

Project Title: Development of a banker plant system to improve the biological control of two-spotted spider mite in hardy nursery stock

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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

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

Dr John Buxton
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Signature Date

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Grower Summary

Headline

Lime trees show potential for use as banker plants in Hardy Nursery Stock. They are a good host for the Lime mite, which breeds up well and provides prey for a range of predators of two-spotted spider mite.

Background and expected deliverables

The biological control of two-spotted spider mite in protected nursery stock relies on regular introductions of the predatory mite *Phytoseiulus persimilis* during the summer months. This approach can work well, but can be expensive on a large scale and *P. persimilis* is less effective during periods of hot weather. Applications of acaricides are therefore often needed to correct the predator/prey balance and prevent crop damage. Biological control could be improved by rearing up non-pest mites on a suitable 'banker' plant as a food source for predators. These predators could then move on to crop plants to prey on two-spotted spider mites. This would provide a low cost, augmentative biological control technique that would reduce pest damage and be environmentally sustainable.

The expected deliverables from this project are:

- An evaluation of several types of host plant/mite combinations to determine their suitability for culturing under protection.
- An evaluation of the suitability of the prey mite species as food for a range of commercially available biological control agents.
- Determination of the degree of movement of beneficials from the banker plant onto nearby nursery stock plants.

Summary of the project and main conclusions

- A potential banker plant system using fruit tree red spider mite (*Panonychus ulmi*) was evaluated. Mite numbers increased initially on *Sorbus aria lutescens* (whitebeam) host plants, but then decreased during the summer months. Although low numbers of native *Amblyseius andersoni* predatory mites were found on this host, the poor establishment of prey mites meant that it was unsuitable for use in a banker plant system.
- Laboratory trials showed however that the predatory mites *A. andersoni*, *A. californicus* and *Phytoseiulus persimilis*, and the predatory midge larva *Feltiella acarisuga* could all prey successfully on fruit tree red spider mite.
- Lime trees (*Tilia cordata*) were infested with lime mite (*Eotetranychus tiliarium*) and after an initial period of low population growth, mite numbers increased rapidly and supported populations of predators such as *Amblyseius* spp., *P. persimilis* and *F. acarisuga*. Two-

year old lime trees were easy to cultivate and withstood high temperatures inside a polythene tunnel during summer 2006.

- A laboratory method was developed which allowed the predation rates of individual predator species to be evaluated in a standardised system, allowing statistical comparisons of the data to be made. The results from these tests were reinforced by direct observation of predator and prey in sealed Petri dishes.

The main conclusions from the first year of this project were:

- Fruit tree red spider mite is an unsuitable prey mite on whitebeam under protection, as mite numbers do not increase sufficiently for use in a banker plant system.
- Lime mite and lime trees have considerable potential for use in a banker plant system, as lime mite acts as a highly reproductive prey source for a range of beneficial insects and mites. Further work is required to develop systems for establishing banker plants (lime trees), lime mites and predator populations as early as possible in the season.
- Lime trees are inexpensive to buy bare root and are easy to cultivate under protection.
- The banker plant system, using lime trees and lime mites, shows promise and, because lime mite is specific to this host plant, it cannot damage other nursery stock plant species.

Financial benefits

- This work has the potential to reduce the overall cost of the biological control programme for two-spotted spider mite control, but this cannot be quantified as yet.
- There is still no data on the movement of predators from the banker plant onto adjoining crop plants, and this is needed before the practicality of the system (and therefore its financial benefits) can be quantified.

Action points for growers

- Growers would need to plan a production schedule in order to make a banker plant system work on a larger scale, so that clean host plants could be produced in sequence over the summer, as older banker plants became less effective.

Science Section

Introduction

In the UK, there has been a trend toward growing nursery stock under protection (polythene tunnels or glasshouses) for several years, and this trend, together with warmer summers and the presence of a wide range of susceptible plants, has allowed the two-spotted spider mite (*Tetranychus urticae*) to become a very damaging pest. Host plants such as *Weigelia*, *Choisya*, *Ceanothus*, *Buddleia* and *Crinodendron* can suffer serious damage, leading to loss of quality and reduced marketability.

Most growers follow an integrated pest management (IPM) programme, using introductions of the predatory mite *Phytoseiulus persimilis* on a regular basis, and spraying “hot spots” with acaricides when necessary. This approach can work well, but warm weather reduces the activity of the predator. Force (1967) found that *P. persimilis* was most effective at 20 °C; above this level, its effectiveness decreased rapidly. However, as it is a Type I predator (McMurty & Croft, 1997), it specialises in feeding on tetranychid mites, and so is unable to exist in times when prey is scarce. By contrast, predatory mites such as *Amblyseius andersoni* and *A. californicus* are Type II predators, able to feed on a range of mites and also pollen, fungal spores and other small organisms such as Collembola (springtails). This enables them to survive even when numbers of two-spotted spider mite are low.

A banker plant system is already in use by growers of ornamental foliage plants in Florida. This utilises the Banks grass mite (*Oligonychus pratensis*) as prey, reared on maize (*Zea mays*) host plants (Osborne, pers.comm.). In this system, regular introductions of the predatory mite *A. californicus* are made to the banker plants, which are placed at intervals amongst the crop. This results in increased density of predators as well and also an influx of wild predators. The overall effect is improved control of two-spotted spider mite on the foliage plants. This technology cannot be transferred to the UK as Banks grass mite is not native to Europe. However the apparent success of this system does demonstrate that the principle is sound.

The aim of this project was to find a host plant which supported a species of mite that was host specific, and therefore did not pose a risk to adjoining nursery stock plants, but could also act as a food source for generalist predators of mites such as *A. andersoni*. Once the prey mite was established on these ‘banker’ plants, high numbers of predators could provide a reservoir of biocontrol agents that could move onto surrounding plants and improve the biological control of two-spotted spider mite.

Materials and Methods

Experimental sites

Field experiments were done between April and October 2006 at Bransford Webbs Plant Company, Worcester. Laboratory work was done at ADAS sites at Boxworth (Cambs) and Rosemaund (Herefordshire).

Field experiments

Work was done to evaluate two possible banker plants systems. All work was done in a polythene tunnel (30m x 5m) with side ventilation.

Banker plant system 1 – Fruit tree red spider mite/whitebeam tree

The first banker plant system investigated used fruit tree red spider mite (*Panonychus ulmi*) as prey and the whitebeam tree (*Sorbus aria lutescens*) as the host plant. The combination was chosen because this cultivar of *Sorbus* is known to be very susceptible to infestation by *P. ulmi*. Containerised trees in 5 L pots (c. 2.2 m high) from the outdoor stock beds at Bransford Webbs Plant Company were examined in April 2006 for the presence of winter eggs of *P. ulmi*. These were visible as dark red eggs clustered around the base of terminal shoots. Ten heavily infested trees with more than 50 eggs per terminal shoot were selected and placed in the polythene tunnel. Once the leaves of the whitebeam tree had expanded, they were examined for *P. ulmi* mites at regular intervals. Once mites had started to hatch from overwintered eggs (mid-May), a random sample of 20 leaves was examined *in situ* on each tree (using a x 10 hand lens) for the presence of *P. ulmi* and any naturally occurring predators. This was done on 5 and 23 June, 3 and 18 July, 2 and 16 August and 15 September. An additional five plants with high *P. ulmi* populations were brought into the tunnel from outdoors on 25 July as *P. ulmi* numbers on the original plants had declined to low levels.

Banker plant system 2 – lime mite/lime tree

Two year old bare root lime trees (*Tilia cordata*) c. 1.5 m high from the outdoor stock beds at Bransford Webbs Plant Company were potted up in 5 L pots in March 2006 and maintained in the polythene tunnel. As the leaves expanded, the trees were examined for the presence of lime mite (*Eotetranychus tiliarium*), but initially none were present. To ensure an infestation of *E. tiliarium*, on the experimental plants, a natural infestation of *E. tiliarium* was located on a mature *T. cordata* tree in Cripplegate Park, Worcester on 5 July 2006. Infested leaves were removed, examined under the microscope for predatory mites and then stapled to the experimental plants at the trial site. A further introduction of *E. tiliarium* was made on 18 July 2006. All visible predators were removed to reduce any natural predation, but it is likely that predatory mite eggs were missed by this method. A random sample of 20 leaves was examined *in situ* on each tree (using a x 10 hand lens) for the presence of *E. tiliarium* and any naturally occurring predators. This was done on 3 and 18 July, 2 and 16 August and 2 and 10 October. In contrast to *P. ulmi* on whitebeam, numbers of *E. tiliarium* on lime increased rapidly from mid-July onwards and the latter mite/tree combination was chosen for more detailed work with commercially available predators.

Introduction of predators to banker plants

To investigate whether predators would survive using *E. tiliarium* as a prey source, commercially available predators were introduced onto the lime trees once the *E. tiliarium* population had built up. This was done as follows:

1. *Phytoseiulus persimilis*: one introduction of 2,000 mites was made on 26 September. These were applied from a shaker bottle in a bran carrier.

2. *Feltiella acarisuga*: 250 pupae of this predatory midge were introduced as pupae in plastic pots with emergence holes on each of three dates (29 August, 7 September and 13 September).

Introduction of pest (two potted spider mite) to banker plant system The aim of this work was to determine whether predators raised on banker plants would influence the level of *T. urticae* infestation on a susceptible plant subject. On 12th May 2006, 100 plants of *Choisya ternata* in 3 L pots were placed into the tunnel, grouped into five blocks of 20 plants. One lime banker tree was placed between each block. The *Choisya* were infested with two-spotted spider mite on 18 and 21 August and 15 September, by placing infested leaves directly onto the test plants, using mites obtained from *Musa*, *Choisya* and *Crocasmia* respectively.

Laboratory experiments

No-choice predatory mite and midge feeding experiments

Laboratory bioassays were done to obtain quantitative data on the consumption of *P. ulmi* and *E. tiliarium* by commercially available predators. The following predators were tested:

1. *Phytoseiulus persimilis*
2. *Amblyseius andersoni*
3. *Amblyseius californicus*
4. *Feltiella acarisuga*

Samples of these predators were obtained from Syngenta Bioline Ltd, Essex. Detached lime or whitebeam leaves were checked under a binocular microscope before use and any naturally occurring mites, predators or eggs were removed. Eight adult *P. ulmi* or *E. tiliarium* were placed onto each of 40 whitebeam or lime leaves respectively. Individual infested leaves were placed onto damp filter paper in screw-top tubes (7.0 x 3.0 cm). One adult predatory mite, or one *F. acarisuga* larva was added to the tube, using ten replicate tubes per predator. Tubes with no predators were used as untreated controls. The tubes were incubated at 21 °C, and 18:6 hours light: dark for 24 h (*P. ulmi*) or 48 h (*E. tiliarium*), after which the number of surviving prey mites in each tube were counted.

Visual observation of interactions between predators and prey.

In order to help confirm the results of the laboratory experiments, additional observations of predator behaviour were done. Samples of *Phytoseiulus persimilis*, *Amblyseius andersoni* and *Amblyseius californicus* were obtained from Syngenta Bioline Ltd, Essex. Lime leaves infested with *E. tiliarium* were removed from the trees and placed into a Petri dish on damp filter paper. Predatory mites were introduced (five adult mites per dish, three replicate dish per mite species). Five *F. acarisuga* predatory larvae were taken directly from those present on lime trees at the experimental site were also introduced into three replicate dishes. Observations were made using a binocular microscope for periods of 30 minutes. Quantitative data was not obtained during this work, but records of the feeding behaviour were made.

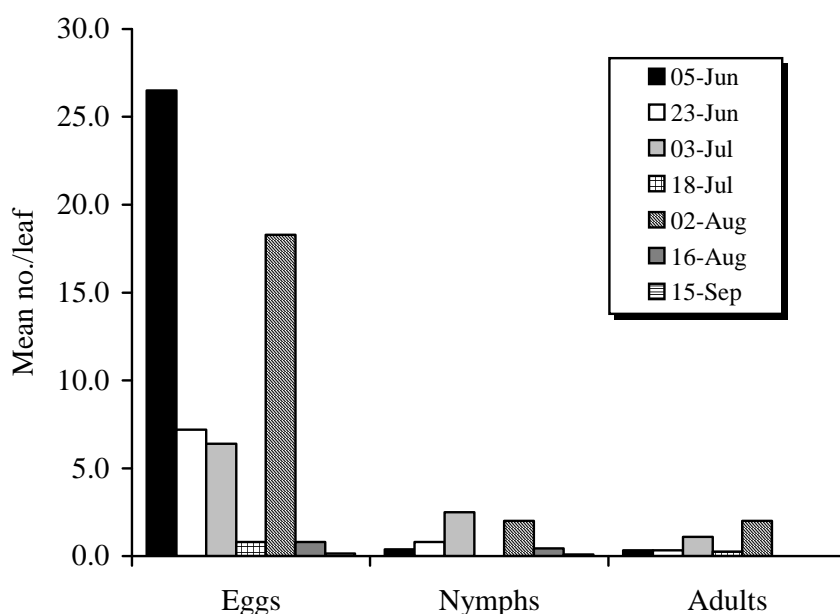
Results and Discussion

Field experiments

Banker plant system 1 - Fruit tree red spider mite/whitebeam tree

The mean numbers of *P. ulmi* found on whitebeam banker plant trees between early June and mid September are shown in Figure 1. Although high numbers of mites were initially present, the population of all stages soon declined rapidly during June and July. The introduction of new, infested plants on 25 July increased mite numbers temporarily in early August, but by mid-August populations had again substantially decreased.

Figure 1. Mean numbers of *P. ulmi* on whitebeam tree leaves June to September 2006.



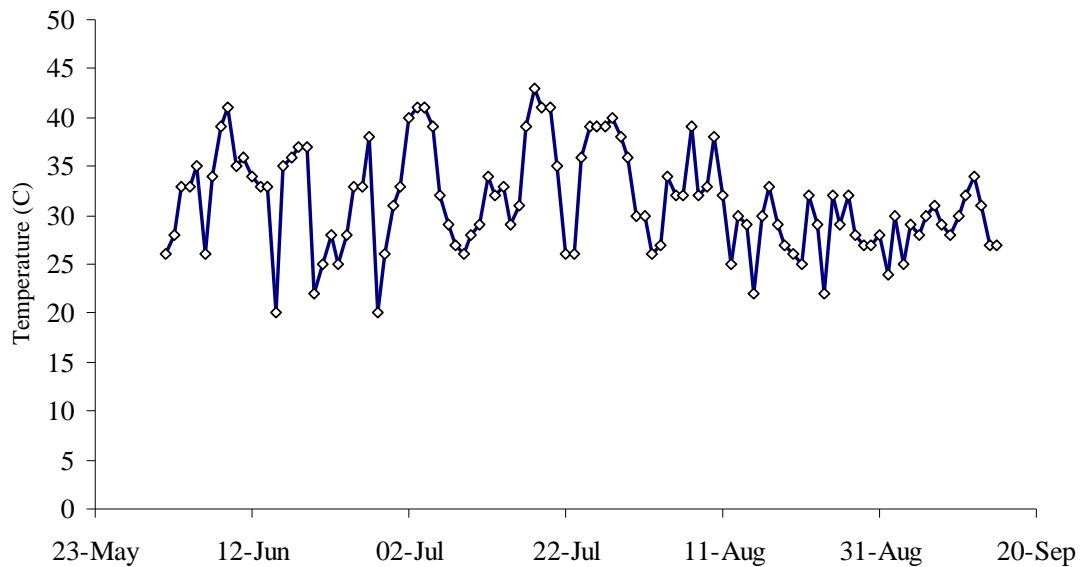
Counts of naturally-occurring (i.e. not introduced) predatory mites made during the assessments for *P. ulmi* showed that the predatory mite *A. andersoni* could be easily found throughout the season, though only in low numbers (Table 1). *A. andersoni* has been previously reported as part of a complex of predatory mite species on whitebeam (Herbert, 1981).

Table 1. Mean numbers of *A. andersoni* naturally found on whitebeam tree leaves.

Date	Mean number per leaf
16 June 06	0.2
23 June 06	0.1
18 July 06	0.25
25 July 06	0.35
2 August 06	0.25
16 August 06	0.3
15 September 06	0.35

As the counts were made *in situ* using a hand lens, it is probable that some mites were missed, and the counts of *P. ulmi* given in Figure 1 are probably an underestimate. The factors responsible for the decline in numbers of *P. ulmi* are not precisely known, but are most likely to relate to high temperatures in the tunnel. Figure 2 shows the maximum daily temperature inside the tunnel during the experimental period. The mean maximum temperature was >30 °C throughout June and July, and only marginally lower in August and September. On individual days, the temperature sometimes exceeded 40 °C. When whitebeam trees left outdoors were examined, *P. ulmi* was often present in moderate to high numbers, despite the presence of predatory mites. Outside maximum air temperatures averaged c. 23 °C in June and July, some 9 °C lower than in the tunnel, and closer to the range of 25-28 °C given by Helle & Sabelis (1985) as the optimum temperature for *P. ulmi*.

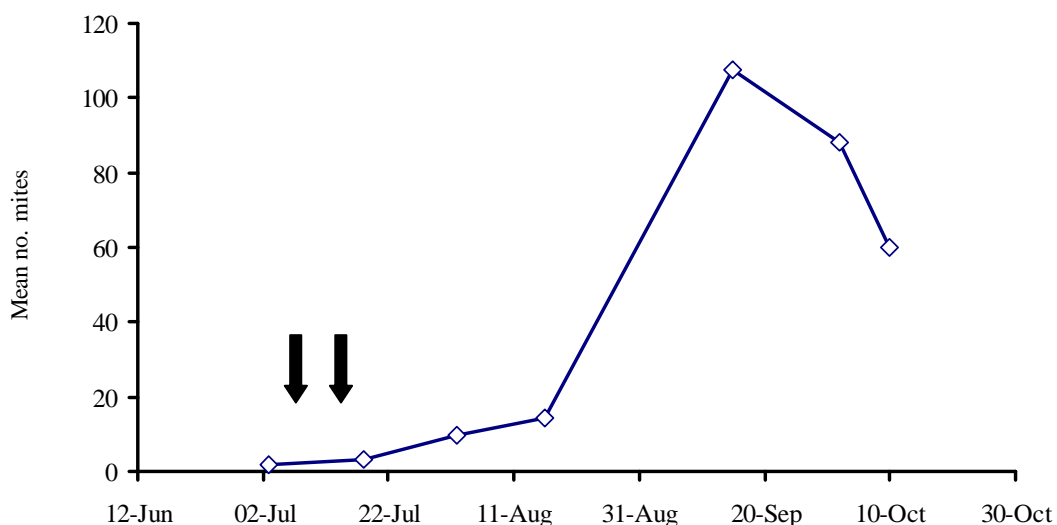
Figure 2. Maximum daily temperatures (°C) inside the experimental trial polytunnel, June – September 2006.



Banker and prey mite system 2 - lime mite/lime tree

Numbers of lime mites increased slowly on lime trees from early July onwards, increased more rapidly during August, and reached a peak of >100 mites per leaf in mid-September. Populations then decreased gradually as natural leaf senescence occurred (Figure 3).

Figure 3. Mean numbers of *E. tiliarium* on lime tree leaves June to October 2006 (arrows indicate dates of introductions of *E. tiliarium*)



Infestation of lime leaves with the lime mite caused white spots on the dorsal leaf surface initially, but as mite numbers increased, infested leaves turned yellow or brown and showed signs of necrosis. Lime mites tended to cluster in large colonies around the leaf veins. At the time of the final assessment on 10 October 2006, leaf senescence had begun and some mites had turned brown ready to enter the diapause stage. Lime mite overwinters as diapause adult mites, and can cause damaging infestations to broad leaf lime trees, especially street trees (Strouts & Winter, 2005).

As with the whitebeam, the most common naturally-occurring predator observed during sampling (Table 2) was *A. andersoni* (this identification was made by Mike Lole, ADAS Entomologist, and confirmed by Dr Anne Baker of the British Museum). *A. andersoni* has been recorded regularly as the dominant predator in lime trees (van der Linden, 2005, Kropczynska-Linkiewicz, 2005).

Because the whitebeam/*P. ulmi* banker plant system was unsuccessful, all other work used the lime/*E. tiliarium* system.

Introduction of predators to banker plants

Larvae of *F. acarisuga* were first seen on 15 September, and after the application of *P. persimilis* on 26 September, this predator was recorded at the next two assessments in October. This indicated that both predators were able to feed on lime mite, and this was confirmed by the laboratory experiments. It is believed to be the first record of predation of lime mite by these species.

Table 2. Mean numbers of naturally occurring *A. andersoni* and introduced *F. acarisuga* and *P. persimilis* on lime tree leaves.

Mean number <i>A.</i>	Mean number <i>F.</i>	Mean number of <i>P.</i>
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Date	<i>andersoni</i> per leaf	<i>acarisuga</i> per leaf	<i>persimilis</i> per leaf
3 July 06	0.10	0.00	0.00
18 July 06	0.15	0.00	0.00
2 August 06	0.20	0.00	0.00
16 August 06	0.25	0.00	0.00
15 September 06	0.35	0.15	0.00
2 October 06	0.00	0.25	0.50
10 October 06	0.00	0.55	0.45

Introduction of pest (two potted spider mite) to banker plant system

T. urticae failed to establish on the *Choisya ternata* test plants, so no data on the effect of the banker plants on *T. urticae* was obtained.

Laboratory experiments

No-choice predatory mite and midge feeding experiments

The results of the experiments on predation of *P. ulmi* and *E. tiliarium* are given in Figures 4 and 5 respectively.

For *P. ulmi* the data indicate that after 24 h, only *F. acarisuga* larvae and *P. persimilis* significantly reduced the proportion of *P. ulmi* surviving in comparison with the control ($P = 0.05$). For *E. tiliarium*, only *F. acarisuga* larvae significantly reduced the proportion of *E. tiliarium* surviving in comparison with the control ($P = 0.05$) after 48 h.

Figure 4. The proportion of prey mites (*P. ulmi*) remaining after exposure to predatory mite adults and *F. acarisuga* larvae after 24 h (error bars are 95% confidence limits)

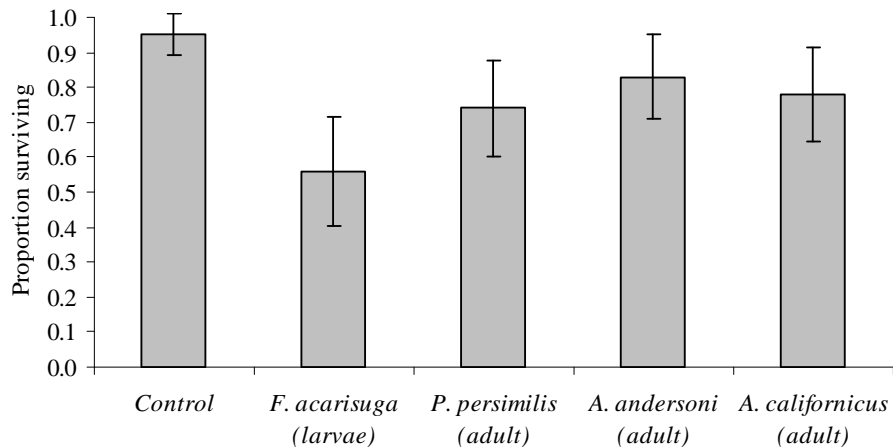
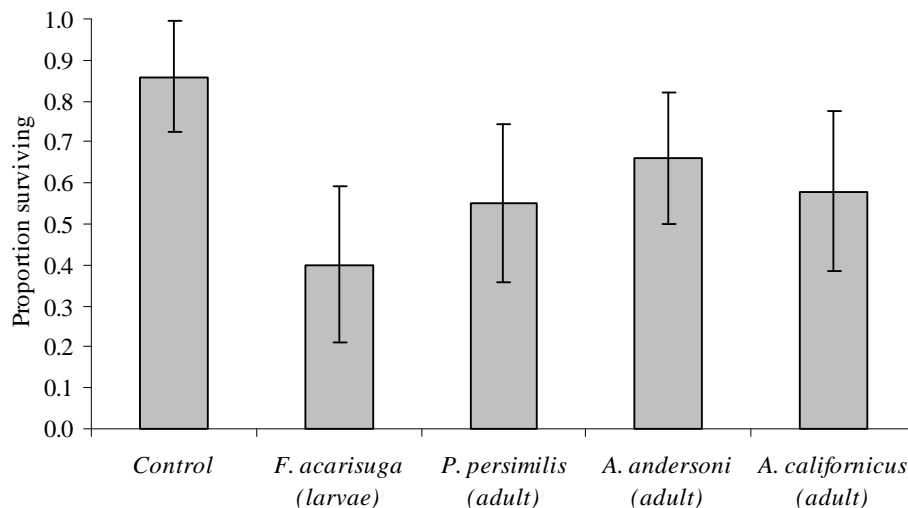


Figure 5. The proportion of prey mites (*E. tiliarium*) remaining after exposure to predatory mite adults and *F. acarisuga* larvae after 48 h (error bars are 95% confidence limits)



Visual observation of interactions between predators and prey

1. *Feltiella acarissuga*: larvae predated eggs, larvae and adults of *E. tiliarium*. Larvae were relatively immobile on the leaf compared to predatory mites, and tended to stay in the area where a colony of *E. tiliarium* was present and feed until all the prey was consumed. When feeding, the mouth hook of the larva penetrated the mite prey, and the contents were then sucked out. This species was seen to complete its development with no prey other than lime mites, and successfully pupate on the leaf.
2. *Phytoseiulus persimilis*: this species actively searched the leaf surface. Feeding on *E. tiliarium* was observed in all replicates within 5 minutes, although the proportion of searching time was high compared to the time spent handling prey.
3. *Amblyseius andersoni*: this species was much slower to locate prey than the other predatory mite species observed. No feeding was observed for the first 20 minutes after the introduction of prey. Mites collected from the wild seemed to be more active than the commercially-reared strain.
4. *Amblyseius californicus*: this species was smaller and paler in colour than *A. andersoni*, but much more active on the leaf. Predation of *E. tiliarium* was seen within 3- 5 minutes. Adult *A. californicus* seized prey more rapidly than *P. persimilis*, grasping the prey at one end and sucking out the contents, although this process itself was slow.

Conclusions

- *Panonychus ulmi* (fruit tree red spider mite) and whitebeam trees are unsuitable for use as a prey mite/banker plant, as it was not possible to culture the mites under protection at prevailing summer temperatures.
- *Eotetranychus tiliarium* (lime mite) and broad leaf limes are suitable for use as a prey mite/banker plant system; the trees are cheap to purchase bare root, easy to cultivate under protection, and the lime mite increases well.

- *E. tiliarium* numbers increased from mid season onwards, but to be effective as a prey mite, they need to build up earlier in the season, from end June onwards. Further work is needed to devise a system to achieve this.
- Three predatory mite species (*Phytoseiulus persimilis*, *Amblyseius andersoni* and *Amblyseius californicus*) and one predatory midge (*Feltiella acarisuga*) were shown to feed on *E. tiliarium* and so could be utilised in a biological control banker plant system. Of these, *F. acarisuga* was shown in laboratory experiments to be the most effective predator.
- It was not possible to demonstrate movement of predators from the banker plant to adjoining pest infested test plants in the first year's experiments. Further work on this topic is required.

Technology transfer

None to date.

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