information on the foraging of a single species, encompassing all three levels.

Foraging by coccinellids for individual prey items has received some attention but the detailed behaviour has not been fully elucidated. There have been conflicting reports of coccinellids being able to detect individual prey visually and olfactorily over short distances (Allen *et al.*, 1970; Stubbs, 1980; Obata, 1986) versus detection only through physical contact (Fleschner, 1950; Kehat, 1968; Storch, 1976). Coccinellids maximise their encounter rate of aggregated prey, by adjusting movement patterns following prey encounters (Hassell & Southwood, 1978; Carter & Dixon, 1984). This behaviour is characterised by a decrease

in speed (involving orthokinesis) and an increase in tortuosity (involving klinokinesis) plus scanning for a short period following prey consumption (Laing, 1937; Banks, 1957; Dixon, 1959; Carter & Dixon, 1982 & 1984; Nakamuta, 1985; Podoler & Henen, 1986). This behaviour is known as intensive foraging, and reverts to extensive foraging after a period of unsuccessful searching (Carter & Dixon, 1982).

Chilocorus nigritus (Fabricius), which originates from the Indian sub-continent, is an economically valuable biocontrol agent of scale insects (Hemiptera: Diaspididae) on several crops in numerous countries (Samways, 1984). It is particularly valuable in controlling red scale *Aonidiella aurantii* (Maskell) on citrus in southern Africa (Samways, 1984, 1986). In this geographical region the adult beetles shuttle between citrus orchards and giant bamboo *Dendrocalamus giganteus* Munro, on which large numbers of the diaspidid scale *Asterolecanium miliaris* (Boisduval) are encountered (Samways, 1984; Hattingh & Samways, 1991). The success of *C. nigritus* in southern Africa has been linked to this behaviour (Hattingh & Samways, 1991).

The effectiveness of shuttling by *C. nigritus* between alternative biotopes in the field, suggests that biotope location is based on the recognition of cues associated with profitable biotopes. This means that the beetles move directly towards and in immediate response to, environmental cues, as opposed to settling in prey-rich areas simply after a random search.

This study, through laboratory experimentation, investigates the role played by visual cues in biotope location by *C. nigritus*. The role of sight and olfaction in the location of prey patches, and the detection of individual prey items over short distances is also investigated. Alternate intensive and extensive foraging modes, in response to prey consumption and the time since the last prey encounter, are also demonstrated.

Materials and methods

Three spatial scales of location were studied: 1) biotope, 2) patch, and 3) individual prey items.

Biotope location

Walls and ceiling of a room (3 m × 3 m × 3.5 m) were covered in white, opaque paper. A clear perspex cylindrical tunnel (2.4 m long and 1 m in diameter), closed at both ends, was placed inside the room, on a white platform (Fig. 1). The temperature was between 25 °C and 30 °C. Four incandescent flood-lights and two fluorescent lights were positioned beneath the platform so that the light intensity at both ends of the tunnel was 1000 lux.

The trials involved 60 *C. nigritus* adults per replicate, starved for 24 h, and released into the centre of the perspex tunnel. Numbers of beetles in the terminal 0.5 m of each end of the tunnel were periodically recorded for up to 135 min after release. The tunnel was rotated through 180 ° between replicates to eliminate any bias towards either end of this apparatus. Attractiveness of images painted on white paper screens, using chrome oxide green paint (GLS 013, 'dekade paints'), were compared. In each comparison, equivalent surface areas were covered in paint on the two screens at opposite ends of the tunnel.

Eight parallel vertical stripes, each 1.2 m long and 84 mm broad, were painted on one screen. Five horizontal stripes, 2 m long and 84 mm broad on another. A horizon with a tree line was painted, having a solid base area 2 m wide and 0.3 m high, above which various tree shapes protruded. Another screen had a solid base area of 2 m × 0.5 m without protruding tree shapes. The shape of a tree was painted on another screen, with a 0.15 m broad and 0.34 m high stem, on top of which the foliar portion was in the shape of a circular disk, 1.1 m in diameter, with the lower fifth cut off (Fig. 1). Twenty one leaves, the shape and size of a typical large citrus leaf, were painted on a screen 1.7 m high and 1 m wide. On another screen of the same size, 21 solid squares 80 mm × 80 mm were painted.

The attractiveness of the following images were compared under five conditions: a) control: no images; b) vertical versus horizontal stripes; c) the flat horizon versus the horizon with a tree line; d) the shape of a tree versus vertical stripes; e) paintings of citrus leaves versus squares; f) real *Citrus sinensis* cv. Valencia leaves versus *Jacaranda mimosifolia* D. Don leaves. The leaves were suspended from cotton threads against the walls at either end of the tunnel. Six citrus twigs, with 75 leaves in total, were used per replicate. The total surface areas of *C. sinensis* and *J. mimosifolia* leaves used were equivalent. Comparisons a) to d) were made with the permutation test for related samples and because of the computational cumbersomeness of this test with numerous replicates, comparisons e) and f) were made with the Wilcoxon signed ranks test, which is the permutation test based on ranks not integers (Siegel & Castellan, 1988).

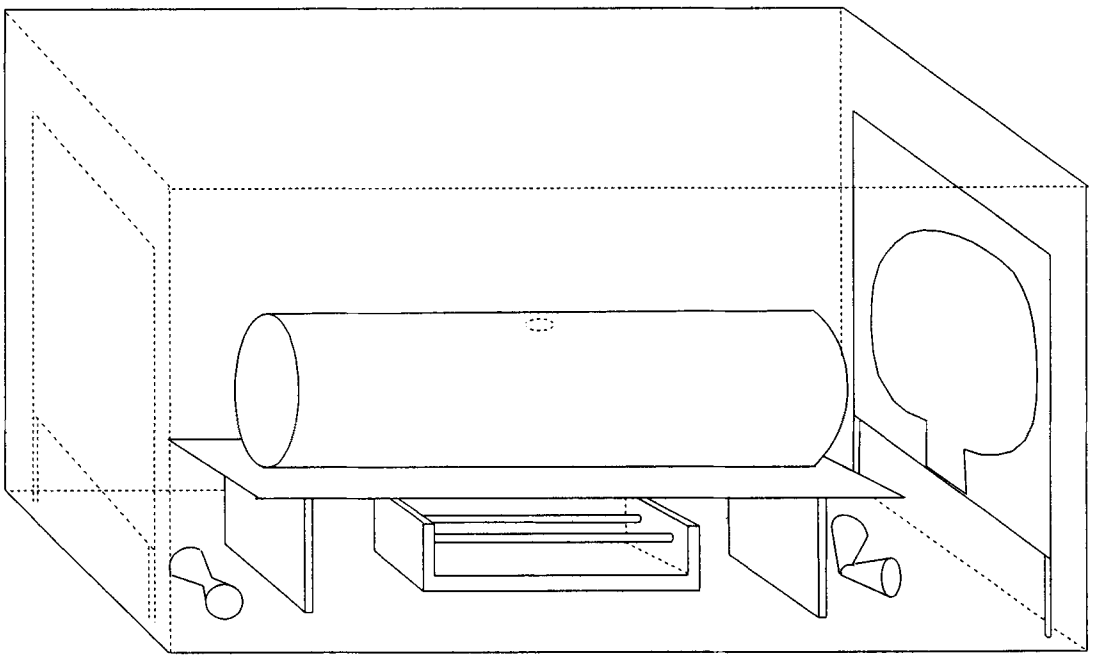

Fig. 1. Flight chamber in which visual aspects of biotope location were investigated.

Patch location
Experiments 1 to 5 were conducted inside the white room as described above, but without images on the walls and without the perspex tunnel.

Experiment 1: vision and olfaction. Four arenas were arranged in a square on top of a platform (Fig. 2). The arenas were wooden-framed cages 0.2 m × 0.2 m × 0.2 m covered in fine white netting with wooden floors. In each of the centrally-positioned corners of the arenas, a butternut *Cucurbita moschata* cv. Waltham was placed. In the first trial these were uninfested, and in the second, they were encrusted with *Aspidiotus nerii* Bouché (Hemiptera: Diaspididae) which is routinely used for rearing *C. nigritus* (Samways & Tate, 1986).

Ten *C. nigritus* adults, which had been starved for 24 h, were placed in a glass petri dish, for each replicate. These were placed on the floors of the arenas in the corners opposite the butternuts. Five minutes later, the lids of the petri dishes were removed and the time it took for the first beetle to make contact with the butternuts in each arena was recorded. The experiment was also conducted with fourth-instar larvae, starved for 16 h, five larvae per replicate. Times were compared using a Mann-Whitney U-test, $\alpha = 0.05$.

Experiment 2 (choice): vision and olfaction. Four butternuts, infested with *A. nerii*, and four uninfested butternuts, were arranged on opposite sides of 0.4 m high arenas with 0.5 m × 0.4 m floors covered in brown paper. Ten adult beetles per replicate, starved for 24 h, were released on the middle of the floor. Times in excess of 30 s for the first beetles to encounter an uninfested or an infested butternut were recorded and were compared using the Wilcoxon signed ranks test, $\alpha = 0.05$.

Experiment 3: olfaction. As in experiment 1, ten beetles per replicate were released on the arena floors in the corners opposite the butternuts. Times in excess of 30 s for the first individual in each replicate to make contact with an uninfested butternut in the presence or absence of prey odours were compared using a one tailed Mann-Whitney U-test, $\alpha = 0.05$. The arenas were placed on top of a wire gauze box containing either clean butternuts or *A. nerii*-infested butternuts.

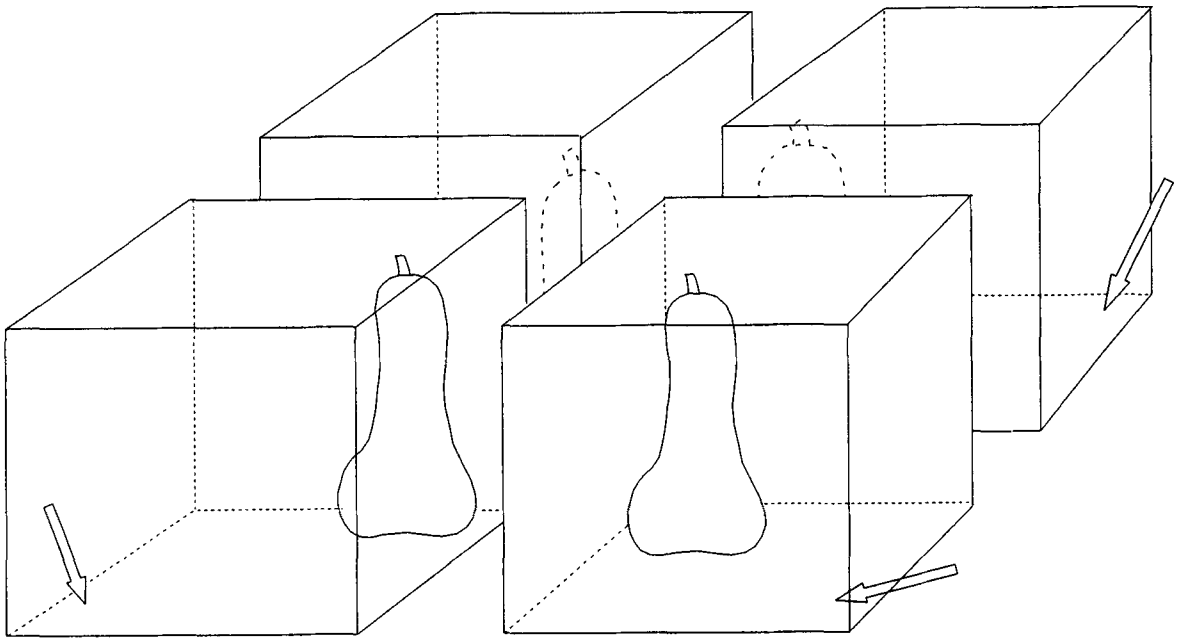

Fig. 2. Four arenas in which the sensory modalities involved in the location of prey patches were investigated. The arrows indicate the sites for release of the beetles.

Experiment 4: olfaction and foraging paths. The arena had a circular perspex disk covered in opaque white paper as a floor (Fig. 3). A 50 mm high PVC collar with gauze-covered circular holes, supported a perspex disk above the floor. The arena was placed on top of the wire box described above, which contained either clean butternuts or *A. nerii*-infested butternuts. Air was extracted from a hole in the floor, and beetles were inserted through a hole in the lid. Adult beetles were starved for 24 h prior to being placed in the arena. Their foraging paths were video taped, from commencement of movement until contact was made with another individual, the perimeter of the arena, or the hole in the centre of the arena floor. The speed of movement and number of turns in excess of 20 ° in the presence and absence of prey odour were compared using Mann-Whitney U-tests, $\alpha = 0.05$.

Experiment 5: olfactometers. A Y-tube olfactometer was made of 40 mm diameter glass tubing with a 185 mm long stem and two arms of 110 mm which were 47 ° apart. Air was extracted from the base of the stem. Two glass chambers of 5 l each, contained 0.5 l of water to balance the humidity of air drawn through each into a branch of the Y-tube. Mixing of the air from the two arms was observed using white NH_4Cl gas drawn through one arm and clean air through the other. The minimum flow rate whereby contamination of the air in one arm with air from the other, could be avoided, was 15 l min^{-1}. The apparatus was surrounded by 0.75 m-high walls of white polystyrene to eliminate visual aspects of the surroundings. Fluorescent and incandescent lighting was provided vertically above the apparatus and the room temperature was between 25 °C and 27 °C.

Forty *C. nigritus* adults, starved for 24 h, were inserted at the base of the stem and the numbers in the two branches periodically recorded. A control was run where clean air was drawn through both branches. A trial was conducted in which one glass chamber contained only the flask with water, while the other additionally contained three butternuts encrusted with *A. nerii*.

Another olfactometer consisted of a straight pipe of clear perspex, 53 mm in diameter and 1 m long (Fig. 4). Both ends were covered in gauze and inserted into cardboard boxes. Clean butternuts were placed in one box and *A. nerii*-infested butternuts in the other. Air was extracted from the centre of the tube at 0.4 l min^{-1}. The apparatus was also surrounded by polystyrene walls. Forty *C. nigritus* adults, starved for 24 h, were inserted at the centre of the tube. The numbers in the terminal 0.3 m of each end were periodically recorded.

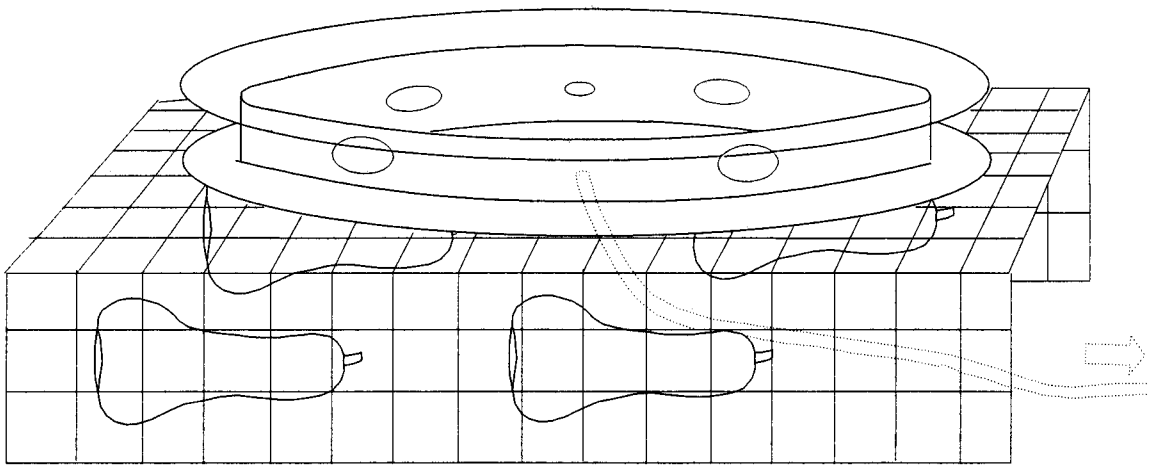

Fig. 3. Foraging arena in which the effect of prey odour on the searching paths of *C. nigritus* was investigated. Air was extracted from the arena through a pipe as indicated by the arrow.

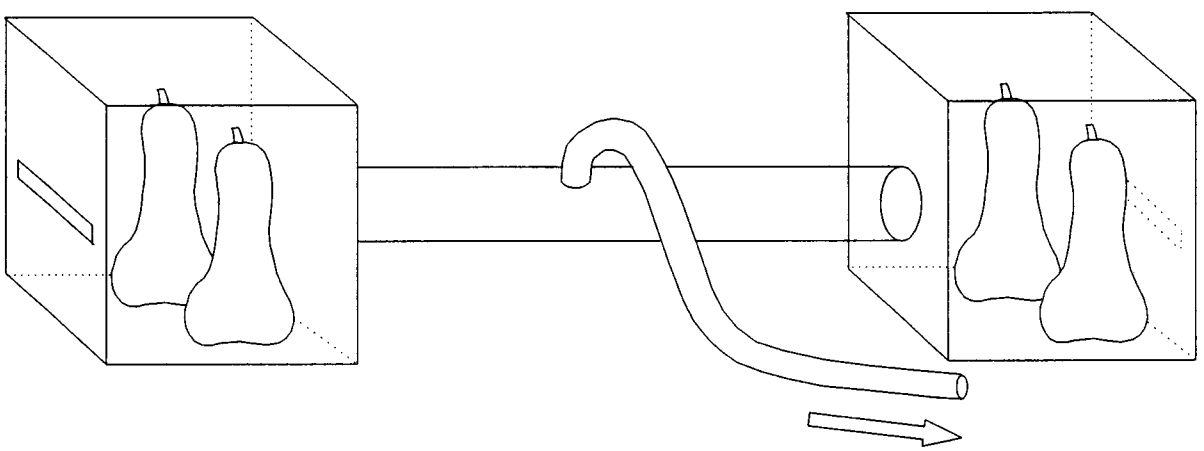

Fig. 4. Olfactometer in which the response of *C. nigritus* to prey odour was investigated. Air was extracted through a pipe as indicated by the arrow.

Movement patterns

Search patterns of *C. nigritus* adults and larvae on the upper surface of a large pumpkin *Cucurbita maxima* cv. Flat White Boer, with a circular boundary of polyester fibre padding, 250 mm in diameter, were video taped. Recordings of extensive foraging after 24 h of starvation, commenced 5 min after introduction into the arena. Each individual was then presented with a mature female *A. nerii* and the after-feeding foraging patterns filmed. Paths were recorded until the foragers either made contact with an obstacle or flew away. The arena was wiped with 90% ethanol between replicates.

The distances travelled per 10 s were calculated. The ratio of distances travelled to straight line distances between positions, at 10 s intervals, quantified tortuosity of the path (Nakamuta, 1985). Sizes of individual turns larger than 20 ° were measured. Long sweeping turns were divided into sections equivalent to 1.5 body lengths of a fourth instar larva, as the majority of discrete turns were completed over a shorter distance.

Detection of individual prey

Experiments were conducted in the white room as described under patch location. Arenas comprised plastic petri dishes, 87 mm in diameter and 15 mm deep, with white filter paper on the base. Each arena was placed on a wooden platform with a hole through which a glass vial was inserted (Fig. 5). One of the following was placed at the prey site: an intact mature female *A. nerii* (visual and olfactory aspects of prey

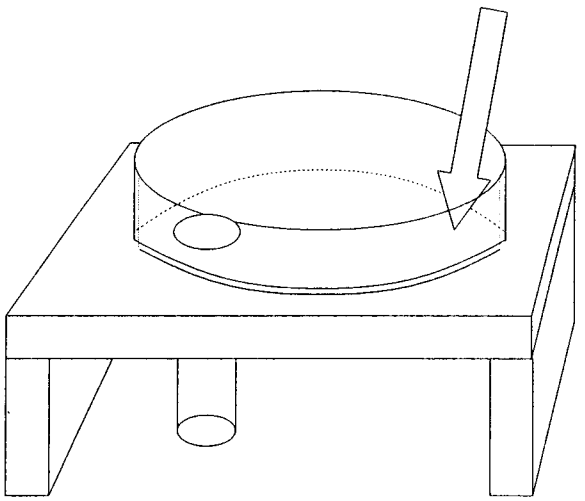

Fig. 5. Arena in which the sensory modalities involved in the detection of individual prey items by *C. nigritus* were investigated. The arrow indicates the prey site.

present); an imitation of a scale insect made from bees wax (only visual aspects); a drop of macerated *A. nerii* absorbed by the filter paper (only olfactory aspects); a faint pencil mark (control).

C. nigritus adults and fourth-instar larvae were starved for 24 h. Five individuals were placed in a vial with a polyester fibre plug in the neck and allowed 10 min to settle after handling. The plug was removed with minimal disturbance and the vial inserted into the petri dish arena. The time was recorded from emergence of the first individual into the petri dish to contact with the prey site (Fig. 5).

Results

Biotope location

a) In the control, there were no significant differences between the numbers of beetles at either end of the perspex tunnel at various times after introduction (Table 1, Fig. 1).

b) There were significantly more beetles at the end of the tunnel facing the vertical stripes than at the opposite end facing the horizontal stripes at 45 min and 75 min after introduction, but not at other times (Table 1).

c) In all the counts, significantly more beetles occurred at the end facing the painting of a horizon with a tree line than at the opposite end facing a flat horizon (Table 1).

d) There were significantly more beetles at the end facing vertical stripes than at the end facing the tree shape at 135 min, but not after shorter periods of exposure (Table 1).

e) There were significantly more beetles at the end facing the painting of citrus leaves, than at the end facing the squares, at 30 min and 60 min, but not after longer periods of exposure (Table 1).

f) There were significantly more beetles at the end facing the *C. sinensis* leaves than at the end facing the *J. mimosifolia* leaves at 60 min, but not at other times (Table 1).

Patch location

Experiments 1, 2 and 3. Larvae did not locate infested butternuts significantly more rapidly than uninfested butternuts (Table 2, Fig. 2). Infested butternuts were located by adults more rapidly than uninfested butternuts when each was presented separately. Infested butternuts were not located more rapidly than uninfested butternuts by adults in the choice experiment. Uninfested butternuts were located more rapidly by adults in the presence of prey odour than in the absence of prey odour.

Experiment 4. There was no significant difference between the mean speed [$=6.2$ mm s^{-1} ± 0.4 ($\pm$1SE), $n=33$] and the mean number of turns per mm travelled multiplied by 100 [$=4.3 \pm 0.3$ ($\pm$1SE), $n=33$], in the presence of scale odour, and without prey odour [6.1 mm s^{-1} ± 0.3 ($\pm$1SE), $n=31$ and 3.8 ± 0.2 ($\pm$1SE), $n=31$], (Mann-Whitney U-tests, $P>0.05$, Fig. 3). When using movement paths only from the first 5 min after release, there were also no significant differences (Mann-Whitney U-tests, $P>0.05$): a) In the presence of scale odour, mean speed was 7.7 mm s^{-1} ± 0.5 ($\pm$1SE), $n=10$ and mean $100\times$ (turns mm^{-1}) was 3.3 ± 0.3 ($\pm$SE), $n=10$; b) In the absence of prey odour, speed was 7.2 mm s^{-1} ± 0.7 ($\pm$1SE), $n=11$ and $100\times$ (turns mm^{-1}) was 3.2 ± 0.3 ($\pm$SE), $n=11$.

Experiment 5: olfactometers. In the control, counts of beetles in each branch of the Y-tube were not significantly different at any of the times after introduction (Table 3). There were also no significant differences between counts in the two arms when clean air was drawn through one, and air carrying *A. nerii* odour through the other (Table 3).

In the control with the straight line olfactometer, there were no significant differences between the

Table 1. Mean numbers of *C. nigritus* adults in the terminal 0.5 m of either end of a cylindrical tunnel, at various times after introduction into the apparatus, with various visual images presented at opposite ends, against the walls of the experimental room

Images compared		Replicates *n*	Mean numbers of beetles at either end of the tunnel at various times after release				
			15 min	45 min	75 min	105 min	135 min
a)	control: no images vs. no images	10	4.6 ; 5.8	8.7 ; 10.3	10.8 ; 12.2	–	12.8 ; 14.4
b)	vertical lines vs. horizontal lines	11	7.6 ; 5.8	14.3* ; 10.8*	18.6* ; 12.6*	19.5 ; 15.0	–
c)	horizon + tree line vs. flat horizon	10	11.2* ; 6.7*	15.3* ; 9.4*	19.1*; 10.8*	23.6* ; 10.4*	24.4* ; 11.4*
d)	vertical lines vs. tree shape	12	7.3 ; 7.4	13.1 ; 12.2	15.6 ; 12.9	16.2 ; 10.8	17.2* ; 13.5*
			30 min	60 min	90 min	120 min	
e)	painted ovate leaves vs. squares	16	9.2* ; 6.7*	13.4* ; 9.1*	13.5 ; 11.8	13.6 ; 12.3	
f)	real leaves: *C. sinensis* vs. *J. mimosifolia*	17	12.4 ; 9.8	15.5* ; 10.7*	17.1 ; 13.6	15.2 ; 13.2	

* significantly different, $\alpha = 0.05$, permutation test for related samples for the first four comparisons, Wilcoxon signed ranks test for the last two comparisons.

Table 2. Time taken for first *C. nigritus* to make contact with a butternut, infested with *A. nerii* or uninfested, mean (s) ± 1SE (n)

Life stage	Experiment 1	
	Uninfested butternut	Infested butternut
Adults	351a ± 90 (15)	132b ± 30 (15)
Larvae	2132a ± 460 (20)	1727a ± 291 (20)
	Experiment 2: Choice	
	Uninfested butternuts	Infested butternuts
Adults	306a ± 84 (17)	246a ± 66 (17)
	Experiment 3	
	Uninfested butternut + odour	Uninfested butternut × odour
Adults	174a ± 36 (24)	348b ± 72 (24)

A different letter following numbers in the same line indicates a significant difference, $\alpha = 0.05$, Mann-Whitney U-tests for experiments 1 & 3 and Wilcoxon Signed Ranks Test for the choice experiment.

counts at the two ends of the tube (Table 4, Fig. 4). No significant differences were found between counts at the end attached to the source of scale insect odour, and the opposite end without prey odour (Table 4).

Movement patterns

The search paths of *C. nigritus* adults and larvae are summarised in Table 5. For the first 60 s after feeding, the speed, tortuosity of search path and turns/distance travelled, for adults and larvae were significantly different from the measurements before feeding. During the period 12 min to 14 min after feeding, these measurements had reverted back to levels similar to those prior to feeding.

Table 3. Mean numbers of *C. nigritus* adults in the two branches of a Y-tube olfactometer at various times after introduction of 40 beetles per replicate into the apparatus, n=10 (Experiment 5)

Time after introduction (min)	Mean (±1 SE) numbers of beetles in the two arms of a Y-tube olfactometer			
	Control		Treatment	
	Branch A no prey odour	Branch B no prey odour	Branch A no prey odour	Branch B + prey odour
5	2.2 ± 0.5	2.5 ± 0.7	3.7 ± 0.5	2.8 ± 0.7
10	2.4 ± 0.6	2.9 ± 0.7	4.6 ± 0.7	2.8 ± 0.8
15	2.4 ± 0.7	2.9 ± 0.5	4.0 ± 0.8	2.5 ± 0.7
20	2.6 ± 0.5	2.7 ± 0.5	3.5 ± 0.8	2.6 ± 0.7
25	2.9 ± 0.4	2.8 ± 0.5	3.3 ± 0.8	2.6 ± 0.9
30	2.5 ± 0.5	2.8 ± 0.7	4.1 ± 0.6	2.7 ± 0.9
40	2.8 ± 0.5	3.0 ± 0.7	3.4 ± 0.6	2.9 ± 0.8
50	3.6 ± 0.8	4.0 ± 0.7	3.0 ± 0.7	3.7 ± 0.9
60	–	–	3.0 ± 0.9	3.4 ± 1.1
75	–	–	3.2 ± 0.7	2.7 ± 0.5
100	4.3 ± 0.8	4.1 ± 1.0	3.7 ± 0.7	2.5 ± 0.6
115	5.2 ± 1.0	5.1 ± 1.1	–	–
130	4.8 ± 0.9	5.6 ± 1.0	4.3 ± 0.7	2.8 ± 0.7
200	5.8 ± 0.6	5.4 ± 0.8	–	–

No significant differences between counts in the two branches at the different times after introduction in both control and treatment, permutation test for related samples, $\alpha = 0.05$.

Table 4. Mean numbers (±1 SE) of *C. nigritus* adults in the terminal 0.2 m and 0.3 m at either end of an olfactometer, at various times after introduction of 40 beetles per replicate into the apparatus, n=20 (Experiment 5)

Time after introduction (min)	Mean numbers at either end of the olfactometer in the terminal 0.2 m		terminal 0.3 m	
	Side A no prey odour	Side B no prey odour	Side A no prey odour	Side B no prey odour
10	3.5 ± 0.5	3.6 ± 0.5	3.8 ± 0.5	3.7 ± 0.5
20	3.8 ± 0.5	3.5 ± 0.5	4.3 ± 0.5	3.9 ± 0.7
30	4.0 ± 0.7	3.7 ± 0.6	4.6 ± 0.6	4.2 ± 0.6
	Side A no prey odour	Side B + prey odour	Side A no prey odour	Side B + prey odour
10	4.2 ± 0.5	3.6 ± 0.5	4.5 ± 0.5	4.0 ± 0.6
20	4.7 ± 0.7	3.0 ± 0.5	4.9 ± 0.6	3.3 ± 0.5
30	4.1 ± 0.6	3.2 ± 0.5	4.5 ± 0.6	3.3 ± 0.5

No significant differences between counts at either end of the apparatus, at the different times after introduction, in both control and treatment, Wilcoxon signed ranks test, $\alpha = 0.05$.

Detection of individual prey

Adults located the complete scale insect (vision and olfaction), the wax imitation (vision) and the site of absorbed macerated scale (olfaction) more rapidly than the control site (Table 6). Larvae did not locate the

Table 5. Analysis of *C. nigritus* search paths before and after feeding; speed, tortuosity factor (path distance/straight line distance for each 10 s period) and turns/path distance

Measurement	Before feeding	First 60 s after feeding	12 to 14 min after feeding
Adult			
speed (mm s^{-1})	8.9a ± 0.8 (6)	4.3b ± 1.1 (6)	8.1a ± 1.1 (4)
tortuosity factor	1.21a ± 0.05 (6)	2.26b ± 0.31 (6)	1.37a ± 0.9 (4)
100 × (turns mm^{-1})	4.1a ± 0.8 (6)	19.5b ± 2.3 (6)	7.3a ± 1.3 (6)
Larvae			
speed (mm s^{-1})	6.3a ± 0.2 (5)	3.5b ± 0.3 (5)	5.8a ± 1.0 (4)
tortuosity factor	1.42a ± 0.9 (5)	1.86b ± 0.2 (5)	1.50a ± 0.1 (4)
100 × (turns mm^{-1})	3.6a ± 1.2 (5)	10.6b ± 1.7 (5)	7.3a ± 1.9 (4)

A different letter following numbers in a column (adults and larvae separate), indicates a significant difference, Friedman ANOVA followed by a nonparametric multiple comparison, $\alpha = 0.05$.

Table 6. Time taken for first *C. nigritus* adults and fourth instar larvae to locate the prey site, mean (s) ± 1SE (n)

Life stage	Prey site			
	A. nerii	Wax imitation	Macerated *A. nerii*	Control
Adults	217a ± 58 (20)	311a ± 75 (20)	326a ± 76 (19)	694b ± 117 (20)
Larvae	536a ± 112 (20)	–	–	633a ± 139 (20)

A different letter following numbers in a column indicates a significant difference, $\alpha = 0.05$, Kruskal Wallis ANOVA followed by a nonparametric multiple comparison for adults, and a Mann-Whitney U-test for larvae.

complete scale insect significantly more rapidly than the control site.

Discussion

Aggregation

Aggregation of large numbers of individuals is a common feature among coccinellids (Hagen, 1962). This behaviour often coincides with weather conditions which are unfavourable for the beetles. Most aggregating species are attracted to prominent objects silhouetted on the horizon (Hagen, 1962; Hodek, 1973). A characteristic of aggregation behaviour is that they do not feed at these sites and enter a state of diapause (Hagen, 1962). Tirumala *et al.* (1954), Ketkar (1959) and Ahmad (1970) observed congregation of *C. nigritus*, mostly on the undersurface of leaves, but also on fruits and branches of banyan trees *Ficus benghalensis* in India and Pakistan. This behaviour was accompanied by a form of diapause during which they did not feed (Tirumala *et al.*, 1954).

In South Africa, this species congregates on giant bamboo *D. giganteus* during the winter months, but it does not form tight groups (Samways, 1984). This behaviour is also different in that the beetles continue feeding and reproducing, although qualitative observations indicate that the reproductive rate is considerably lower. *C. nigritus* were also found congregating on the underside of *Ficus sur* Forssk. leaves, where such plants were adjacent to clumps of *D. giganteus*. They formed small groups of two to five beetles and were immobile. There was no prey on these plants, and there were no immature stages.

Qualitative observations suggest that seasonal rhythmicity persists to a degree, during rearing in the laboratory under controlled environmental conditions (26±1 °C, 14L:10D). Reduction in reproductive rate was observed in South Africa from late summer to late winter. In Pakistan and India, congregation and

diapause occurred during winter or summer depending on the region (Tirumala *et al.*, 1954; Ketkar, 1959; Ahmad, 1970). The geographical origins of the material which originally entered southern Africa is unknown (Samways, 1984), and therefore it is not possible to determine whether rhythmicity of the South African biotype developed through local adaptation, or was inherent in the founder population. However, the aggregation on bamboo appears to be a remnant of the aggregation behaviour observed in India and Pakistan.

Location of landscape patterns, biotopes and prey patches

The physical handling of the beetles during introduction into the perspex tunnel was probably disruptive. There may also have been a delay in the response of the beetles to the visual cues on initial exposure. Therefore the absence of significant responses to visual cues during the first 15 min of exposure, were not regarded as meaningful.

The importance of visual aspects of the landscape in the location of biotopes by these predators was demonstrated in this study. During foraging, *C. nigritus* was optically attracted to prominently silhouetted features, such as a horizon with a tree line and individual trees. Vertically oriented parallel lines were also more attractive to these predators than horizontal lines. This explains their congregation on *D. giganteus* which are up to 20 m tall and grow in dense stands, in which the vertical stems are visually prominent. The same process may apply to *C. nigritus* settling in the crowns of palm trees.

Most of the plants on which *C. nigritus* is regularly encountered, have leaves of similar shape, being ovate, elliptic or oblong, e.g. citrus, guava, mango, banyan, coffee and *Ficus* spp. This suggests that leaf shape, in addition to tree or biotope shape, is an important visual cue for fine aspects of biotope location.

In this study, *C. nigritus* showed a preference for ovate leaves above compound bipinnate leaves. To eliminate the possibility of leaf size and subtle colour differences having effected this response, the attractiveness of paintings of leaves were compared with equal-sized squares. The leaf paintings were again preferred, indicating that it was the shape of the leaf which was recognised.

Habituation and a responsive hierarchy of cues

It was established that vertical lines were attractive to *C. nigritus*. The absence of a preference for vertical lines above the painting of a tree, indicated that the tree image was also attractive. However, after 135 min of unsuccessful foraging in the presence of these images, a preference for the vertical lines became evident. Similarly the preference for painted leaves above painted squares, and for *C. sinensis* leaves above *J. mimosifolia* leaves, and for vertical lines above horizontal lines, was no longer evident after various periods of exposure without additional stimulus. This may be explained by habituation to these cues.

The preference for the vertical lines above the painting of a tree after prolonged exposure, may indicate a hierarchy in their responsiveness to cues. The vertical lines may be perceived as a longer-range cue than the tree. In the event of unsuccessful foraging, it would be advantageous to habituate to shorter-range cues more rapidly than to longer-range cues. The absence of a significant difference between the counts in the ends of the tunnel facing the vertical and horizontal lines at 105 min after introduction, is only marginally insignificant, $P = 0.055$. This indicates that this apparent anomaly may not be real.

Responses to shapes of cues at various spatial scales

The shapes of visual cues to which the coccinellids responded were all similar. Apart from vertical lines, all the other attractive cues, were either dome-shaped or in the shape of two domes with abutting flat sides. These are tree shapes on a horizon, individual trees, leaves and the profile of individual scale insects. Hodek (1973) illustrates landscape features at which aggregations of coccinellids were found. These too are characterised by dome shapes. Further, the profile of individual coccinellids also has this shape, which may be significant in their aggregation behaviour.

Simple feature-detector neural circuits are common controllers of behaviour in flying insects (Gould, 1982). The similar shape of cues utilised by coccinellids, suggests that such a feature-detector is involved. The shapes of the cues do not vary but the distance from the cue and therefore the time required to reach the site varies. Further, as the distance at which the cue may be perceived decreases, so the number of items increases disproportionately, giving the occurrence of cues a partially fractal nature (see Mandelbrot, 1983; Morse *et al.*, 1985).

Further, the fractal nature of the cues means that the short-range cues are more plentiful, and therefore in a profitable biotope will be encountered at a higher rate than longer-range cues. Response to a short-range cue (such as the image of an individual scale insect) is reinforced by a contact stimulus from the prey item. This contact stimulus occurs shortly after the visual stimulus as both occur at short range. This explains the value of a positive correlation between the duration of responsiveness to a cue and the distance from it.

Olfactory versus visual cues in the location of prey patches

In Experiment 1, where infested and uninfested butternuts were presented to adult *C. nigritus* on separate occasions, the beetles located infested butternuts more rapidly than uninfested butternuts. This could have been in response to visual or olfactory stimulation. In Experiment 2, where individuals were presented with a choice between infested and uninfested butternuts, infested butternuts were not located more rapidly than uninfested butternuts. This suggests that it is unlikely that the response was a taxis (source-oriented).

In Experiment 3, the times to locate uninfested butternuts in the presence and absence of prey odour were compared. The presence of prey odour, although not emanating from the uninfested butternut, resulted in a more rapid discovery thereof than in the absence of prey odour. This confirms that patch location is facilitated by olfactory stimulation, but that the response is not source-oriented (i.e., it is a nondirectional sensory cue). This was again confirmed by *C. nigritus* adults not selecting the odour-carrying branches of the olfactometers.

Experiment 4 indicated that there were no differences, characteristic of alternations between intensive and extensive search, between movement patterns in the presence or absence of prey odour. A possible explanation for these findings is that prey odour stimulated the commencement of, and sustained, foraging behaviour, precluding other activities such as cleaning and resting.

The discovery of prey in the presence of prey odour may also have involved increased sensitivity to a visual key, triggered by this olfactory stimulation through cross-channel potentiation. If this cue, to which the beetles had become sensitive, was not specific enough to discriminate between scale-infested and uninfested sites, it would explain their inability to select infested butternuts above clean butternuts in the choice experiment.

There is still a great deal which remains to be discovered about the stimuli effecting biotope and patch location and selection. Possible interactions between visual and olfactory stimuli, and the role played by learning, should be investigated.

Prey location by adults versus larvae

Experiment 1 (patch location), indicated that prey odour was involved in the location of prey patches by adults but not by larvae. *C. nigritus* adults were capable of locating individual prey visually and olfactorily over short distances (detection of individual prey). Fourth-instar larvae relied on physical contact to detect individual prey. Adults and larvae altered their movement patterns from typically extensive search to intensive search following prey consumption (movement patterns). This reverted back to extensive search with time when subsequent prey were not encountered. This type of foraging maximises the encounter rate of patchily-distributed prey (Hassell & Southwood, 1978; Carter & Dixon, 1984).

Biotope selection for coccinellid larvae is performed by adults (Blackman, 1967; Hodek, 1973).Therefore, there is less likelihood of larvae having to locate prey in an unsuitable biotope than adults. Further, adults being active dispersers are readily able to locate prey. It seems then that mobility is coupled to visual sensitivity and that adults can locate prey patches and individual prey much more effectively from a distance than can larvae.

Significance of adult responses relative to biocontrol

As *C. nigritus* readily locates *D. giganteus*, and shuttles between alternative biotopes, it would be valuable to promote planting *D. giganteus* in close proximity to orchards. During winter, when the prey population levels are low in the citrus orchards, and stringent chemical control is applied, such sites would serve as valuable reservoirs of the biocontrol agents. *D. giganteus* is an alien species to southern Africa, but it is not an aggressive invader and is highly dependent on plentiful water, restricting it to damp areas, such as beside farm dams.

Provided that the scale-insect prey is present, the beetle is maintained in the area by its response to the visual cues. At the largest spatial scale, it is responding to the citrus-tree or bamboo-clump tree line. Then at

the next level down it is responding to the ovate leaves. While at the smallest level not only is it responding to the shape of other conspecifics but also to the shape and olfactory stimuli from individual prey items.

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