Project Title	Protected herbs, ornamentals and celery: development of an on-nursery rearing system for *Atheta coriaria* for reduced cost biological control of sciarid and shore flies.
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Project leader:	Jude Bennison, ADAS
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Location of project:	ADAS Boxworth, STC and commercial nurseries
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The results and conclusions in this report are based on a series of experiments conducted over a one-year period. The conditions under which the experiments were carried out and the results have been reported in detail and with accuracy. However, because of the biological nature of the work it must be borne in mind that different circumstances and conditions could

produce different results. Therefore, care must be taken with interpretation of the results, especially if they are used as the basis for commercial product recommendations.

AUTHENTICATION

I declare that this work was done under my supervision according to the procedures described herein and that the report represents a true and accurate record of the results obtained.

Jude Bennison
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GROWER SUMMARY

Headline

Atheta adults significantly reduced sciarid flies in parsley pots and in soil grown celery however the high release rates needed would be too expensive if they were bought commercially. A nursery based rearing release system may produce sufficiently high rates and release rates from the rearing boxes can be greatly increased by not feeding the units for one week before release.

Background and expected deliverables

Sciarid and shore flies are common pests and contaminants respectively on many protected edible and ornamental crops. Sciarid fly larvae cause crop losses by damaging roots and stems and shore fly adults can cause marketing problems in herbs, potted ornamentals and celery due to the presence of flies or their droppings on the marketed plants or pots. Although entomopathogenic nematodes (e.g. *Steinernema feltiae*) and predatory mites (*Hypoaspis* spp.) are available for sciarid fly control, they are expensive and do not always give reliable control of sciarids on protected herbs. Neither of these biological control agents gives effective control of shore flies. An alternative control is needed as reliable and cost-effective biological control options are necessary for both pests for use within IPM programmes.

The predatory beetle, *Atheta coriaria* is known to feed on both sciarid and shore fly eggs and larvae, and has recently become commercially available. Direct releases of *Atheta* have reduced numbers of both sciarid and shore flies in experiments on ornamental crops. However, grower experience has been variable. Recent grower-funded research has indicated that *Atheta* can contribute to reducing shore flies and crop losses on celery. Grower releases of *Atheta* adults have given unreliable control of shore flies on pot herbs, where control needs to be very effective to meet stringent retail standards. Releases of *Atheta* as either adults or mixed life stages did not control shore flies in a trial on a pot herb nursery within project PC 210 in 2004. An improved and cost-effective release method for establishing high numbers

of *Atheta* in susceptible crops, leading to improved predator performance and more effective and economic control of target pests, is needed.

Following research in Canada in 2002 on using an artificial diet (trout pellets) to rear *Atheta*, a UK commercial nursery (WJ Findon & Son) experimented with using 'breeding boxes' for *Atheta* in crops of poinsettia and cyclamen during 2004. The system produced large numbers of the predators at very little cost and good control of sciarid flies was achieved in both crops. Other growers of ornamentals tried the system with variable success. Fungal contamination in the 'breeding boxes', variable production rates of *Atheta* and potential negative interactions between *Atheta* and other biological control agents and naturally-occurring invertebrates need to be resolved. Further scientific development and testing of the system is needed.

The overall expected deliverable of the project is to develop an effective, reliable and practical rearing system for *Atheta*, to enable growers to rear large numbers of the predator on their own nurseries, for improved, low-cost biological control of sciarid and shore flies.

Work in year 1 of the project identified additional fundamental gaps in knowledge on *Atheta* behaviour and biology that need to be filled before further experiments are planned on commercial nurseries. Work in year 2 thus focussed on filling key gaps in knowledge, as agreed with HDC and the project co-ordinators.

The expected deliverables in year 2 were:

1. Determine numbers of *Atheta* needed to control relevant densities of sciarid and shore flies
2. Refine the detail on *Atheta* rearing-release units
3. Identify key aspects of *Atheta* behaviour in selected crops and growing substrates
4. Investigate the interactions between *Atheta* and other ground-dwelling biological control agents
5. Review current knowledge on compatibility of pesticides with *Atheta* in IPM programmes.
6. Identify options for further work on commercial nurseries.

Summary of the project and main conclusions

1. Numbers of Atheta needed to control relevant densities of sciarid and shore flies

- Adding *Atheta* adults at five or 10 per parsley pot (equivalent to 500 or 1000 *Atheta* per m² before pot spacing) reduced mean numbers of sciarid flies 22 days later, from 11.4 per pot in untreated pots, to 3 and 1.7 per pot respectively (75% and 85% reductions respectively), see Figure 1. Adding *Atheta* at one or two per pot (equivalent to 100 and 200 per m² before pot spacing) did not significantly reduce numbers of sciarid flies.

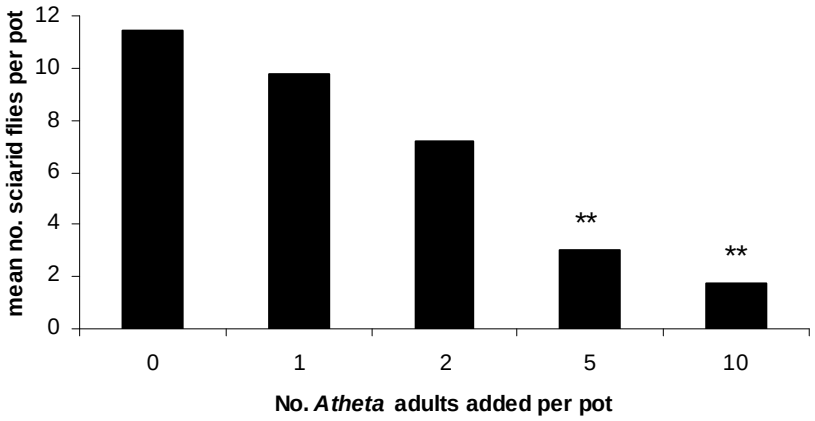


Figure 1. Mean numbers of sciarid flies and *Atheta* per pot of parsley, 22 days after adding *Atheta* adults at different rates per pot. ** significantly lower numbers than in untreated controls ($P<0.01$).

- Adding *Atheta* at rates of 25 per m² or higher, to samples of shore fly-infested soil removed from sequentially planted celery crops during weeks 14-19, led to significant reductions in numbers of adult shore flies emerging from samples taken in weeks 15, 16 and 19, when compared with numbers emerging from soil samples treated with the grower's standard release rate of *Atheta* at 5 per m², see Figure 2. On the final sampling date in week 19, when mean shore fly populations in control samples (treatment E) had increased to 85 per tray, equivalent to 1700 per m² soil, *Atheta* at 25 per m² reduced the numbers of shore flies to

10.5 per tray, equivalent to 210 per m^2 soil (88% reduction), see Figure 2. Adding *Atheta* at higher rates than 25 per m^2 (45, 205 and 305 per m^2) gave similar reductions in numbers of shore flies as adding the predators at 25 per m^2.

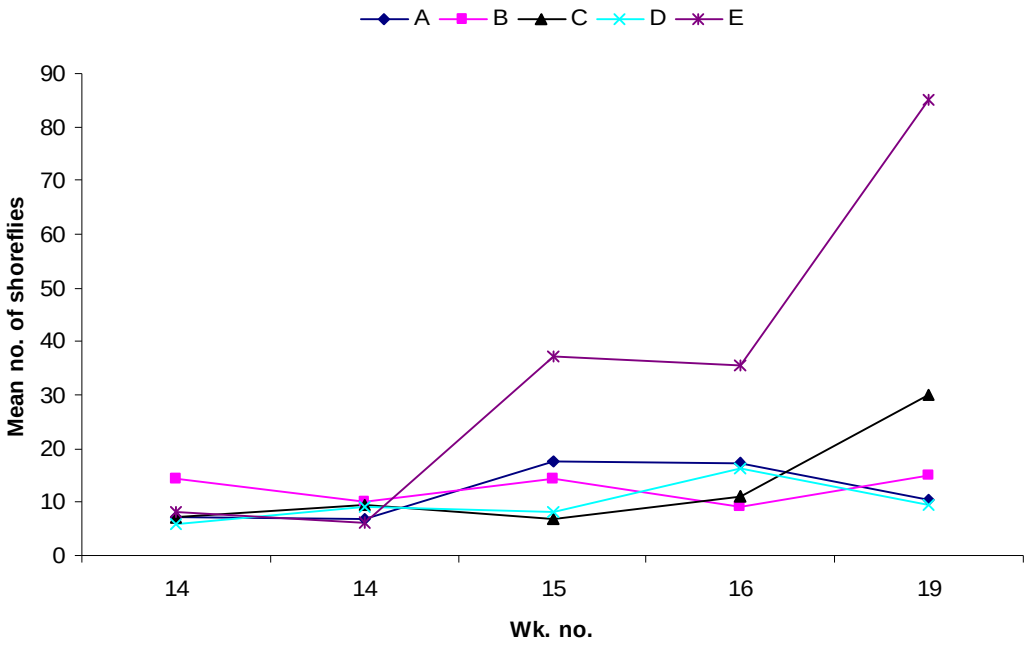


Figure 2. The effect of different rates of *Atheta* on numbers of shore flies emerging per tray from soil samples taken on weeks 14 (2 samples), and weeks 15, 16 and 19.
A, B, C, D and E are *Atheta* released at 25, 45, 205, 305 and 5 per m^2 respectively.

- The effective release rates in both the pot herb and celery experiments would be commercially unacceptable if using direct releases of *Atheta* bought from commercial suppliers. Further research is needed to determine whether the *Atheta* rearing/release system could enable sufficient predators to be released into these crops for improved, low cost control of sciarid and shore flies.

2. *Refine the detail on Atheta rearing-release units*

- *Atheta* rearing-release boxes (plastic boxes containing *Atheta* cultures in 1.5 litres of coir and vermiculite substrate, and fitted with ventilation/exit holes in the lids) were used in a 3-week glasshouse

experiment to determine whether food shortage could stimulate *Atheta* to leave the boxes. During the first week, a significantly higher percentage of beetles (47%) left boxes that had not been fed for one week, than boxes that had been recently fed with turkey grower crumbs (6%). On day 1, 18% of the *Atheta* left the unfed boxes and 2% left the fed boxes.

- On day 1 of the experiment, a significantly higher percentage of *Atheta* emerged from boxes with higher 'starting' numbers (e.g. 2,000 per box) than from boxes with lower densities of beetles (e.g. 1000 per box).
- Thus, if a quick-release of high numbers of *Atheta* are needed, e.g. when placing newly germinated herb plants in a production glasshouse infested with sciarid flies, boxes with high densities of *Atheta* should be used, and the boxes should not be fed for a week before the predators are released.

3. Key aspects of Atheta behaviour in selected crops and growing substrates

- *Atheta* adults and larvae were shown to successfully enter and breed in the compost in young pots of parsley and also in older pots with more dense compost and root systems.
- In soil-grown crops e.g. celery, bait pots sunk into the ground, with the rims level with the soil surface, are a better method for trapping and monitoring *Atheta* than those stood on the soil surface.

4. Interactions between Atheta and other ground-dwelling biological control agents

Aphidoletes is a predatory midge that is used for biological control of aphids. The larvae drop to the ground to pupate, where they might be potential prey for *Atheta*. *Atheta* adults ate *Aphidoletes* larvae when offered as the only prey in a Petri dish. However, *Atheta* did not significantly reduce numbers of *Aphidoletes* larvae and pupae in the compost of aphid-infested parsley plants, when alternative prey species e.g. sciarid flies and mites were present in or on the compost. The results indicate that *Atheta* and *Aphidoletes* are compatible in IPM. However, *Atheta* may have a negative impact on

Aphidoletes if alternative prey are not available or if the prey are in shorter supply than in this experiment.

5. Compatibility of pesticides with Atheta in IPM programmes

Growers need information on pesticide compatibility with *Atheta*, particularly for use within IPM on ornamentals. Current knowledge was collated on the side-effects on *Atheta* of key pesticides, commonly used against whitefly on poinsettia:

- Imidacloprid (Intercept 70WG applied as a compost drench, and Imidasect 5GR incorporated in the compost before potting) is harmful to both *Atheta* adults and larvae and should not be used together with *Atheta* in an IPM programme.
- No specific information is available on the side effects of thiacloprid (Calypso applied as a foliar spray, and Exemptor incorporated in the compost) on *Atheta*. However, this insecticide is known to kill other beetle species and both products are likely to have adverse effects on *Atheta*. Calypso may be less harmful to *Atheta* than Exemptor, due to the beetle's preference for living in the compost. However, when Calypso is applied as a high volume spray, some will reach the compost. Information is required on the specific side effects of Calypso and Exemptor on *Atheta*.
- Spiromefisen (Oberon) has no known effects on beetle species. It is likely that Oberon may be used together with *Atheta* in IPM programmes, although specific information on its compatibility is needed.
- Acetamiprid (Gazelle) is known to kill beetle adults and larvae, but there is no specific information available on the side effects on *Atheta*, and this information is now needed.

6. Options for further work on commercial nurseries

- As a result of progress in the project during year 1, several growers of pot herbs, ornamentals and HNS have adopted the *Atheta* rearing-release system developed in this project and have adapted it to their own needs and circumstances.

- The system has given promising commercial results against shore flies in bedding plant propagation and against sciarid flies in the propagation of herbs and ornamentals. Feedback from growers of protected HNS will be reported in the final project report.
- Further research progress during year 2 of the project has filled some key fundamental gaps in knowledge on *Atheta* biology and behaviour and about the potential of the rearing-release system. Knowledge gained on numbers of *Atheta* required to control known densities of sciarid and shore flies, methods for manipulating release of known numbers of the predators from rearing units, and improved methods for monitoring dispersal of *Atheta* in soil-grown crops will enable an improved system to be tested in commercial crops.

Financial benefits

It will not be possible to fully quantify the cost-benefits of using the *Atheta* rearing-release system until further experiments are done to test the system on commercial nurseries. The financial benefits of the system will depend on:

- The size and number of rearing units used per unit area of glasshouse.
- The lifespan, production and release rate of the units.
- The pest population size and the level of control given.
- Whether the system is used for rearing-release or for rearing followed by direct release (by hand).
- Staff time needed for maintenance of the rearing units, and for direct release if this option is used.

Action points for growers

- Some growers are already experimenting with their own *Atheta* rearing or rearing release units, on nurseries growing protected herbs and ornamentals. However, the full project results should be awaited before the system is considered for adoption on a large scale.
- Any growers interested in trying the rearing system should contact Jude Bennison for details, tel. 01954 228225, email jude.bennison@adas.co.uk

SCIENCE SECTION

Introduction

Sciarid and shore flies are common pests and contaminants respectively on many protected edible and ornamental crops. Sciarid fly larvae cause crop losses by damaging roots and stems, and shore fly adults can cause crop rejections or marketing problems in herbs, potted ornamentals and celery due to the presence of flies or their droppings on the marketed plants or pots. Shore flies can also spread root diseases e.g. *Pythium*.

Although entomopathogenic nematodes and *Hypoaspis* spp. predatory mites are available for sciarid fly control, these are expensive and do not always give reliable control of sciarids on protected herbs. Neither of these biological control agents gives effective control of shore flies at economically viable application rates. An alternative control is needed as reliable and cost-effective biological control options are necessary for both pests for use within IPM programmes.

The predatory beetle *Atheta coriaria* is known to be an effective predator of both sciarid and shore fly eggs and larvae and has recently become commercially available. *Atheta* has been shown to reduce numbers of both sciarid and shore flies in ornamental crops when released as mixed life stages (R. GreatRex, personal communication). Recent grower-funded research by Stockbridge Technology Centre (STC) has indicated that *Atheta* can contribute to reducing shore flies and crop losses on celery, but further work is needed to confirm consistent control. A system to reduce the cost of the predators and to improve their performance on various protected crops is desirable.

To date, commercial releases of *Atheta* adults have given unreliable control of shore flies on susceptible pot herbs, where control needs to be very effective to meet the stringent standards set by the retailers. Releases of the predator as either adults or mixed life stages did not give successful control in a trial on a commercial herb nursery in HDC project PC 210 in 2004 (Bennison,

2005; Bennison *et al*, 2005). This was possibly due to poor beetle survival after their initial release due to variable availability of shore fly prey eggs before adult fly releases were made to the trial plots. An improved release method for successful predator establishment in the crop and thus more effective control is needed.

Following research in Canada on using an artificial diet (trout pellets) to rear *Atheta* (Carney *et al.*, 2002), a UK commercial nursery (W J Findon & Son) experimented with using 'breeding boxes' for *Atheta* in crops of poinsettia and cyclamen during 2004. The system produced large numbers of the predators at very little cost and good control of sciarid flies was achieved in both crops. Other growers of ornamentals also tried the system with variable success.

Fungal contamination in the 'breeding boxes', potential problems with interactions between *Atheta*, other ground-dwelling biological control agents and other invertebrates attracted to the trout pellets, fungi or *Atheta* in the rearing boxes need to be resolved. With further scientific development and testing, a reliable on-nursery rearing system for maintaining a constant supply of large numbers of *Atheta* at very little cost has good potential for giving improved, low-cost control of both sciarid and shore flies.

The overall objective of the project is to develop an effective, reliable and practical rearing system for *Atheta coriaria*, to enable growers to rear large numbers of the predators on their own nurseries, for improved, low-cost biological control of sciarid and shore flies on various protected crops.

Work in year 1 of the project identified additional gaps in knowledge on *Atheta* behaviour and biology that needed to be filled before any further experiments are planned on commercial nurseries. Thus, the original work plan for year 2 was revised, and designed to fill some key fundamental gaps in knowledge that were agreed with HDC and the project co-ordinators. The need for further trials work on commercial nurseries, as originally intended for 2007, will be reviewed in autumn 2007 with HDC and the project co-ordinators.

Objective 1: Determine numbers of *Atheta* needed to control relevant densities of sciarid and shore flies

Sciarid fly experiment on herbs

Materials & methods

Sixty pots of young parsley plants were collected from a commercial pot herb nursery on 6 June 2007 and brought back to ADAS Boxworth. The newly emerged plants had been in the production glasshouse for only one day after being brought out of the germination room. Plants with similar numbers of sciarid fly eggs visible on the surface of the compost were selected. These eggs would not have had time to hatch into larvae at the time of collection, as sciarid fly egg hatch occurs in about 3-4 days. At ADAS Boxworth, each pot was checked to make sure that no sciarid fly adults were present, to prevent any further egg-laying, so that each pot had similar numbers of sciarid fly eggs. Each pot (8 x 8 cm) was then was placed inside its own 'fly emergence pot', i.e. a larger (one-litre) white plastic pot (12 cm diameter). *Atheta* adults were added to each pot at the following rates:

1. No *Atheta* per pot (untreated control)
2. One per pot (equivalent to 100 per m^2 as there are 100 pots per m^2 on the nursery, before pot spacing)
3. Two per pot (200 per m^2)
4. Five per pot (500 per m^2)
5. Ten per pot (1,000 per m^2)

There were 12 replicate pots per rate of *Atheta*.

A small yellow sticky trap (10x5 cm) was attached to the inside of each of the control pots, using a paper clip. Each pot was covered with a 'lid' of insect-proof mesh, secured with a rubber band. The pots were placed in a randomized block design on a bench in a glasshouse. The glasshouse temperature was set at a mean of 19°C, consistent with that used in the commercial herb glasshouse. A Tinytalk ® datalogger was placed in an empty white plastic 'emergence pot' covered with insect-proof mesh, to

monitor mean, maximum and minimum temperatures in the pots throughout the experiment.

The pots were watered as necessary through the mesh lid, to keep the compost moist. The sticky traps in the control pots were monitored to determine when the first sciarid fly adult emergence occurred. Two weeks after the experiment was set up, on 20 June, once the first sciarid flies were seen on the control pot traps, sticky traps were added to the remainder of the pots (sticky traps were not added to the pots containing *Atheta* at the start of the experiment, to avoid trapping the predators). The pots were then left for a further eight days, to allow all the sciarid flies to emerge.

Assessments

Numbers of sciarid fly and *Atheta* per pot were recorded using the following assessment methods:

- Numbers of sciarid and *Atheta* adults on each sticky trap were counted.
- Each plant, the compost in each pot and the inside of each 'emergence' pot was examined, and any sciarid flies and *Atheta* were counted.
- Any remaining sciarid fly larvae and *Atheta* adult and larvae in the compost were counted after Tullgren funnel extraction over a 48-hr period.
- The total numbers of sciarid flies and *Atheta* per pot, from the three assessment methods above were calculated.

Statistical analysis

The mean numbers of sciarid flies per pot when treated with the different rates of *Atheta* were compared using analysis of variance (ANOVA).

Results and Discussion

Glasshouse temperatures

Mean temperatures inside the emergence pots averaged *c*. 21°C throughout the experiment (Figure 1). Minimum temperatures were *c*. 17°C and maximum daily temperatures fluctuated between 21°C and 33°C, the latter

maximum temperature occurred only on one date, on 9 June. These temperatures were within the known temperature range of *Atheta* (12-35°C, Syngenta Bioline information; 15-32°C, Miller & Williams, 1983).

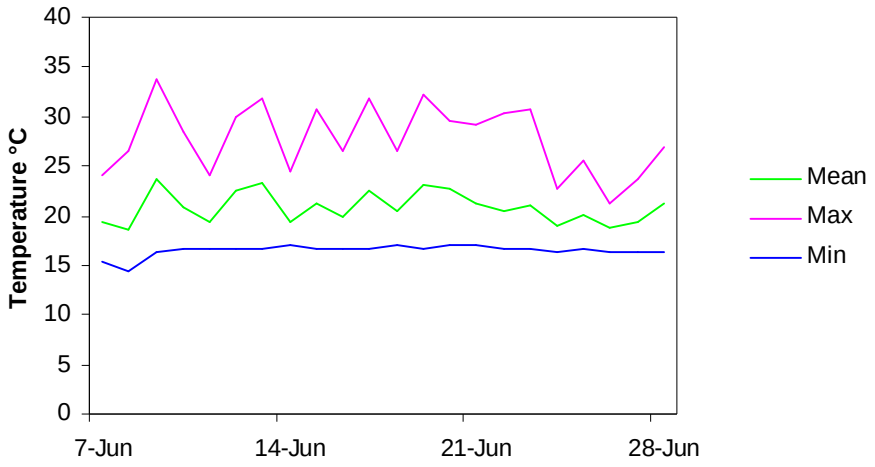


Figure 1. Mean, maximum and minimum temperatures in the emergence pots in the glasshouse, during the experiment to determine the numbers of *Atheta* needed to control relevant densities of sciarid flies on potted parsley.

Sciarid fly emergence

The first sciarid fly adults were seen on traps in untreated control pots 14 days after the experiment was set up (15 days after the pots were collected, when the sciarid fly eggs were up to one day old). Thus, the first sciarid adults emerged in the glasshouse 15-16 days after egg-laying. The sciarid flies were confirmed as *Bradysia difformis* (formerly known as *Bradysia paupera*, which is regarded as the most common damaging sciarid fly species occurring in UK glasshouses). Although no published information is available on the development rate of this species at different temperatures, the time taken in this experiment for eggs to develop into adults at mean glasshouse temperatures of 19-24°C is consistent with ADAS data for the same species when reared in the laboratory at 21-23°C, when egg to adult time was 14-18 days. This result highlights the need for sciarid fly infestations to be controlled during the first two production weeks of each herb crop, to reduce numbers of new generation adults which will lay further eggs on the crop, or infest newly emerging herbs in the same glasshouse.

Control of sciarid flies

In untreated control pots with no *Atheta* there was a mean of 11.4 sciarid flies per pot, 22 days after the experiment was set up (Table 1 and Figure 2). In pots treated with five and 10 Atheta adults per pot, there were significantly fewer ($P<0.01$) sciarid flies per pot (means of 3 and 1.7 per pot respectively), than in control pots. These reductions represented 74% and 85% control of sciarid flies by *Atheta*, when added at five and 10 adults per pot respectively. Sciarid fly numbers were not significantly reduced in pots treated with one or two *Atheta* adults per pot. Although *Atheta* added at five or 10 adults per pot had significantly reduced numbers of sciarid flies, 'zero tolerance' of the pest was only achieved in 25% and 33% of the pots respectively (Table 1). However, it is likely that if *Atheta* was used at the same rates on sequential crops of parsley, the sciarid fly population would be further reduced over time, and that zero tolerance of the pest would be achievable on a higher proportion of the crop. The effective rates of *Atheta*, five and 10 per pot, were equivalent to 500 and 1,000 per m^2 respectively (Table 1), as on the commercial nursery, there are 100 pots per m^2 before spacing. Such high rates would be uneconomic if the grower bought the *Atheta* from a commercial supplier, but might be possible if the *Atheta* were reared on the nursery. Such high rates of *Atheta* are unlikely to be necessary once the sciarid fly density on the nursery was reduced. Further work would be necessary to determine the rates of *Atheta* needed to reduce lower 'starting' numbers of sciarid fly eggs.

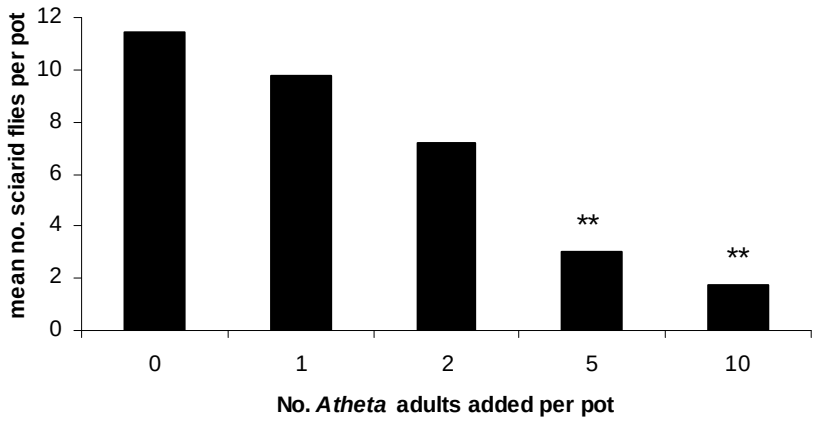


Figure 2. Mean numbers of sciarid flies and *Atheta* per pot of parsley, 22 days after adding *Atheta* adults at different rates per pot. ** significantly lower numbers than in untreated controls (P<0.01).

Table 1. Mean numbers of sciarid flies and *Atheta* per pot of parsley, 22 days after adding *Atheta* adults at different rates per pot. ** significantly lower numbers than in untreated controls (P<0.01).

Number of *Atheta* adults added per pot on day 0 (equivalent per m²)	Mean number of sciarid flies per pot on day 22 (equivalent per m²)	% pots with sciarid flies on day 22	Mean numbers of *Atheta* adults per pot on day 22	Mean numbers of *Atheta* larvae per pot on day 22	% pots with *Atheta* adults on day 22	% pots with *Atheta* larvae on day 22
0 (0)	11.4 (1140)	100%	0	0	0	0
1 (100)	9.8 (980)	100%	0.8	0.8	75%	33%
2 (200)	7.2 (720)	100%	1	1.2	75%	42%
5 (500)	3.0** (300**)	75%	2.9	1.3	100%	50%
10 (1000)	1.7** (170**)	67%	3.8	0.6	92%	25%

Reproduction of *Atheta* in herb pots

During the 22-day period of the experiment, the *Atheta* adults had produced larvae in up to 50% of the pots (Table 1). The absence of *Atheta* larvae in some pots is likely to have been due to only male adults having been added (it is not possible to determine the sex of *Atheta* on live adults due to them being so active). In addition, some *Atheta* larvae may have been eaten by the adults, particularly in the pots to which 10 *Atheta* adults had been added, in which most of the sciarid flies had been eaten and thus were less available as prey. Fewer *Atheta* adults were present in the pots at the end of the experiment than the number added 22 days earlier (Table 1). This is likely to have been due partly to natural mortality of the adults, which were of unknown age when added to the pots (adults live for approximately 21 days at 25°C, Carney *et al*, 2002), and also to the next generation of adults not yet

having developed (the time taken for adult to next generation of adults is 19-23 days at 25°C, Bennison 2007, and thus would take longer at the mean of 21°C during this 22-day experiment).

Shore fly experiment on celery

Materials & methods

Samples of soil with natural shore fly infestations were collected from the perimeter of glasshouses with recently planted celery crops. The soil samples were collected from the same nursery, from each of five crops that were planted between weeks 14 and 19, in order to test *Atheta* against increasing densities of shore flies. The soil samples were taken from two different glasshouses in week 14 and from other glasshouses in weeks 15, 16 and 19 respectively. Each of the samples was placed into small seed trays measuring 0.2 x 0.25m (0.05 m^2 surface area). There were four replicate samples for each of the five treatments A-E (see below).

Treatments

Atheta adults were released onto each tray at different rates (Table 2). Single releases of *Atheta* had been made by the grower to the glasshouse perimeter one week prior to the soil collection. Therefore the 'baseline' treatment for the experiment (and the control) were a standard release rate of *Atheta* of 5 per m^2. Therefore actual release rates for each treatment were as indicated in Table 2.

Table 2. Nominal and actual release rates of *Atheta* adults per tray (and equivalent per m^2) of soil collected from celery glasshouse celery

Code	Nominal release rate	Actual release rate
A	1 (20 per m^2)	25 per m^2
B	2 (40 per m^2)	45 per m^2
C	10 (200 per m^2)	205 per m^2
D	15 (300 per m^2)	305 per m^2
E (control)	0	5 per m^2

Each tray was then covered and sealed with a clear ventilated cover, and a yellow sticky trap was placed on the lip of the tray above the soil surface to trap emerging shore fly adults. The trays were then placed in a glasshouse (at STC) and kept at the same temperature regime as in the celery crops (minimum 8 ^{0}C, venting at 23^{0}C). The number of emerging adult shore flies was recorded over a 14-day period.

In addition to monitoring numbers of shore flies emerging from the soil samples, ten yellow sticky traps were placed around the perimeter of a commercial celery crop and were replaced weekly. Numbers of shore flies on the traps were recorded weekly between weeks 16 and 21.

Statistical analysis

Analysis of the data on numbers of shore flies in the soil samples was done on the square root transformed means, and means were compared using LSD at the 95% level of significance.

Results and Discussion

Control of shore flies

The effects of the different rates of *Atheta* introduced to soil samples removed from the commercial crops during weeks 14-19 against increasing

populations of shore flies are shown in Table 3 and Figure 3. There was no interaction of temperature as there was little variation in mean daily temperatures within the glasshouse at STC during this period (18.2 to 18.6 ^{0}C). Analysis of the data at each sampling date shows that from week 15 onwards, significantly more (P<0.05) shore flies emerged from soil samples with treatment E (standard release rate of *Atheta* at 5 per m^2) than from samples with all the higher introduced rates of the predator (Table 3).

Table 3. Mean numbers of adult shore flies (square root transformed means) emerging from soil collected from protected celery crops in successive weeks (14-19) and treated with different rates of *Atheta* (A-E).

Treatments	Week 14	Week 14	Week 15	Week 16	Week19
A (25 per m^2)	7.25 (2.59)	7.00 (2.59)	17.75 (4.17)	17.25 (4.09)	10.50 (2.99)
B (45 per m^2)	14.25 (3.70)	10.0 (3.11)	14.50 (3.47)	9.25 (2.90)	15.0 (3.84)
C (205 per m^2)	7.25 (2.19)	9.50 (2.83)	7.00 (2.55)	11.0 (3.28)	30.0 (5.46)
D (305 per m^2)	5.75 (2.25)	9.0 (2.97)	8.00 (2.71)	16.25 (3.98)	9.50 (2.87)
E (5 per m^2)	8.0 (2.99)	6.25 (2.403)	37.25 (6.06)	35.50 (5.86)	85.0 (9.18)
LSD(p>0.05)	(1.790)	(1.295)	(1.656)	(1.406)	(1.558)

Significant differences between treatments are calculated from transformed means using LSD at 95% significance.

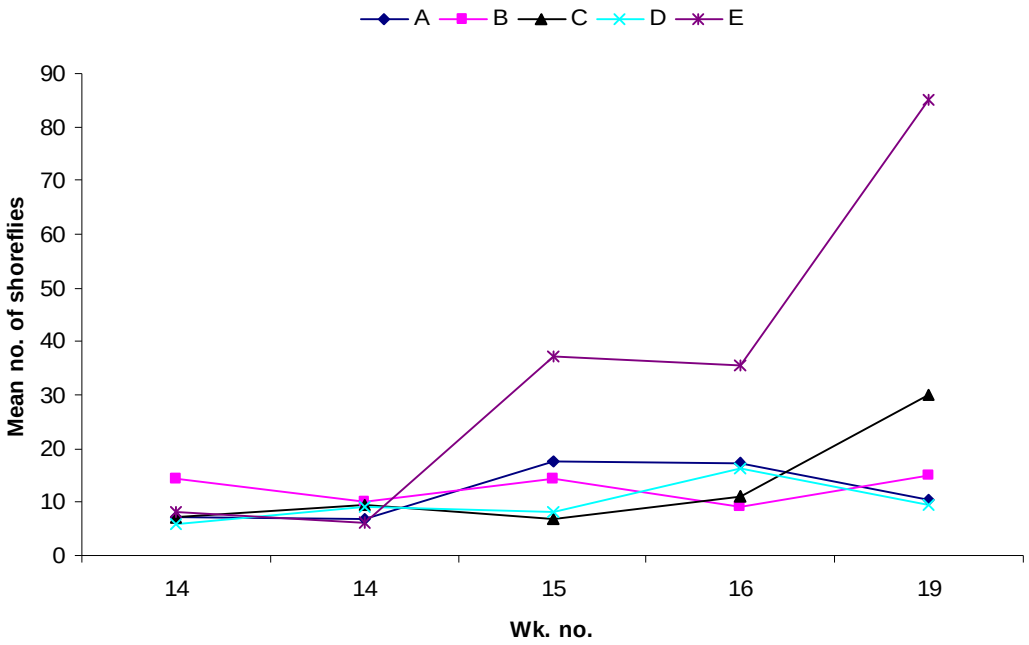


Figure 3. The effect of different rates of *Atheta* on numbers of shore flies emerging per tray from soil samples taken on weeks 14 (2 samples) and weeks 15, 16 and 19.
A, B, C, D and E are *Atheta* released at 25, 45, 205, 305 and 5 per m^2 respectively.

The results in Figure 3 also show that over time there was an increasing population of shore flies collected in the soil samples from the sequentially planted crops that had previously been treated with the standard release rate of *Atheta* (treatment E, 5 per m^2). This pattern of population growth corresponds to the increasing shore fly population observed in the celery crops at the commercial site (Figure 4). The results demonstrate that during this period of shore fly population growth, the higher rates of *Atheta* (treatments A-D) were able to significantly reduce the numbers of flies emerging from soil, when compared with the standard release rate. A mean of 85 shore flies per tray (1700 per m^2) emerged from control soil samples (treatment E, 5 per m^2) taken in week 19, whereas a mean of 10.5 shore flies per tray (210 per m^2) emerged from soil samples treated with treatment A, 20 per m^2 (Figure 3). This represents an 85% reduction in numbers of shore flies emerging from soil samples treated with 20 *Atheta* per m^2. The standard release rate of *Atheta* (5 per m^2) has previously been observed to reduce

shore fly populations within commercial celery crops; however the levels of control achieved required further improvement. In the experiment reported here, all rates of *Atheta* above the standard release rate were able to significantly reduce the shore fly population.

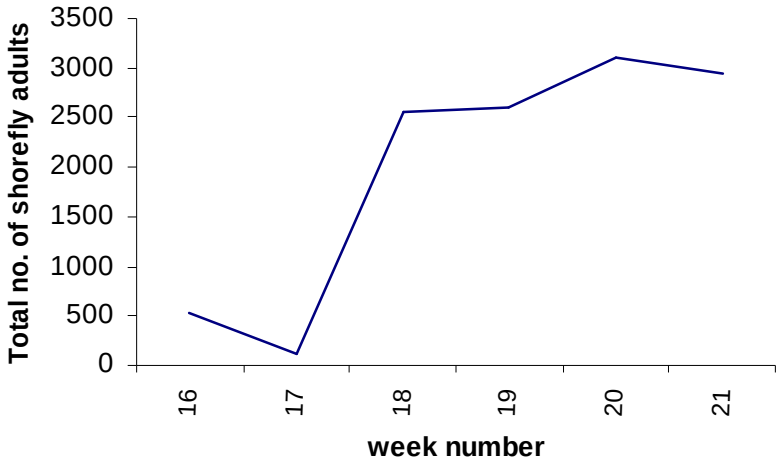


Figure 4. The total numbers of shore flies trapped (per 10 traps) in a commercial celery crop between weeks 16 and 21.

The results showed that the different rates of *Atheta* (Table 2) gave similar reductions in numbers of shore flies, when compared with that given by the standard release rate (Figure 3), despite the introduced rates varying considerably. Such release rates would be commercially unacceptable in celery if using direct releases of *Atheta* bought from commercial suppliers, and would only therefore be practical if a rearing/release system were used. Further research is required to clarify whether this latter approach would be viable in celery.

Objective 1: conclusions

- In potted parsley, *Atheta* adults added at the rates of five or 10 per pot (equivalent to 500 or 1000 per m^2 before pot spacing) to 1-day old plants naturally infested with sciarid fly eggs, significantly reduced mean

numbers of sciarid flies per pot 22 days later, when compared with untreated control pots. Mean numbers of sciarid flies per control pot after 22 days were 11.4, whereas in the pots treated with five or 10 *Atheta* per pot, they were 3 and 1.7 respectively (75% and 85% reductions respectively). *Atheta* rates of one or two adults per pot (100 and 200 per m^2 before pot spacing) did not significantly reduce numbers of sciarid flies at the 'starting' density of the pest in this experiment.

- In soil samples naturally infested with shore flies, taken from the perimeter of glasshouses growing celery, *Atheta* adults added at the rates of 20, 40, 200 and 300 per m^2 (actual rates of 25, 45, 205 and 305 per m^2 respectively) significantly reduced numbers of shore flies 14 days later, when compared with control samples of soil that the grower had treated with *Atheta* at five per m^2. The actual *Atheta* rate of 25 per m^2 was equally as effective as the higher rates tested. When shore fly populations increased in the celery glasshouses in week 19, mean numbers of shore flies emerging per tray of soil after 14 days in control trays were 85 (1700 per m^2), whereas in the trays treated with 25 *Atheta* per m^2, they were 10.5 per tray (239 per m^2), representing an 88% reduction.

- Such release rates would be commercially unacceptable in pot herbs and celery if using direct releases of *Atheta* bought from commercial suppliers. Further research is needed to determine whether the *Atheta* rearing/release system could enable sufficient predators to be released into these crops for improved, low cost control of sciarid and shore flies.

Objective 2: Refining the detail of the *Atheta* rearing-release units

Amount of food needed per Atheta adult and larva per week

Knowledge of how much food (turkey grower crumbs) are needed per *Atheta* adult and larva per week would ensure that sufficient food is added to cultures of a known beetle density, and may help to manipulate feeding regimes to encourage *Atheta* to leave the rearing-release units.

Materials & methods

Two sets of laboratory bioassays were carried out. In the first set, one *Atheta* adult was added to each of 20 replicate tightly-fitting Petri dishes, lined with damp filter paper to which 1g (fresh weight) of turkey grower crumbs had been added, and containing a specimen tube lid with a piece of damp cotton wool. The dishes with *Atheta* were left for one week in an incubator at 21°C, 16-hr photoperiod. After one week, the remaining turkey crumbs in each dish were weighed and records were made of whether the *Atheta* in each dish was still alive or dead.

In the second set of bioassays, the same method was used as in the first set, but ten *Atheta* adults were added to each of 20 replicate dishes, so that greater, more measurable amounts of food would be eaten after one week. In addition, an additional 20 1g-samples of turkey grower crumbs were air dried in an oven to determine dry weights when added to the dishes, and the amount of turkey crumbs remaining in the dishes after one week was weighed before and after drying, to determine fresh and dry weights.

Results and Discussion

In the first set of bioassays, only 11 of the 20 *Atheta* adults were still alive after one week. The deaths could have been partly due to natural mortalities as the *Atheta* were of unknown ages when taken from the culture and added to the dishes, and *Atheta* adults are known to live for only up to 21 days at 25°C (Carney *et al*, 2002). The mean weights of the turkey crumbs were slightly higher than the 'starting' weight (1g) after one week. This is likely to have been partly due to the turkey crumbs absorbing moisture from the damp filter paper, thus gaining weight, and to very small quantities of food being eaten by individual beetles, some of which did not survive the 7-day bioassay.

In the second set of bioassays, only 25 of the 200 *Atheta,* in five of the 20 dishes were still alive after one week, thus the results with remaining weights of turkey crumbs were inconclusive. It is possible that the high mortalities of *Atheta* were due to them finding the turkey grower crumbs unpalatable, when offered as dry food in the specimen tube lids. Although the turkey

crumbs absorbed some moisture from the damp filter paper and cotton wool, they remained relatively dry, when compared with their texture after incorporation in the damp substrate used in the *Atheta* rearing units.

A different bioassay or experiment would have to be designed in order to determine the optimum amount of food needed by a known number of *Atheta* adults and larvae over a selected time period.

Factors stimulating release of Atheta from rearing units

Potential factors stimulating *Atheta* to leave the rearing/release units could include warm temperatures, high *Atheta* density and shortage of food. An experiment was set up at ADAS Boxworth to test the effect of food availability and *Atheta* density on release of the beetles from rearing-release units.

Materials & methods

Selection of Atheta rearing boxes

On 21 August, eight 7-week old *Atheta* rearing boxes were selected from those that had been set up with 60 *Atheta* adults in each box between 4 and 6 July. The *Atheta* had been reared on turkey crumbs at 25°C. Numbers of *Atheta* per box were estimated by taking six replicate 30 mL sub-samples of the substrate from each box, turning the samples onto a large white plastic tray and counting the numbers of *Atheta* adults and larvae. All the counted *Atheta* were returned to each appropriate box. Four replicate pairs of boxes, each pair with similar numbers of *Atheta* were then selected, one box of each pair was fed with 5 g of turkey crumbs and the other box in the pair was not fed. All the boxes had previously been fed one week earlier in the *Atheta* rearing laboratory, thus the 'unfed' boxes had not been fed for a week. The 'starting' numbers of *Atheta* adults and larvae in each of the paired boxes are given in Table 3.

Table 3. Starting numbers of *Atheta* in each of the pairs of fed/unfed boxes used in the *Atheta* release experiment

Pair number	Box	Fed/unfed	Starting no. of *Atheta* adults per box	Starting no. of *Atheta* adults plus larvae per box

1	A	Fed	1525	2000
	B	Unfed	1542	1967
2	C	Fed	1108	1192
	D	Unfed	1292	1608
3	E	Fed	1067	1367
	F	Unfed	1150	1592
4	G	Fed	817	1450
	H	Unfed	975	1242

Experiment set-up

On 22 August, each box was placed in the middle of an insect-proof mesh cage (0.5 x 0.5 x 0.5 m). The cages were placed on damp capillary matting on a bench in a research glasshouse, and the substrate in the boxes was kept damp using the self-watering system developed in year 1 of the project. This entailed drilling four holes (7mm diameter) in the bottom of each box before adding the substrate and *Atheta*, and plugging the holes with cotton wool, which acted as a wick. Blue sticky traps were placed on the floor of the cage around the edges of each box, to trap any *Atheta* adults and larvae crawling or jumping from the boxes. One yellow sticky trap was hung from the roof of each cage, to trap any flying *Atheta* adults. The lid of each box was covered in tin foil to reflect direct sunlight. The two ventilation holes (2.5 cm diameter) in the box lids were opened by removing the insect-screening mesh, thus allowing the beetles to leave the boxes. The 'fed' boxes were fed with 5 g of turkey crumbs every week, and the 'unfed' boxes were left unfed throughout the experiment period.

Assessments

1. The sticky traps in each cage were checked every working day for three weeks after the experiment was set up. Numbers of *Atheta* adults and larvae were recorded on each assessment date.
2. Numbers of *Atheta* adults and larvae remaining in the release boxes at the end of the experiment were estimated, using the same method as used to estimate 'starting' numbers of beetles per box.
3. Glasshouse temperatures were recorded using a Tiny Talk ® datalogger placed in an extra *Atheta* box placed adjacent to the cages.

Statistical analysis

The percentage of *Atheta* leaving the fed/unfed boxes after one day and one week respectively were compared using analysis of variance (ANOVA). Regression analysis was used to compare the percentage of *Atheta* leaving fed and unfed boxes of different densities after one day and one week respectively.

Results and Discussion

Numbers of Atheta leaving fed and unfed boxes

Mean numbers of *Atheta* adults plus larvae leaving fed and unfed boxes on day 1 and after weeks 1, 2 and 3 are shown in Figure 5. Most of the *Atheta* leaving the boxes were adults, but low numbers of larvae were also released. Much higher numbers of *Atheta* left the unfed boxes than the fed boxed on day 1 and during week 1 (Figure 5). However, due to the different 'starting' numbers of *Atheta* in individual boxes (Table 3), statistical analysis was done to compare the percentage of beetles leaving each box, rather than the actual numbers. The highest numbers of *Atheta* left the unfed boxes during the first week of the experiment, whereas the numbers leaving the fed boxes was more similar in each of the three weeks (Figure 5). The total numbers of *Atheta* leaving fed and unfed boxes over the 3-week period was 315 and 919 respectively (Figure 5).

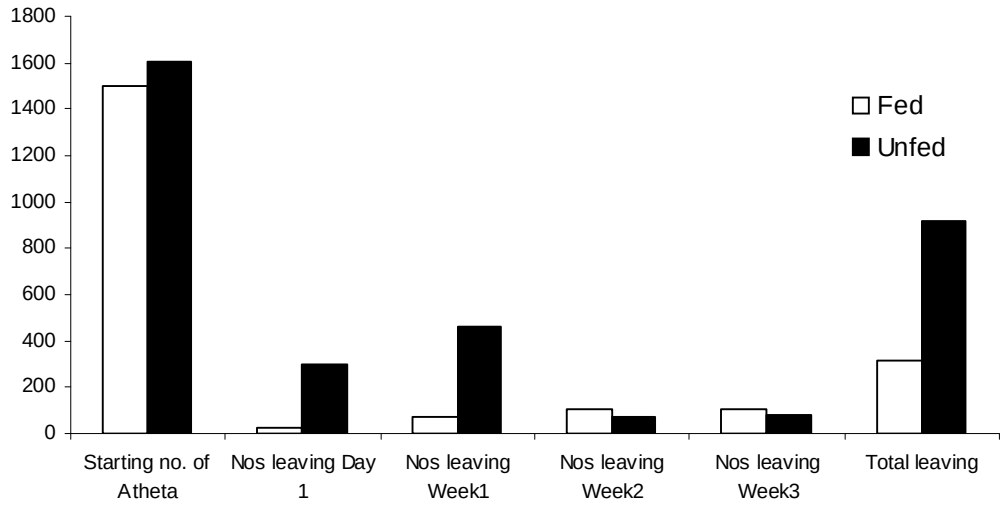


Figure 5. Mean starting numbers of *Atheta* adults plus larvae per release box, mean numbers leaving each box after day 1 and weeks 1-3, and total numbers leaving over the 3-week period.

Percentage of Atheta 'starting' numbers leaving fed and unfed boxes

When comparing the percentage of the 'starting' numbers of *Atheta* leaving fed and unfed boxes, it was only valid to do this during the first week of the experiment, as after that period, numbers of *Atheta* would be constantly changing, due to *Atheta* leaving the boxes and to the remaining *Atheta* breeding inside the boxes. On day 1, significantly greater percentages of *Atheta* had left the unfed boxes (17.9%) than the fed boxes (1.6%), $P<0.01$ (Figure 6). Similarly, during week 1 (including day 1), significantly greater percentages of *Atheta* had left the unfed boxes (46.7%) than the fed boxes (6.3%), $P<0.001$.

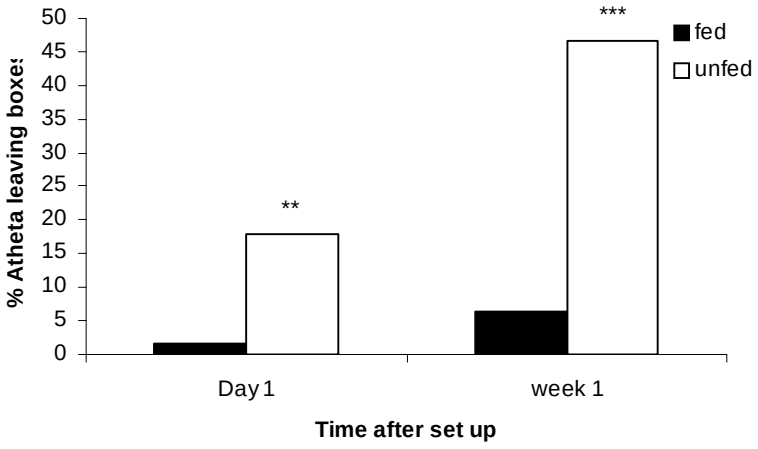


Figure 6. % *Atheta* leaving fed and unfed boxes on day 1 and during week 1.

Percentage of Atheta leaving boxes with different 'starting' numbers

Regression analysis was used to compare the percentage of the 'starting' numbers of *Atheta* emerging from fed and unfed boxes of different densities after one day and one week respectively. On day 1, 83% of the variance was accounted for. For the fed boxes, the fitted % emergence is 0.48% for a box with 1,000 *Atheta* and 2.88% for a box with 2000 *Atheta*. For the unfed boxes, the fitted % emergence is 7.98% for a box with 1,000 *Atheta* and 24.46% for a box with 2,000 *Atheta*. This shows that a significantly higher percentage of *Atheta* emerged from boxes with higher starting numbers than

from boxes with lower densities of beetles, and that the difference in percentage emergence is greater for unfed boxes than for fed boxes. Thus, if a quick-release of high numbers of *Atheta* are needed, boxes with high densities of *Atheta* should be used, and the boxes should not be fed for a week before the *Atheta* are released.

The regression analysis on the percentage of *Atheta* emerging from boxes after one week indicated a poorer correlation between % emergence and 'starting' density of beetles, with 76.5% of the variance accounted for. For the fed boxes, the fitted % emergence is 2.5 for a box with 1000 *Atheta* and 7.1 for a box with 2000 *Atheta*. For the unfed boxes, the data indicates that beetle density had no effect on % emergence. This is probably due to large numbers of beetles already having left the boxes by the end of week 1.

Atheta remaining in boxes at end of experiment

At the end of the 3-week experiment, the mean number of *Atheta* adult plus larvae in unfed boxes was only 44, whereas the mean number in fed boxes was 481. The unfed boxes thus only had a 3-week lifespan. The lifespan of the fed boxes will be determined by leaving them in the cages with the sticky traps for several more weeks, and continuing to monitor beetle emergence.

Glasshouse temperatures

Glasshouse temperatures during the 3-week experiment (22 August to 12 September) are shown in Figure 6. Mean temperatures averaged c. 19-23°C, with maximum and minimum temperatures reaching 33°C and 12°C respectively. These temperatures were within the known temperature range of *Atheta* (12-35°C, Syngenta Bioline information; 15-32°C, Miller & Williams, 1983).

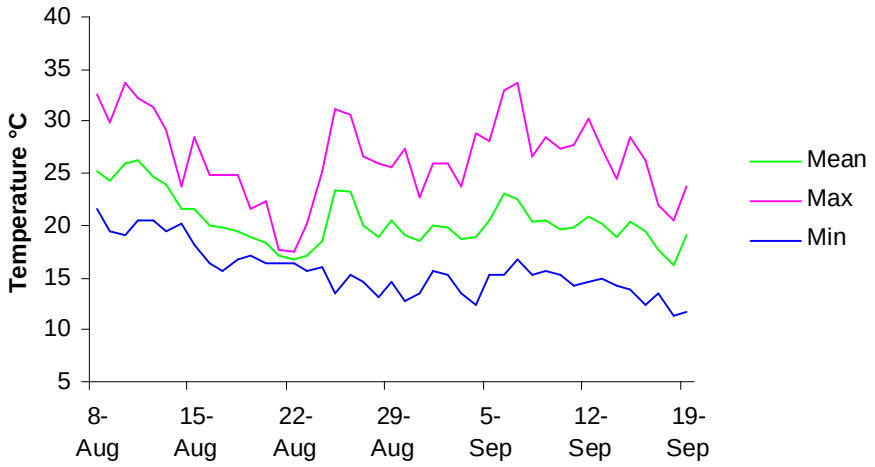


Figure 6. Mean, maximum and minimum temperatures during the experiment comparing release of *Atheta* from fed and unfed rearing-release boxes

Factors stimulating Atheta flight

Flight is likely to be the main method of adult dispersal in glasshouses. Increased temperature is likely to be the major factor stimulating flight, but light may also be involved. An experiment to determine the effect of temperature and light on *Atheta* flight has been done this year in PSD-funded project PS 2120 and the results will be referred to in the final report for PC 239.

Atheta development rate at low temperatures

So far in PC 239, the development rate of *Atheta* has only been determined at a constant 25°C. Knowledge of the development rates at lower temperatures is relevant for early bedding plant and celery crops. Experiments to determine the development rates of *Atheta* at both lower constant and fluctuating glasshouse temperatures are being done this year in PSD-funded project PS 2120 and the results will be referred to in the final report for PC 239.

Efficacy of self-watering rearing units on woven ground-cover matting

The efficacy of the self-watering rearing *Atheta* units has only been tested so far in the project on capillary matting and in ebb and flood gutters. The methods designed for both these irrigation systems have proved successful, and has minimized the *Atheta* rearing-release unit maintenance time for growers. Bedding plants are often grown in trays on the glasshouse floor,

stood on woven ground-cover matting, and watered with overhead irrigation. Sample *Atheta* rearing-release boxes designed for use on capillary matting were given to a grower to try on woven ground-cover matting, to test whether the substrate in the boxes remained damp enough without additional watering. It is possible that the boxes may need to be stood on a section of capillary matting stood on the woven ground-cover matting. No results are available as the grower did not have time to test the system. The system will be tested at ADAS Boxworth and the results will be given in the final project report.

Lifespan of Atheta rearing-release units in the glasshouse

As reported above, the 3-week experiment to determine *Atheta* release rates from fed and unfed boxes is being continued to determine the lifespan of the fed boxes under ideal conditions. Lifespan will be partly dependent on glasshouse environment, potential contaminants and grower maintenance regime. As this experiment is being done in insect-proof mesh cages, there will be minimal contaminants e.g. predatory mites that may adversely affect *Atheta*, and the rearing units are being maintained under optimum conditions for *Atheta*. Thus the results of this experiment should be validated in any further trials on commercial nurseries, should the current project be extended.

Objective 2: conclusions

- Laboratory bioassays to determine the amount of turkey grower crumbs needed by individual *Atheta* were inconclusive due to high *Atheta* mortalities during the 1-week bioassays. The mortalities may have been partly due to the *Atheta* finding the turkey crumbs unpalatable when offered dry in a dish, rather than being incorporated in damp substrate, as used in the rearing boxes.
- In a 3-week glasshouse experiment comparing numbers of *Atheta* leaving either fed or unfed rearing-release boxes, a significantly higher percentage of beetles left the unfed boxes (47%) than the fed boxes (6%) during the first week. On day 1, 18% of the *Atheta* left the unfed boxes and 2% left the fed boxes. On day 1, a significantly higher percentage of *Atheta* emerged from boxes with higher 'starting'

numbers (e.g. 2,000 per box) than from boxes with lower densities of beetles (e.g. 1000 per box). Thus, if a quick-release of high numbers of *Atheta* are needed, boxes with high densities of *Atheta* should be used, and the boxes should not be fed for a week before the *Atheta* are released.

- An experiment to determine the effect of temperature and light on *Atheta* flight (which is likely to be the main method of dispersal in a large glasshouse) has been done this year in PSD-funded project PS 2120 and the results will be referred to in the final report for PC 239.
- Experiments to determine the development rates of *Atheta* at both low constant and fluctuating glasshouse temperatures are being done this year in PSD-funded project PS 2120 and the results will be referred to in the final report for PC 239. The results will be relevant for early season bedding plant and celery crops.
- The efficacy of the self-watering systems for the *Atheta* rearing units (successful when used on capillary matting and in ebb and flood gutters) will be tested on woven ground-cover matting during the remainder of the project. The results will be relevant to bedding plants that are grown in trays stood on the glasshouse floor.
- At the end of the 3-week experiment, the mean number of *Atheta* adult plus larvae in unfed boxes was only 44, whereas the mean number in fed boxes was 481. The unfed boxes thus only had a 3-week lifespan. The lifespan of the fed boxes will be determined by leaving them in the cages with the sticky traps for several more weeks, and continuing to monitor beetle emergence. However, the lifespan of the rearing-release units in this experiment, under ideal conditions, should be validated further on commercial nurseries, as lifespan will be partly dependent on glasshouse environment, potential contaminants and grower maintenance regime.

Objective 3: Identify key aspects of *Atheta* behaviour in selected crops and growing substrates

Atheta entry of compost in crop pots

In the herb trial in year 1 of the project, much lower mean numbers of *Atheta* (0.2 per pot) were found in 4-week old pots of parsley infested with sciarid fly

larvae , than in bait pots (pots containing damp compost with a small amount of turkey grower crumbs incorporated) used for monitoring dispersal of the predators (9 per pot). This result could have been due to the more compact compost and dense root system making the parsley pots a less favourable environment than the compost in the bait pots, or to the *Atheta* favouring or breeding more successfully in the bait pots. The comparative entry and breeding of *Atheta* in crop pots (e.g. parsley, mint and poinsettia) and bait pot would be best addressed in any future research on commercial nurseries, when *Atheta* numbers could be determined using Tullgren funnel extraction. However, some information was gained during year 2 of the project, in the work done in Objectives 1 and 3 as follows:

Objective 1

When different rates of *Atheta* adults were added to 1-day old parsley plants infested with sciarid fly eggs, after 22 days, *Atheta* adults had successfully entered the compost as they were present in 75-100% of the pots and they had produced larvae in up to 50% of the pots (Table 1). The absence of *Atheta* larvae in some pots could have been partly due to only male beetles being added to some pots (it is not possible to determine the sex of live adults due to them being so active) and partly due to some larvae being eaten by the adults.

Objective 3

In the experiment in Objective 4, to determine the interaction of *Atheta* with Aphidoletes, 10 *Atheta* adults and 10 larvae were added to each mature parsley plants on two occasions and after four weeks, means of 13.3 adults and 29.8 larvae per pot were recovered (Table 6). This result indicates that *Atheta* adults and larvae successfully entered the compost of mature parsley plants with dense compost and root systems. However, further research is required to compare the numbers of *Atheta* entering and breeding in crop pots and in bait pots on a commercial nursery.

Trapping method for Atheta in soil-grown crops

In order to monitor the presence and dispersal of *Atheta* within a crop it is important to be able to trap the predator. In year 1 of the project, *Atheta*

were successfully trapped in 'bait pots'. These were pots filled with damp compost baited with turkey grower crumbs, stood on capillary matting on the glasshouse floor or on benches with potted ornamentals, or placed in ebb and flood gutters in pot herb crops. In the protected celery crop grown in the soil, this method was unsuccessful at trapping any *Atheta*. It was not clear from the year 1 trial whether the failure to trap *Atheta* was due to insufficient numbers of predators leaving the rearing-release boxes, or whether this trapping method was ineffective in a soil environment. In year 2, the following experiment was designed to identify a successful method to trap and hence monitor *Atheta* in a soil-grown crop.

Material and methods

Within an empty glasshouse (120m^2) at STC, 16 baited plant pots (3 inch plant pots filled with compost and a small volume of turkey grower crumbs) were distributed on the soil surface as in Figure 7. Eight replicate pots were stood on the soil surface and eight were sunk so that the tops of the pots were level with the soil surface. Two *Atheta* rearing-release units with large visible numbers of larvae and adults in the substrate were then placed at one end of the glasshouse, 11 metres away from the furthest bait pots. The numbers of *Atheta* adults and larvae in each pot were recorded seven days after being placed in or on the soil. The experiment was done three times, repeated at one week intervals from weeks 26-29. The soil and pots were kept damp with overhead watering.

Statistical analysis

Analysis was done on the square root transformed data using analysis of variance (ANOVA) and the means were compared using LSD.

Results and discussion

The results showed that pots below the soil surface were significantly more successful at trapping *Atheta* adults and larvae (Table 5). *Atheta* were found in 31% of traps below the soil surface, with a total of 67 adults and 41 larvae, whereas *Atheta* were found in only 10% of baited pots stood on the soil surface, with a total of 10 adults and six larvae. Although some larvae may have left the *Atheta* rearing-release units, it is likely that some were also

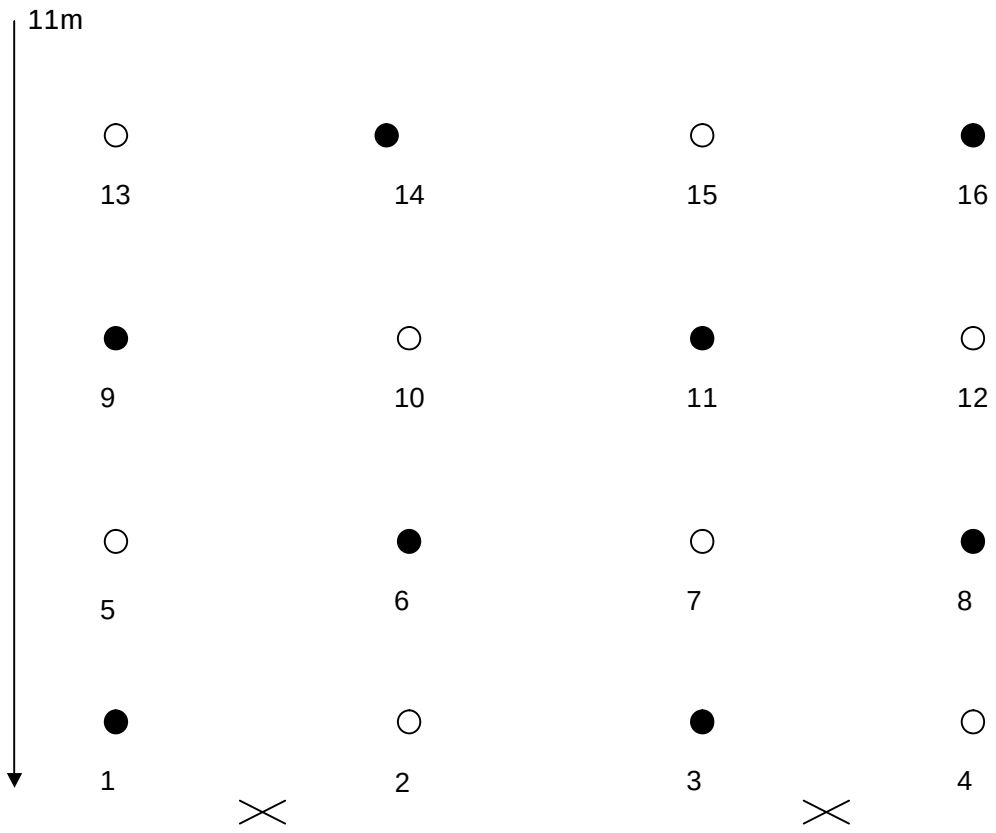
offspring of the adults entering the pots. The results show that in a soil-grown crop, a better method for monitoring the predator is to place the bait pots so that the rims of the pots are just below the soil surface. Such pots are akin to pitfall traps, a standard method used in the field for trapping ground-dwelling beetles.

Table 5. The mean number and (sqrt transformed mean) of *Atheta* adults and larvae recorded in bait pots stood on or sunk below the soil surface in a glasshouse

Bait pot	Adults	Larvae
Below surface	2.83 (1.18)	1.73 (0.90)
On surface	0.43 (0.28)	1.26 (0.10)
LSD (47df)	(0.45)	(0.53)

In year 1 of the project, no *Atheta* were trapped in bait pots placed on the soil surface in a commercial celery glasshouse, whereas in this experiment, small numbers were retrieved. A major factor contributing to the greater success of the bait pots placed on the soil surface in the year 2 experiment was that glasshouse temperatures (mean 19°C) were within the known activity range of *Atheta* (15-32°C, Miller & Williams, 1983), whereas in the year 1 experiment, temperatures remained below 15°C for much of the experimental period. Other factors that could have contributed to the lack of success of the bait pots stood on the soil surface in the commercial celery crop in year 1 was that the soil contained large numbers of invertebrates, including shore fly larvae, providing the beetles with alternatives to the turkey grower crumbs used as bait. In addition, in the commercial celery glasshouse, there was also a larger dilution effect in terms of soil surface area to bait pot ratio, than in the small glasshouse at STC used in the experiment in year 2. However, in the herb and poinsettia trials in year 1, *Atheta* were successfully trapped in the bait pots despite there being high numbers of sciarid fly prey in the crop pots, and despite a similar 'dilution effect' of the large surface area of compost in the crop pots compared with the bait pots.

In the year 2 experiment, 74% of the *Atheta* were retrieved from pots in the front half of the glasshouse nearest the rearing units (pots 1-8), (see Figure 1). However, the remaining 26% of the beetles were found in the furthest pots, and as the pots were only set up for one week before being sampled, it could be expected that with more time there would be an increased dispersal of the predator, as shown in the trial in a poinsettia glasshouse in year 1. Further research is required to test the success of bait pots sunk in the soil in commercial celery crops, for monitoring *Atheta* presence and dispersal.



● = bait pot below soil level

○ = bait pot on soil surface

X = *Atheta* rearing-release box

Figure 7. The layout of bait pots used for trapping and monitoring *Atheta* in soil within a glasshouse (120m^2)

Objective 3: conclusions

- Results of work in Objective 1 demonstrated that *Atheta* adults can successfully enter and breed in the compost in young pots of parsley.
- Results of work in Objective 4 demonstrated that *Atheta* adults and larvae can successfully enter the compost of mature parsley plants with dense compost and root systems.
- Further research would be required to compare the numbers of *Atheta* entering and breeding in crop pots and bait pots on a commercial nursery.
- In soil-grown crops e.g. celery, bait pots sunk into the ground, with the rims level with the soil surface, are a better method for trapping and monitoring *Atheta* than those stood on the soil surface.

Objective 4: Investigate the interaction of Atheta with other ground-dwelling biological control agents

*Interaction of **Atheta** with **Aphidoletes aphidimyza***

Aphidoletes aphidimyza is a predatory midge used for aphid control, commonly used on certain ornamental crops and also on protected herbs such as parsley and mint, which are attacked by aphid species that are not susceptible to parasitism by commercially available parasitoids. *A. aphidimyza* larvae feed on aphids on the plants, then drop to the ground to pupate, thus the late larval stage and the pupae are potential prey for *Atheta*. An experiment was done at ADAS Boxworth to determine whether *Atheta* and *Aphidoletes* are compatible within an IPM programme.

Materials and methods

Pots of parsley infested with hawthorn-parsley aphid were collected from a commercial herb nursery.

Pilot bioassay

A pilot bioassay was set up to test whether *Atheta* will predate *Aphidoletes* larvae and hawthorn-parsley aphids. One *Atheta* adult was placed into each of four tightly fitting ventilated Petri dishes lined with damp filter paper. In two of the dishes, the *Atheta* was offered 10 hawthorn-parsley aphids as prey and in the other two dishes, the predator was offered six *Aphidoletes* larvae. The dishes were left for 24 hours at 21°C, after which time the numbers of aphids and *Aphidoletes* larvae were counted and recorded.

Glasshouse experiment

Eight 'clean' parsley plants (bought from a supermarket) were infested with approximately equal numbers of hawthorn-parsley aphids that had been collected from the commercial herb nursery. Each of the eight replicate plants was placed in an individual insect-proof mesh cage, 50 x 50 x 50 cm. The cages were placed on damp capillary matting on a bench in a research glasshouse at ADAS Boxworth. The plants were sub-irrigated through the mesh base of the cage. Glasshouse temperatures were monitored with a Tiny Talk ® datalogger placed in a white plastic pot in one of the cages. On 10 July, when the aphids had developed large colonies on the plants, a commercial 'blister' pack of *Aphidoletes* was added to each of the eight cages. Each pack contained approximately 250 *Aphidoletes*, most of which were pupae, with a few adults having emerged. The *Aphidoletes* were allowed to lay eggs on the parsley plants, and larvae were allowed to develop in the aphid colonies. On 11 and 18 July, 10 *Atheta* adults and 10 larvae were added to the compost in each pot in four of the replicate cages, and the other four were left untreated with *Atheta* and thus acted as controls. The plants were monitored for the presence of the first *Aphidoletes* larvae. Published information on the development of *Aphidoletes* at the temperatures occurring in the glasshouse during the experiment was used to estimate when these *Aphidoletes* larvae would drop to the ground to pupate, and when the next generation of adults would emerge from the pupae. On 25 July (15 days after adding the Aphidoletes pupae and adults), the foliage from each pot was cut off and the pot with compost (8 cm diameter) was placed inside a larger (one-litre) white plastic 'emergence' pot (12 cm diameter). Two small yellow sticky traps (each 10x5 cm) were attached to the inside of each 'emergence' pot, then each emergence pot was covered

with a 'lid' of insect-proof mesh, secured with a rubber band. The eight emergence pots were left in a controlled temperature laboratory (20°C, 16-hr photoperiod) for two weeks (until 8 August), to allow any *Aphidoletes* adults to emerge from the pots and get caught on the sticky traps.

Assessments

- On 8 August, numbers of *Aphidoletes, Atheta* and sciarid fly adults on the sticky traps were counted and recorded.
- On 8 August, the surface of the compost in each pot and the inside of each emergence pot was checked for any remaining *Aphidoletes*, *Atheta* and sciarid fly adults.
- On 8 August, the compost in each pot was placed in a Tullgren funnel for 48 hours to extract any remaining *Aphidoletes* larvae, *Atheta* adults or larvae, sciarid fly larvae, and any other invertebrates that could have been potential prey for *Atheta*.
- Total numbers of *Aphidoletes* emerging from each pots were calculated.

Statistical analysis

Mean numbers of *Aphidoletes* emerging in pots treated with *Atheta* and in control pots untreated with *Atheta* were compared using analysis of variance (ANOVA).

Results and discussion

Pilot bioassay

After 24 hours, there were no *Aphidoletes* larvae remaining in one dish and only two of the six larvae remained in the other. This result demonstrated that *Atheta* will eat *Aphidoletes* larvae if they are offered as the only available prey in an artificial environment i.e. in a Petri dish on damp filter paper. After 24 hours, all ten hawthorn-parsley aphids were still alive in each dish. This

result indicated that although these aphids commonly occur at the base of parsley stems, and frequently fall from heavily infested plants to the compost, they are not eaten by *Atheta,* even when offered as the only available prey.

Glasshouse experiment

Atheta adults and larvae (means of 13.3 and 29.8 respectively) were found in all the pots treated with *Atheta* (Table 6). One of the four control pots had become accidentally contaminated with *Atheta*, and contained one adult and 17 larvae at the end of the experiment. There was no significant difference between mean numbers of *Aphidoletes* emerging in pots treated with *Atheta* (27 per pot) and those emerging in control pots untreated with *Atheta* (37 per pot), see Table 6. Sciarid flies were present in all pots, but there was no significant difference between mean numbers in pots treated with *Atheta* (10 per pot) and in control pots untreated with *Atheta* (31). Thrips and mites were also present in both treated (means of 3 and 22 per pot respectively) and untreated pots (means of 1 and 100 per pot respectively). The mites were not identified to species but under a low power microscope, looked like a mixture of *Hypoaspis* spp., *Amblyseius* spp. and others. As numbers of thrips and mites were so variable between replicate pots, the analysis of variance was done on $\log_{10}$ data as well as actual numbers per pot. There were no significant differences between mean or $\log_{10}$ numbers of thrips and mites in treated and untreated pots.

Table 6. Mean numbers of *Aphidoletes*, sciarid flies, thrips and mites and *Atheta* per parsley pot ($\log_{10}$ means of thrips and mites per pot shown in brackets).* One *Atheta* adult and 17 larvae were found in one control pot only.

Invertebrate species	Mean nos per pot treated with *Atheta*	Mean nos per control pot untreated with *Atheta*
Aphidoletes	26.8	36.8
sciarid flies	10.0	30.5
thrips	3.0 (0.38)	1.0 (0.23)
mites	22 (1.33)	100 (1.86)

Atheta adults	13.3	*
Atheta larvae	29.8	*

The results indicate that although the pilot bioassay showed that *Atheta* will eat *Aphidoletes* larvae when offered as the only prey in a Petri dish, they do not significantly reduce numbers of *Aphidoletes* larvae and pupae in the compost of aphid-infested parsley plants, when alternative prey species e.g. sciarid flies and mites are present in or on the compost. *Atheta* might have a negative impact on *Aphidoletes* if alternative prey are not available or if they are in shorter supply than in this experiment.

Glasshouse environment and effect on Aphidoletes development

As the experiment was done during June and July, natural daylength exceeded 15 hours, the photoperiod needed for *Aphidoletes* larvae to complete their development into pupae. Mean glasshouse temperatures averaged c. 21°C during the experimental period, minimum temperatures were 17-18°C and maximum temperatures reached 33°C (Figure 8). At 21°C, *Aphidoletes* eggs hatch into larvae in two days, the larvae feed on aphids for seven days before dropping from the plants to pupate, and the pupal stage lasts 10 days before the new adults emerge (Malais & Ravensberg, 1992). Thus the *Aphidoletes* larvae and pupae in this experiment would have been available in the compost for predation by *Atheta* from day 9 (when the first larvae would have dropped from the plants) to day 15 (when the foliage was cut off the plants and the pots and sticky traps were placed in the emergence pots). First new adult *Aphidoletes* would have started emerging on day 19, four days after cutting placing the parsley pots in the emergence pots.

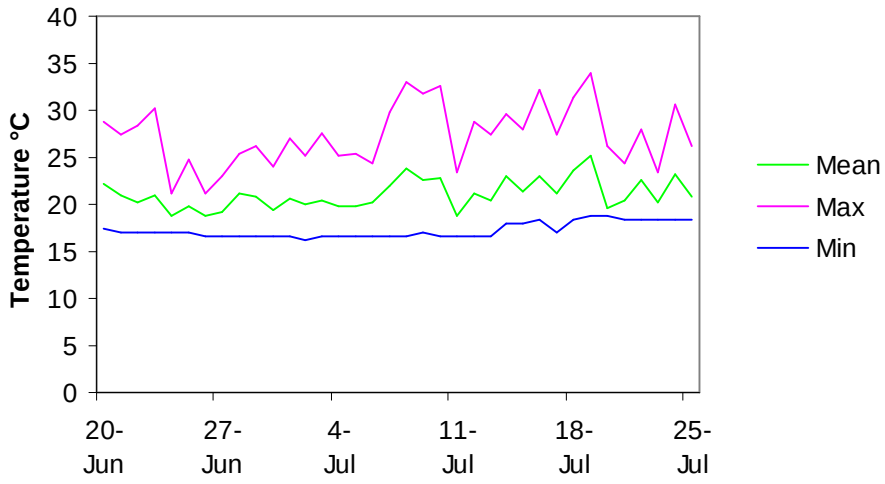


Figure 8. Mean, maximum and minimum temperatures during the glasshouse experiment to determine the interaction of *Atheta* with *Aphidoletes* .

Interaction of Atheta with Hypoaspis sp.

In the year 1 report for this project, the results of Canadian laboratory studies were summarized, which demonstrated that both *Hypoaspis miles* and *H. aculeifer* can eat *Atheta* and vice-versa. The results indicated that the use of *Atheta* in combination with *Hypoaspis* spp. is likely to be incompatible. UK grower experience has indicated that contamination of *Atheta* rearing-release units with *Hypoaspis* might be detrimental to *Atheta* production (Russ Woodcock, personal communication).

During the remainder of the project, a small-scale experiment will be done at ADAS Boxworth, to test the potential negative effects of *Hypoaspis* sp. on *Atheta*, both as a contaminant in rearing-release units and as a competitor in pots of herbs infested with sciarid flies. The results will be included in the final project report.

Objective 4: conclusions

- *Atheta* adults ate *Aphidoletes* larvae when offered as the only prey in a Petri dish. However, *Atheta* did not significantly reduce numbers of *Aphidoletes* larvae and pupae in the compost of aphid-infested parsley plants, when alternative prey species e.g. sciarid flies and mites were present in or on the compost.

Objective 5: Review current knowledge on compatibility of pesticides with *Atheta* in IPM programmes

Results of the poinsettia trial in year 1 of the project highlighted the need for information on pesticide compatibility with *Atheta*, particularly those commonly used for whitefly control, including imidacloprid (Intercept), thiacloprid (Calypso) and spiromesifen (Oberon). Three new products are now available for whitefly control on ornamentals: two compost-incorporated products, imidacloprid (Imidasect 5GR) and thiacloprid (Exemptor) and one product for application as a foliar spray, acetamiprid (Gazelle). Information on the compatibility of these new products with *Atheta* is now also needed. Current knowledge on the effects of these pesticides on *Atheta* is summarised below:

Imidacloprid

Two imidacloprid products are currently approved for use on protected ornamentals, for control of whitefly and other pests. Imidacloprid is a soil-acting systemic neonicotinoid insecticide with activity against a wide range of insects. Intercept 70WG is applied as a compost drench, and Imidasect 5GR is incorporated in the compost before potting. Information on side effects on *Atheta*:

- Canadian research tested the effects of both a direct spray of imidacloprid (laboratory test) and a drench to pots of compost (growth room experiment). Both application methods proved to be harmful (causing over 50% mortality) to both *Atheta* adults and larvae (Jandricic *et al*, 2005). The conclusion of the researchers was that imidacloprid should be avoided if *Atheta* is used in an IPM programme.
- The Biobest and Koppert websites gives information on the side effects of pesticides on biological control agents, that is largely based on the results of the testing scheme of the International Organisation for Biological Control (IOBC). No specific information is given on *Atheta*, but the Biobest website reports that imidacloprid applied a foliar spray is reported to be 'toxic' (causing over 75% mortality,

according to IOBC classification) to coleoptera (beetles). Imidacloprid applied as a drench is reported as 'non-toxic' (causing less than 25% mortality) to beetle adults and 'moderately toxic' (causing 50-75% mortality) to beetle larvae. No information on persistence is given.

- Imidacloprid is recommended for persistent vine weevil control on protected ornamentals, giving up to six months persistence (Intercept 70WG) and up to 12 months persistence (Imidasect 5GR). Although vine weevils are in a different group of coleoptera to *Atheta*, it is possible that the persistent effects against *Atheta* will be similar to those against vine weevils.

Thiacloprid

Two thiacloprid products are currently available for use on protected ornamentals, for control of whitefly and other pests. Calypso has a Specific Off-Label Approval (SOLA) for use on ornamentals and is applied as a foliar spray. Exemptor is approved for use on protected ornamentals for incorporation in the compost before potting. Like imidacloprid, thiacloprid is a systemic neonicotinoid insecticide with activity against a wide range of insects. Information on side effects on *Atheta*:

- There is no specific information yet available on the effect of thiacloprid on *Atheta*. However, American research is currently being done on the side effects of pesticides on *Atheta*, including the effects of drenches of neonicotinoid insecticides (Raymond Cloyd, personal communication).
- Thiacloprid is known to have activity against other coleoptera, e.g. Calypso is effective against apple blossom weevil, and Exemptor is effective against vine weevil in the compost and also leaf beetles.
- The Biobest website reports that foliar sprays of thiacloprid are moderately toxic (causing 50-75% mortality) to adult coleoptera and 'toxic' (causing over 75% mortality) to larvae. Drenches of thiacloprid are reported as 'slightly toxic' (causing 25-50% mortality) to adult coleoptera and 'toxic' to larvae. No information is given on persistence.

- Foliar sprays of Calypso may be less harmful to *Atheta* than compost-incorporation with Exemptor, due to the beetle's preference for living in the compost. However, when Calypso is applied as a high volume spray, some will reach the compost.
- Further information is required on the side effects of Calypso and Exemptor on *Atheta*, but it is likely that some adverse effects will occur.

Spiromesifen

Spiromesifen (Oberon) has a SOLA for use as a foliar spray on protected ornamentals, for the control of whiteflies and spider mites. Information on side effects on *Atheta*:

- Oberon has no known efficacy against beetles.
- The Biobest website reports that foliar sprays of spiromesifen are 'non toxic' to coleoptera larvae but no information is given for adults.
- It is likely that Oberon may be used in conjunction with *Atheta* in IPM programmes, although specific information on its compatibity is required.

Acetamiprid

Acetamiprid (Gazelle) is a neonicotinoid insecticide, approved for use as a foliar spray on protected ornamentals, for the control of whiteflies and aphids. Information on side effects on *Atheta*:

- There is no specific information yet available on the effect of acetamiprid on *Atheta*.
- The Biobest website reports that foliar sprays of acetamiprid are moderately toxic (causing 50-75% mortality) to coleopteran adults and larvae.
- Foliar sprays of Gazelle may be less harmful to *Atheta* than to foliar-dwelling beetles, due to the preference by *Atheta* for living in the compost. However, when Gazelle is applied as a high volume spray, some will reach the compost.
- Further information is required on the side effects of Gazelle on *Atheta*, but it is likely that some adverse effects will occur.

Objective 5: conclusions

- Imidacloprid (Intercept 70WG applied as a compost drench, and Imidasect 5GR incorporated in the compost before potting) is harmful to both *Atheta* adults and larvae and should not be used together with *Atheta* in an IPM programme.
- No specific information is available on the side effects of thiacloprid (Calypso applied as a foliar spray, and Exemptor incorporated in the compost) on *Atheta*. However, this insecticide is known to kill other beetle species and both products are likely to have adverse effects on *Atheta*. Information is required on the specific side effects of Calypso and Exemptor on *Atheta*.
- Spiromesifen (Oberon) has no known effects on beetle species. It is likely that Oberon may be used together with *Atheta* in IPM programmes, although specific information on its compatibility is needed.
- Acetamiprid (Gazelle) is known to kill beetle adults and larvae, but there is no specific information available on the side effects on *Atheta* and this information is needed.

Objective 6: Identify options for further work on commercial nurseries

Any further detailed experiments on commercial nurseries would require further funding, for the project to be extended beyond the current end date of 28 February 2008. In addition to the work reported above in Objectives 1-5, selected growers were contacted or visited to discuss their interest or experience in using *Atheta* rearing-release systems as follows:

Protected ornamentals

Regular contact was made with a grower using large-scale *Atheta* rearing-release units in bedding plant and poinsettia production. The grower reported that very low numbers of shore flies were recorded during 2007 on plug trays of both early season and autumn bedding plants (Russ Woodcock, personal communication). ADAS assessed the numbers of *Atheta* in the nursery open rearing units in June 2007, and found only low numbers of the predators. The rearing units had been left on the nursery for several months

and had become contaminated with large numbers of mites which could have been detrimental to *Atheta*. In addition, staff changes on the nursery had meant that there had been little time for maintenance of the rearing units. The rearing-release units were used in some poinsettia crops later in the season, and in September 2007, the grower reported that plenty of *Atheta* were visible in and under the pots. However, the grower was also using drenches of nematodes for control of sciarid flies and was still experiencing some problems with control. Use of pesticides e.g. Gazelle for control of whiteflies on the poinsettia crop may have had some adverse effects on *Atheta*. The grower was very interested in the project research results during year 2 and would be willing to host a further trial on the nursery during 2008 if requested, to test methods for improving the use of rearing-release systems on protected ornamentals.

Protected herbs

A grower of containerised herbs and ornamental garden plants was visited in August 2007 to discuss progress with rearing and releasing *Atheta* in the propagation house. The grower had attended one of the HDC/Defra-funded ADAS workshops on Integrated management of pests and diseases on protected herbs in September 2006. (The workshops were done as part of HDC project PC 210 and Defra project HH3118TPC). During the workshops, progress to date with PC 239 was presented, and the *Atheta* rearing-release system was demonstrated. As a result of this, the grower adopted the rearing system in the propagation house in autumn 2006, with the aim of improving biological control of sciarid flies. The grower is experienced in IPM and had been using *Hypoaspis miles* for biological control of scarid flies, but control had been inadequate. The grower has kept *Atheta* rearing boxes on capillary matting on the heated concrete floor (25°C) of the propagation house. Instead of using the boxes as rearing-release units, the grower allowed the *Atheta* to breed in the boxes, then used them for direct release, i.e. they were broadcast together with the substrate over the top of the plants, just as they would be if packs of *Atheta* had been bought from a commercial supplier. The *Atheta* were distributed over the plants every few weeks and two boxes of every batch of *Atheta* were used for starting off the next batch.

The grower considered that control of sciarid flies had been excellent, and planned to stop using *Hypoaspis* and to remove the long 'curtain' yellow sticky traps which had been previously used for mass trapping of sciarid fly adults (Martin Emmett, personal communication). Although shore flies were less of a problem than sciarid flies on the nursery, since adopting the *Atheta* system, any intermittent problems with shore flies had no longer occurred.

This system for rearing and releasing *Atheta* is likely to be more suitable for nurseries with small propagation or production houses, or on larger nurseries with enough staff time available to distribute the predators as well as to rear them. On many large nurseries with limited staff resources, a larger-scale rearing-release system is likely to be more practical and feasible.

2. A grower of AYR pot herbs was visited to discuss progress with PC 239 . The grower is currently using *Hypoaspis miles* for control of sciarid flies, but control has been inadequate (Rob Grundy, personal communication). The grower had stopped using drenches of nematodes due to application difficulties on the nursery. The grower is very keen to adopt the use of *Atheta* rearing-release units in a large glasshouse and would be keen to host a trial next year.

Hardy nursery stock

Two growers of protected HNS have set up *Atheta* rearing-release units in their propagation houses during 2007. The growers will be contacted later this year to discuss progress and their experience will be reported in the final project report.

Objective 6: conclusions

- As a result of progress in the project during year 1, several growers of pot herbs, ornamentals and HNS have adopted the *Atheta* rearing-release system and have adapted it to their own needs and circumstances.
- The system has given promising commercial results against shore flies in bedding plant propagation and against sciarid flies in the propagation of herbs and ornamentals.

- Further research progress during year 2 of the project has filled some key fundamental gaps in knowledge on *Atheta* biology and behaviour and about the potential of the rearing-release system. Knowledge gained on numbers of *Atheta* required to control known densities of sciarid and shore flies, methods for manipulating release of known numbers of the predators from rearing units, and improved methods for monitoring dispersal of *Atheta* in soil-grown crops will enable an improved system to be tested in commercial crops.
- Options for further work in commercial crops of ornamentals, herbs and celery will be discussed with HDC at a project review meeting in October 2007.

Technology transfer

At the request of HDC, most of the technology transfer for this project is being deferred until the final year of the project.

Consultancy to growers

- The *Atheta* rearing-release unit system was discussed and demonstrated to several growers of herbs and ornamentals, who have now set up their own rearing systems, in both propagation and production glasshouses.

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