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# **FINAL REPORT**

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To:  
Horticultural Development Council  
Bradbourne House  
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East Malling  
Kent  
ME19 6DZ

**Tomato: Further development of  
sustainable mealybug control strategies**

January 2007

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**Commercial – In Confidence**

The results and conclusions in this report are based on a series of carefully monitored applied studies in experimental facilities and large-scale commercial glasshouses. The conditions under which the studies were carried out and the results have been reported with detail and 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 the interpretation of the results especially if they are used as the basis for commercial product recommendations.

**Authentication**

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

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Report: Final report

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## GROWER SECTION

### HEADLINES

- A sustainable IPM programme was designed for use against obscure mealybug (*Pseudococcus viburni*) on organic tomato and pepper crops but did not provide adequate control of the mealybug population in the commercial tomato crop and more effective control measures are required.
- Monitoring crop invasion and establishment of mealybugs on young plants enabled early season control measures to be timed more accurately.
- Two species of parasitoids (*Leptomastix epona* and *Pseudaphycus maculipennis*) were shown to be capable of locating, attacking and completing their development in most life cycle stages of *P. viburni* on both tomato and pepper plants but in the commercial tomato crop both species of parasitoid failed to sustain high levels of parasitism.
- Eradicoat T offered the best potential for a second line of defence treatment in an organic tomato crop, but was commercially unviable.
- The insect growth regulator, buprofezin (Applaud), remains the most effective second line of defence for conventional crops.

### BACKGROUND AND EXPECTED DELIVERABLES

Mealybugs belong to the insect family, Homoptera, which also includes aphids, whiteflies and scale insects. They are one of the most significant pest groups, with over 3000 species known to feed on a wide range of plant families in habitats varying from the soil to tree tops. HDC funded studies have been prompted by an increase in the incidence of the obscure mealybug (*Pseudococcus viburni*) on protected tomato crops in the UK. In addition to tomatoes, it has been recorded in the UK on other edible crops, including peppers, and on glasshouse-grown ornamental plants.

A previous HDC project (PC 161), which was completed in 2002, investigated the increase in incidence of infestations of *P. viburni* in UK tomato crops and began to formulate a control strategy. The most effective and IPM compatible method of controlling mealybugs on tomato plants during the production season was shown to be the insect growth regulator, buprofezin (Applaud), but this could not be applied to organic crops. Furthermore, this dependence on a single chemical insecticide was not consistent with the TGA's long term goal of pesticide-free crop production.

To become sustainable, the control strategy required a biological component that would act continuously throughout the growing season. Project PC 161 evaluated several biological control agents (eg *Hypoaspis* spp. and *Beauveria bassiana*) but none proved to be very promising. However, preliminary investigations indicated that the parasitoids, *Leptomastix epona* and *Pseudaphycus maculipennis* had potential to fulfil this role. *Leptomastix epona* could be released in UK crops but *P. maculipennis* was not indigenous and therefore required a licence before it could be used in trials in glasshouses.

The overall aim of this project was to formulate sustainable strategies, based on a combination of new and existing control measures, for the control of *P. viburni* on UK tomato and pepper crops. The specific objectives were:

1. To develop a robust biological control measure based on parasitoids.
2. To construct sustainable mealybug control strategies tailored to the specific requirements of conventional and organic tomato and pepper crops.
3. To test and refine the control strategies in commercial crops.

## SUMMARY OF WORK PRIOR TO 2006

It was considered highly unlikely that any one sustainable control measure would be successful against *P. viburni* on commercial crops. Therefore, a programme was designed consisting of a suite of compatible control measures that could be used to combat the pest at four distinct stages throughout the growing season:

- Survival on the glasshouse structure between crops. These control measures were developed in Project PC 161.
- Initial crop invasion by overwintered survivors.
- Season-long suppression of mealybug population growth.
- A compatible second line of defence.

### Initial crop invasion by overwintered survivors:

It had been shown that mealybug eggs on the structure of the glasshouse hatch within three weeks of the glasshouse being heated and they quickly migrate to the new plants. A 2% dilution of Savona had been effective against first instar *P. viburni* as they colonised tomato plants, reducing numbers by up to 93%. However, there was some concern over the effect of this product on young plants. Alternative products were tested in this project in 2006.

### Season-long suppression of mealybug population growth:

Much of the work in the early stages of this project was aimed at developing a biological control measure for *P. viburni* that would provide season-long suppression of the pest's population growth. A series of laboratory-based experiments provided important information about the key biological parameters that governed the ability of three species of parasitoids (*Leptomastix epona*, *Anagyrus pseudococci* and *Pseudaphycus maculipennis*) to successfully locate, attack and complete their development in the mealybugs on tomato and pepper plants. There appeared to be no fundamental reasons why these parasitoids would not become established in populations of *P. viburni* on both types of plants. *Anagyrus pseudococci* was the weakest candidate due to its poorer performance at the temperatures that are common in crops in the early season. The stronger candidates, *L. epona* and *P. maculipennis*, were tested in larger scale experiments in 2005.

*Leptomastix epona* could be released in UK crops but *P. maculipennis* was a non-native species and did not have official approval. As part of this project, an application for a licence to release *P. maculipennis* in the UK was successfully obtained from Defra.

The studies in the second year of the project consisted of detailed examinations of the population growth of *L. epona* and *P. maculipennis* against that of their host, *P. viburni*, within experimental glasshouse crops at STC RF. The licence for release of *P. maculipennis* was not issued in time for the larger scale trials and so only *L. epona* could be evaluated in the commercial tomato and pepper crops in 2005.

*Tomatoes:*

In contained glasshouse tomato trials at STC RF, results showed that 80-90% parasitism was achieved from a single release of either *P. maculipennis* or a combination of *P. maculipennis* / *L. epona*. A maximum level of 50% was achieved by *L. epona* by itself. While both species could have the potential to become components in a mealybug control programme for tomato crops, *P. maculipennis* appeared to show the greatest promise. However, it was several weeks before these parasitoids began to reduce the pest populations and there remained doubts as to whether these control measures would be successful in commercial crops.

In commercial tomato crops *L. epona* became established in the mealybug population on plants following four weekly releases at the rate of two parasitoids per m<sup>2</sup> and achieved a level of 15% parasitism 70 days after the first release.

#### *Peppers:*

The parasitoids did not perform so well in the contained glasshouse pepper trials at STC RF. In this case, a maximum of 40% parasitism was achieved by *P. maculipennis* but no parasitism was detected by *L. epona* throughout the trial. The differences in the results between tomatoes and peppers may be explained by the habitats occupied by *P. viburni* on these crops. On tomato plants, the mealybugs are situated primarily on the lower stems while on peppers they are usually found beneath the calyx of the fruits. It would appear that the parasitoids are more successful when attacking mealybugs situated on the open stems than when the pests are tightly encrypted beneath the calyx of the peppers. This may simply be because the parasitoids are unable to locate their hosts.

Observations of *L. epona* within a commercial pepper crop recorded a level of 23% parasitism 77 days after the first release of the parasitoids. In both cases, the speed of establishment was not adequate to bring the pest population under control.

The generation time for *P. viburni* is slow, taking 50 days from egg to adult at 21±2°C. This provides an advantage to the parasitoids, which complete their development in about the half the time. However, the pest has very high fecundity rates (adult female *P. viburni* can produce up to 500 eggs in its egg sac), which means its population can continue to grow rapidly while the parasitoids are becoming established. There was insufficient resource within this project to allow more detailed studies of the population dynamics within this biological system but we believed that the parasitoids would have to be released in the early part of the growing season while pest numbers are still relatively small. However, there were still concerns over the parasitoids' ability to control *P. viburni* at the population level due to the huge numbers of offspring produced by the pest.

#### **A compatible second line of defence:**

This component was required for use mid-season to redress the balance between pest and parasitoid should control with the primary control agent falter. Applaud remains an effective second line of defence for conventional crops but an alternative was required for organic crops. The effect of Savona on first instar larvae is mentioned above. This product had also been shown to have a direct effect on other life cycle stages on the plant but the results had been variable; 2% and 4% dilutions giving 30-60% and 40-100% control respectively. Control would therefore be dependant on a series of applications.

Some UK growers who used the fungal pathogen, *Verticillium lecanii*, against glasshouse whiteflies, reported incidental control of mealybugs but this had not been confirmed in controlled experiments. Laboratory bioassays done under ideal conditions showed that the pathogen reduced numbers of first instar nymphs but only by about 10%. It was presumed that the mealybugs were protected from infection by their waxy covering.

However, it was hypothesised that the effect of the fungus could be enhanced by applying it after Savona because the soap would breakdown the waxy protection and allow more fungal spores to come into contact with the insect's body. Preliminary results were variable but 100% mortality of mealybugs was achieved in some laboratory experiments. The studies were scaled up to a commercial tomato crop in 2004. These results showed only a small advantage in using Mycotal in addition to Savona over three applications at approximately seven day intervals. This was contrary to the previous laboratory scale studies and was attributed to the difficulty in obtaining good spray cover among the horizontal spray bundles.

The studies were repeated in 2005 with Eradicoat T incorporated in the trial. This was a new formulation of a starch-based material with a physical mode of action. All three treatments (*i.e.* 4% Savona, 4% Savona plus 0.1% Mycotal, Eradicoat T) reduced the numbers of mealybugs, whereas the numbers of mealybugs increased in the untreated controls. There was no apparent difference between the Savona and Savona plus Mycotal treatments (except in the number of viable egg sacs) thus indicating that there had been no additional effect by the fungus. This was supported by the fact that no fungal growth was evident on the dead mealybugs on the plants, nor did any develop when the cadavers were removed and incubated under more ideal conditions in the laboratory. Eradicoat T performed significantly better than the Savona on all development stages of the mealybug and currently offers the best potential for a second line of defence treatment in organic tomato crops. However, the effectiveness will always depend on the contact of the product and the pest.

## **SUMMARY OF WORK COMPLETED IN 2006**

### **Mealybug crop invasion studies**

This work was completed in an organic tomato crop (cv Capri). The plants were stood out on plastic covered soil when delivered from the propagators in week 51 2005 and planted into the soil in week 4 2006. The presence of mealybugs was first noted on plants in the first week of January 2006 with the majority being young nymphs (instars 1 / 2). At this stage, there were up to 50 per stem in the monitored area with even larger numbers in other specific areas. Mealybugs were also present on volunteer tomato seedlings that had germinated beneath the plastic. A second flush of invaders reached the plants between the assessments in weeks 4 and 5 2006 (*i.e.* 5-6 weeks into the crop), which followed the plastic being opened up for planting in the soil. Adults and egg masses were first seen in significant numbers from week 9 2006 (crop week 10).

### **Control of mealybug crop invaders**



The intention of the spray programme was to reduce numbers of mealybugs to a level that could be managed with a more sustainable control measure based on parasitoids. Three treatments (4% Savona, 1.5% Eradicoat T and 3% Eradicoat T) were compared to untreated controls. The original programme of 2-3 sprays was extended to control the second flush of mealybugs that emerged from below the floor plastic and to compensate for poor coverage of early applications caused by the lower leaves shielding the stems. A total of seven sprays were applied before crop week 10 when the surviving mealybugs began to form egg masses. At that time, there was no difference between the three treatments and numbers were reduced to approximately 4% of the untreated controls. There was no evidence of acute phytotoxicity from sprays in any treatments.

Although this spray programme was considered adequate to pave the way for the more sustainable control strategy based on parasitoids, the number of sprays applied would have to be reduced in the future by improving the efficacy of each application. It was anticipated that this could be achieved by using a different deleafing strategy and improved spray equipment.

### **Evaluation of a season-long mealybug control programme**

A lot of consideration was given to how the parasitoids should be released into the crop. Ideally, we would have followed a programme of routine releases of large numbers of parasitoids throughout the crop starting at the beginning of the growing season. Initial studies showed that *L. epona* and *P. maculipennis* were active and would produce offspring in conditions (ie. Short day length), akin to those experienced in the glasshouses.

The routine releases of large numbers of parasitoids throughout a crop starting at the beginning of the growing season is a very successful approach for *Encarsia formosa* against glasshouse whitefly, and is possible because the parasitoids are inexpensive (£2-£3 per thousand). However due to the early development of the commercial supply, *L. epona* and *P. maculipennis* are considerably more expensive to produce and it was necessary to devise a completely different strategy to keep the cost within sensible parameters. It was proposed that we should establish intensive breeding areas (IBAs) in the early season from which the parasitoids would disperse as conditions became more suitable.

Parasitoids were released in the IBA every week from weeks 10 to 19 (*i.e.* 10 releases) and every two weeks from weeks 20 to 24 (*i.e.* 2 releases). On each occasion, the releases consisted of 200 *L. epona* and 800 *P. maculipennis*. Numbers of mealybugs and levels of parasitism were monitored within the IBA and at distances of 3m, 8m and 16m from the IBA.

Both species of parasitoid established quickly and appeared to be doing well (60% parasitism) until the first generation of mealybug eggs started to hatch in week 14. By week 17, this generation of mealybugs were reaching the third instar stage and the percentage parasitism was approximately 5% in the IBA with no apparent spread to the adjacent rows. From this point onwards, the mealybug population growth rapidly outstripped the parasitoids and the application of second line of defence treatments became necessary.

In fact, Eradicoat T was applied at approximately 2-3 week intervals throughout the remainder of the growing season. The mealybug numbers were suppressed to non damaging levels until late-August but the populations then grew even more rapidly and

this resulted in some plant loss from late-September onwards. Furthermore, the cost of the spray programme was unacceptable.

One of the main concerns after the previous stage of this project was whether these parasitoids would be able to suppress the rapid population growth of their mealybug host. The results in the commercial trial suggest that both species of parasitoid were unable to achieve a sustainable reduction in the mealybug populations. There seems little doubt that additional control measures against mealybugs are still required for organic crops.

### **Overall summary:**

Studies in another HDC funded project (PC 240) have shown that mealybugs are now the most difficult pest to control within the whole IPM programme for organic tomatoes. In fact, more than twenty IPM compatible control measures or combinations of control measures that are acceptable in organic production have now been evaluated against mealybugs. Many have shown potential when tested against individual mealybugs in the laboratory but have failed when scaled up in commercial crops. This is particularly true in the middle of the season when mealybugs are protected from treatments among the horizontal tomato stem bundles. Furthermore, the reproductive capacity of the survivors of any treatments is such that the population is soon replenished.

In conventional crops, the greatest success against mealybugs has been with the insect growth regulator, buprofezin, applied before mealybug invaders begin to lay eggs at the start of the season.

## **FINANCIAL BENEFITS TO GROWERS**

The cost of control measures applied against patchy infestations of mealybugs on two monitored nurseries throughout 2002 varied from £2,000 to £4,500 per hectare. This was comparable to estimates received in response to a grower survey in the late 1990s, which averaged £3,100 per hectare. Despite these intensive control measures, the growers still suffered financial losses due to mealybug damage. These losses have been difficult to quantify but one grower estimated them to be over £1,000 per hectare in 2005. The total cost of a mealybug infestation to a tomato business is therefore in excess of £4,000 per hectare.

The most effective control measures identified to date are heavily dependant on chemical insecticides. The development of successful biological or physical control against mealybugs will therefore provide another step towards the TGA's long-term goal of pesticide-free crop production. This will increase the desirability of TGA members' produce and strengthen their marketing position. Furthermore, the most effective existing control measures are not compatible with the standards that govern organic production. The development of successful biological control will therefore fill an important gap in organic growers' overall pest management armoury.

## **ACTION POINTS FOR GROWERS**

- Monitor and target early season invasions.
- Back up with second line of defence treatment such as buprofezin.

- Strategy against mealybugs in organic tomato crops must shift from control measures applied against established populations of the pest during the season to control measures that prevent the initial colonisation of the plants.

# **SCIENCE SECTION**

## **PART 1:**

### **GENERAL INTRODUCTION**

#### **Background:**

Mealybugs belong to the insect family, Homoptera, which also includes aphids, whiteflies and scale insects. They are one of the most significant pest groups, with over 3000 species known to feed on a wide range of plant families in habitats varying from the soil to tree tops.

HDC funded studies have been prompted by an increase in the incidence of *Pseudococcus viburni* (Signoret) (the obscure mealybug) on protected tomato crops in the UK. This species is a polyphagous cosmopolitan pest (Ben-Dov, 1994). In addition to tomatoes, it has been recorded in the UK on other edible crops, including peppers, and on glasshouse-grown ornamental plants.

HDC project, PC 161, which was completed in 2002, investigated the increase in incidence of infestations of *P. viburni* in UK tomato crops and began to formulate a control strategy (Jacobson & Croft, 2002). The most effective and IPM compatible method of controlling mealybugs on tomato plants during the production season was shown to be the insect growth regulator, buprofezin (Applaud), but this can not be applied to organic crops. Furthermore, this dependence on a single chemical insecticide was not consistent with the TGA's long term goal of pesticide-free crop production.

To become sustainable, the control strategy required a biological component that would act continuously throughout the growing season. Project PC 161 evaluated several biological control agents (eg *Hypoaspis* spp., *Chrysoperla* spp., *Beauveria bassiana*) but none proved to be very promising. However, preliminary investigations indicated that the parasitoids, *Leptomastix epona* (EPPO, 2002) and *Pseudaphycus maculipennis* (Charles, 2001) had potential to fulfil this role. *Leptomastix epona* could be released in UK crops but *P. maculipennis* was not indigenous and therefore required a licence before it could be used in trials in glasshouses.

The overall aim of this project was to formulate sustainable strategies, based on a combination of new and existing control measures, for the control of *P. viburni* on UK tomato and pepper crops.

The specific objectives were:

1. To develop a robust biological control measure based on parasitoids.
2. To construct sustainable mealybug control strategies tailored to the specific requirements of conventional and organic tomato and pepper crops.
3. To test and refine the control strategies in commercial crops.

### **Summary of work to date:**

It was considered highly unlikely that any one sustainable control measure would be successful against *P. viburni* on commercial crops. Therefore, a programme was designed consisting of a suite of compatible control measures that could be used to combat the pest at four distinct stages throughout the growing season:

- Survival on the glasshouse structure between crops. These control measures were developed in Project PC161 (Jacobson & Croft, 2002).
- Initial crop invasion by overwintered survivors.
- Season-long suppression of mealybug population growth.
- A compatible second line of defence.

### **Initial crop invasion by overwintered survivors:**

It has been shown that mealybug eggs on the structure of the glasshouse hatch within three weeks of the glasshouse being heated and they quickly migrate to the new plants. A 2% dilution of Savona has been effective against first instar *P. viburni* as they colonise tomato plants, reducing numbers by up to 93%. However, there is some concern over the effect of this product on young plants. Alternative products were tested in this project in 2006.

### **Season-long suppression of mealybug population growth:**

Much of the work in the early stages of this project was aimed at developing a biological control measure for *P. viburni* that would provide season-long suppression of the pest's population growth (Croft & Jacobson, 2006). A series of laboratory-based experiments provided important information about the key biological parameters that governed the ability of three species of parasitoids (*Leptomastix epona*, *Anagyrus pseudococci* and *Pseudaphycus maculipennis*) to successfully locate, attack and complete their development in the mealybugs on tomato and pepper plants. There appeared to be no fundamental reasons why these parasitoids would not become established in populations of *P. viburni* on both types of plants. However, there were questions over their ability to control *P. viburni* at the population level due to the huge numbers of offspring produced by the pest. *Anagyrus pseudococci* was the weakest candidate due to its poorer performance at the temperatures that are common in crops in the early season. The stronger candidates, *L. epona* and *P. maculipennis*, were tested at the population level in larger scale experiments in 2005.

*Leptomastix epona* could be released in UK crops but *P. maculipennis* was a non-native species and did not have official approval. As part of this project, an application for a licence to release *P. maculipennis* in the UK was successfully obtained from Defra. The studies in the second year of the project consisted of detailed examinations of the population growth of *L. epona* and *P. maculipennis* against that of their host, *P. viburni*, within experimental glasshouse crops, and larger-scale observations of the establishment of *L. epona* in commercial tomato and pepper crops.

In contained glasshouse tomato trials at STC Research Foundation, results showed that 80-90% parasitism was achieved from a single release of either *P. maculipennis* or a combination of *P. maculipennis* / *L. epona*. A maximum level of 50% was achieved by *L. epona* by itself. While both species could have the potential to become components in a mealybug control programme for tomato crops, *P. maculipennis* appeared to show the greatest promise. However, it was several weeks before these parasitoids began to reduce the pest populations and there remain doubts as to whether these control measures would be successful in commercial crops.

The parasitoids did not perform so well in the contained glasshouse pepper trials at STC Research Foundation. In this case, a maximum of 40% parasitism was achieved by *P. maculipennis* but no parasitism was detected by *L. epona* throughout the trial. The differences in the results between tomatoes and peppers may be explained by the habitats occupied by *P. viburni* on these crops. On tomato plants, the mealybugs are situated primarily on the lower stems while on peppers they are usually found beneath the calyx of the fruits. It would appear that the parasitoids are more successful when attacking mealybugs situated on the open stems than when the pests are tightly encrypted beneath the calyx of the peppers. This may simply be because the parasitoids are unable to locate their hosts.

The licence for release of *P. maculipennis* was not issued in time for the larger scale trials and so only *L. epona* could be evaluated in the commercial tomato and pepper crops. *L. epona* became established in the mealybug population on tomato plants following four weekly releases at the rate of 2 parasitoids per m<sup>2</sup> and achieved a level of 15% parasitism 70 days after the first release. Similar observations of *L. epona* within a commercial pepper crop recorded a level of 23% parasitism 77 days after the first release of the parasitoids. In both cases, the speed of establishment was not adequate to bring the pest population under control.

The generation time for *P. viburni* is slow, taking 50 days from egg to adult at 21±2°C (Heidari, 1989). This provides an advantage to the parasitoids, which complete their development in about the half the time. However, the pest has very high fecundity rates (adult female *P. viburni* can produce up to 500 eggs in its egg sac), which means the population could continue to grow rapidly while the parasitoids are becoming established. There was insufficient resource within this project to allow more detailed studies of the population dynamics within this biological system. However, to be successful, we believed that the parasitoids would have to be released in the early part of the growing season while pest numbers are still relatively small. The studies were continued in 2006.

#### **A compatible second line of defence:**

This component was required for use mid-season to redress the balance between pest and parasitoid should control with the primary control agent falter. Applaud (Buprofezin) remains an effective second line of defence for conventional crops but an alternative was required for organic crops. The effect of Savona on first instar larvae is mentioned above. This product has also been shown to have a direct effect on other life cycle stages on the plant but the results had been variable; 2% and 4% dilutions giving 30-60% and 40-100% control respectively. Control would therefore be dependant on a series of applications.

Some UK growers who used the fungal pathogen, *Verticillium lecanii*, against glasshouse whiteflies, reported incidental control of mealybugs but this had not been confirmed in controlled experiments. Laboratory bioassays done under ideal conditions showed that the pathogen reduced numbers of first instar nymphs but only by about 10%. It was presumed that the mealybugs were protected from infection by their waxy covering. However, it was hypothesised that the effect of the fungus could be enhanced by applying it after Savona because the soap would breakdown the waxy protection and allow more fungal spores to come into contact with the insect's body. Preliminary results were variable but 100% mortality of mealybugs was achieved in some laboratory experiments. The studies were scaled up to a commercial tomato crop in 2004. These results showed only a small advantage in using Mycotal in addition to Savona over three applications at approximately seven day intervals (Jacobson & Croft, 2005). This was

contrary to the previous laboratory scale studies and was attributed to the difficulty in obtaining good spray cover among the horizontal spray bundles.

The studies were repeated in 2005 with Eradicoat T incorporated in the trial (Croft & Jacobson, 2006). This was a new formulation of a starch-based material with a physical mode of action. All three treatments (*i.e.* 4% Savona, 4% Savona plus 0.1% Mycotal, Eradicoat T) reduced the numbers of mealybugs, whereas the numbers of mealybugs increased in the untreated controls. There was no apparent difference between the Savona and Savona plus Mycotal treatments (except in the number of viable egg sacs) thus indicating that there had been no additional effect by the fungus. This was supported by the fact that no fungal growth was evident on the dead mealybugs on the plants, nor did any develop when the cadavers were removed and incubated under more ideal conditions in the laboratory. Eradicoat T performed significantly better than the Savona on all development stages of the mealybug and currently offers the best potential for a second line of defence treatment in organic tomato crops. However, the effectiveness will always depend on the contact of the product and the pest.

## **PART 2**

### **MEALYBUG CROP INVASION STUDIES - 2006**

#### **Background:**

The overall objective of this study was to monitor crop invasion by mealybugs at the beginning of the season, and their subsequent development on the plants, to help time key actions in the control programme.

Previous studies have shown that young mealybugs, emerging from egg masses that have survived between crops begin to colonise plants soon after the glasshouse heating is switched back on. The most likely survival sites are:

- Posts
- Irrigation lines
- Sprinkler heads
- Soil

Young mealybugs are free to move from the first three sites straight onto the plants, so colonisation may be expected from these sites within three weeks. However, the soil is completely covered by plastic until the “stood-out” plants are ready to be planted in the ground. This restricts the mealybugs movement from the soil for the first few weeks and they can only reach the plants via the plastic overlaps and gaps around dollies. The fate of these mealybugs was unknown.

#### **Materials and method:**

Location: Rows 145-147, House 25, New Site, WSG, Isle of Wight.

Crop: Tomato, cv Capri  
Grown in soil to organic standards  
Heat switched on one day before plants arrived from propagator  
Plants stood out – 20 December (week 51, 2005)  
Planted in soil – week 4, 2006

Assessments: Initially, the crop was walked twice per week paying particular attention to the plants near posts. When mealybugs were first seen, eight assessment stations, each consisting of four plants, were established in each row. At weekly intervals, the numbers of small nymphs (instars 1 and 2), large nymphs (instar 3) and adults were recorded separately.

#### **Results and discussion:**

##### **Initial invasion:**

The presence of mealybugs was first noted on plants in the first week of January 2006. The most advanced stage seen on 10 January was nymph 3 but these were in the minority; most being first and second instars. At this stage, there were up to 50 per stem in the monitored area with even larger numbers in other specific areas. Two egg masses, which must have originated from surviving adults, were found on plants on 11 January but these were the exception and not yet hatching. Mealybugs were also present on volunteer tomato seedlings that had germinated beneath the plastic (especially those that were emerging through gaps in the plastic). These observations triggered the start of the more detailed monitoring.



### **Subsequent mealybug development:**

The mealybug development from young nymphs (instars 1 / 2), through final stage nymphs (instar 3), to adults / eggs masses on plants in the assessment stations is shown in Figure 1. Mealybugs were first seen on the plants two weeks after the heating was switched on but the main invasion occurred the following week, which was consistent with previous observations. A second flush of invaders reached the plants between the assessments in weeks 4 and 5 (*i.e.* 5-6 weeks into the crop), which followed the plastic being opened up for planting in the soil. Increasing numbers of third instars were found through weeks 3 and 4 (crop weeks 4 and 5), with a large increase in week 5 (crop week 6). Adults and egg masses were first seen in significant numbers from week 9 (crop week 10).

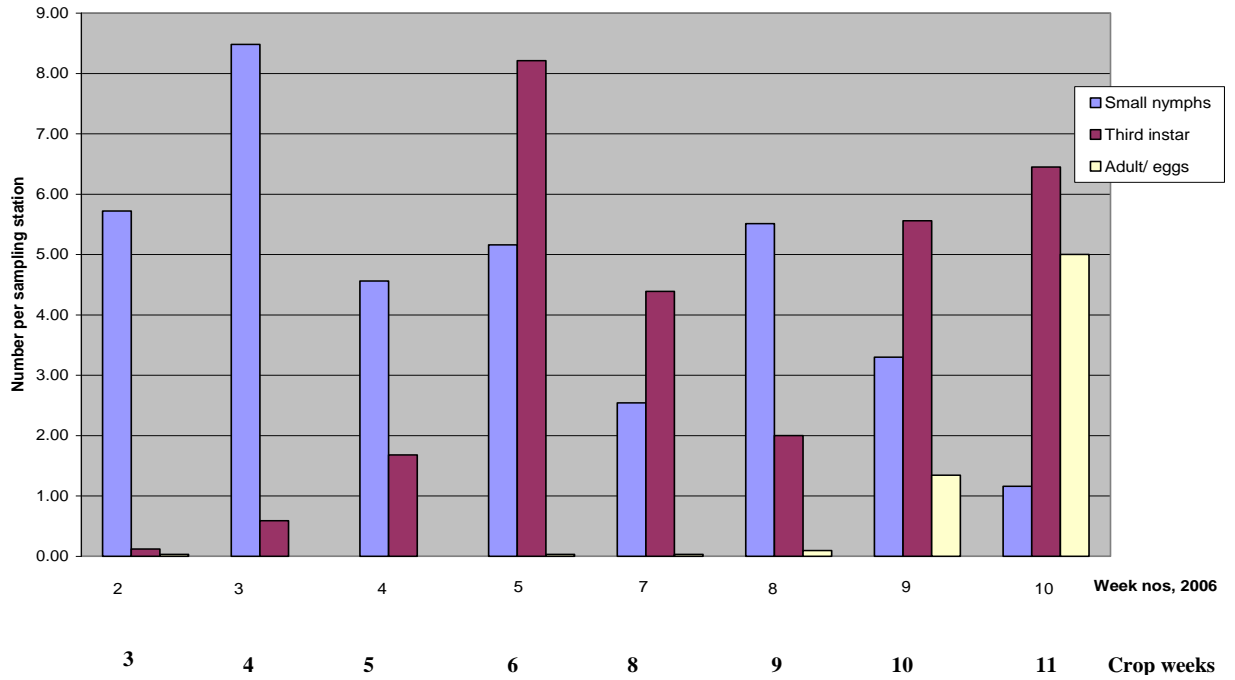
### **Timing of sprays:**

The ideal time to apply sprays would be 1-2 weeks after the second flush of invaders (in this case crop weeks 5-6). However, at this time the lower stems were still masked by foliage and there was some conflict between deleafing, to the possible detriment of plant growth, and obtaining the maximum impact from the spray applications. It is vital that sprays are effective before the tenth week of the crop to prevent the production of egg masses and the potential for a huge increase in numbers.

### **Summary of events / actions (Starting week 51, 2005):**

<b><u>Crop week</u></b>	<b><u>Event</u></b>	<b><u>Action</u></b>
<b>2-3</b>	<b>Appearance of instars 1 &amp; 2</b>	
<b>4-5</b>	<b>Developing to third instar</b>	
<b>5-6</b>	<b>Anticipate start of second flush of invaders</b>	
<b>6</b>		<b>Remove lower leaves</b>
<b>6-8</b>		<b>Start spray applications</b>
<b>9-10</b>	<b>Developing to adults and egg masses</b>	

**Figure 1. Mealybug development from young nymphs (instars 1 and 2), through final stage nymphs (nymph 3), to adults / egg sacs on plants.**



## PART 3

### CONTROL OF MEALYBUG CROP INVADERS - 2006

#### Background:

The overall objective of this study was to compare three treatments against nymphal stages of mealybugs following their invasion of young tomato plants. The work was done in a commercial tomato crop among routine crop management practices.

Invasion and establishment of mealybugs on the plants were monitored in a parallel study (Part 2 of this report) and spray applications were timed according to those results. It was originally intended to apply a series of 2-3 sprays but this programme was extended to i) control a second flush of mealybugs that emerged from below the floor plastic and ii) compensate for the poor coverage of early applications caused by the lower leaves shielding the stems.

The intention of the spray programme was to reduce numbers of mealybugs to a level that could be managed with a more sustainable control measure based on parasitoids (Part 4 of this report).

#### Materials and method:

Location: House 25, New Site, WSG, Isle of Wight.

Crop: Tomato, cv Capri  
Grown in soil to organic standards  
Heat switched on one day before plants arrived from propagator  
Plants stood out – 20 December (week 51, 2005)  
Planted in soil – week 4, 2006

Trial area: Rows 135-169.  
Total area of approx 3,800m<sup>2</sup> (54m x 70m)

Treatments: 1. Untreated control - Rows 145 / 147  
2. Savona 4% - Rows 137 / 143  
3. Eradicoat T – 30l / 1000l water - Rows 149 / 155  
4. Eradicoat T – 15l / 1000l water - Rows 157 / 169

Application: Sprays were applied to the lower stems with a “North Star” sprayer fitted with a lance and single nozzle. The treatments were applied weekly on Monday (Eradicoat T) and Tuesday (Savona) from week 3 to week 10 2006 (*i.e* a total of 7 sprays).

Assessments: Initially, the crop was walked twice per week paying particular attention to the plants near posts. When mealybugs were first seen, eight assessment stations, each consisting of four plants, were established in the central two rows of each plot. Thereafter, the numbers of mealybugs were recorded at each sample station at weekly intervals.

#### Data

analysis: Analysis of data was done using ANOVA of the square root transformed data and treatment means were compared using the LSD values given.

#### Results and discussion:

Mealybugs were first seen on the plants two weeks after the heating was switched on but the main invasion occurred the following week, which was consistent with previous observations. This triggered the start of the spray programme, which began in week 3 2006 (crop week 4). There was a second flush of invaders between weeks 4 and 5 2006 (*i.e.* 5-6 weeks into the crop), which followed the plastic being opened. Increasing numbers of third instars were found through weeks 3 and 4 2006 (crop weeks 4 and 5), with a large increase in week 5 2006 (crop week 6). Adults and egg masses were first seen in significant numbers from week 9 2006 (crop week 10).

In the initial stages of the trial, spray coverage was impeded by the presence of lower leaves. There was conflict between the need to remove these leaves to improve spray coverage and the detrimental effect it would have on plant growth. The offending leaves were removed in week 5 2006 (crop week 6) with the exception of a single leaf that anchored the plastic clip attached to the support string. This leaf continued to provide some direct harbourage for mealybugs and shielded others on the stems, thus reducing the efficacy of the sprays and prolonging the spray programme. Towards the end of this trial, an alternative spray lance with four nozzles was brought into use in the adjacent commercial crop and this appeared to improve both spray coverage and control of the pests.

The mean numbers of mealybug instars 1/2 and 3 recorded at the sample stations between weeks 2 and 10 2006 are shown in Tables 2a and 2b respectively. The numbers fluctuated as the mealybugs developed from instars 1/2 to 3, and as the second flush of young mealybugs reached the plants. To aid clarity, the combined data for all instars are illustrated in Figures 2– 5. The three insecticidal treatments provided an immediate decline in mealybug numbers but there was a resurgence after the plastic was split and more invaders were able to reach the plants. By week 10 2006, the numbers of mealybug survivors were quite small in all treated plots with no significant difference between treatments. At that time, there was an overall mean of 0.14 third instar mealybugs per sample station in the sprayed plots compared to 3.84 per sample station in the untreated control. To help put this into perspective, 0.14 mealybugs per sample station was equivalent to 0.03 mealybugs per plant or 1 mealybug per 33 plants.

This spray programme was considered adequate to pave the way for a more sustainable control strategy based on parasitoids. However, the number of sprays applied must be reduced in the future by improving the efficacy of each application. It is anticipated that this will be achieved by using a different deleafing strategy and improved spray equipment.

There was no evidence of acute phytotoxicity from sprays in any treatments.

**Table 2a. Mean numbers (square root transformed) of first and second instars recorded at weekly intervals after Savona and Eradicoat T treatments (two rates).**

Crop week	Control	Savona	Eradicoat T (high rate)	Eradicoat T (low rate)	LSD
2*	3.41 (1.84)	3.81 (1.93)	3.38 (1.81)	4.50 (2.11)	(0.517)
3	5.06 (2.23)	1.22 (1.10)	1.94 (1.38)	2.41 (1.55)	(0.447)
4	2.56 (1.52)	0.16 (0.28)	0.69 (0.68)	0.75 (0.80)	(0.743)
5	1.91 (1.38)	2.50 (1.49)	0.84 (0.91)	0.75(0.86)	(0.484)
7	1.88 (1.34)	2.16 (1.35)	1.47 (1.21)	0.72 (0.84)	(0.518)
8	4.69 (2.15)	0.56 (0.74)	0.53 (0.71)	0.69 (0.79)	(0.469)
9	1.91 (1.28)	0.03 (0.09)	0.16 (0.20)	0.16 (0.27)	(0.553)
10	0.25 (0.35)	0.19 (0.35)	0.06 (0.13)	0.00 (0.00)	(0.465)

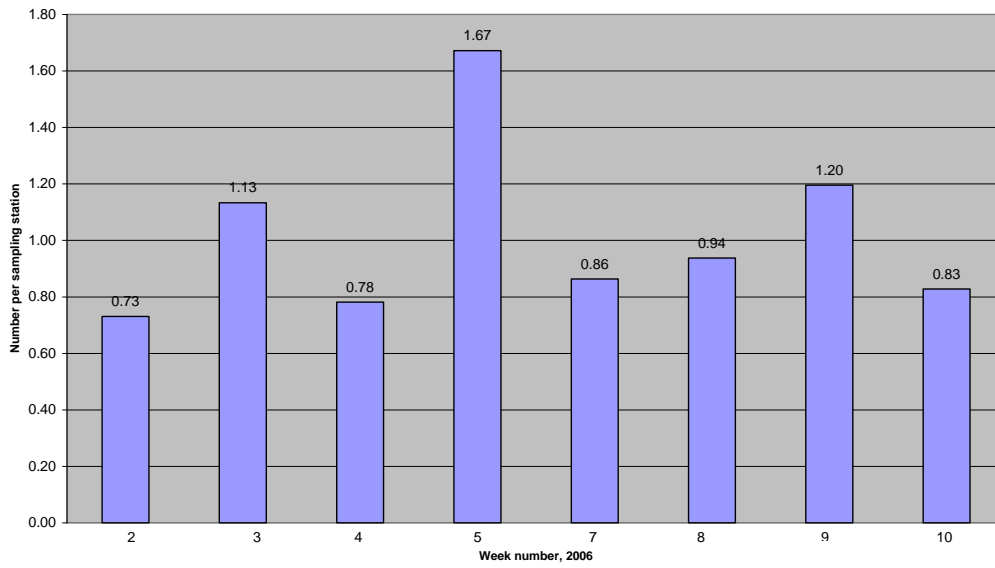
\* Pre-treatment

**Table 2b. Mean numbers (square root transformed) of third instars recorded at weekly intervals after Savona and Eradicoat T treatments (two rates).**

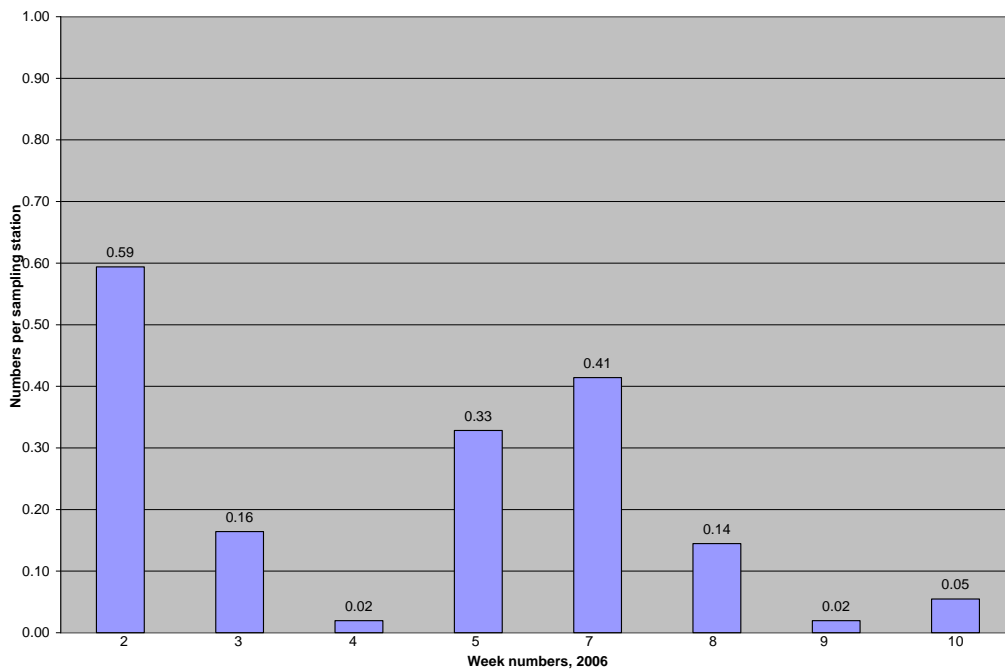
Crop week	Control	Savona	Eradicoat T (high rate)	Eradicoat T (low rate)	LSD
2*	0.03 (0.09)	0.94 (0.80)	0.53 (0.62)	0.56 (0.72)	(0.584)
3	0.315 (0.55)	0.10 (0.22)	0.20 (0.22)	0.03 (0.09)	(0.400)
4	0.84 (0.87)	0.00 (0.00)	0.25 (0.42)	0.66 (0.76)	(0.403)
5	5.09 (2.25)	0.13 (0.24)	1.38 (1.15)	1.44 (1.20)	(0.353)
7	3.34 (1.82)	1.16 (1.04)	1.03 (0.99)	1.94 (1.38)	(0.354)
8	1.66 (1.23)	0.60 (0.75)	0.43 (0.57)	0.97 (0.94)	(0.564)
9	3.09 (1.75)	0.13 (0.25)	0.29 (0.51)	0.66 (0.78)	(0.360)
10	3.84 (1.94)	0.25 (0.42)	0.09 (0.21)	0.09 (0.21)	(0.451)

\* Pre-treatment

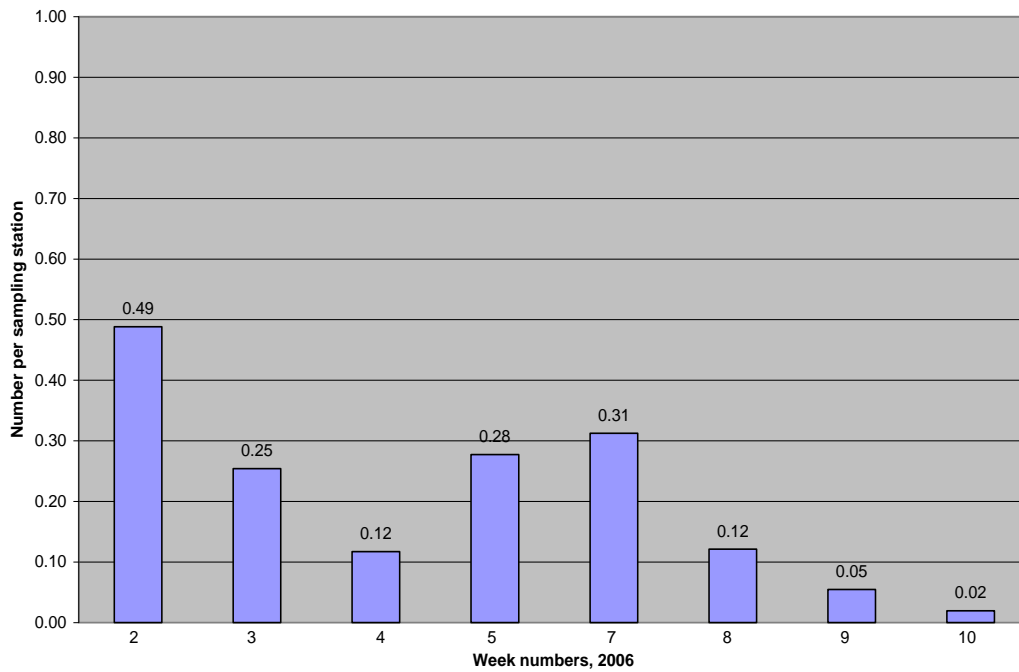
**Figure 2. Numbers of mealybugs per sampling station in untreated controls on eight assessment dates.**



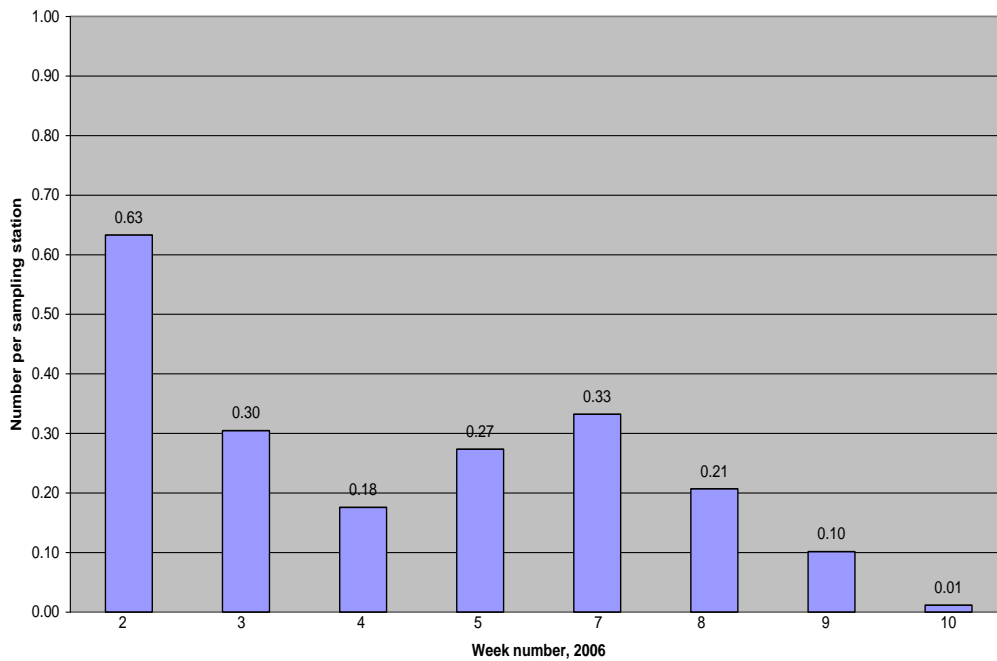
**Figure 3. Numbers of mealybugs per sampling station in Savona treatments on eight assessment dates.**



**Figure 4. Numbers of mealybugs per sampling station in Eradicoat T (higher rate) treatments on eight assessment dates.**



**Figure 5. Numbers of mealybugs per sampling station in Eradicoat T (lower rate) treatments on eight assessment dates.**



## PART 4

### EVALUATION OF A SEASON-LONG MEALYBUG CONTROL PROGRAMME

#### Background:

A sustainable IPM programme, consisting of a suite of compatible control measures, has been designed for use against obscure mealybug (*Pseudococcus viburni*) on organic tomato. The components of the programme, which were developed in a series of independent studies in earlier stages of this project, were combined and evaluated in a commercial crop in this trial.

The proposed IPM programme begins with a series of sprays of soft chemicals (soap or starch-based products) applied 5-8 weeks after heating the glasshouse (Parts 2 and 3 of this report). The aim was to reduce the number of mealybug invaders to a level that could be controlled biologically. Two species of parasitoids, *Leptomastix epona* and *Pseudaphycus maculipennis*, have been shown to be capable of locating, attacking and completing their development in most life cycle stages of *P. viburni* on tomato plants (Croft & Jacobson, 2006). It was proposed that these parasitoids be used to suppress the mealybug population throughout the season. The same soft chemicals would be held in reserve to be used as a last resort to redress the balance between pest and natural enemies should this become necessary.

Prior to the release of the parasitoids it was important to first establish if the parasitoids would be active in the short day lengths experienced in the glasshouses in the early part of the season. For many species of insect short day lengths can significantly reduce activity. Therefore small scale experiments were conducted prior to their release.

A lot of consideration was given to how the parasitoids should be released into the crop. Ideally, we would have followed a programme of routine releases of large numbers throughout the crop starting at the beginning of the growing season. Such an approach is very successful for *Encarsia formosa* against glasshouse whitefly, partly because the relatively large releases compensate for the poor performance of *E. formosa* in the less than ideal conditions that prevail through January and February. The approach is possible with *E. formosa* because the parasitoids are inexpensive (£2-£3 per thousand). However, *L. epona* and *P. maculipennis* are considerably more expensive to produce and it was necessary to devise a completely different strategy to keep the cost within sensible parameters. It was proposed that:

- When the early season sprays were applied to combat mealybug invaders, every 35th double row would be left untreated.
- Parasitoid releases would begin about 5-6 weeks into the crop as the invaders began to reach the third instar. We anticipated that the searching ability of the parasitoids would be relatively poor under the early season conditions and so all releases were concentrated close to the infested plants in the untreated rows.
- Although the parasitoids would be released at relatively high rates, it was anticipated that this would be cost effective because it would only be done over a proportion of the crop.

The aim was to establish intensive breeding areas (IBAs) in the glasshouse from which the parasitoids would disperse as conditions became more suitable.



## **Materials and method:**

### *Parasitoid activity:*

The activity of *L. epona* and *P. maculipennis* were recorded from the following two temperature and two daylength regimes in controlled environmental chambers:

1. 22°C, 16 hours daylength
2. 22°C, 8 hours daylength
3. 17°C, 16 hours daylength
4. 17°C, 8 hours daylength

Activity was monitored for ten separate females of each species for two minutes every 30 minute interval over a period of eight hours. Activity was recorded as the percentage of time the parasitoid was observed walking. In addition the ability of the parasitoids to produce offspring under the four different environments was also recorded. Each parasitoid was given two mealybugs (2<sup>nd</sup> instars) on a tomato leaf on damp filter paper. After 24 hours the mealybugs were removed and placed in a CE room (16L:8D, 21 ± 2°C). Each female parasitoid was then given new mealybugs. The numbers of offspring emerging from the mealybug were recorded for each day. The above procedure was repeated three times.

### *Commercial trial:*

Location: House 25, New Site, WSG, Isle of Wight.

Crop: Tomato, cv Capri  
Grown in soil to organic standards  
Plants stood out – week 51, 2005  
Planted in soil – week 4, 2006

Trial area: Rows 121 to 151; total area 1568m<sup>2</sup> (22.4m x 70m).

#### Details of

IBA: Rows 145 & 147.  
Total area of 224m<sup>2</sup> (3.2m x 70m)  
Number of plants: Approx 250  
Number of heads: Initially two heads per plant, so 500 heads. Additional heads were taken from week 11 and these stems were taken into account in assessments from week 18.

Parasitoids: Parasitoids were released in the IBA every week from weeks 10 to 19 (*i.e.* 10 releases) and every two weeks from weeks 20 to 24 (*i.e.* 2 releases). On each occasion, the releases consisted of 200 *L. epona* and 800 *P. maculipennis*. It is difficult to convert these numbers into rates per m<sup>2</sup> due to the open nature of the IBAs but it would be approximately 4 parasitoids (*i.e.* 2 males and 2 females)/m<sup>2</sup>/week. At the proposed frequency of IBAs, this was equivalent to 0.2/m<sup>2</sup>/wk overall. Parasitoid dispersal was monitored to the south of the IBA in rows 121 to 143.

Assessments: The trial began with rigid procedures for assessments but these were modified to take into account our findings as the work progressed. The procedures and modifications are detailed below:

#### 1. Mealybug counts within the IBA:

Eight sample stations, each consisting of 4 heads were established in each double row. The numbers of mealybug nymphs, adults and egg masses were recorded separately in each station at intervals shown in Figure 6. Any evidence of parasitism was recorded although this was difficult to quantify within the glasshouse.

**2. Parasitism within the IBA:**

Additional sample stations, also consisting of 4 heads, were established for destructive sampling. At intervals from week 14 (see Table 3), all mealybugs were removed from one of the sample stations and specimens sent to STC RF for emergence tests.

**3. Mealybug counts outside the IBA:**

To obtain more general information about mealybug populations in the rest of the glasshouse, an additional 8 sampling stations were set up (as above) at distances of approx 3m (row 141), 8m (row 135) and 16m (row 125) to the south of the IBA. The numbers of mealybug nymphs, adults and egg masses were recorded separately in each station at time intervals shown in Figures 7, 8 and 9. In addition, any evidence of parasitism was recorded.

**4. Parasitism outside the IBA:**

At intervals from week 17 (see Table 3), the rows either side of rows 141, 135 and 125 were walked and 20-30 mealybugs collected and sent to STC RF for emergence tests.

**Results and discussion:**

*Parasitoid activity:*

Table 3. The mean percentage activity ( $\pm$ sd) of *L. epona* and *P. maculipennis* at two different daylengths and temperatures

	16 hours		8 hours	
	22 °C	17 °C	22 °C	17 °C
<i>L. epona</i>	78.43 (5.45)	81.38 (8.58)	87.33 (10.11)	81.40 (7.59)
<i>P. maculipennis</i>	63.77 (16.07)	60.28 (5.17)	60.03 (11.95)	69.06 (6.58)

Table 4. The mean number ( $\pm$ sd) of offspring produced by *L. epona* and *P. maculipennis* at two different daylengths and temperatures

	16 hours		8 hours	
	22 °C	17 °C	22 °C	17 °C
<i>L. epona</i>	0.50 (0.57)	0.53 (0.82)	0.34 (0.55)	0.27 (0.52)
<i>P. maculipennis</i>	0.60 (1.35)	0.77 (1.87)	0.70 (1.28)	0.60 (1.33)

The results in Tables 3 and 4 show there is no difference in either the activity or the oviposition of *L. epona* and *P. maculipennis* in either long or short daylengths. Numbers of offspring were low, but this is possibly due to the low numbers of mealybugs offered which can inhibit oviposition. The results suggest that the environment experienced in an early release of the parasitoids would not affect the performance of the parasitoids.

*Commercial Trial:*

The numbers of mealybugs per sample station in the IBA and more distant rows (125, 135 and 141) during the course of the trial are shown in Figures 6-9. The levels of parasitism detected in the samples sent to STC RF are shown in Table 5.

Full details of mealybug crop invasion between week 51 2005 and week 10 2006 are provided in Part 2 of this report. In summary, two peaks of mealybug invasion occurred between weeks 2 and 5 2006. Significant numbers of adults and egg masses were recorded from week 8 2006.

A comparison of the efficacy of programmes of three soft chemicals against mealybug invaders was done in parallel to this study. The full results are provided in Part 3 of this report. In summary, the products provided similar levels of control but the intended spray programmes had to be extended to cope with the second flush of mealybug invaders and to compensate for the poorer than anticipated effect of the of the first sprays.

The following notes support the data in Figures 6 to 9 and Table 5, providing a more revealing commentary on the experiences with the experimental control programme between January and August 2006. The notes explain why and how the proposed strategy was modified on several occasions in response to crop monitoring.

#### **Notes to support data:**

It was originally intended to stop applications of soft chemicals when the parasitoids were first released. However, it was necessary to continue these applications at 2-3 week intervals throughout the trial to suppress the mealybug population growth. Only the IBA remained unsprayed during the first few weeks of the trial. In week 13, three weeks after the first releases of parasitoids, random sampling and on-site dissection of large mealybug nymphs and adult females revealed approx 20% with *P. maculipennis* and 7% with *L. epona* in the IBA. There were up to seven immature *P. maculipennis* per mealybug; the most advanced being translucent pupae while most were still orange / brown larvae. The *L. epona* were orange / brown larvae. Samples sent the following week from the IBA to STC RF for emergence tests revealed 60% parasitism.

By this time, most of the mealybug invaders were engulfed in waxy egg masses and there were relatively few at a receptive stage for parasitoids. With hindsight, it would have been sensible to stop releasing adult parasitoids at this stage to enable us to gain an understanding of the potential of the offspring from this early establishment. However, the releases continued until week 24.

Mealybug eggs started to hatch from week 14 and numbers of motile stages in the IBA increased at an alarming rate over the next few weeks. By week 17, this generation of mealybugs were reaching the third instar stage. About 40 such individuals were dissected on-site but no immature parasites were found. Samples sent to STC RF the same week revealed much reduced percentage parasitism (approx 5%) in the IBA and no apparent spread to the adjacent rows. At this stage, the mealybug population growth appeared to be rapidly outstripping the parasitoids.

As this generation of mealybugs completed their life cycle, a relatively large number of male pupae were found on the plants. This was the only time that this was observed during the season and its significance is not fully understood.

A decision was made to apply a second line of defence spray to the IBA and the adjacent rows before this generation of mealybugs started producing eggs. The latter began in week 18.

From week 20, the level of parasitism determined from checks done on the nursery remained very poor, with only about 2% *P. maculipennis* (c5 immature parasitoids per

mealybug) and no *L. epona* being found in the IBA. The emergence tests at STC RF were broadly comparable although a small number of parasitised mealybugs were detected 8m and 16m from the IBA during this period.

The sprays with Eradicoat T continued at about 2-3 week intervals. This suppressed the mealybug population growth and as a probable consequence the percentage parasitism improved. This reached about 60% in the IBA by week 24 and was still above 40% at week 31. Some dispersal was detected to 3m from the IBA during that period but not any further. When interpreting these results, it must be remembered that 9,000 *P. maculipennis* and 3,000 *L. epona* had been released into the IBA during the season.

It is important to note that 2006 was an unusual growing season, with two extended periods of exceptionally hot weather in July and September. In the remainder of this glasshouse (*i.e* 30,000m<sup>2</sup> of mixed tomato varieties) control of mealybugs was totally dependant on the use of sprays of soft chemicals applied at a frequency determined by continuous crop monitoring. In fact, this resulted in Eradicoat T being applied at approximately 2-3 week intervals throughout the whole season. The mealybug numbers were suppressed to non damaging levels until late-August but the populations then grew rapidly resulting in some plant loss from late-September onwards. Furthermore, the cost of the spray programme was unacceptable.

**Overall comments:**

- Mealybugs remained present on the stems of the plants throughout the season. The pest’s population growth was suppressed until late August but then increased and some crop loss was experienced from late-September onwards.
- Expenditure on Eradicoat T was unacceptable.
- With the benefit of hindsight, the release of parasitoids should have begun about two weeks earlier but it is doubtful that this would have made a great deal of difference over the course of the season.
- Parasitoids were still found in the IBA after the spray programme began indicating some compatibility with Eradicoat T.
- The main concern after the previous stage of this project was whether the parasitoids would be able to suppress the rapid population growth of their mealybug host. These results suggest that they are more likely to coexist with the pest than to control it.
- Additional control measures against mealybugs are still required for organic crops.

**Figure 6. Mealybug development in the IBA.**

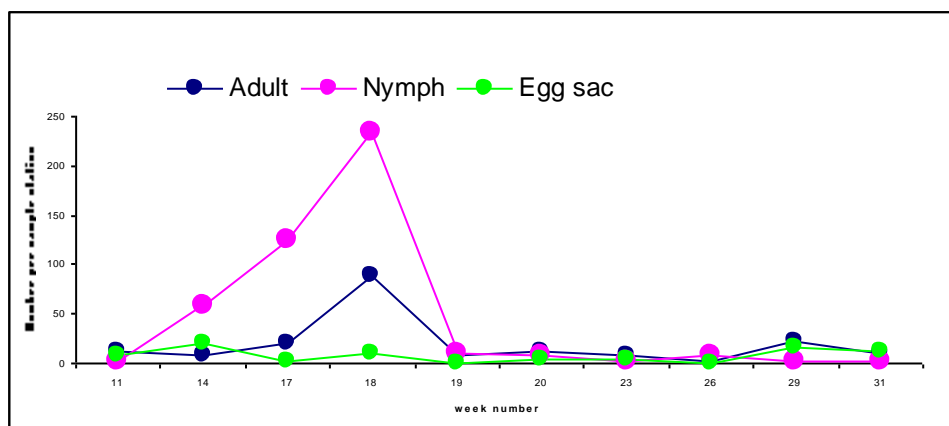


Figure 7. Mealybug development in row 141 (approx 3m from IBA)

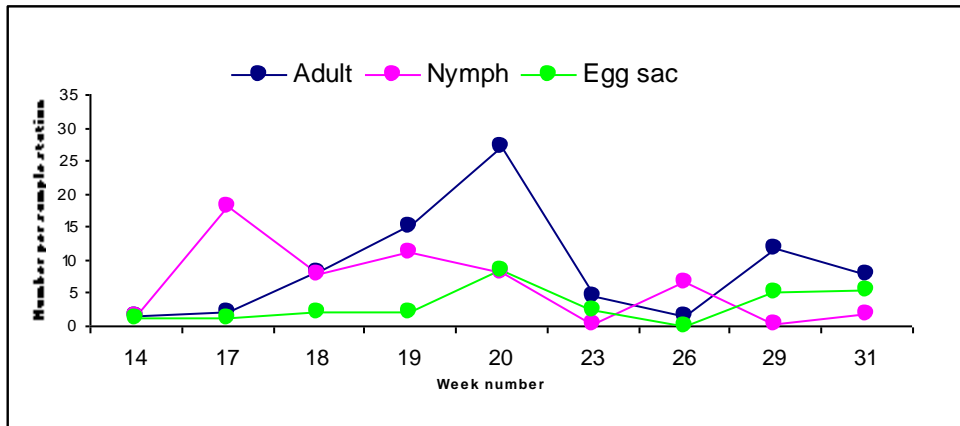
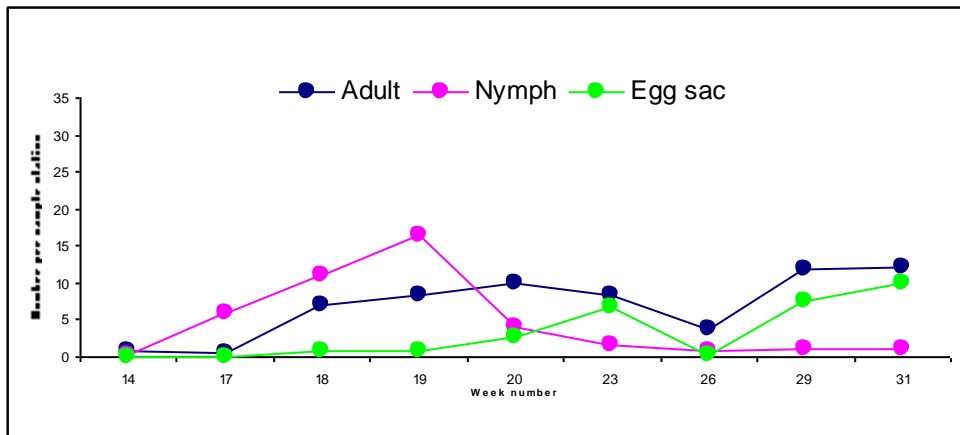
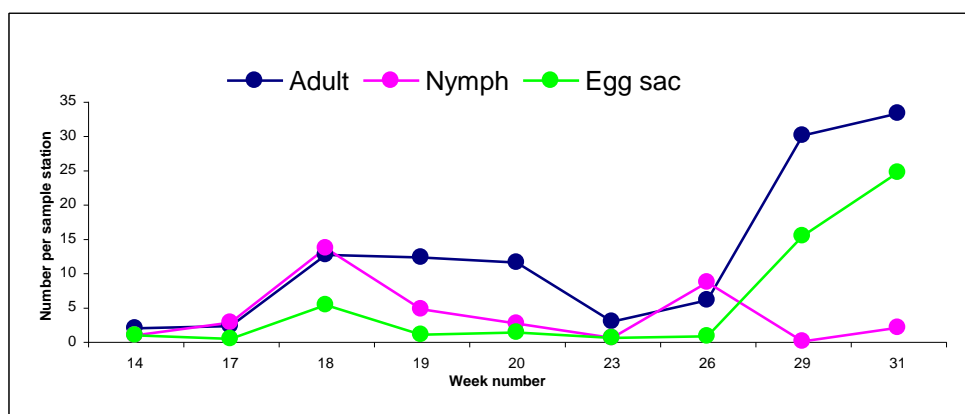


Figure 8. Mealybug development in row 135 (approx 8m from IBA)



**Figure 9. Mealybug development in row 125 (approx 16m from IBA)**



**Table 5. Results of parasitoid emergence tests**

Week Number	Approximate percentage parasitism (number of <i>P. maculipennis</i> [P] and <i>L. epona</i> [L] emerged)			
	Within IBA	3m from IBA	8m from IBA	16m from IBA
14	60%	-	-	-
17	5% (21P + 2L)	0	0	0
19	0	-	0	4% (12P)
20	3% (5P)	0	10% (5P)	0
24	64% (63P + 1L)	28% (5P)	0	0
28	30% (15P)	3% (3P)	0	-
31	45% (84P + 1L)	0	0	0

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