# FINAL REPORT

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# Mushrooms: Integrated pest management of sciarid flies M 42

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May 2006

Commercial - In Confidence

Project title:	Mushrooms: Integrated pest management of sciarid flies
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Report :	Final report, May 2006
Previous reports:	Interim report, 31 <sup>st</sup> January 2005
	First annual report, 31 <sup>st</sup> March 2005
	Supplementary report, 31st September 2005
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Date commenced:	1 <sup>st</sup> April 2004
Date completion due:	31 <sup>st</sup> May 2006
Key words:	sciarids, mushroom, insect pest, Decis, Spinosad,
	Gnatrol, Thiacloprid, Nemasys, AGARIguard, Dimilin.

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The results and conclusions in this report are based on an investigation conducted over one year. The conditions under which the experiment was carried out and the results obtained 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 interpretation of the results especially if they are used as the basis for commercial product recommendations.

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# **Grower Summary**

# M 42

# Mushrooms: integrated pest management of sciarid flies

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Mushrooms: integrated pest management of sciarid flies

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# Headline

Acceptable control of sciarid flies was achieved in a mushroom crop by treating the casing with Dimilin (diflubenzuron) [NB. Dimilin susceptible population], Calypso (thiacloprid) or Nemasys M (*Steinernema feltiae*). Although the casing treatments of Nemasys M and Calypso gave good sciarid control the compost treatments were variable and generally not very effective.

## **Background and expected deliverables**

Mushroom sciarid flies are important pests of commercial mushroom production. The main species causing damage in the UK are *Lycoriella castanescens and Lycoriella ingenua*. The latter is of particular concern because it has started to develop resistance to diflubenzuron, the most widely used chemical insecticide for sciarid fly control. At present, only one other chemical insecticide is available, the pyrethroid deltamethrin. A biological control agent, the insect pathogenic nematode *Steinernema feltiae*, is also being sold, but it is reputed only to work when applied to mushroom casing.

In order to develop sustainable methods of controlling sciarid flies, new control agents need to be evaluated. More information is also required about the effectiveness of insect pathogenic nematodes, particularly as a compost treatment for control of first generation sciarids.

The aim of this project was to investigate an IPM strategy for controlling sciarid fly pests for the UK mushroom industry. The expected deliverables from this work included:

- An assessment of potential control agents and a control strategy based on current control methods.
- New information on control methods that could rapidly become available to the industry.
- Information made available to industry, via fact sheets, popular articles, training events and a project advisory group.
- Consideration of a strategy to minimise the development of insecticide resistance in sciarid flies.

# Summary of the project and main conclusions

#### Laboratory evaluation of different control agents

Laboratory experiments were done to evaluate the effect of seven different control agents on *L. ingenua* populations in mushroom compost and casing. Control agents were applied as drenches and by mixing (= admix) into the substrate. *Lycoriella ingenua* were tested in two ways: (a) by applying eggs to the substrate three days before or after the application of the control agent; (b) allowing adult females to lay their eggs on substrate treated with the control agents. The control agents used are set out in the following table:

Control agent	Туре				
AGARIguard (ECOspray Ltd.)	Novel insecticide, based on food-grade garlic extract				
Decis (AgrEvo)	Chemical insecticide (deltamethrin) approved in UK for				
	mushrooms				
Dimilin (Uniroyal)	Chemical insecticide (diflubenzuron) approved in UK				
	for mushrooms				
Gnatrol (Valent Biosciences)	Biological control agent, insect pathogenic bacterium				
	(Bacillus thuringiensis), sold for use on mushroom				
	crops in USA				
Nemasys M (Becker	Biological control agent, insect pathogenic nematode				
Underwood)	(Steinernema feltiae) available in UK				
Spinosad (Dow)	Novel chemical insecticide approved in UK as				
	Conserve for other crops				
Thiacloprid granules (Bayer)	Novel chemical insecticide (thiacloprid) approved in				
	UK for other crops				

• <u>AGARIguard</u> gave poor control in compost and casing. However, in previous work, it caused high levels of mortality when drenched directly onto *L. ingenua* eggs. It also gave better control when fly infestation occurred immediately after treatment. The poor control in this study may be due to short persistence of AGARIguard in the substrate. It may have some ovipositional deterrent effect but this would also be detrimentally affected by short persistence.

- <u>Decis</u> gave poor control in casing and compost.
- <u>Dimilin</u> gave good control in casing and compost.
- <u>Gnatrol</u> gave good control in casing and compost.
- <u>Nemasys</u> gave good control in casing and compost.
- <u>Spinosad</u> gave poor control in casing, and mixed results in compost.
- <u>Thiacloprid</u> gave good control, except when mixed into compost against eggs.

#### Crop scale evaluation of selected control agents

From the results of the laboratory experiments, Nemasys, Gnatrol, and thiacloprid (as Calypso) were selected for evaluation on a crop scale. In accordance with commercial practice, the treatments were mixed into compost at spawning, or applied to casing as drenches. Dimilin was also included as a standard casing treatment, and the experiment included untreated controls.

- None of the replicated treatments gave good control of the first generation of sciarid larvae (all gave less than 35% control compared to the untreated trays).
- Control of the second generation was improved in all treatments. Overall, good sciarid control was achieved by treating the casing with Dimilin, Calypso and Nemasys, and by treating the compost with Nemasys.
- Dimilin gave 80% control of the second generation.
- Control of the second generation with Calypso was less in the compost (46% control) than the casing (70% control).
- Nemasys performed equally well in compost and casing (70% control of second generation in both cases).
- Gnatrol gave about 50% control of the second generation in compost and casing. It is possible that control could have been improved if a higher concentration had been used.
- Mushroom yield was increased with all treatments over untreated substrate. When compared to the Dimilin treated trays, higher yields were achieved where casing had been treated with Gnatrol and Calypso and where compost had been treated with Gnatrol and Nemasys.

### Dose response trial

The effect of different rates of Nemasys M and Gnatrol applied as a drench to mushroom compost for control of sciarids was determined in a pot (2.5 kg compost) trial on the

Warwick HRI mushroom unit. Dose rates 0.75N, N, 1.5N, 2N and 4N were used, where N = recommended rate (Nemasys M = 3 x 10<sup>6</sup> nematodes per m<sup>2</sup>; Gnatrol = 14.6 g product/m<sup>2</sup>). In addition, the effect of Calypso applied to compost as a single N rate drench treatment (N = 115 ppm w/w product/substrate) was investigated. This was one to follow up the results from the laboratory experiments where Calypso had been more effective as a drench treatment than when admixed.

The mean percentage levels of controls were as follows:

- Nemasys M gave 30%, 63% and 87% control at rates of 1.5N, 2N and 4N respectively.
- Gnatrol gave 63% at N rate and >76% control at 1.5N and higher dose rates.
- Calypso applied as a drench at N dose rate gave >80% control.

A regression analysis was carried out on the results to determine the relationship between dose rates and sciarid control. Dose rates were then selected for evaluation on a crop scale based on the efficacy of the treatments balanced against the costs of using higher doses. The intention was that increasing the dose rate of the control agents should improve their efficacy when applied to compost on a crop scale.

#### Crop scale trial (2)

A crop scale trial was done to verify the efficacy of the control agents using the dose rates and application method for compost based on the results from the previous experiment. All treatments were applied separately to compost and casing. Calypso (N) was drenched onto the compost. Gnatrol (1.5N) and Nemasys M (2N) were mixed into the compost at spawning. For comparison, Calypso (N), Gnatrol (1.5N) and Nemasys M (N) were also assessed as drench treatments on the casing. Dimilin (N) was drenched onto the casing as a positive control and there were also untreated controls. Percentage control of first generation sciarids and both first and second generations together to give overall percentage control was measured by comparison of fly emergence from treated trays with emergence from the untreated trays. Mushroom yield and quality for each treatment was also recorded. All the results were subjected to an analysis of variants. Mean percentage control was as follows:

#### First generation sciarid control:

- Dimilin as a casing drench gave 42% control.
- Calypso on compost gave 51% control and as a casing drench gave 34% control.
- Gnatrol in compost gave 44% control and as a casing drench gave 46% control.
- Nemasys M in compost gave 38% control and as a casing drench gave 44% control. *Overall sciarid control:* 
  - Dimilin as a casing drench gave 77 % control.
  - Calypso on compost gave 45 % control and as a casing drench gave 64% control.
  - Gnatrol in compost gave 26% control and as a casing drench gave 56% control.
  - Nemasys M in compost gave 37% control and as a casing drench gave 68% control.

Dimilin, Calypso and Nemasys M all gave acceptable overall control of sciarids when used as a casing drench. For an acceptable level of control, it is likely that Gnatrol would need to be used at a higher dose rate (2N). This rate was found to be effective in previous work carried out at Warwick HRI. None of the treatments gave effective control of the first generation of sciarids. With the casing treatments this is possibly due to the time of application of the casing. Sciarids develop into pupae about 18 days after oviposition. This means that they would be protected from treatments applied at casing at 17 or 18 days after spawning. Applying the casing and thus the casing treatments earlier might give better first generation sciarid control. With the compost treatments, the poor control might be due to the nonhomogenous nature of the substrate which makes it difficult to ensure that there are no pockets of untreated compost in which sciarid larvae can develop. This may explain the difference between the dose response trial and the later crop scale experiment. However, a combined compost and casing treatment might be useful where there is a particularly high sciarid infestation. Used on a succession of mushroom crops, this could break the cycle of infestation. Calypso and Gnatrol give new options to improve grower choice which is vital in the battle to prevent resistance developing to existing products.

Mushroom yield and quality (shown as mean weight per tray of 50 kg compost):

• Dimilin as a casing drench gave 9.4 kg mushrooms of which 13% were buttons, 67% open and 20% closed cups.

- Calypso on compost gave 11.6 kg mushrooms of which 16% were buttons, 54% open and 30% closed cups. As a casing drench it gave 10.5 kg mushrooms of which 12% were buttons, 24% open and 64% closed cups.
- Gnatrol in compost gave 12.3 kg mushrooms of which 16% were buttons, 53% were open and 31% closed cups. As a casing drench it gave 11.1 kg mushrooms of which 13% were buttons, 56% open and 31% closed cups.
- Nemasys M in compost gave 11.9 kg mushrooms of which 16% were buttons, 54% open and 30% closed cup. As a casing drench it gave 10.3 kg mushrooms of which 14% were buttons, 64% open and 22% closed cups..
- The untreated trays gave 10.1 kg mushrooms. Of these, 14% were buttons, 35% closed and 52% open mushrooms.

### **Financial benefits**

The total cost of pest infestations to the UK mushroom industry is estimated at £11-12 million per annum, including direct loss from insect attack, transmission of mushroom pathogens, and costs of buying and applying insecticides. The financial benefits of this study cannot be ascertained at this stage, as they will depend upon a number of factors including the future availability of insecticides and the development and spread of insecticide resistance. However, a sustainable and effective integrated pest management system for sciarid flies should be of financial benefit to growers. Monitoring flies around the farm highlights the times when pest treatment is necessary and when there are no flies, no treatments should be applied. Following a practice of good hygiene, removing debris from farm surrounds and keeping compost as far from the cropping houses as possible will all help to eliminate places where sciarids can breed. These basic practices reduce the need for applying costly fly control treatments.

The efficacy of compost treatments to control sciarids varied between experiments. Nemasys M (N) in compost gave effective overall control in the first unit trial and at 2N in the dose response experiment. Treating the compost with either Gnatrol (1.5N) or Nemasys M (2N) increased both the yield and quality of the mushrooms. If a compost treatment was used, maybe in conjunction with a casing treatment, the yield increase would help to offset the extra cost of treatment. In the longer term, the development of methods that reduce the use of

chemical insecticides, or enable insecticide-free production, may well give a marketing benefit.

### Action points for growers

- In these experiments, compost treatments of Calypso (N), Gnatrol (1.5N) and Nemasys M (2N) all gave some control of first generation sciarids but results on the overall efficacy were variable from one experiment to another.
- Mushroom yield was increased with the compost treatments, especially with Gnatrol.
- Casing treated with Dimilin effectively controlled sciarids but our populations have not yet developed resistance. It did not give good control of first generation sciarids.
- Calypso (N), Gnatrol (1.5N) and Nemasys M (N) gave reasonable control of sciarid fly populations in mushroom casing and gave some increase in yield when compared to both the Dimilin treatment and the untreated control.
- Although compost treatments alone did not give effective sciarid control, they might be useful if used in combination with a casing treatment. This strategy could be effective in breaking the cycle of very heavy sciarid infestations on a mushroom farm if used on sequential crops for a short period.
- Fly monitoring is essential to determine when and where there are sciarids. Control measures are not needed when there are no flies around. This makes treatment cost effective and helps to avoid build up of resistance.
- With any treatment, it is good practice to vary the control strategy at intervals using a rotation of control agents throughout the farm to avoid the possibility of fly resistance.
- Care needs to be taken to ensure that pesticides are stored as per the instructions. Particular care needs to be taken with nematodes as they are living organisms.
- Neither Calypso nor Gnatrol have PSD approval as yet.
- A cost assessment of all the treatments would be needed especially when used at the higher rates or if both a compost and casing treatment was used. The increase in yield might help offset the cost of the control agents.

# SCIENCE SECTION

#### Introduction

Mushroom sciarid flies are important pests of the commercial mushroom, *Agaricus bisporus* (Lange) Imbach. The main species causing damage in the UK are *Lycoriella castanescens* and *Lycoriella ingenua*. The latter is of particular concern because it is extending its range and has started to develop resistance to diflubenzuron (Dimilin), the most widely used chemical for sciarid fly control. The total cost of pest infestations to the UK mushroom industry is estimated at £11-12 million per annum, including direct loss from insect attack, transmission of pathogens and insecticide costs (R. Gaze pers. comm.; DEFRA Basic Horticultural Statistics for the UK, 2001). Sciarids can sometimes cause complete crop failures. Less quantifiable, but often just as important, are 'fly factors' such as nuisance to mushroom pickers, the presence of flies in mushroom pre-packs and flies getting into people's homes adjacent to the farm.

The aim of this project was to investigate an IPM strategy for controlling sciarid fly pests for the UK mushroom industry. With the withdrawal of many insecticides for use in mushroom crops, the need for new methods of control and for a greater choice of control agents to help growers prevent build up of fly resistance, is becoming urgent. At present, the only chemical with approval for use against mushroom sciarid larvae is Dimilin (diflubenzuron) and the most readily available biological control agent is the nematode *Steinernema feltiae*. Efficacy with both of these can be variable, possibly due to incorrect application, different susceptibility to diflubenzuron in sciarid populations and to insufficient grower experience in use of biological agents (R. Gwynn, pers. comm.).

Where infestations of sciarids start early in the crop, and when sciarid numbers are high, it is important to get good control of the first generation. Sciarids can infest the compost immediately after pasteurisation, at spawning and through the early stages of crop growth, and where infestation is occurring at these stages, it can result in huge second and third generations of sciarids. To reduce the number of sciarids, it would be beneficial to target the first generation of sciarid larvae and therefore to use control agents in the compost. There is no control at present for use against the first generation sciarid larvae in compost. Sciarids need about 17 days to develop from eggs into flies at 25°C so flies infesting the compost at or

before spawning lay eggs that mature into the first generation of sciarids that emerge shortly after casing. As each female sciarid can lay 100-200 eggs (Binns, 1973) reducing the number of flies from the first generation would be a valuable step in sciarid control.

The objectives of the project were as follows:

- 1. To evaluate the susceptibility of sciarid flies to a range of control agents in the laboratory, including novel insecticides and biological control agents, applied to both mushroom compost and casing (target date November 2004).
- To assess selected control agents for their ability to control populations of sciarid flies in a crop scale experiment (target date May 2005).
- 3. To evaluate a range of dose rates for Gnatrol and Nemasys M when used as a compost treatment and to determine efficacy of Calypso as a drench on compost (target date September 2005).
- To devise and evaluate an integrated pest management strategy which is intended to reduce the selection pressure for the development of resistance to chemical insecticides (target date March 2006).
- 5. To transfer knowledge to the industry (target date May 2006).

#### **Objective 1: Laboratory evaluation of control agents against** *L. ingenua*

#### Materials and methods

#### Lycoriella ingenua cultures

Cultures of *L. ingenua* originated from a grower's holding and were reared on a mixture of 25 g soya flour (The Health Store, Coventry, UK) and 500g Irish moss peat (35 % moisture content) (Vitax, Coalville, Leicestershire, UK) within a plant propagator (25 x 18 x 20 cm, Stewart Plastics, Croydon, UK) at 25°C in darkness. The lid of each propagator was vented with two 2 cm diameter holes plugged with cotton wool. Cultures were initiated by introducing 50 gravid adult female *L. ingenua* into a propagator using a pooter. The filial generation of adult *L. ingenua* emerged in approximately 21 days.

A method for obtaining eggs of *L. ingenua* was adapted from Binns (1973). Gravid adult female *L. ingenua* (180 - 220) were incubated for 3 d at 25°C within an oviposition chamber, which consisted of a cylindrical plastic frame (5 cm high x 6 cm diameter) covered in fine

mesh nylon gauze, placed on the base of a Petri dish (7.5 cm diameter) which contained 25 g Vitax Irish moss peat (35% moisture content). The oviposition chamber was placed on moistened tissue paper in a plant propagator (23 cm high x 9 cm diameter, Stewart Plastics, Croydon, UK) to maintain a humid atmosphere. Eggs were extracted by flotation on water.

#### Mushroom cultivation substrates

Phase II mushroom compost was prepared at the Warwick HRI mushroom unit. Casing treatments comprised moist moss peat (35% moisture content) supplemented with 5% soya flour as a food source for sciarid larvae. Note that bioassays were not done using compost inoculated with *A. bisporus*, as high levels of mycelium are known to inhibit sciarid development (Binns, 1975), which could confound the results of experiments.

#### Laboratory bioassay

A bioassay was done to evaluate the effects of seven control agents (Table 1) on the survival to adulthood of L. ingenua eggs. Control agents were applied at the manufacturer's recommended rate (Table 1). The treatments were applied to both compost and casing substrates as drenches and by mixing (= admix). Controls were treated with distilled water. For drench treatment of compost, 100 g of compost was pressed into plastic pots (10 cm high x 8 cm diameter, with white polythene lids vented with a 1 cm hole plugged with cotton wool (A W Gregory & Co. Ltd., London). Treatments were then drenched onto the compost surface using a Gilson pipette at a rate of 9 ml per pot (equivalent to 1 L  $/m^2$ ). For admix treatment of compost, treatments were mixed into compost at a rate of 9 ml per 100g compost. Compost was then pressed into plastic pots (as above) at a rate of 100 g per pot. For drench treatment of casing, 25 g casing substrate was pressed into plastic pots (8 cm high x 6 cm diameter, vented lids as above). Treatments were then drenched onto the casing surface using a Gilson pipette at a rate of 5 ml per pot (equivalent to 1  $L/m^2$ ). The casing was then pressed down using a second pot until 3 cm deep. For admix treatment of casing, treatments were mixed into casing at a rate of 5 ml per 25 g casing substrate. The substrate was then placed into plastic pots (as above), 25 g per pot, and pressed to a depth of 3 cm. Lycoriella ingenua eggs were pipetted onto the surface of cultivation substrates at a rate of 25 per pot for casing and 50 per pot for compost. For drench treatments, eggs were applied either three days before or three days after the drench was applied. This was done to prevent eggs becoming immersed in liquid suspensions of the control agents at the substrate surface, as previous work has indicated that this can give distorted results. For admix treatments, the

eggs were applied three days after the application of the control agents. Each pot was covered with a vented sticky trap (Oecos 10 cm<sup>2</sup> with 1 cm hole in the centre plugged with cotton wool) and incubated at 25°C darkness in a controlled environment room for 17 days, after which the sticky traps were inspected daily for 14 days and the number of adult *L. ingenua* caught on each trap was recorded. There were three replicate pots of each treatment and three controls, and the experiment was repeated three times.

Following this, a second a laboratory bioassay was done which provided a compound measure of the oviposition of adult female *L. ingenua* and the subsequent development and survival of their offspring to adulthood. Control agents were admixed into cultivation substrates as described previously. For each treatment, six replicate pots were placed in a plant propagator (Stewart Plastics, Croydon, England, 25 x 18 x 20 cm). Thirty gravid female *L. ingenua* collected with a pooter from a culture 1 - 2 days after emergence were transferred to each propagator. After four days the pots were covered with a vented sticky trap and incubated at 25°C in darkness in a controlled environment room for 17 days, after which the sticky traps were inspected daily for 14 days and the number of adult *L. ingenua* caught on each trap was recorded. The experiment was repeated on three separate occasions.

Treatment	Active ingredient	Application rate
AGARIguard (ECOspray Ltd.)	garlic	1% substrate weight
Dimilin (Uniroyal)	diflubenzuron	30 ppm a.i. 40%
Decis (AgrEvo)	deltamethrin	3 ml product / 100 m <sup>2</sup>
Gnatrol (Valent Biosciences)	Bacillus thuringiensis	3 lbs /1000 ft <sup>2</sup> (= 1465 g / 100 m <sup>2</sup> )
Nemasys M Becker Underwood)	Steinernema feltiae	3,000,000 / m <sup>2</sup>
Spinosad (Dow)	spinosad	30 ppm (a.i. 48 %)
Thiacloprid granules (Bayer)	thiacloprid	600 ppm product

Table 1: Control agents evaluated in laboratory experiments against L. ingenua

#### Results

Results of the bioassays are given in Tables 2 and 3.

<u>AGARIguard</u> gave poor control in both compost and casing when eggs were introduced regardless of time of treatment. As previous experiments had shown the garlic treatment to be effective when applied on the same day as the egg infestation, this suggests that the garlic has poor persistence. As there was also poor control in these bioassays when the drench treatment

was applied three days after egg infestation, it is possible that the larvae had migrated from the substrate surface before the treatment was applied. However, when the substrates were infested with flies, the control given by AGARIguard was higher (72.7% in casing and 84.5% in compost). This suggests that: (a) a large proportion of eggs were oviposited while AGARIguard was still active; and/or (b) it may have a negative effect on oviposition. The latter could be useful if used on compost immediately post pasteurisation, at spawning or on the casing.

Decis gave poor control in casing and compost throughout.

<u>Dimilin</u> was effective against sciarid larvae in casing and compost but in commercial practice its use is limited to casing treatment as it is otherwise too expensive. In these bioassays a Dimilin susceptible culture of the mushroom sciarid *Lycoriella ingenua* was used. Where the sciarid population has become resistant, Dimilin will not be effective.

<u>Gnatrol</u> gave good control of *L. ingenua* in both casing and compost. Previous work on the Warwick HRI mushroom unit showed that Gnatrol only gave good control of *L. castanescens* when used at double the recommended dose rate (Valent Biosciences commercial report), which may indicate that this species exhibits less susceptibility than *L. ingenua*, although caution is required as the two experiments were done on very different spatial scales. Crop scale experiments in the USA have indicated that Gnatrol is effective against *L. ingenua* (Keil 2002) and hence further investigation would appear to be warranted.

<u>Nemasys M</u> was effective as a control measure in both casing and compost. However, it has been suggested that colonised compost is too dry for nematode activity (Roma Gwyn, agricultural consultant, personal communication). As control of the first generation of sciarids is most important to prevent a large infestation in the crop, Nemasys M might have a window of opportunity for control of this generation before the mycelium fully colonises the compost. In practice, growers often use Nemasys M as a split dose at casing and then again between mushroom flushes. It could be worthwhile to investigate sciarid control using the split dose to compost at spawning and to casing so that the compost is protected from the larvae from the start of a crop.

<u>Spinosad</u> gave poor control in casing throughout. With compost it gave good control both when used as a drench treatment three days before egg infestation and when used as an admix before infestation with flies. However, it gave poor control when admixed into the compost and when used as a drench three days after egg infestation. Thus eggs laid in the compost a

few days after an admix treatment or eggs already in the compost when a drench treatment was applied would not be controlled.

<u>Thiacloprid</u> granules gave good control, apart from the compost admix treatment with egg infestation.

**Table 2.** Susceptibility of L. ingenua to different control agents applied to compost and casing. The results are given as % control compared to untreated substrate.

	% control							
		Cas	sing		Compost			
Infestation type:	Egg			Adult	Egg			Adult
Introduced:	Day 0	Day 3	Day 3	Day 0	Day 0	Day 3	Day 3	Day 0
Application:	Drench	Drench	Admix	Admix	Drench	Drench	Admix	Admix
Applied at:	Day 3	Day 0	Day 0	Day 0	Day 3	Day 0	Day 0	Day 0
AGARIguard	11.5	5.2	2.3	72.7	41.1	7.0	14.2	84.5
Decis	22.4	38.3	8.1	64.1	37.8	47.8	22.6	32.8
Dimilin	79.8	94.2	94.2	97.7	68.6	85.9	83.8	89.1
Gnatrol	87.3	98.8	100	93.8	80.7	99.4	68.7	75.6
Nemasys	95.9	96.5	96.5	95.6	96.7	91.9	94.8	90.7
Spinosad	39.2	8.1	4.0	-11.0	25.8	89.6	36.2	77.6
Thiacloprid	88.5	95.9	94.9	95.9	68.8	87.8	37.1	79.4

 Table 3. Mean % control from all treatments

	AGARIguard	Decis	Dimilin	Gnatrol	Nemasys	Spinosad	Thiacloprid
Casing	22.9	33.2	91.5	95.0	96.1	10.1	91.3
Compost	36.7	35.3	81.9	81.1	93.5	57.3	68.3

# **Objective 2:** Crop scale evaluation of control agents in casing and compost against *L. ingenua*

#### Materials and methods

From the results of Objective 1, Gnatrol, Nemasys and thiacloprid were selected for evaluation on a crop scale and used at the manufacturers' recommended rates for casing (N). (The compost has a larger volume than the casing, thence could require a larger total amount of control agent to be applied to it. However, this has to be balanced against the greater cost

that would be entailed. In addition, some control agents are applied on a per area basis, rather than a per volume basis). As the thiacloprid granules had been used in solution for the laboratory experiments, Bayer decided it would be best to use it in the form of Calypso at an equivalent dose rate = 150 ppm product. The compounds were applied to casing and compost as separate treatments. One tray of Gnatrol was included at 1.5N. In accordance with commercial practice the treatments were mixed into compost at spawning or applied to casing as drenches. Dimilin was also included as a standard casing treatment and the experiment included untreated controls. L. ingenua cultures were prepared as described previously. Trays of HRI mushroom compost (50 kg per tray) were spawned with Agaricus bisporus (Sinden A15) spawn at 0.5% w/w. Eight trays were spawned for each treatment with a further eight for the untreated control. Compost admix treatments were added at spawning. All the trays were covered with muslin held in place with staples and then sealed around the tray with 5 mm wide paper tape. The muslin was used to contain sciarid flies for the infestation and to ensure that equal numbers of flies were used to infest each tray. The trays were arranged in a random order in one growing chamber and 10 gravid female sciarids introduced into each corner under the muslin over each tray (40 flies per tray). The small holes made in the muslin to allow fly entry were plugged with cotton wool. The cropping chamber was operated according to Warwick HRI standard procedure (HRIW027). The trays were left covered until day 18 when the muslin was removed and the trays cased. The trays for each treatment were then separated into a different house to avoid emerging flies confounding results between treatments. Where necessary, casing treatments were applied by drench and then an inverted sticky trap (Oecos, Ltd, Kimpton, Hertfordshire UK, 10 cm x 20 cm) was placed on the casing of each tray to trap a sample of emerging flies. Fly numbers were recorded regularly throughout the life of the crop as well as yield and quality of mushrooms and the results were analysed by ANOVA.

#### Results

Data for the effect of control agents on the numbers of adult sciarid flies trapped from mushroom trays are shown in Table 4. There was a significant difference (P < 0.001) in sciarid emergence between treatments. None of the replicated treatments gave good control of the first generation of sciarids (all less than 35% control compared to the untreated trays). Control of the second generation was better in all treatments. Control of the second generation with Calypso was less in the compost than the casing, however Nemasys M performed equally well in compost and casing. Although Gnatrol gave only about 50%

control in compost and casing, the 1.5x dose applied to the compost gave the best final control (care has to be taken with this result as it was not replicated) so it is possible that sciarid control from a casing treatment could have been improved if a higher concentration had been used.

		Number of sciarids per tray				% control	
			Generati	on	Generation		
Treatment	Substrate	1st	2nd	Total	1st	2nd	Final
Control		632	2887	3519			
Calypso	compost	480	1555	2036	24.0	46.1	42.2
Gnatrol	compost	423	1397	1820	33.0	51.6	48.3
Gnatrol x 1.5	compost	133	602	735	79.0	77.1	77.5
Nemasys	compost	506	819	1325	20.0	71.6	62.3
Dimilin	casing	991	569	1559	-3.3	80.3	65.3
Calypso	casing	432	842	1274	31.7	70.8	63.8
Gnatrol	casing	618	1554	2172	2.3	46.2	38.3
Nemasys M	casing	439	772	1211	30.6	73.2	65.6

 Table 4. Sciarid emergence per tray and percent control compared with untreated compost.

Data for mushroom yields are shown in Table 5. There was a significant difference (P < 0.001) in yield between treatments. The untreated control had the lowest mean yield (10.815 kg/tray) and the highest percent of damaged mushrooms (19.3%). Compared with the yield from trays with the standard sciarid treatment Dimilin, Gnatrol in both compost and casing, Calypso in casing and Nemasys in compost increased the yield of mushrooms (4.8%, 9.8%, 9.8% and 2.5% respectively). There were significant differences (P < 0.001) in the percentage of damaged mushrooms between treatments, but there was no correlation between damage and sciarid control apart from the untreated control trays which had the highest percent of fly damaged mushrooms and the largest emergence of sciarids. This may be because there was some sciarid control by all treatments and sciarid larvae tend to burrow into the stipes only when the infestation level is very high (Hussey & Gurney 1968).

Treatment	Substrate	Yield (kg)	% damaged	% buttons	% closed	% open
		per tray	by sciarids			
Control		10.815	19.3	7.5	55.5	17.7
Calypso	Compost	11.116	11.9	5.3	60.9	22.0
Gnatrol	Compost	12.307	5.7	3.3	54.5	36.6
Nemasys	Compost	12.042	7.4	5.3	54.5	32.8
Gnatrol x 1.5	Compost	13.478	4.1	7.4	68.8	19.6
Dimilin	Casing	11.745	9.8	8.6	66.5	15.2
Calypso	Casing	12.897	0.6	7.5	37.8	54.0
Gnatrol	Casing	12.903	3.4	8.1	48.8	39.7
Nemasys	Casing	11.170	5.4	7.7	54.4	32.5

Table 5. Yield and quality of mushrooms per tray from all treatments.

## Conclusions

Good sciarid control was achieved by treating the casing with Dimilin, Calypso and Nemasys M, and by treating the compost with Nemasys M. Apart from the unreplicated compost treatment with Gnatrol at 1.5x rate, none of the treatments gave good control of the first generation of sciarids. This may have been caused by the fact that the compost has a much larger volume compared to the casing and therefore needs an increased dose of the control agent. Further investigation of Gnatrol at a higher concentration would be warranted, given that it may give good levels of control and, critically, be able to control the first generation of sciarid flies. Further investigation is also warranted of the effects of Nemasys M in combination with other treatments or used as a split dose in compost and casing. A split dose used in casing has been shown to be more effective than a single dose. The extra cost of applying a compost treatment at a higher dose might be partially offset by the increase in yield of mushrooms, and could be valuable where there is heavy pest pressure used in conjunction with a casing treatment.

# **Objective 3: Dose response of Nemasys M and Gnatrol in compost and evaluation of Calypso as a compost drench treatment against** *L. ingenua*

#### Materials and methods

#### Mushroom unit trial

The aim of this semi-crop scale experiment was to evaluate the effect of using different dose rates of control agent on sciarid control in compost. The data could then be used to identify dose rates for use in compost on a crop scale. Mushroom compost was infested with gravid female sciarids on two occasions to simulate natural infestation conditions. HRI Phase II mushroom compost was placed into large polythene bags (25 kg per bag, 1 bag per treatment) and sealed with a cotton wool plug. Fifteen females were introduced into each bag and the bags were left on the floor of a growing chamber for three days. The compost was then spawned with Agaricus bisporus (Sinden A15) spawn at 0.5% and placed into pots (plastic plant pots 25 cm x 30 cm diameter, 2.5 kg compost per pot). There were eight replicate pots for each treatment, including the untreated control. All treatments were applied to the compost immediately after spawning as a drench using a spray bar attached to a Zipette (Jencons Scientific, Bedfordshire, UK. 50 ml). Nemasys M and Gnatrol were applied at different rates (0.75N, N, 1.5N, 2N and 4N) where N represents the manufacturer's recommended rate (Nemasys M, N = 3 x  $10^{6}/m^{2}$ , Gnatrol, N = 14.6 g/m<sup>2</sup>). In addition, one treatment of Calypso was included at the manufacturer's recommended rate (150 ppm w/w product substrate). This was one to follow up the results from the laboratory experiments where Calypso had been more effective as a drench treatment than when admixed. The recommended rate was used in spite of the volume of compost being larger than that of the casing for cost and residue implications.

The pots were enclosed in polythene bags to contain the sciarids used to infest the compost and to avoid cross infestation of the filial generation of sciarid flies between treatments. The bags were sealed with a rubber band around a 5 cm wide plug of non-absorbent cotton wool to allow gaseous exchange. The pots were arranged in a random order on shelves in the growing chamber and, after two days, 5 gravid female sciarids were introduced into each bag. The cropping chamber was operated according to Warwick HRI standard procedure (HRIW027). After 18 days the pots were cased and an inverted sticky trap (Oecos, Ltd, Kimpton, Hertfordshire UK, 10 cm x 20 cm) was placed on the casing of each to trap the emerging flies.

During the course of the experiment, carbon dioxide levels within the bags rose above the normal cropping level (ca. 1000 ppm). Consequently, holes were cut in the top and bottom of the bags, and one day after this the bags were opened. The high  $CO_2$  delayed mushroom development and there is a risk that it may have affected the activity of the control agents. It was decided, therefore, to continue the experiment to record the second generation of sciarids.

A regression analysis was done on the relationship between the numbers of adult sciarid flies counted on sticky traps (transformed to the  $log_{10}$  scale) and the dose rate of the control agents, which was expressed as multiples of N. A test for curvature on the log transformed data was carried out but proved insignificant. Estimates were made from the regression model of the dose rates required to give 50%, 75% and 95% control of sciarid populations, 75% being considered effective control.

#### **Results and Discussion**

Data on the effect of the control agents on the numbers of adult sciarid flies trapped from mushroom pots are shown in Table 6.

Control acout	Sciarid		Dose rate						
Control agent	generation	0	0.75 N	Ν	1.5N	2N	4N		
Untreated	1st	54	-	-	-	-	-		
Untreated	2nd	378	-	-	-	-	-		
Calypso	1st	-	-	10	-	-	-		
Calypso	2nd	-	-	89	-	-	-		
Gnatrol	1st	-	29	20	13	32	16		
Gnatrol	2nd	-	254	273	323	332	326		
Nemasys M	1st	-	41	52	38	20	7		
Nemasys M	2nd	-	208	163	179	151	79		

**Table 6.** Mean numbers of sciarids caught on sticky traps placed over mushroom compost

 treated with control agents at different rates

Manufacturer's recommended rate (N) as follows:

Calypso N = 115 ppm w/w product/substrate;

Gnatrol N = 14.6 g/m<sup>2</sup>;

Nemasys N =  $3 \times 10^6 / m^2$ .

<u>Calypso</u> gave good control of both 1st and 2nd generation sciarids (82% and 76% respectively).

#### Gnatrol

There was a significant effect of dose rate on the emergence of the 1st generation of sciarids (p=<0.01) but not for the 2nd generation. From the regression analysis, the dose rates needed for 50%, 75% and 95% control of the 1st generation were 0.2 N, 2.8 N and 7.5 N respectively, where N = 14.6 g m-<sup>2</sup>. However, emergence from the 2N rate was unexpectedly high and this will have influenced the results of the analysis, raising the estimated dose rate needed for effective control. None of the dose rates gave good control of the second generation based on these findings. It would appear that Gnatrol does not give persistent control in mushroom compost and therefore a supplementary treatment to the casing would be required.

#### Nemasys M

There was a significant effect (p=< 0.01) of dose rate on control of sciarid emergence for both 1st and 2nd generations. The estimated dose rates needed for 50%, 75% and 95% control of the 1st generation were 1.3 N, 2.4 N and 5.2 N respectively, where N = 3 x  $10^6$  m<sup>-2</sup>. For control of the second generation, rates of 1.1 N (50% control), 2.8 N (75%) and 7.0 N (95%) were estimated.

The numbers of sciarids from the control pots were lower than expected and this may have artificially reduced apparent percentage control given by treatments. In the first crop scale trial (Objective 2), Nemasys M (N) mixed into the compost gave better sciarid control of the 2nd generation (72%) than observed here. In this experiment, the regression analysis showed that the recommended rate (N) gave 49% control of sciarids from the 2nd generation. It is also possible that the nematodes are more effective when mixed into the compost than when applied as a drench.

The high levels of  $CO_2$  created by containment of the pots in polythene bags may have influenced the results of this experiment. However, the bags were opened soon after emergence of the first generation of sciarids and  $CO_2$  levels quickly returned to normal.

#### Conclusions

Calypso gave good control of first and second generation sciarids when used as a compost drench.

Gnatrol gave good control of the first generation of sciarids. Our analysis showed that for 75% control of the first generation, the dose rate would need to be 2.2N. However, Gnatrol did not persist to control the second generation even at higher dose rates. This could mean that, in a crop, any sciarids surviving from the first generation, or sciarids infesting after casing, would not be controlled. If used as a compost treatment to control some first generation sciarids, it would need to be supplemented with a casing treatment which may not be economically feasible.

Overall, Nemasys M also gave encouraging levels of control. Our results indicate that 2.4 and 2.8 times the recommended dose rate would be required to give 75% control of first and second generation sciarids respectively. We would consider this to be a reasonable level of sciarid control. It is noteworthy that the nematodes applied to compost continued to control the second generation of sciarid larvae, which result from eggs laid into the casing layer. The cost of the increased dose rate could make this treatment too expensive. However, there is an increased yield associated with the use of Nemasys M in compost and, where there is heavy pest pressure, it might be worthwhile to break the cycle of sciarid infestation.

The extra cost involved in using Gnatrol and Nemasys M at the higher rates could be offset by the higher yields these treatments are known to produce. In this trial, mushroom yield was not assessed but the previous HDC trial showed that compost treated with Gnatrol (N & 1.5N) and Nemasys M (N) and yielded 13.8%, 24.6% and 11.3% more mushrooms respectively than the untreated compost.

# Objective 4: Second crop scale evaluation of control agents against *L*. *ingenua*

#### Introduction

This objective was to evaluate Calypso, Gnatrol and Nemasys M as separate compost and casing treatments at the most effective rates determined from the previous experiments. The aim of the compost treatments was to control the first generation of sciarids and determine whether the treatment exerted lasting sciarid control. The best method of application for each control agent had been identified previously in this project and also in laboratory experiments using Calypso (Bayer funded). The final dose rate for each treatment was chosen to reflect acceptable sciarid control balanced with cost.

#### Materials and methods

The dose rates for Calypso, Gnatrol and Nemasys M and methods of application used are shown in Table 7. Gnatrol and Nemasys M were mixed into compost at spawning, Calypso was drenched onto the compost immediately after spawning and the casing treatments were all applied as drenches immediately after casing. Dimilin was included as a standard casing treatment and the experiment included untreated controls. Lycoriella ingenua cultures were prepared as described previously. Trays of HRI mushroom compost (50 kg per tray) were spawned with Agaricus bisporus (Sinden A15) spawn at 0.5% w/w. Eight trays of compost were spawned for each treatment and 16 trays for the untreated controls. The compost treatments were separated into individual cropping houses for infestation to avoid any deterrent effects that may be caused by the treatment and the trays of untreated compost were arranged in a random order in another cropping house. Twenty five female sciarids were introduced onto each tray over a period of 10 days. The flies were partially contained by aspirating them from culture into a small plastic pot (8 x 6 cm, A W Gregory & Co. Ltd., London) which was then inverted over the compost. The cropping chambers were operated according to Warwick HRI standard procedure (HRIW027). The trays were cased after 17 days and the casing treatments applied by drench. The trays were then separated into three cropping houses - compost treatments were placed in one, casing treatments in a second and the untreated controls in a third. This was done to avoid emerging flies confounding results between the untreated controls and the compost and casing treatments. An inverted sticky trap (Oecos, Ltd, Kimpton, Hertfordshire UK, 10 cm x 20 cm) was placed on the casing of each tray to trap a sample of emerging flies. As the sticky side was face down, it was assumed that the majority of the flies caught would come from the substrate beneath it and not from passing flies from a different treatment. As a check that the numbers of flies caught were a true reflection of sciarid emergence from the treatment beneath the trap and not from other treatments, the sticky traps on the top trays of each treatment were covered with a net fly cover. It was impractical to cover all the traps as the nets seriously impeded watering and crop harvesting. Fly numbers were recorded over time as well as yield and quality of mushrooms. The emergence data was subjected to a square root transformation and analysed by ANOVA. The yield and mushroom crop quality data was treated by ANOVA.

Treatment	Compost tre	atment	Casing treatment		
Traiment	Application	Dose rate	Application	Dose rate	
Dimilin	-	-	Drench	30 ppm a.i. (N)	
Calypso	Drench	150 ppm product (N)	Drench	150 ppm product (N)	
Gnatrol	Admix	2198 g / 100 m <sup>2</sup> (1.5N)	Drench	2198 g / 100 m <sup>2</sup> (1.5N)	
Nemasys M	Admix	6,000,000 / m <sup>2</sup> (2N)	Drench	3,000,000 / m <sup>2</sup> (N)	

Table 7: Dose rates of control agents evaluated in the unit trial against L. ingenua

#### **Results and discussion**

#### Sciarid emergence and treatment control:

Data for the effect of control agents on the numbers of adult sciarid flies trapped from mushroom trays are shown in Table 8 and Figs. 1a and 1b. There was a significant difference (P < 0.001) in sciarid emergence between treatments and a significant effect of tray position (= < 0.001). Emergence was greatest from the highest trays on the stack and smallest from the lowest trays. None of the replicated compost treatments gave very good overall sciarid control nor good control of the first generation of sciarids (from 30.1% to 50.1% control compared to the untreated trays). This may be due to the non-homogenous nature of the compost which makes it difficult to ensure there are no pockets of untreated material in which sciarid larvae can survive. This might explain the difference between this experiment and the smaller scale multiple dose experiment. With the casing treatments, overall sciarid control was 76.5% Dimilin, 64.1% Calypso, 55.6% Gnatrol and 68.2% Nemasys M. The casing treatments did not give good control of the first generation of sciarids but there was no significant difference in first generation control between compost and casing treatments. It is possible that the casing treatment can control less mature sciarid larvae from the first generation as they migrate towards the surface to pupate. However, sciarid larvae that infested the compost early on in the crop cycle would already have formed protective pupae and would therefore not be killed by the control agent. This is particularly the case where the control agent is an insect growth regulator e.g. Dimilin.

		No. sciarids per tray		Amplused	% control		
		Generation		Analysed data *	Generation		
Treatment	Substrate	1 st	2nd	uuu	1st	2nd	Overall
Control	-	240.2	1312.4	-	-	-	-
Calypso	Compost	119.9	742.3	29.16	50.1	44.5	44.5
Gnatrol	Compost	141.6	1002.6	33.48	41.0	23.6	26.3
Nemasys M	Compost	153.1	820.5	30.93	36.2	37.5	37.3
Dimilin	Casing	129.6	235.0	18.79	46.0	82.1	76.5
Calypso	Casing	168.0	388.8	23.21	30.1	70.4	64.1
Gnatrol	Casing	128.6	561.0	25.84	46.4	57.3	55.6
Nemasys M	Casing	123.1	370.0	22.10	48.7	71.8	68.2
				lsd=3.23			

**Table 8**. Sciarid emergence per tray and percent control compared with untreated compost.

\* Analysed data was square root transformed; p=5%; df=61

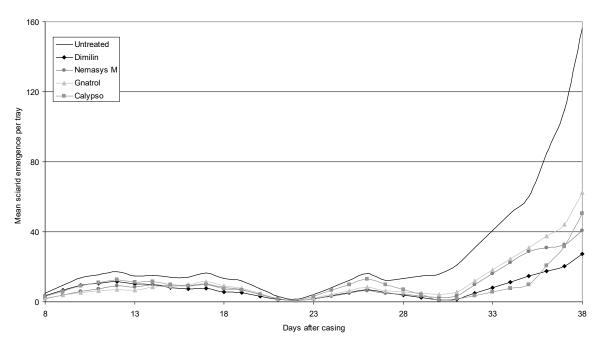
#### Yield of mushrooms:

Data for mushroom yields are shown in Table 9. There was a significant difference (P < 0.005) in yield between treatments but no significant effect of tray position on overall yield. All three compost treatments gave significantly (P < 0.005) higher mean yields than the other treatments. The Dimilin treatment and the untreated controls had the lowest mean yield (9.4 and 10.1 kg/tray respectively).

		Total yield (kg)	% Buttons	% Closed	% Open	
Untreated		10.11	13.5	34.7	51.8	
		11.50	16.7	52.0	20.4	
Calypso	Compost	11.56	15.7	53.8	30.4	
Gnatrol	Compost	12.28	16.0	52.9	31.1	
Nemasys M	Compost	11.88	15.9	53.9	30.3	
Dimilin	Casing	9.42	13.2	67.2	19.7	
Calypso	Casing	10.52	12.1	24.0	63.9	
Gnatrol	Casing	11.08	13.4	56.0	30.6	
Nemasys M	Casing	10.33	14.0	64.3	21.7	
p=<5% lsd=1.54; df=61						

Table 9. Yield (per tray) and grade (%) of mushrooms from all treatments.

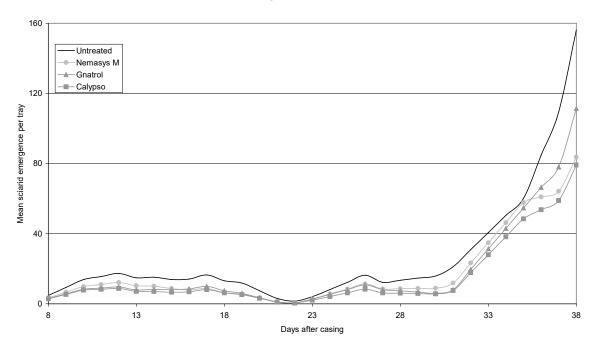
Fig. 1a. Mean sciarid emergence from casing treatments. (Data subjected to a weighted moving average).



Sciarid emergence - casing treatments

Fig. 1b. Mean sciarid emergence from compost treatments. (Data subjected to a weighted moving average).

#### Sciarid emergence - compost treatments



#### Mushroom quality:

The compost treatments gave the highest percentage of button mushrooms (16%) but this was not statistically significant. There was a significant effect of treatment on the yield of closed and open mushrooms (P <0.001). The casing treated with Dimilin, Gnatrol and Nemasys M gave more closed cup mushrooms (67, 56 and 64% respectively) but the casing treated with Calypso and the untreated controls had the lowest quality of mushrooms (64% and 52% open cups). There was a highly significant effect of tray position on yield of button mushrooms (p <0.001), the yield being greater on the lower stacks. The stack position did not significantly influence yield of closed and open mushrooms.

#### Conclusion

Good sciarid control was achieved by treating the casing with Dimilin (77%). Calypso gave effective control in casing (64%) but was not effective as a compost treatment nor did it give control of the first generation of sciarids. Gnatrol at 1.5 x the recommended rate did not give effective sciarid control in either casing nor compost and would need to be used at a higher dose rate to effect adequate control. However, in the first crop scale experiment, Objective 2, good control of was achieved with a compost treatment of Gnatrol at this rate (78% control) and in previous work funded by Valent Biosciences, Gnatrol in casing at 2N gave good overall control of the mushroom sciarid *L. castanescens* (72%). Treating a crop at this rate would probably be expensive but the yield of mushrooms from compost treated with Gnatrol was significantly higher than the untreated control, and that could offset the cost of treatment. Nemasys M gave effective sciarid control when applied to casing (68%) but was not effective as a compost treatment in this trial even though used at double the recommended rate. This was different from the results from both the previous experiments (Objectives 2 and 3).

#### Discussion

#### Calypso

Throughout the experiments in this project, Calypso has given effective control of sciarids when used as a casing treatment. If this can be taken forward and granted approval for use in mushrooms, its will give growers another tool in their armoury against mushroom sciarids and will help to reduce the risk of sciarid resistance by the continual use of one compound. As a compost treatment Calypso did not give effective control at the recommended rate.

#### Gnatrol

Gnatrol did not give effective sciarid control when used at the recommended rate in compost but at 1.5 x the recommended rate, the results from the compost treatments were variable between experiments. In both the unreplicated tray in the first crop scale experiment and in the dose response trial (Objective 3), it gave > 70% control of the first generation sciarids. However, in the second crop scale trial (Objective 4) the level of control fell to 41%. The level of control remained constant throughout the life of the crop in the first crop scale experiment but fell in the other two trials. Therefore this effect was probably due to a consequence of the reduced numbers from the first generation rather than persistence of the active ingredient in the compost.

Gnatrol was not effective as a casing treatment at either the recommended rate nor at 1.5 x the recommended rate. However, in previous work carried out at HRI, Gnatrol at 2N gave good control in casing (72%). In the USA, Gnatrol is being used commercially by mushroom growers for sciarid control. The reasons for the variability in results in the present study, and between the results from the present study and previous studies, are not known. It may have been caused by variation in the control agent, the sciarid population or the experimental conditions. Overall, it appears that Gnatrol has potential for sciarid control but more work is required on it.

Yield of mushrooms from compost treated with Gnatrol in both experiments was significantly higher (p = <0.05) than the untreated control (Table 11).

#### Nemasys M

The efficacy results from Nemasys M between these experiments are conflicting, although some variability might be expected with a biological control agent. In the initial crop scale trial (Objective 2), first generation sciarid control with Nemasys M (N) in compost was poor (20%) but it improved to give 72% control of the second generation, giving overall control of 62% (Table 10). In the dose response trial (Objective 3), Nemasys M (2N) gave >60% control of both first and second generation sciarids in the compost, thus showing persistence throughout the crop. In the second crop scale trial however, even though it was used at twice the recommended rate (2N), the level of control was poor (<40%) throughout the experiment. This variation in results may have been caused by variation in experimental conditions or in the control agent and further work is warranted. Overall, Nemasys M appears to have

potential as a compost treatment in some situations, for example, when a sciarid infestation on a farm is particularly high.

Nemasys M (N) gave consistent, effective sciarid control when applied to the casing (Table 10).

Yield of mushrooms from compost treated with Nemasys M in both experiments was significantly higher (p = <0.05) than the untreated control (Table 11).

 Table 10. Summary of the percentage control of treatments in the three crop-scale

 experiments

Treatment	Substrate	Crop scale trial 1		Dose response trial		Crop scale trial 2	
Treatment		1st gen	Overall	1st gen	Overall	1st gen	Overall
Calypso(N)	compost	24.0	42.2	82.3	79.3	50.1	44.5
Gnatrol(N)	compost	33.0	48.3	62.9	32.1	-	-
Gnatrol(1.5N)	compost	79.0	77.5	76.2	22.2	41.0	26.3
Nemasys M(N)	compost	20.0	62.3	2.3	50.2	-	-
Nemasys M(2N)	compost	-	-	63.2	60.3	36.2	37.3
Dimilin(N)	casing	-3.3	65.3	-	-	46.0	76.5
Calypso(N)	casing	31.7	63.8	-	-	30.1	64.1
Gnatrol(N)	casing	2.3	38.3	-	-	-	-
Gnatrol(1.5N)	casing	-	-	-	-	46.4	55.6
Nemasys M(N)	casing	30.6	65.6	-	-	48.7	68.2

Table 11. Yield of mushrooms (kg per tray) from both crop scale experiments

		Crop scale trial 1	Crop scale trial 2
Untreated		10.82	10.11
Nemasys M(N)	compost	12.04	-
Nemasys M(2N)	compost	-	11.88
Calypso(N)	compost	11.12	11.56
Gnatrol(N)	compost	12.31	-
Gnatrol(1.5N)	compost	13.48	12.28
Dimilin(N)	casing	11.75	9.42
Nemasys M(N)	casing	11.17	10.33
Calypso(N)	casing	12.90	10.52
Gnatrol(N)	casing	12.90	-
Gnatrol(1.5N)	casing	-	11.08

On the basis of these results, compost treatments would need to be supplemented with a further treatment at casing to control a heavy sciarid infestation. Although this would be expensive, the increased yield associated with compost treatments of either Nemasys M or Gnatrol would offset some of the costs and the combined treatment, applied to a succession of crops, should be effective in breaking the cycle of sciarid infestation on a farm. This needs to be thoroughly tested and a cost analysis carried out. At present, Gnatrol does not have PSD approval and Valent Biosciences have not costed it for the UK market.

Some facts to be considered when evaluating these results:

- The sciarid infestation used in these experiments was high and as we infested directly onto each treatment, there was maximum pressure possible for treatments to exert control. In a commercial practice the initial infestation would be far lower and this may affect the effectiveness of control, especially with the biological control agents. In addition we did not knock down the adults at any stage of the trial.
- The culture of *L. ingenua* used in this study was susceptible to Dimilin. Dimilin would not have good control if the fly population was resistant to diflubenzuron.
- The effect of Gnatrol on sciarids has been thought to be species related. However, these experiments were carried out using *Lycoriella ingenua* and in both these and in previous experiments with *L. castanescens*, Gnatrol needed to be used at 2N to give effective sciarid control.

## **Recommendations for IPM against mushroom sciarids**

- 1. Farm hygiene is a basic step to help minimise fly pest problems. Sciarids breed in organic waste, compost and rotting vegetation. Keeping areas around the cropping houses clean and clear from debris, clearing away grass cuttings, removing spent mushroom compost from the farm and storing the materials for compost production as far away as is practicable all reduce possible sciarid breeding grounds.
- 2. Fly infestations should be monitored with sticky traps around the farm to determine where and when sciarids are present.
- 3. Control agents should only be applied when necessary. When there are no sciarids, controls should not be used. This helps to prevent resistance build up.
- 4. A rotation of control agents should be used. This will also help to prevent resistance build up.
- 5. Effective casing treatments to control normal sciarid infestations (i.e. none or very few caught on the traps during the spawn run):

Dimilin (for susceptible sciarid populations)

Nemasys M (N)

Calypso (N) (if given PSD approval)

Gnatrol (2N) (if given PSD approval)

6. For heavy, early or persistent sciarid infestation, possible compost treatments are:

Nemasys M (2N) followed by a casing treatment.

Calypso (N) (if given PSD approval) followed by a casing

treatment.

Gnatrol (2N) (if given PSD approval) followed by a casing treatment.

The treatment should be repeated for a succession of crops until the sciarid population is under control.

7. A cost analysis should be carried out and should take into account:

the value of crop losses if no treatment is done,

the cost of treatment,

extra income from higher yields and better quality mushrooms with some treatments.

# **Technology Transfer**

- HDC news article May 2005.
- HDC news article May 2006.
- Scientific paper will be written at end of project.
- IPM strategy leaflet could be written for growers if funding is available.

## References

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