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Mass rearing of thrips and assay method for screening of insecticides

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ABSTRACT

Mass rearing of the thrips species, *Frankliniella occidentalis*, *F. intonsa*, *Thrips palmi* and *T. tabaci* has been carried out. Thrips fed with pollen laid eggs into water through a thin membrane. Eggs were collected on filter paper using an aspirator and kept moist in a petri dish until just before hatching. For collecting newly hatched larvae, a very small amount of pollen and a leaf disc or broad bean seed were placed in a petri dish. Large numbers of larvae gathered at the food in two to three hours and were removed to a rearing cage for mass production. Larvae were fed pollen directly and honey solution through a thin membrane and developed to adult without additional food. As an alternative food, germinated broad bean seeds stripped of the seed coat could be used for both leaf living and flower living thrips. As an assay method for screening liquid insecticides, insect dipping is recommended. More than 30 insecticides were tested against *F. occidentalis*.

INTRODUCTION

Since the 1980s, several thrips pests such as *Thrips palmi* and *Frankliniella occidentalis*, have become serious pests of vegetable and ornamentals crops. Because of their low susceptibility to many kinds of insecticides and wide distribution, these thrips may be some of the most serious pests in the world. To study life histories, behavior, relationships between thrips and their natural enemies and virus transmission it is necessary to rear thrips in the laboratory. Mass production of thrips is also needed for screening insecticides. Various rearing methods of thrips have been described, but none for mass rearing of thrips species or strains with differing susceptibility to insecticides or geographic characteristic.

No thrips has been reared artificially on a completely synthetic diet. Murai & Ishii (1982) developed an artificial method which used pollen and honey solution available through a stretched thin membrane (Sealon film or Parafilm). Using this method the whole life cycle of several thrips species, *F. intonsa*, *F. occidentalis*, *T. tabaci*, *T. hawaiiensis*, *T. coloratus*, *T. flavus* (Murai, 1988) and *T. nigropilosus* (Nakao, 1993) could be completed.

This paper describes a mass rearing method for thrips and an insecticide assay method.

MATERIALS AND METHODS

Insects

F. intonsa originated from adults collected in white clover flowers in Shimane pref. (Honshu, Japan) in June, 1990. *T. tabaci* originated from adults collected in an onion field in Shimane pref. (Honshu, Japan) in June, 1990. *F. occidentalis* originated from adults collected in

chrysanthemum flowers in Shizuoka pref. (Honshu, Japan) in March, 1994. These three species were reared on pollen using the Murai and Ishii (1982) method for successive generations. *T. palmi* originated from adults collected in an egg plant field in Okayama pref. (Honshu, Japan) in October, 1993, and were reared on kidney bean leaves for successive generations. *F. intonsa* and *F. occidentalis* were assayed for susceptibility to insecticides.

Rearing Method

Two types of cages were used, one for oviposition and larval rearing, and one for rearing larvae and adults. The former consisted of a polyacrylate cylinder (80 mm diameter, 50 mm high), with the bottom end covered by a double stretched membrane. In the oviposition units, gauze (60 to 100 μm) glued to the bottom replaced the membrane. The latter cage, for mass rearing on alternative food, was a tight box (120x98x46 mm) with a ventilation hole (10 mm in diameter) covered with gauze in the lid. To assess susceptibility to insecticides a small glass ring (36 mm diameter, 40 mm high) was used.

Two kinds of pollen, tea pollen (*Thea sinensis*) and pine pollen (*Pinus thunbergii*) were used for collecting eggs and rearing larvae respectively. Pollen was collected in autumn and late spring and stored for more than 3 months in a -20°C freezer. As an alternative food, broad bean seeds (*Vicia faba*) were provided for thrips larvae rearing. Seeds were germinated in running tap water for 3-4 days at room temperature and their seed coats removed.

Adults or larvae (300 to 400 per cage) and pollen were placed in cages and the top end covered with stretched film. A few millilitres of water was deposited on top and covered with a small Petri dish lid or another layer of film for oviposition and rearing, respectively. Thrips were reared at 20°C and 16:8 LD.

Eggs laid in water were collected on filter paper (No.2, 55 mm diameter) in a water suction funnel bottle every 1 to 3 days. The moist filter paper and eggs were placed on a piece of unstretched film floating on water in a closed Petri dish until hatching. The efficiency of collecting eggs on filter paper was tested.

Newly hatched larvae were removed to a rearing cage, with suitable pollen and an uneven paper disc (such as kitchen paper) providing cavities, inserted as a pupation site on the bottom. Honey solution, encapsulated between two stretched membranes, was provided. Alternatively, sugar solution and water could be used. For *T. palmi*, freeze dried sweet pepper powder was provided with pollen for larval rearing. The larvae could thus feed on pollen and pepper powder directly and honey solution through the membrane.

Rearing on alternative food

For the mass rearing of even aged thrips, *T. palmi* larvae were reared on broad bean seeds at different densities (100 to 1500 individuals) in box cages. Several germinated broad beans were provided as food. A sheet of kitchen paper was placed on the bottom of the box to provide pupation sites as with the pollen rearing method. Larvae were reared at a density of 20 to 40 larvae per bean and additional beans were provided if beans dried out at pupation. Thrips were reared at 24°C and 16:8 LD

During insecticide susceptibility tests insects were allowed to feed on 10% honey solution.

Available assay method for screening of insecticides

Three organophosphate formulations, malathion, methidathion and dichlorvos were tested by three methods. All tests were conducted at 22°C.

Insect dipping method

Female adults were dipped in several concentrations of formulation for 20 seconds. Treated thrips were placed in glass ring cage with 10% honey solution. Mortality was observed 24 and 48 h after treatment. A non-ionic surfactant solution (1/10000) was added as spreader.

Film method

Thrips were allowed to feed on several solutions (0.5 ml) of insecticide formulation through a thin membrane. Mortality was observed 4, 24 and 48 h after treatment.

Filter paper method

Solutions (0.5 ml) were dropped onto filter paper (36 mm in diameter) and dried for 60 minutes. The filter paper was placed in a glass cage with 20 adult thrips. Thrips were allowed to touch the filter paper and feed on 10% honey solution through a thin membrane. Mortality was observed 24 and 48 h after treatment.

Application of insect dipping method for insecticide screening for *F. occidentalis*

Field concentrations of 30 insecticide formulations were tested against larvae and adults using the insect dipping method. For the insect growth regulators, thrips were allowed to feed on dipped cabbage leaves and mortality of thrips was observed 96 h after treatment. For other insecticides, mortality was observed 48 h after treatment. Four day old larvae and 10 day old adults were used for screening and tests were conducted at 24°C.

RESULTS

Rearing procedure

Collecting eggs and newly hatched larvae

Numbers of *F. intonsa*, *F. occidentalis* and *T. palmi* eggs collected are shown in Table 1. *F. occidentalis* and *T. palmi* feeding on tea pollen could lay eggs in water through a thin membrane as well as *F. intonsa* but the numbers of eggs per female were fewer for *F. intonsa*.

Table 1. Number of eggs laid in water through a thin membrane.

Species	Eggs per 100 females
<i>F. intonsa</i>	815.3 ± 77.5
<i>F. occidentalis</i>	269.7 ± 30.6
<i>T. palmi</i>	270.6 ± 17.5

The efficiency of egg collection was higher than 80% when large numbers of eggs were laid. Eggs could develop and hatch on moistened filter paper. The emerging larvae walked onto the film from where they could be picked up individually with a fine brush. A quicker way to collect large numbers of larvae was to attract them to a small leaf disc (c. 1 cm diameter) or a very small amount of pollen (< 1 mg) placed on a piece of unstretched film in the Petri dish. Large numbers gathered on this food in 1 or 2 hours and could be collected and counted quickly. The efficiency of collecting hatched larvae by the food trap method was tested on *F. intonsa* and *F. occidentalis*. Larvae gathered on the food within 3 hours and could be collected and counted quickly. The efficiency of collecting larvae was very high for both species.

Mass rearing of larvae

Larvae pupated in the cavities of the paper disc and developed to adults without any additional food. *F. intonsa* larvae developed well in ring cages at all densities. Developmental duration from hatching to adult eclosion was 10 to 12 days and mortality was less than 20% except with 400 introduced larvae.

When reared on the alternative food in box cages, larvae developed to adults without an additional supply of water and pupated in the cavities of the paper. At all rearing densities, *T. palmi* larvae developed as well in box cages on broad bean as in ring cages. Developmental duration from hatching to adult eclosion was 9 to 12 days and mortalities were less than 15% in ring cages and except for a density of 1000 larvae less than 10% in box cages.

Assay method for susceptibility to insecticides

Insect dipping was the most effective evaluation method for determining the susceptibility of thrips to the tested insecticides. Effects were determined within 24 h of treatment. Because of the fumigant effect of dichlorvos it was not possible to evaluate the effect of this pesticide on thrips using the film method or filter paper method.

Screening of insecticides against *F. occidentalis*

Most of the organophosphate insecticides resulted in high mortality of larvae but not adults. Pyrethroids, carbamates and chloronicotinyls resulted in less mortality of larvae and adults than organophosphates. The insect growth regulators flufenoxuron and chlorfluazuron resulted in high mortality of larvae. Teflubenzuron and tebufenozide were not effective against larvae. Thiocyclam and cartap resulted in higher mortality of larvae than pyrethroids, carbamates and chloronicotinyls. Of the 23 tested insecticides none were effective against adults (Table 2).

DISCUSSION

It is difficult to collect large number of eggs of terebrantian species. Artificial membranes have been used for feeding and as oviposition sites for thrips (Sakimura & Carter, 1934; Murai & Ishii, 1982; Kirk, 1985; Teulon & Penman, 1986). Many species can be induced to lay eggs in water through a thin membrane, and their eggs can be collected in this way.

Table 2. Susceptibility of *F. occidentalis* to several insecticides.

Insecticide	Concentration (ppm)	Larval Mortality (%)	Adult Mortality (%)
flufenoxuron	50	100.0	-
chlorfluazuron	25	93.5	-
teflubenzuron	25	8.4	-
tebufenozide	100	0.0	-
dichlorvos	500	100.0	9.4
phenthoate	500	100.0	3.3
prothiofos	450	100.0	0.0
chlorpyrifos-methyl	250	100.0	-
sulprofos	500	96.8	2.4
malathion	500	94.4	0.0
profenofos	400	90.8	-
fenitrothion	500	85.4	6.3
methidathion	400	83.8	3.6
pirimiphos-methyl	450	77.8	13.2
cyanophos	500	63.7	3.3
chlorfenvinphos	250	41.7	9.4
acephate	500	34.5	18.8
thiocyclam	500	85.0	14.8
cartap	500	69.1	7.1
permethrin	100	24.6	0.0
tau-fluvalinate	100	0.0	0.0
bifenthrin	20	0.0	0.0
methomyl	450	21.1	0.0
thiodicarb	750	3.2	6.7
alanycarb	400	0.0	11.8
carbaryl	500	-	3.6
pirimicarb	240	0.0	-
pyridaben	200	14.3	0.0
imidacloprid	100	1.2	8.8
acetamiprid	100	0.0	0.0

With *F. occidentalis* and *T. palmi* the number of eggs laid per female using this method was similar or greater than the techniques presented by Robb (1989) or Kawai (1985). Pollen has a big effect on egg production of flower thrips (Murai and Ishii, 1982; Kirk, 1985; Teulon & Penman, 1991). Many kinds of pollen can be used for rearing thrips. Only a few types of pollen can be collected in sufficient amounts to sustain stock cultures or for mass propagation. Flowers of plants such as pine and maize can be collected and spread on paper in the laboratory. After 2 or 3 days, pollen is shed and collected by sieving. Pollen can be stored at -20°C for more than 2 years and still be used for rearing thrips. In the oviposition unit a little tea pollen (20 to 40 mg) is provided on a piece of tissue paper every 2 to 3 days. In the larval rearing unit 100 to 200 mg is supplied only once. Tea pollen is suitable for oviposition and pine pollen for rearing larvae.

It was previously difficult to collect large numbers of newly hatched larvae quickly. The method described here is very simple and useful for mass rearing and did not damage the thrips larvae through handling. The results suggest that 100 mg pine pollen is enough to rear 500 thrips larvae. More than 500 larvae might damage the thin membrane through the number of feeding punctures. Crushed and dried bee-pollen can also be used for rearing larvae (Murai, unpublished) or dried fruit and leaf powder to rear larvae and adults (Koyama & Matsui, 1992) but they easily moisten and become mouldy in damp conditions. When thrips larvae are

reared on these foods, an oviposition cage with gauze on the bottom should be used to prevent excess moisture.

At all rearing densities *T. palmi* larvae developed as well in box cages on broad beans as in the ring cage. The result suggest that it is possible to rear more than 1000 larvae in a small box cage and to produce a cohort of even aged thrips.

This mass rearing method for thrips is a closed system and has big advantages for maintaining stock cultures of populations from different locations and susceptibility to insecticides and also prevents contamination with other thrips species or strains. It was easy to produce even aged larvae by this method and is therefore suitable to produce insects for insecticide screening.

It was possible to evaluate the effects of liquid formulations within 24 h of treatment using the dip method. For other formulations alternative methods are required. The susceptibility of *F. occidentalis* was relatively low. Pyrethroids, carbamates and chloronicotinyls had no effect on larvae or adults. *F. occidentalis* had not been exposed insecticides for over 20 generations. Therefore it is suggested that the resistance to these insecticides of *F. occidentalis* is stable.

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Mycoinsecticides in thrips management

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ABSTRACT

Liquid and powder formulations of the entomopathogenic fungus *Beauveria bassiana* strain GHA were evaluated for management of the western flower thrips *Frankliniella occidentalis* in covered ornamental and vegetable crops. Weekly applications to moderate thrips populations provided control comparable to chemical insecticides and considerably better than releases of predator mites. The level of efficacy is higher on larval thrips than adults, and several days are required to infect and kill target insects. Accordingly, there is some lag time between initiation of treatment and population reduction. Sampling by direct counts detected subtle population differences in treated plots, while sticky traps rendered thrips outbreaks more apparent. The addition of sucrose to the spray did not enhance the efficacy of the *B. bassiana* sprays.

INTRODUCTION

Western flower thrips, *Frankliniella occidentalis*, populations often increase exponentially, quickly reaching levels that cause severe economic losses. Thrips populations are very difficult to control under these conditions, even with repeated applications of chemical insecticides. Such repeated insecticide applications inevitably lead to resistance and present worker exposure hazards, particularly in confined spaces of protected floriculture and vegetable production.

Mycotech Corporation has obtained US EPA registration for formulated preparations of a potent and prolific strain of the entomopathogenic fungus *Beauveria bassiana*, strain GHA. The US EPA classified these formulations as "reduced risk" insecticides and, based on toxicology studies, exempted the strain from the requirement to establish residue tolerances on food crops.

Beauveria bassiana acts as a contact mycoinsecticide. The contact unit is an asexual, nonmotile spore called a conidium. Conidia adhere to the insect integument, where they germinate and penetrate causing an invasive infection that kills the insect in 3-7 days.

Laboratory bioassays have shown that *B. bassiana* GHA is efficacious against *F. occidentalis* in addition to aphids and whiteflies (Murphy et al. 1998). Beginning in 1995, Mycotech Corporation has collaborated with university and contract researchers in a series of field trials conducted in commercial floriculture and vegetable greenhouses. This paper presents results from 4 trials that were selected because they illustrate strategies for effective use of *B. bassiana* in thrips population management in protected crops.

METHODS AND MATERIALS

Beauveria bassiana preparations used in these trials are commercial formulations marketed under the trade name BotaniGard®. The active ingredient in the formulations is a dry conidia powder recovered from a solid substrate production system. This powder contains 1.4×10^{11} conidia per gram. Two formulations were used in the trials, an oil based emulsifiable suspension (ES) containing 2.1×10^{13} conidia per litre and a water dispersible powder (WP) containing 4.4×10^{13} conidia per kg.

Trials were conducted in commercial greenhouses with collaborating researchers in California and Maryland, USA, and in Morocco. *Beauveria* formulations were applied with grower wand sprayers using typical spray volumes for specific crops and local grower practices. Spray concentrations ranged from 2.5×10^7 to 1.1×10^8 conidia per ml, representing 125–500 ml of ES formulation or 62–250 grams of water dispersible powder formulation per 100 litres.

Thrips populations were assessed by two methods. Direct counts were made by tapping flowers over white paper, with results expressed as average number of thrips per flower from multiple flowers. Adult and nymphal thrips could be distinguished in direct counts. In other trials, adult thrips populations were also monitored by counts of thrips on yellow sticky cards placed over the plants.

Table 1. Trial scheme for evaluation of *B. bassiana* formulations in thrips control programmes.

Trial	Treatments (100 litres ⁻¹ spray)	Number of applications	Plots
Gerbera daisies California, US	<i>B. bassiana</i> ES 250 ml Neem oil 1 liter <i>Phytoseiulus persimilis</i> one release	3	18 m ² (2 benches)
Roses California, US	<i>B. bassiana</i> ES 500 ml <i>B. bassiana</i> WP 250 g Methiocarb 21 g	3	50 m ² (3 replicates)
Bedding Plants Maryland, US	<i>B. bassiana</i> WP 125 g <i>B. bassiana</i> WP 250 g Fenoxycarb 113 g <i>Neoseiulus cucumeris</i> one release	7	186-246 m ² (whole houses)
Peppers Inezgane, Morocco	<i>B. bassiana</i> ES 250 ml <i>B. bassiana</i> ES 250 ml with 1 kg sucrose	3	300 m ² (6 rows × 30 m)

In most trials, *B. bassiana* formulations were compared with an untreated control and chemical insecticides used by the grower. In two trials, predatory mites were used as an additional comparison. Chemical insecticides were applied at recommended label rates with the same frequency of application, spray equipment and volume as *B. bassiana* formulations. The predatory mites were applied as single releases in both trials where they were used.

RESULTS AND DISCUSSION

Gerbera daisies, Watsonville, California, USA

The gerbera trial results suggest a greater susceptibility to *B. bassiana* of larval thrips than adults (Table 2). As a practical matter, this can result in a delay in reduction of the adult population. Accordingly, vigilant scouting for early infestations is paramount, and judgment of the level of control requires experience and patience. The thrips in gerberas are concentrated in upright flowers and accessible to overhead sprays, making this a good system for a contact material such as *B. bassiana*.

Table 2. Average number of western flower thrips per flower on gerbera daisies treated with *Beauveria bassiana*, neem oil, or *Phytoseiulus persimilis* - Watsonville, California, USA.

Treatment		Weeks after first application			
		0	1	2	3
<i>B. bassiana</i>	adults	1.5	4.6	5.9	2.7
	larvae	1.5	1.5	0.2	0.8
	total	3.0	6.1	6.1	3.5
<i>P. persimilis</i>	adults	1.7	3.2	9.3	10.1
	larvae	2.0	1.6	7.9	3.3
	total	3.3	4.8	17.2	13.4
Neem oil	adults	2.0	2.1	6.7	5.4
	larvae	1.3	1.3	3.8	3.9
	total	3.3	3.4	10.5	9.3

The predatory mites *Phytoseiulus persimilis* appeared to have had little effect on the thrips. Neem oil (Triac®) was less effective than the *B. bassiana*.

Roses, Goleta, California, USA

In the rose trial, both of the *B. bassiana* formulations at equivalent active ingredient rates were comparable to MesuroI® (methiocarb) in control. As in most trials, the treatment effects with *B. bassiana* were most apparent two weeks after treatment initiation. Two surprising results were the lack of rapid effect of methiocarb and the apparent strong effect of the WP formulation of *B. bassiana* one week after the first application. None of the differences was statistically significant, but this is another case of maintaining a moderate thrips population at a low level by early treatment (Figure 1.)

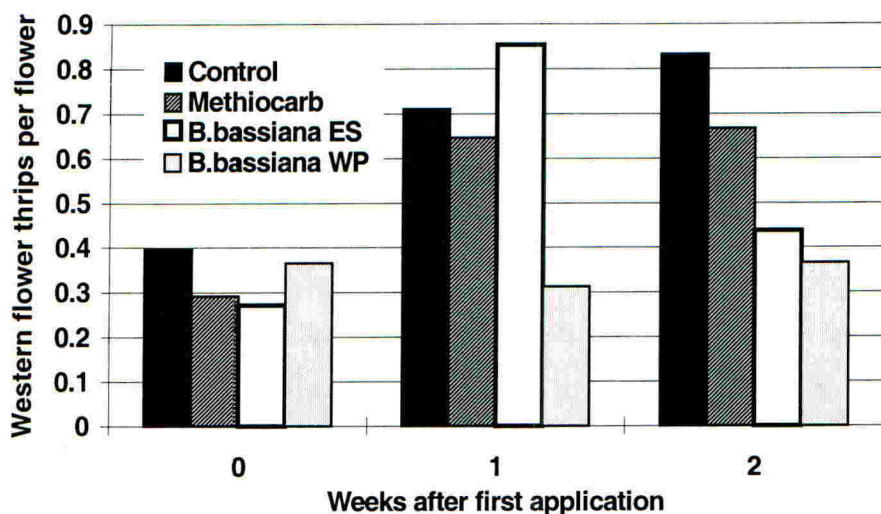


Figure 1. Western flower thrips per flower on Cardinal roses treated with two formulations of *Beauveria bassiana* or methiocarb, Goleta, California.

Bedding plants, Burtonsville, Maryland, USA

Thrips management in mixed bedding plants in Maryland was evaluated with weekly applications of two rates of the WP formulation of *B. bassiana* and Precision® (fenoxycarb). The trend again demonstrated that a moderate population of thrips can be held in check with regular applications of *B. bassiana* alone. Fenoxycarb provided comparable performance and may be a complementary soft material to be used in conjunction with *B. bassiana*. The predatory mites alone did not prevent a thrips outbreak (Figure 2).

The population patterns displayed by the two sampling methods indicate that subtle differences are best detected by the direct tap counts, while thrips outbreaks may be more readily detected by sticky card traps.

Sweet peppers, Inezgane, Morocco

As an additional comparison in the pepper trial, a sugar bait was included as a bait to bring the thrips into contact with the fungus conidia. Lower adult thrips counts relative to the control were statistically significant for the *B. bassiana* with sugar plot on day 6 after the first application and for the plots treated with *B. bassiana* with and without sugar for day 13 ($\alpha=0.05$, Duncan's MRT). Significantly lower means for larvae counts occurred for *B. bassiana* without sucrose on day 13 and for *B. bassiana* with and without sucrose on day 21. When compared to the control plot, the combined larval and adult data for days 13 and 21 show 71% less thrips in the *B. bassiana* without sucrose plot and 47% less thrips in the *B. bassiana* with sucrose plot (Table 3). It should be noted that, since the plots were not replicated, the statistical differences apply to the plots *per se* rather than the treatments.

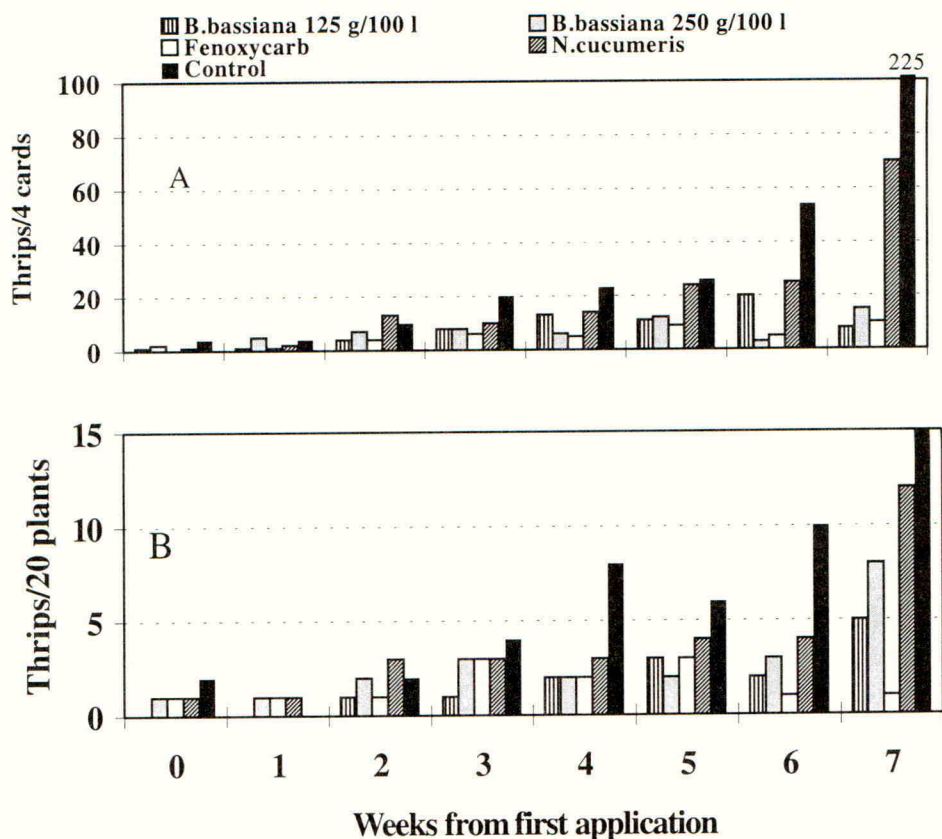


Figure 2. Population trends of thrips in bedding plant greenhouses (Burtonsville MD) as measured by yellow sticky cards (A) and flower tap counts (B).

Table 3. Western flower thrips/flower on sweet peppers treated with *Beauveria bassiana* in Inezgane, Morocco.

Treatment		Days after first application			
		0	6	13	21
Control	adults	4.2a	2.9a	3.1a	5.6a
	larvae	6.1a	3.5a	1.8a	6.1a
	total	7.9a	6.4a	4.9a	11.7a
<i>B. bassiana</i>	adults	5.2a	2.9a	0.4b	3.6a
	larvae	5.0a	3.3a	0.1b	0.7b
	total	10.2a	6.2a	0.5b	4.3b
<i>B. bassiana</i> with sucrose	adults	3.9a	1.7b	0.9b	4.3a
	larvae	2.9a	4.6a	1.3a	2.3b
	total	6.8a	6.3a	2.3ab	6.6ab

Means within the same column, followed by the same letter are not significantly different (Duncan's MRT, $P = 0.05$).

There was sudden increase in adults at the last evaluation. This is likely to be due to an influx from outside of the greenhouse. Entomopathogens are best used to manage a resident population rather than to deal with a large immigration.

CONCLUSIONS

Western flower thrips can be managed by using *Beauveria bassiana* (GHA strain) treatments to maintain populations at moderate levels, primarily by attacking the larval stage. Large immigrations of adult thrips will require a fast acting knockdown material. The common practice of adding a sugar attractant to enhance the efficacy of thrips control materials was not effective in the trial presented here.

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Density dependent regulation of western flower thrips, *Frankliniella occidentalis*, in field peppers by the insidious flower bug, *Orius insidiosus*

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ABSTRACT

The western flower thrips, *Frankliniella occidentalis*, is a very important pest damaging vegetable crops, and is a vector of tospoviruses. In experimental field plots of peppers in 1996 and 1997, the insidious flower bug, *Orius insidiosus*, invaded in sufficient numbers to significantly suppress western flower thrips adults and immatures. Local extinction occurred within days once predator to prey ratios in the pepper flowers were about 1:40. Populations of *F. tritici* and *F. bispinosa* were also significantly suppressed. The biological insecticide spinosad reduced prey density without significant effects on predator populations, and this resulted in more rapid extinction of western flower thrips. Exclusion of insidious flower bug in treatments of broad-spectrum, synthetic insecticides resulted in greatly increased densities of thrips compared to untreated plots. Biological control of thrips by insidious flower bugs was successful in commercial pepper fields when broad-spectrum insecticides were not used.

INTRODUCTION

Thysanoptera are opportunistic species exploiting temporary or intermittently occurring environments. Pest thrips are *r*-selected with population characteristics that include vagility, a short generation time, polyphagy, a tendency toward parthenogenesis, and possibly a competitive breeding structure that promotes aggregation and exploitation of localized optimal conditions (Mound, 1997). Although there are many records of mortality by natural enemies, there is little information indicating density-dependent regulation of thrips under natural conditions (Sabelis and Van Rijn, 1997; Loomans *et al.*, 1997). Nagai (1990) used an emulsion of fenthion on eggplants in a field to suppress populations of an anthocorid predator *Orius* sp. with little effect on *Thrips palmi* populations. Thrips populations were numerically greater in plots treated with fenthion compared to untreated plots, possibly due to exclusion of the predator's populations.

Parella & Lewis (1997) summarized information on biological control, and concluded that natural enemies play an insignificant role in regulating thrips in field crops. Population attributes of rapid colonization and growth are believed to possibly outstrip the capacities of natural enemies to regulate opportunistic thrips species (Mound & Teulon, 1995). Biotic agents have been somewhat successful for thrips control in greenhouses when integrated with other tactics (Jacobson, 1997). Kirk (1997) questioned whether the lack of reported cases of density dependence under field conditions reflected genuine lack of much effect of natural enemies on thrips populations or simply a shortage of quantifiable information. Sabelis & Van Rijn (1997) used a simple one-predator one-prey model to estimate the intrinsic capacities of predatory arthropods to reduce thrips populations. Model simulations predicted suppression and extinction of western flower thrips, *Frankliniella occidentalis*, by several predators, including anthocorid predators.

The western flower thrips was originally distributed throughout the semi-arid parts of southwestern North America, but the pest is now almost cosmopolitan (Mound, 1997). Other native thrips species commonly inhabit vegetable crops in Florida region including *F. tritici* and *F. bispinosa*. Application of broad-spectrum insecticides was noted in commercial pepper fields to increase rather than decrease thrips populations. During 1996 and 1997 replicated field experiments were conducted to determine if exclusion of predators might be responsible for this observed phenomenon.

METHODS AND MATERIALS

'Camelot' sweet peppers were transplanted to field plots on Julian dates 84 and 83 in 1996 and 1997, respectively. Experimental design each year was a randomized complete block with four replicates. Each plot consisted of two raised beds of black plastic mulch 13 m long with beds spaced 0.9 m apart. Each bed consisted of two linear rows with a 30 cm spacing between and within rows. Treatments included an untreated control, spinosad 2SC at 0.1 kg a.i./ha, fenpropathrin 2.4EC (Valent USA Corp.) at 0.22 kg a.i./ha, and acephate 75S (Valent USA Corp.) at 1.1 kg a.i./ha. Treatments were applied in a water solution at 432 litres/ha using a gas-pressurized backpack sprayer at 413 kPa equipped with three hollow-cone nozzles (D7-45). Side nozzles were directed straight into the plants of the 2-row bed and a nozzle was placed over the top of the bed. Treatments were applied 3 times at weekly intervals each year and 10 flowers sampled per plot 1, 4, and 7 DAT. Flowers were placed in 70% alcohol and thrips and natural enemies were extracted under a stereo scope at 40X. Adult thrips in the samples were separated to species. Immature thrips in the samples could not be separated to species; therefore, immature thrips species composition was assessed by collecting immature thrips from the plots on various sample dates each season and allowing them to develop to adult.

RESULTS

Western flower thrips and *F. tritici* comprised over 99% of adult and immature thrips in pepper flowers in 1996. These species and *F. bispinosa* were abundant in 1997. Insidious flower bugs comprised over 95% of predators both years. Adults and nymphs were observed feeding on adult and immature thrips. Effects of insecticides and predators on adult and immature thrips were determined each year using analyses of covariance over time. Insidious flower bugs significantly ($P < 0.002$) suppressed western flower thrips adults, *F. tritici* adults, and immature thrips in 1996 and 1997. Adults of *F. bispinosa* were significantly ($P < 0.001$) suppressed by insidious flower bugs when abundant in 1997.

Densities of adult western flower thrips and immature thrips were greatest in the untreated plots during the first week of flowering each year, adult and nymphal insidious flower bugs were most abundant during the last week of sampling each year (Figure 1). The one-predator one-prey model of Sabelis and Van Rijn (1997) of the intrinsic capacities of insidious flower bugs to suppress western flower thrips predicted eventual extinction at a predator to prey ratio above 1:217 and extinction in 6.5 d at a ratio above 1:51. The ratio of insidious flower bugs to total thrips in untreated peppers was 1:44 on date 145 in 1996; densities of adult western flower thrips and immature thrips were less than 0.5/flower 5 d later (Figure 1). Ratios of insidious flower bugs to total thrips ranged between 1:54 and 1:40 in samples taken between dates 132 and 142 in 1997. Densities of the predator were greater on date 144 with a predator to prey ratio of 1:35; immature

-* - *F. occidentalis* -○- larval thrips --▽-- *Orius insidiosus*

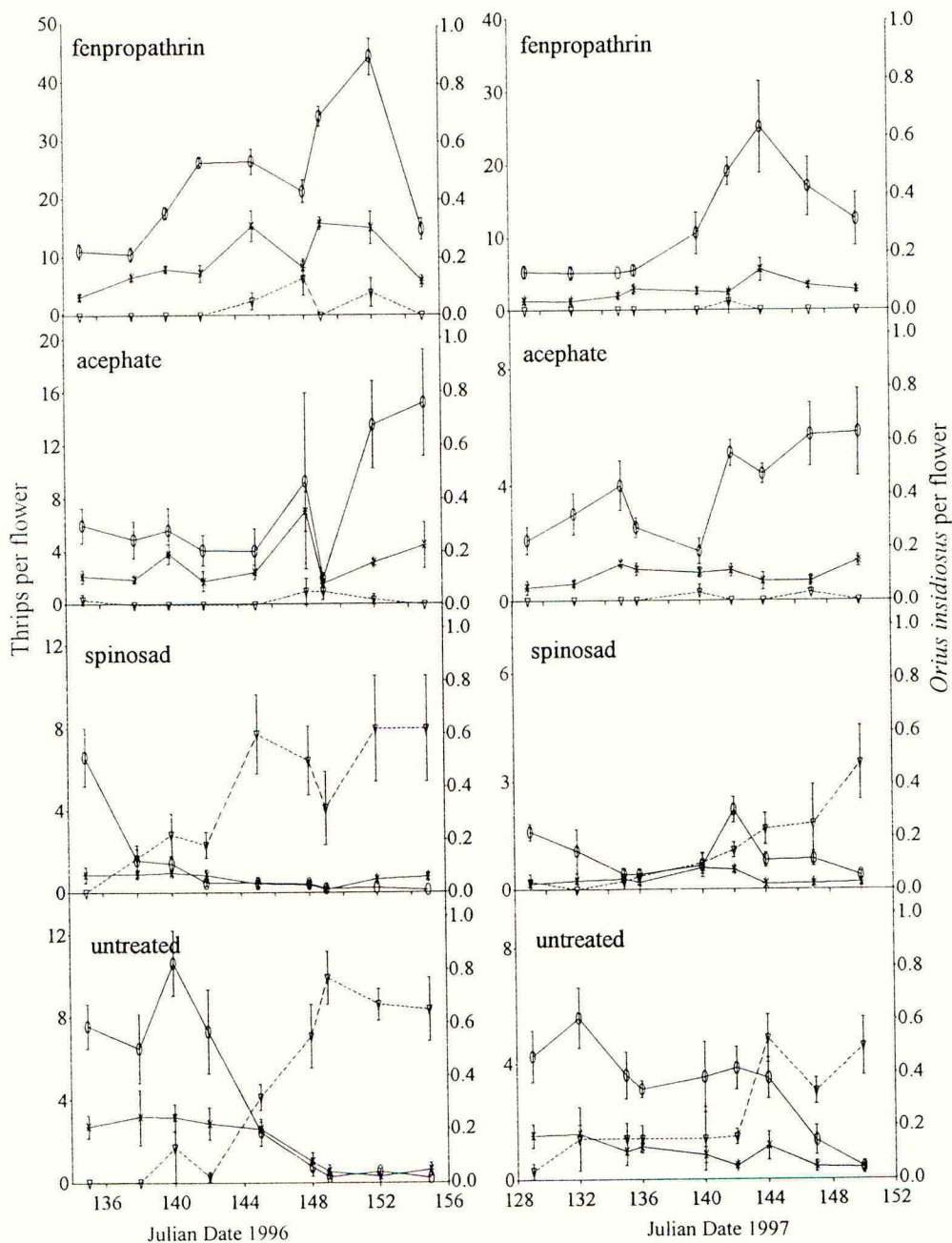


Figure 1. Effect of insecticide treatment on mean density (\pm SEM) of adult *F. occidentalis*, immature thrips and adult and immature *O. insidiosus*.

thrips and western flower thrips adults were less than 0.5/flower 6 d later (Figure 1).

The effects of various insecticide treatments on thrips populations were compared using orthogonal comparisons. Densities of adult western flower thrips and immature thrips were significantly ($P < 0.001$) less in acephate than in fenpropathrin treatments both years. Whether these results were due to better efficacy for acephate, thrips hormologosis for fenpropathrin, or both of these effects is unclear. Few insidious flower bugs survived in acephate and fenpropathrin treatments (Figure 1). Spinosad was significantly ($P < 0.001$) more effective both years than the broad-spectrum insecticides in suppressing western flower thrips adults and immature thrips. Application of spinosad was not detrimental to buildup of insidious flower bug populations.

Densities of thrips and insidious flower bugs were estimated twice weekly in 8-ha commercial pepper fields during the spring of 1997 and 1998. No broad-spectrum insecticides were applied; 3 applications of spinosad 2SC at 0.1 kg a.i./ha were made in 1998. Patterns of population abundance of prey and predator were similar to those observed in the replicated small-plot experiments with insidious flower bugs invading in sufficient densities to provide biological control and thrips densities declining to low levels for the last 4 weeks of each growing season.

Our research revealed density dependence of thrips in peppers under field conditions. We are examining the effects of insidious flower bugs on thrips inhabiting other plant hosts in the agroecosystem.

ACKNOWLEDGEMENTS

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Biology and prospects for enhancing biocontrol of the western flower thrips *Frankliniella occidentalis* in cut roses

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ABSTRACT

The within-crop distribution and pupation biology of the western flower thrips, *Frankliniella occidentalis*, was studied in two growth substrates of cut roses, open peat and rockwool bags. The number and proportion of thrips pupating successfully was significantly smaller in rockwool. In peat, the thrips emerged evenly from the substrate, whereas in rockwool more thrips emerged from the cubes in which roses grow than from the plastic covered areas. Within-crop distribution of thrips in flowers and buds and on leaves of erect and bent shoots was similar in both substrates. A preliminary experiment on pot roses was conducted to study the interaction between soil-dwelling *Hypoaspis* mites and crop-dwelling *Amblyseius cucumeris*. Evidence of indirect food competition was found between the two species when they occurred together, resulting in a marked decrease of *Hypoaspis* numbers in the pots. The results are discussed in terms of the feasibility of using more than one biocontrol agent of thrips in different substrate types of cut roses.

INTRODUCTION

Two kinds of predatory mites are currently recommended for the control of the western flower thrips (WFT) in cut roses: crop-dwelling *Amblyseius cucumeris* preying on larvae and soil-dwelling predatory mites of *Hypoaspis* spp. preying on pupae. However the potential interactions and feasibility of the simultaneous use of these predators has not been studied in this crop. Two types of growth substrates, rockwool and peat (the latter either as open beds or in bags) are currently used to grow cut roses in Finland. The type of growth substrate may affect the pupation success of WFT particularly and also the efficacy of their biocontrol agents. Studies were therefore initiated on the biology of WFT in greenhouse cut roses (cultivar White Escimo) grown in rockwool and peat (Experiment 1) and on the use of the two predatory mite species simultaneously in roses (Experiment 2) to find out ways of enhancing biocontrol success of WFT in this crop.

METHODS AND MATERIALS

Experiment 1: The within-crop distribution and pupation biology of WFT was studied in four 38 m² greenhouse compartments, each of them with one bed of open peat and one bed of rockwool bags. The thrips were allowed to multiply for 2.5 - 3 months inside net cages (n = 6 per substrate). The crop was then harvested and the number of adults, larvae and pupae in flowers/buds, erect shoots and bent shoots was determined microscopically. Thrips that

emerged from the substrate after successful pupation were captured over 2-3 weeks, i.e. until no more emerged, with sticky traps placed inside plexi glass cages that completely covered the bottom of the net cages. Thrips captured within the first 24 hours were assumed to originate from plants after their removal. In some plexi glass cages ($n = 10$ for rockwool and 15 for peat), the distribution of adults emerging from different parts of the substrate was studied in more detail by covering individual open rockwool cubes with 1-litre plastic boxes that had separate sticky traps inside them. In peat beds, plots of the size of one cube around plant stems were covered with boxes to compare the proportion of adults emerging from the total area of open cubes in relation to the rest of the bed area in the two substrate types.

Experiment 2: A cage experiment lasting 10 weeks was conducted using 6 cages per treatment and 5 pot roses per cage. There were three treatments: (1) untreated control; (2) *H. miles* applied in the pots at the rate of 50 per pot at the start of the experiment, and (3) *H. miles* applied as above and *A. cucumeris* applied at the rate of 50 (first application), 25 (second application) and 25 (third application) per plant in weeks 0, 3 and 6 of the experiment. Thrips numbers were monitored by taking leaf samples ($n = 1$ per plant per cage) every two weeks. Leaf area damaged by thrips was measured from the same leaves using an image analyser. At the end of the experiment, the size of the adult thrips population was determined by placing blue sticky traps in the cages for five days. The number of *H. miles* per pot was determined by extracting the mites from the soil in dry Baermann funnels.

RESULTS

Within-crop distribution of thrips

Within the crop, the thrips were distributed similarly in plants grown in either rockwool or peat (Fig. 1). The total number of prepupae and pupae found in all cages on the plants was less than 10 and is not included in calculations. The majority of adults recovered on plants (mean \pm s.e. per cage 219 ± 71 in rockwool, 319 ± 79 in peat) were found in flowers. Over half of the total number of larvae (528 ± 106 in peat and 768 ± 206 in rockwool) were found on the leaves of erect and bent shoots and the rest in flowers and buds.

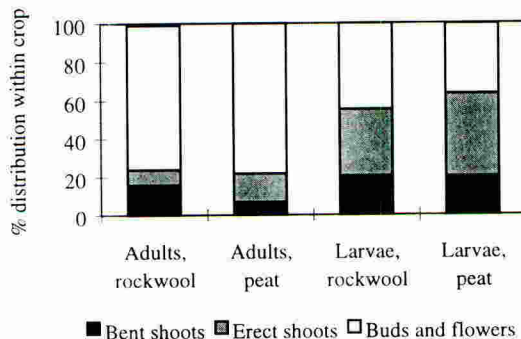


Figure 1. Within-crop distribution of adults and larvae of *F. occidentalis* in two growth substrates of cut roses (White Escimo). $n=6$ cages for both substrates.

Distribution of thrips between plants and substrate

There were two main differences related to thrips biology in peat beds and rockwool bags. First, the proportion of the total thrips population (1727 ± 247 in peat, 1285 ± 298 in rockwool; difference not significant at $p < 0.05$, t-test) in the larval stage at the time of removing the crop was double in rockwool compared to peat, but the proportion of successfully pupated thrips emerging from rockwool was only half of that from peat ($F = 0.76$ with $p = 0.389$ for the substrate effect on the total size of the thrips population, $F = 6.43$ with $p = 0.0047$ for the size of thrips population in different developmental categories, i.e. larvae, pupae or adults, and $F = 3.90$ with $p = 0.0313$ for the interaction of substrate and number of thrips in different developmental categories, analysis of variance; Fig. 2a). Second, the number of thrips that emerged from the total area of open rockwool cubes in peat beds was almost in direct proportion to that area (3.5%), comprising $4.1 \pm 1.2\%$ of the total number that emerged (Fig. 2b). As in peat, the cubes in rockwool cages covered 3.5% of the total area of the beds, but $39 \pm 10\%$ of thrips captured emerged from this area. The rest (61%) emerged from the plastic covered parts of the rockwool beds.

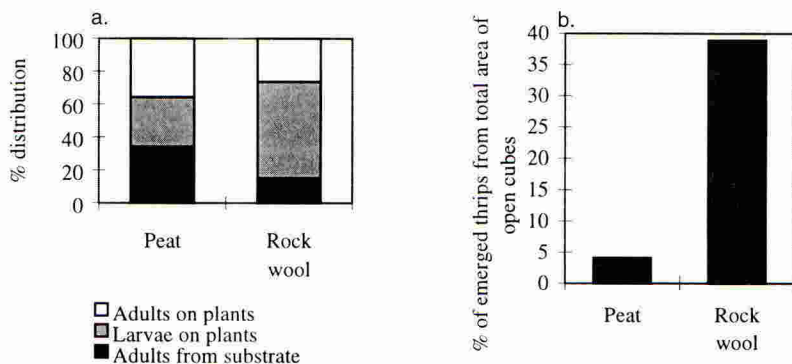


Figure. 2. (a) Distribution of thrips between substrate (=successfully pupated thrips emerging as adults) and plants (larvae or adults) in peat and rockwool cages ($n = 6$ for both) (a); and the percentage of thrips that emerged from the total area of open rockwool cubes in the two substrate types. (b) The proportions emerging from the area of the open cubes were significantly different in the two substrates (t-test, $P = 0.0007$).

Biocontrol of western flower thrips in pot roses with *Amblyseius cucumeris* and *Hypoaspis miles*

Only the combined use of *A. cucumeris* and *H. miles* reduced the number of larvae per leaf significantly compared to untreated controls (Fig. 3). The control efficacy varied between 60 - 78%, depending on the time of sampling. The number of adults was reduced by 72 - 89%. The use of *H. miles* alone also reduced adult numbers significantly, by 30 - 88% on different sampling occasions (Fig. 3). The damaged leaf area was reduced by a maximum of 41% (week 8) by a single application of *H. miles*, and by a maximum of 66% by the combined use of the predators. At the end of the experiment, the number of adult thrips captured was 33% lower in *H. miles* treatments compared to controls, and 49% lower in the combination treatment of the two predators.

At the end of the experiment, the number of *H. miles* per pot in cages where they were used in combination with *A. cucumeris* had declined to 10 ± 4 from the initial 50 per pot. In cages where *H. miles* was used alone, the numbers per pot had increased beyond those applied initially (Fig. 4).

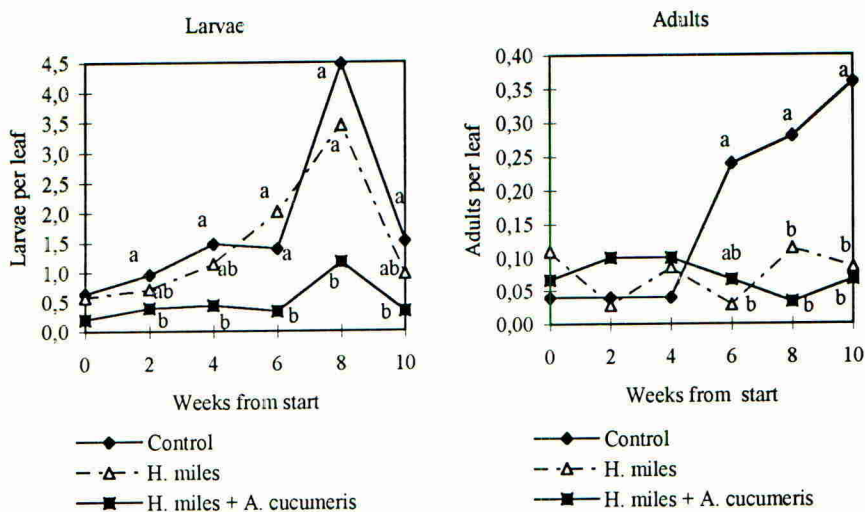


Figure 3. Number of larval and adult thrips per leaf (means of six cages per treatment) on different sampling occasions in the experiment testing the efficacy of *A. cucumeris* and *H. miles* against *F. occidentalis*. Points with same letters on the same day were not significantly different at $P < 0.05$ (analysis of variance). If no letters are given, the numbers were similar in all treatments. Samples in week 0 were taken immediately before the first applications of predators.

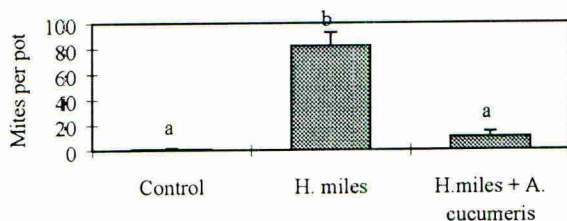


Figure 4. Number of *H. miles* per pot at the end of the experiment testing the efficacy of *H. miles* and *A. cucumeris* against *F. occidentalis* in pot roses. The values, corrected for 70% extraction efficiency, are means \pm SE for 8 pots in control, 12 pots with *H. miles* alone and 9 pots with *H. miles* and *A. cucumeris*. Columns with same letters do not differ significantly from each other at $P < 0.05$ (analysis of variance).

DISCUSSION

The within-crop distribution of thrips is not affected by the substrate of cut roses, but the substrate has a crucial effect on the pupation success of WFT. The peat beds offer homogeneous and favourable pupation conditions for the thrips. In beds of rockwool bags, the open rockwool cubes in which the plants grow, offer the best places for pupation. Either the thrips concentrate in the favourably moist cubes to pupate, or mortality during pupation is lower there compared to that in areas covered with plastic and leaf litter.

The distribution of adults between leaves and flowers may not be quite as the numbers found on plants indicated. Being capable of flying, adults could escape from the plants when they were cut and removed from the cages. This was shown by large numbers of thrips being captured during the first 24 hours after crop removal. Although the greatest damage by thrips from the economic point of view is caused by feeding in buds and flowers, WFT does not avoid laying eggs in rose leaves, either, and the leaves of some rose cultivars are preferred as egg-laying sites compared to petals (Bergh & Le Blanc, 1997). Combining this with the intuitive notion that it must be easier for the adults to escape when disturbed from the open surface of leaves than from inside flowers, the proportion of adults in flowers and buds may have been somewhat overestimated in the sampling.

The reason for the increased proportion of larvae on plants growing in rockwool requires further study. It may be caused by differences in plant biochemistry and/or structure caused by different growth and fertigation conditions. Alternatively, it may have to do with the fact that the size of the adult population was higher in cages in peat beds. This may have resulted in increased competition between adults for feeding and egg-laying places on the leaves, with subsequent reduced reproductive success through decreased number of eggs per female or increased mortality of eggs or small larvae on roses grown in peat.

The differential pupation conditions in peat and rockwool beds may have significant consequences for the success of biocontrol of WFT in cut roses in terms of the number of prey available for predators and the location of the prey items in the substrate. These differences may be coupled with the differential success of predator mite establishment and survival in the heterogeneous substrate comprised of rockwool slabs in bags as compared to the more homogeneous open peat beds.

Based on the declining numbers of *H. miles* in pots in treatments where *H. miles* and *A. cucumeris* were used together, we conclude that in this treatment the major part of the control success was caused by *A. cucumeris*. When the soil dwelling *H. miles* is used simultaneously with the crop-dwelling *A. cucumeris* larvae are removed that would otherwise subsequently pupate, the former species therefore suffers from indirect food competition and its numbers decline with time. However, when *H. miles* is used alone in appropriate numbers, it can control a considerable proportion of the thrips population. This suggests that if the numbers of *Hypoaspis* in the soil could be kept above a certain threshold throughout the growing season, both predator species could contribute to control and the control efficacy might be improved.

Studies on the usefulness and economic feasibility of repeated releases of *Hypoaspis* in combination with *A. cucumeris* is the next logical step following from our results.

Experiments are currently being conducted by us in cut roses to link the differential biology of WFT in peat and rockwool with the success of biocontrol using different release strategies of *Hypoaspis* spp. and *A. cucumeris*.

ACKNOWLEDGEMENTS

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Novel strategies for improving biological control of western flower thrips on protected ornamentals - potential new biological control agents

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ABSTRACT

Three natural enemies were evaluated for their potential in supplementing current methods for biological control of western flower thrips (WFT), *Frankliniella occidentalis*, on protected ornamentals. Adults of the common flower bug, *Anthocoris nemorum*, predated on one adult WFT per day or on 28 second-instar WFT larvae in the laboratory. *A. nemorum* nymphs and lacewing larvae, *Chrysoperla carnea*, each predated on two adult WFT per day or on 24 or 36 larvae respectively. The predation rate of *C. carnea* was reduced when the larvae had to search for WFT on whole verbena plants. Entomopathogenic nematodes, *Steinernema feltiae*, gave a significant reduction in numbers of WFT larvae on verbena leaves in the laboratory and of WFT adults on ivy-leaf geraniums in the glasshouse. Each of the three natural enemies have potential for use in a novel 'push-pull' strategy for improving biological control of WFT.

INTRODUCTION

The commercial uptake of biological control methods within Integrated Pest Management (IPM) on protected ornamentals is increasing, because of problems with pesticide resistance and increasing pressures to reduce pesticide usage. Western flower thrips (WFT), *Frankliniella occidentalis*, is the major pest on many ornamental species and biological control relies heavily on the use of the predatory mite *Amblyseius cucumeris*. This gives good control on a wide range of ornamentals, but predated only on thrips larvae so control is not maintained if adult WFT increase in number in the flowers. More robust biological control programmes are needed, incorporating supplementary natural enemies, to reduce the need for pesticides which disrupt IPM, and to encourage further uptake of IPM on ornamentals.

The common flower bug, *Anthocoris nemorum*, is not yet commercially available but is common in the UK. It feeds on a range of prey including WFT (Bunte *et al.*, 1990) and has shown potential for the control of WFT on chrysanthemums (Wardlow, unpublished data) and on cucumbers (Jacobson, 1991). Lacewing larvae, *Chrysoperla carnea*, are commercially available for aphid control but are not widely used in the UK and, although they are polyphagous predators, little data is available on their efficacy against thrips (Loomans *et al.*, 1995). Entomopathogenic nematodes are widely used on ornamentals for the control of vine weevil and sciarid fly larvae in compost, and have been shown to be effective against WFT pupae in compost (Helyer *et al.*, 1995). Little data is available on the potential of

entomopathogenic nematodes against thrips adults or larvae, although they can control other foliar pests e.g. leaf miners (Williams & Macdonald, 1995).

Commercial use of supplementary biological control agents could be more cost-effective if used on selective 'lure' plants in the glasshouse, chosen for their attractiveness to WFT. Verbena flowers and their volatiles have been shown to attract WFT adults (Pow *et al.*, 1998) and to be potential 'lure' plants in other ornamental crops (Bennison *et al.*, 1998). This paper presents the results of efficacy tests on three natural enemies against WFT on verbena and other ornamental species attractive to WFT, to assess their potential in a novel 'push-pull' strategy for improving biological control on ornamentals.

MATERIALS AND METHODS

Rearing methods and sources of natural enemies

WFT were reared on French bean pods, *Phaseolus vulgaris*, using a method adapted from that reported by other workers (Steiner *et al.*, 1995), to provide synchronised larvae or adults of the same age. *A. nemorum* were reared on French beans, using a method adapted from others reported (Parker, 1981). *C. carnea* were supplied as second-instar larvae by Koppert UK. *S. feltiae* ('Nemasys') were supplied by MicroBio.

Predation rates of *A. nemorum* and *C. carnea* on WFT adults

Verbena 'Sissinghurst Pink' was used as the host plant for all the predation experiments. Three verbena stems, approximately 10 cm long, each with leaves and the central one with an open flower, were placed into a cube of damp 'Oasis'. The simulated 'plant' was placed into a perspex box, 27 x 16 x 9 cm, provided with ventilation holes screened with fine mesh, ensuring that no part of the 'plant' touched the sides of the box. One predator, either an *A. nemorum* adult, a third-instar *A. nemorum* nymph, or a *C. carnea* larva, was placed onto the verbena flower. Five WFT adult females were added to the box and the lid sealed with sticky tape to prevent thrips escape. Ten replicates were set up for each of the three predators and for controls without predators. The boxes were incubated at 21°C with a 16:8 hours light:dark (L:D) photoperiod. After 24 hours, the contents of the boxes were searched carefully and numbers of live and predated thrips were recorded. Predated thrips adults were identified as empty carcasses left after either *A. nemorum* or *C. carnea* had fed on the thrips by sucking out the body contents.

Predation rates of *A. nemorum* and *C. carnea* on WFT larvae

Three verbena leaves, each infested with 20 WFT second instar larvae (L2s) were placed on squares of dampened filter paper in a plastic tube, 6 x 4 cm, giving 60 larvae per tube. One predator was added to each tube, which was sealed with a screw-top lid with a screened ventilation hole. Four replicates were set up for each of the *A. nemorum* adults and third-instar nymphs, *C. carnea* larvae and controls, without predators. The tubes were incubated on their sides at 21°C, 16:8 hours L:D. After 24 hours, the leaves and tubes were examined under a binocular microscope for live, dead or predated thrips larvae. Larvae predated by *A.*

nemorum adults or nymphs were identified as shrivelled empty cuticles, but no evidence of thrips larval carcasses were left by *C. carnea*.

Searching ability of *C. carnea* for WFT larvae

Twenty WFT L2s were placed on the growing tips of four stems of a verbena plant; five larvae on each stem, with each stem separated by an uninfested stem. One *C. carnea* larva was placed on the growing tip of one infested stem. Four replicate plants were set up, and four control plants without lacewings. The plants were placed into large white plastic trays with water and one drop of 'Teepol' added, to catch any thrips larvae or lacewing larvae falling from the plants. The trays were placed into insect rearing cages at 21°C and 16:8 hours L:D. After 24 hours, the plants and water were searched for thrips larvae. The infested stems were detached and examined under a binocular microscope and then washed in 70% alcohol. The remainder of the plant was shaken over a white plastic tray, then detached at the base of the stem and washed in alcohol. Numbers of live thrips larvae per plant were recorded.

Efficacy of *S. feltiae* against WFT larvae the laboratory

Ten WFT L2s were placed on the underside of a verbena leaf, attached to a short piece of stem. The stem was placed into a damp cube of 'Oasis' and both sides of the leaf sprayed with a suspension of *S. feltiae* at 5,000 per ml water, using a hand held sprayer. A fine spray was used, to give good coverage but not run off. Four replicates were sprayed with *S. feltiae* and four sprayed with water as controls. After treatment, each 'plant' was placed inside a plastic tube, 6 x 4 cm, with the screened ventilation hole in the lid taped over for the first night, to increase r.h. and maintain leaf wetness to enhance nematode survival and host location (Williams & Macdonald, 1995). The tubes were incubated overnight in the dark at 21°C. The next morning, the tape was removed from the ventilation hole in the lid of each tube and the tubes were incubated for two more days at 18:6 hours L:D. Three days after treatment, numbers of live and dead thrips larvae were recorded. All thrips recovered were dissected in a drop of water under a binocular microscope and the number parasitised by nematodes recorded.

Efficacy of *S. feltiae* against WFT in experimental glasshouses

Two glasshouse compartments were used for the experiment, each containing eight plots of ten ivy-leaf geranium plants infested with WFT. A pre-treatment assessment of numbers of thrips adults and larvae per plant was made by shaking each plant over a white plastic tray and counting dislodged thrips, which were then returned to the plant. In one compartment, all plants were sprayed with *S. feltiae* at 5,000 per ml water and in the other compartment, all plants were sprayed with water as a control. Sprays were applied to give good coverage but not run off. Immediately after spraying, four of the plots in each compartment were covered with black polythene overnight and the other four were left at ambient glasshouse humidity, natural daylight and photoperiod. The polythene was removed the next morning and the r.h. was recorded overnight both under the polythene and in the glasshouse. The glasshouse temperature was set at 18°C night, venting at 21°C day. After three days, numbers of live thrips adults and larvae per plant were recorded.

RESULTS

Analysis of all data on predation rates and searching ability was carried out using a Generalised Linear Model, treating the data as binomial. When offered five WFT adults on a simulated verbena plant, each of the three predators significantly reduced the numbers of live WFT 24 hours later when compared with the controls ($P < 0.001$). A consistent number of thrips were missing from each treatment, i.e. not all the thrips could be accounted for by the presence of live, dead or predated thrips, therefore the data was adjusted to estimate the total numbers of predated thrips. *A. nemorum* nymphs and *C. carnea* larvae each predated on a mean of 2.14 adults per day (SE 0.382, 18 df) and *A. nemorum* adults predated on a mean of 1.1 adult per day (SE 0.06, 27 df).

When offered 60 WFT larvae on verbena leaves in a tube, each of the three predators significantly reduced the numbers of live thrips larvae 24 hours later, when compared with the controls ($P < 0.001$). As no evidence was left of thrips larvae predated on by *C. carnea*, the numbers eaten by the three predators were estimated by adjusting the numbers of 'missing' thrips, as above. *C. carnea* predated on a mean of 36 larvae per day, which was significantly more than the 24 predated on by *A. nemorum* nymphs, but similar to the 28 predated on by *A. nemorum* adults (SE 1.8, 12 df).

In the *C. carnea* searching experiment on the verbena plants infested with 20 WFT larvae, *C. carnea* significantly reduced the numbers of larvae detected after 24 hours ($P < 0.001$). Mean numbers of larvae were 11 on control plants (SE 1.112, 6 df) and 2.5 on plants treated with one lacewing larva (SE 0.736, 6 df).

The data on efficacy of *S. feltiae* against WFT were tested using analysis of variance. *S. feltiae* significantly reduced numbers of live WFT L2s in the laboratory experiment. Mean numbers of live larvae detected after three days were 6.0 on leaves treated with water (SE 0.774, 6 df) and 1.75 on leaves treated with *S. feltiae* (S.E. 0.601, 6 df). WFT larvae killed by *S. feltiae* appeared as yellow-brown, dried and shrivelled corpses stuck to the leaf. Two of the nine dead larvae and one of the seven live larvae found after treatment with *S. feltiae* contained nematodes when dissected.

In the glasshouse experiment, three days after treatment, mean numbers of WFT adults per plant were significantly lower on plants treated with *S. feltiae* than on those treated with water ($P < 0.001$), and covering the plants with black polythene overnight gave a further significant reduction in numbers of WFT in both nematode and water treatments (Fig. 1). Maximum and minimum r.h. recorded overnight after treatment were 73% and 50% ambient and 78% and 72% under the polythene.

Mean numbers of WFT adults per plant were similar in both compartments before treatment; 6.95 and 6.92 in plots to be treated with water and covered with polythene or left uncovered respectively, and 4.75 and 5.55 in plots to be treated with *S. feltiae* and covered or uncovered respectively (S.E. 1.182, 12 df, N.S.). Three days after treatment, in covered and uncovered plots respectively, water had led to a 28% and 130% increase in WFT numbers, whereas treatment with *S. feltiae* led to an 18% and a 14% decrease.

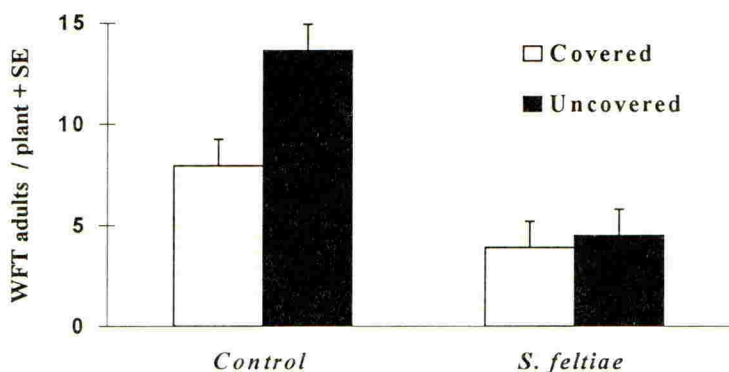


Figure 1. Mean numbers of WFT adults per ivy-leaf geranium 3 days after treatment with water or *S. feltiae* in the glasshouse.

DISCUSSION

The results presented here demonstrate that *C. carnea* were effective predators of both WFT larvae and adults and that daily predation rates on larvae were lower on whole plants than on detached verbena leaves, due to the time spent searching for prey. As *C. carnea* adults are not predatory, inundative releases of larvae would be needed for control in glasshouses and these would be more cost-effective if made only to 'lure' plants attractive to WFT. Current experiments are investigating control of WFT using weekly releases of *C. carnea* to verbena 'lure' plants placed amongst ivy-leaf geraniums.

Bunte *et al.* (1990) reported that *A. nemorum* adults predated on 3.7 adult WFT or 23.2 L2s per day, when offered 20 adult or 40 L2s on French bean leaf discs at 25°C, while here they predated on 1.1 and 28 respectively. The differences between the two sets of data could be due to the difference in temperature, numbers of prey offered and host plant material used. Bunte *et al.* (1990) did not report predation rates of *A. nemorum* nymphs but these results demonstrate that both adults and nymphs are effective predators of WFT adults and larvae. *A. nemorum* could be released inoculatively to 'lure' plants in protected ornamentals, from where adult predators could disperse to the rest of the crop. Future research is planned on using this approach in a 'push-pull' strategy, but using another anthocorid species, *Orius laevigatus*, which is commercially available for thrips control and has given promising initial results on ornamentals (Bertaux, 1993).

The results demonstrate that *S. feltiae* can kill WFT larvae on leaves under optimal laboratory conditions and can reduce WFT numbers when applied in the glasshouse at ambient r.h. Control was slightly enhanced by increasing the r.h. by covering the plants overnight with polythene. The comparative increase in the numbers of WFT adults three days after spraying with water in the glasshouse was probably due to WFT pupae hatching into adults after application. The decrease in numbers of WFT adults after spraying with *S. feltiae* may have been due to mortality of both WFT adults and pupae; efficacy against WFT pupae has been reported previously (Helyer *et al.*, 1995). The rates of *S. feltiae* used here are much higher

than those currently used against other pests in compost, but application could be cost-effective if made only to 'lure' plants in the glasshouse.

Future work is planned which will further investigate the interactions between ornamental host plants, WFT and candidate natural enemies for use in a novel 'push-pull' strategy which will aim to manipulate the movement of WFT in glasshouses, for improved, cost-effective biological control on ornamental crops.

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Pest risk analysis to support and strengthen legislative control of a quarantine thrips: the case of *Thrips palmi*

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ABSTRACT

Analysis of pest risk is now required by the World Trade Organisation to justify phytosanitary measures applied against pests. This paper summarises the key points from a risk analysis of *Thrips palmi*, a plant pest which has spread internationally in recent years. The risks of establishment and economic damage were assessed using a newly developed, internationally agreed, semi-quantitative risk assessment system. Although intercepted on a variety of commodities from different regions of the world, the majority of European *T. palmi* interceptions occur on orchid cut flowers imported from SE Asia. If introduced to protected horticultural cultivation in northern Europe, *T. palmi* would pose a threat to growers of cucumbers, peppers, aubergines and ornamentals, such as chrysanthemums. Following the risk analysis, appropriate phytosanitary measures were enacted by European Community legislation to control the entry of *T. palmi* on orchid cut flowers from SE Asia.

INTRODUCTION**Pest risk analysis**

With the expansion of trade in plant commodities, the probability of introducing plant pests into new geographical areas has greatly increased. In recent years, new pest species have increasingly been introduced into western European glasshouses (Frey, 1993). To prevent the introduction and establishment of pests in new areas, phytosanitary measures are applied to commodities in trade pathways that have been identified as a potential risk. These measures should be designed to reduce the perceived risk to an acceptable level, without providing an unjustified barrier to trade. Pest risk analysis (PRA) is the process of evaluating biological or other scientific and economic evidence to determine whether a pest should be regulated and the strength of any phytosanitary measures to be taken against it (FAO, 1997). Such analyses are now required by the World Trade Organisation to justify phytosanitary measures applied to trade pathways and are part of the processes of cultural and legislative control which is used strategically to tackle plant pests at an international level.

Thrips palmi

Originating in SE Asia, *Thrips palmi*, the palm or melon thrips, spread to India in the 1960s. During the past two decades, it has spread to Africa, Hawaii, the Caribbean, Japan, Australia, South America and Florida. It is a polyphagous plant pest which has been recorded on hosts in 36 plant families including many of horticultural interest, especially the

Cucurbitaceae and Solanaceae. At 18°C, eggs can develop into adults within 30 days (Tsai *et al.*, 1995). Due to the short time required for development, heavy infestations can build up rapidly. Damage to hosts can be severe with adults transmitting viruses and larvae feeding on leaves, near the growing tips of stems and on the surfaces of fruits where scarring is a symptom of damage.

If *T. palmi* did become established in Europe, host crops such as cucumbers, sweet peppers and aubergine grown both in protected cultivation and outdoors in the south would be at risk (CABI / EPPO, 1997). The EC phytosanitary safeguards to prevent entry of this pest were limited to designation as a quarantine organism for all of the EC, together with simple requirements of place of production freedom or pre-export treatments for plants. However, Bartlett (1993) considered that this was not enough, and since then interceptions have steadily increased. Thirty-five outbreaks have been reported from The Netherlands and all have been successfully eradicated (Vierbergen, 1996). The pathway via which most of these *T. palmi* were entering was not efficiently regulated by this legislation. As a consequence the UK authorities undertook to re-examine the PRA for this organism with these new pathways in mind. This paper summarises the key findings from this assessment and the outcome.

MATERIALS AND METHODS

A recently developed semi-quantitative pest risk assessment scheme (EPPO, in press), was used to analyse the risk that *T. palmi* poses to a sector of protected cultivation in England and Wales. The first section of the scheme consists of a series of questions requiring simple "yes" or "no" responses and which are designed to determine whether the organism has potential quarantine pest status. If it does, the assessment moves to the second section of the scheme which is semi-quantitative. Scores from "1" to "9" are given to 49 detailed questions, in three stages, regarding the potential for entry, establishment and economic damage within the area concerned. Low scores represent unlikely or low impact events, high scores represent the opposite. The information used to support the scores given to each question was sourced from published scientific papers and available trade statistics. Upon completion, mean scores for entry, establishment and potential for economic damage were calculated. Such scores are designed to be used in a comparative manner in order to determine the relative importance of different pathways and pests. Quantitative scores are not provided since their interpretation would be difficult without providing examples from other pathways or pest species. Risk management is not presently included in the EPPO risk assessment scheme, but is the last component of PRA. The risk management strategy implemented following the risk assessment is described.

RESULTS

Stage 1 - Potential for entry

By studying the pathways on which interceptions are found, those which present a significant risk can be identified and measures taken to mitigate the risk they pose. The majority of *T. palmi* interceptions in the EC have been on Orchidaceae (L'Her & Roberts,

1998). However, significant interceptions have also been made on Solanaceae and Cucurbitaceae (Table 1). Overall, the greatest proportion of *T. palmi* interceptions have been found on commodities imported from SE Asia (Figure 1).

This assessment focused on the risk posed by orchid cut flowers from SE Asia, conservatively valued at \$25m per annum (Anon., 1997), because at the time of analysis no pre-export inspections were required under EC plant health legislation.

Table 1. The annual number of *Thrips palmi* interceptions by host plant family in the EC from 1995 to 1997.

Host plant family	Year		
	1995 No.	1996 No.	1997 No.
Orchidaceae	13	48	107
Solanaceae	6	12	15
Cucurbitaceae	6	6	29
others	0	2	11
Totals	25	68	162

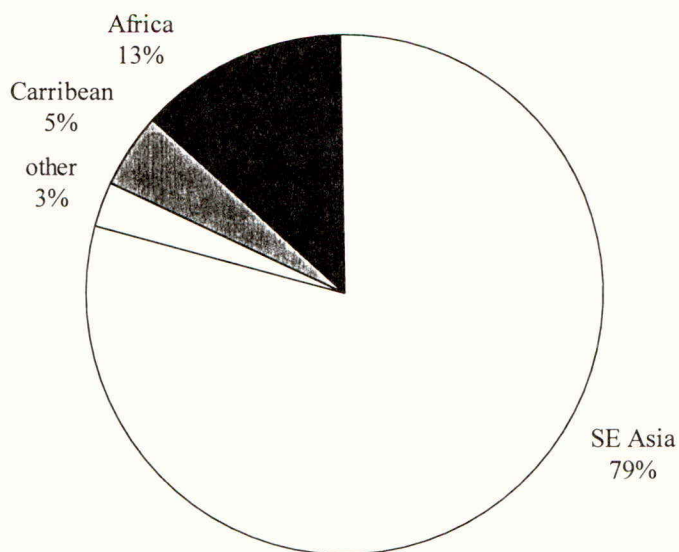


Figure 1. Regions where EC interceptions of *T. palmi* have originated (1995–1997).

Stage 2 - Potential for establishment

Presently, outdoor populations of *T. palmi* are restricted to tropical or semi-tropical regions and previous knowledge of outbreaks in protected cultivation in Japan and The Netherlands suggests that any northern European outbreaks of *T. palmi* are most likely to occur in, and be restricted to, protected cultivation.

One possible scenario for UK outbreaks is the delivery of infested orchids to a glasshouse nursery containing suitable hosts. In 1995, almost 100,000 tonnes of cucumbers, sweet peppers and aubergines were grown under 255 ha of glass, while suitable ornamental hosts (chrysanthemums and carnations), worth £33 million, were grown under 250 ha of glass (MAFF, 1996). The total area of potential host crops represents 18.8% of the total protected horticultural area of England and Wales. Thus a significant proportion of the horticultural industry is threatened by *T. palmi*. This analysis excludes tomatoes because there is conflicting evidence in the literature about the exact risk to them.

Studies on the development of *T. palmi* on different hosts under a variety of temperatures, have shown that the lower threshold for egg development is 7.4°C (Jiajiao *et al.*, 1995). Population development occurs optimally between 24 and 27°C (Tsai *et al.*, 1995). Although sub-optimal, the temperature regime recommended for growing cucumbers (19-22°C) in UK glasshouses (Anon. 1980) would be suitable for the development of *T. palmi* and adults could develop from eggs within 17 to 27 days.

A previously introduced thrips pest of protected cultivation, *Frankliniella occidentalis* is estimated to have a threshold temperature for overall development of 7.9°C (McDonald *et al.*, 1997), which is similar to that of *T. palmi*. However, difficulties experienced when attempting to eradicate *F. occidentalis* are unlikely to be as significant with *T. palmi* due to its lower rate of dispersal (Vierbergen, 1996). Nicotine shreds, deltamethrin, predatory *Amblyseius* mites and *Orius* bugs are already used against *F. occidentalis*, in crops threatened by *T. palmi* and such measures would assist in the inhibition of *T. palmi* establishment and spread in glasshouses.

Stage 3 - Potential to cause economic impact

Although most *T. palmi* interceptions were found on orchids, the largest crop perceived to be at risk in England and Wales is cucumbers. The economic impact section of the PRA therefore concentrated on cucumbers. Aubergines and sweet peppers are also at risk although they are grown less widely in the UK.

T. palmi mainly infests the leaves of cucumber, with only a small proportion attacking flowers or fruit. *T. palmi* can also act as a vector of groundnut bud necrosis tospovirus (GBNV) and watermelon silvery mottle tospovirus (WSMV). These viruses can infect cucumbers and tomatoes experimentally, although the economic consequences from natural infection are unknown. Plants attacked by *T. palmi* show a loss in vigour and have fewer flowers. Annual outbreaks of *T. palmi* in Japan cause severe damage to cucurbits, although specific reports of economic damage are lacking.

A spreadsheet financial model was developed from the financial budget of a standard cucumber producer (ADAS, 1994). Variables within the model included cucumber yield and quality and variable costs such as pest control. Variables could be altered and the consequent effect on sales and gross margin were calculated automatically. In a scenario where *T. palmi* caused yield losses of 10% over the season (as seen in experiments by Kawai, 1986) and control costs doubled, a reduction in gross margin of about 25% (£3,000 per 0.1 ha) could be expected.

In addition to edible crops, ornamentals are also at risk from *T. palmi*. Damage symptoms in the form of leaf bronzing on ornamental hosts such as chrysanthemums and carnations, would reduce the quality and hence the value of ornamentals attacked.

Risk management

Thrips palmi is difficult to detect: adults are small, eggs are laid inside plant tissue and pupae occur in the soil. Females may lay up to 200 eggs and resistance has been reported to many insecticides (Kawai, 1990). Since such factors may allow rapid population build up, it is desirable to implement measures which prevent the entry of the pest rather than those which are designed to control or eradicate it on arrival. Kajita *et al.*, (1992) found *T. palmi* to be common during surveys of Thai orchid nurseries. It is therefore appropriate to target phytosanitary measures against orchid producing nurseries.

After receiving official notice of *T. palmi* interceptions by the EC, the Thai Ministry of Agriculture organised meetings for orchid exporters during April and May 1997. At these meetings, instructions on the correct use and dose of fumigants for control of pests on orchids was given. The Thai Agricultural Regulatory Division also organised fumigation training sessions for orchid exporters.

New EC measures were transposed into UK legislation in February 1998 (Anon., 1998), stipulating that cut flowers of Orchidaceae originating in Thailand must either come from a place of production which has been found to be free from *T. palmi* during official inspections over the previous three months, or have been subjected to an appropriate fumigation treatment prior to export to ensure freedom from Thysanoptera. These phytosanitary measures were developed in co-operation with the Thai authorities and are not regarded as particularly onerous or as unjustifiable barriers to trade, but they do provide safeguards against transport of a potentially serious pest into the UK horticultural industry.

CONCLUSIONS

Thrips palmi has spread in the past and will probably continue to increase its geographic distribution outdoors in tropical regions, e.g. Central and South America and Africa. However, European glasshouse crops and some outdoor crops in the south remain at risk.

Partly as a result of this PRA, improved phytosanitary safeguards have been justified and enacted which aim to prevent entry of *T. palmi* on orchid cut flowers, the trade pathway recognised as posing a high risk to the EC and UK. The trade in cut flowers continues, with

phytosanitary measures in place. Continued monitoring of orchids from SE Asia will show whether the measures applied provide an appropriate level of protection.

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