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#### 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.

...... J A BENNISON Project Leader

Date.....

Report authorised by: .....

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Date.....

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# **GROWER SUMMARY**

# Headline

- Useful control of slugs and snails can be achieved immediately by use of Tex-R ground-cover mattings to prevent their immigration onto uninfested plants.
- The parasitic nematode 'Nemaslug' shows good potential for biological control of slugs and snails and could offer a more effective and environmentally-friendly alternative to slug pellets for control of existing infestations.
- Several novel potential molluscicides were identified.

# **Background and expected deliverables**

Slugs and snails damage a wide range of container-grown HNS species and are difficult to control with slug pellets. Feeding damage is very obvious and causes plant losses or downgrading. This project aims to develop improved, sustainable integrated control strategies for slugs and snails, to reduce plant losses, improve plant quality and satisfy increasing customer demands for ornamental produce showing no physical damage and grown with minimal use of pesticides. Expected deliverables include:

- Identification of the main species of slugs and snails damaging HNS species.
- Identification of plant species most commonly damaged.
- An improved knowledge of the biology of the main slug and snail species so that the optimum timing and components of control strategies can be planned.
- Recommendations for an integrated control strategy, including cultural methods, biological control of existing infestations with a parasitic nematode ('Nemaslug') and use of barriers or repellents to prevent infestations of new plants.

# Summary of the project and main conclusions

# Identification of slug and snail species

The main slug species found on ten selected nurseries throughout England was *Deroceras panormitanum* and the main snail species was the small semi-aquatic snail, *Oxyloma pfeifferi*. Photographs to aid recognition of these species are included in Factsheet 07/02, issued to HNS growers during 2002.

#### Identification of plant species damaged

A wide range of plants were identified on commercial nurseries as being damaged by slugs and snails, including herbaceous and perennial species, alpines, shrubs, grasses and herbs, in plugs, liners and containers, both under protection and outdoors. The snail *O. pfeifferi* was also observed to be associated with algae on the substrate, compost or polythene tunnel coverings. Feeding studies with both *D. panormitanum* and *O. pfeifferi* were carried out, using Hosta and Choisya as representative herbaceous and perennial host plants respectively.

Laboratory tests showed that the snail *O. pfeifferi* grazed the surface of Hosta leaves and stems but did not feed on Choisya. The slug *D. panormitanum* caused leaf holes and severed petioles on Hosta and caused leaf holes and some leaf shredding on young Choisya leaves. It was concluded that *D. panormitanum* damages both herbaceous and tougher-leaved perennial plants. Although *O. pfeifferi* did not damage Choisya, it caused primary damage to other perennial plants e.g. Euonymus and Phormium, to a range of herbaceous plants e.g. Ajuga, lupin, Campanula and Viola and to grasses e.g. Acorus. In some situtations, both *O. pfeifferi* and *D. panormitanum* may survive on a mixed diet of algae, damaged or decaying leaves (including those from perennial plants) and liverworts, although their development and survival was shown to be more successful when a fresh source of suitable plant material is available.

As the snails are active and visible during the day and slugs are mainly nocturnal and tend to hide during the day, the snails may be mistakenly held responsible for slug damage on some plant species e.g. Choisya. However, if present in large numbers on plants or pots, the snails may lead to quality problems as contaminants.

#### Slug and snail biology

The biology of *D. panormitanum* and *O. pfeifferi* was previously almost unknown. Relevant gaps in knowledge of the biology of these species were filled, leading to a much better understanding of both their biology and behaviour which will now enable the optimal timing and components of integrated control strategies to be determined.

The slug *D. panormitanum* was active all year round, with peaks of activity during spring and autumn. Large adult slugs died in late winter/early spring. Egg-laying started in March and large numbers of juveniles were present in April. The minimum temperatures for egg laying and sexual maturation of juveniles were established as 5.4°C and 8.5°C respectively. Under favourable mild, damp conditions, egg laying can continue all summer and the life cycle can be as short as 11 weeks. *D. panormitanum* is well-adapted to the high temperatures and damp conditions present in glasshouses and polythene tunnels, where two or three overlapping slug generations may develop each year. *D. panormitanum* tends to be mainly active at night, hiding

during the day beneath pots and trays and at the base of plants. This tendency for nocturnal activity means that slug numbers are often under-estimated.

The snail *O. pfeifferi* hibernated between September/October and February/March on the sides of pots, on the structure of polythene tunnels and on plants. Snail activity started again during February and most were active by late March. *O. pfeifferi* laid eggs between late March and August, with populations of mixed ages and overlapping generations occurring during this period. Wet conditions stimulated activity and egg-laying and favoured survival. The life cycle from egg to egg was 14 weeks at 20-22°C. *O. pfeifferi* is more active during the day than at night and is easily found on plants and pots, particularly if present in large numbers.

The optimum times for control of both *D. panormitanum* and *O. pfeifferi* are likely to be spring and early autumn. Control in the early spring (March/April) will reduce numbers in any new generation and that in the early autumn will reduce numbers of hibernating snails and over-wintering slugs and reduce damage to evergreen plants. Timing will depend on the control methods used (see sections below).

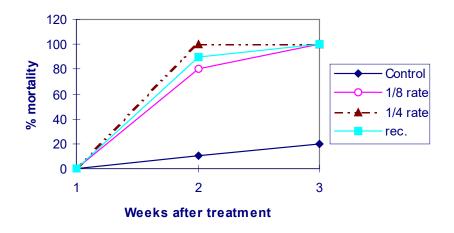
#### **Cultural control**

Experimental work and observations on commercial nurseries have shown that wet conditions favour slug and snail activity and that algae are a source of food, therefore avoiding over-watering will help to reduce infestation levels and damage. Use of Tex-R ground-cover mattings can reduce slug and snail infestations (see section below on barriers/repellents) and these can be even more effective when combined with capillary irrigation. Slugs and snails can feed and shelter amongst weeds, unmarketable plants and trimmed plant material, therefore close attention to nursery hygiene is an important cultural component of an integrated control strategy.

#### Biological control with 'Nemaslug'

In laboratory tests, both full and half-rate 'Nemaslug' killed 80% *D. panormitanum* within four weeks of treatment. Full, 1/4 and 1/8- rates all killed 100% *O. pfeifferi* within three weeks. Two trials comparing different rates of 'Nemaslug' with Draza slug pellets were done on a commercial nursery. None of the treatments significantly reduced numbers of slugs or snails and assessments of relative plant damage were inconclusive. In the first trial during July and August, exceptionally high temperatures are likely to have led to poor nematode survival and efficacy. In the second trial during October, there was a potential problem with nematode quality, although the known avoidance of slugs to nematode-treated compost (as shown in laboratory tests in years 1 and 2) may also have contributed. Further trials with 'Nemaslug' are justified, e.g. to test a 'little and often' approach to application which has recently shown promise in Dutch vegetable crops.

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Mean % mortalities over three weeks in *O. pfeifferi* treated with recommended, 1/8 and 1/4-rates of 'Nemaslug' in laboratory tests.

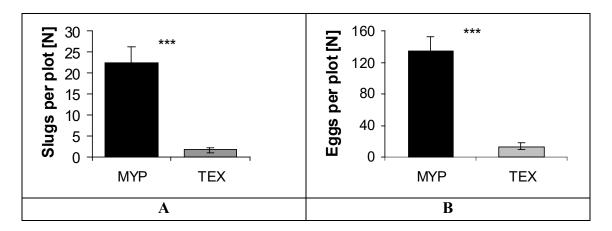
#### Barriers or repellent techniques and novel molluscicides

Fourteen potential barrier, repellent, antifeedant or irritant products were tested in the laboratory against both *D. panormitanum* and *O. pfeifferi*. In addition to their barrier effects, several of these products also killed slugs, reduced egg-laying or survival and delayed egg development. Of the liquid novel molluscicides, cinnamamide, Croptex Fungex and ureaformaldehyde were concluded to be the most promising. Concentrations of 0.1-1% of each product reduced slug and snail activity and damage. Concentrations as low as 0.005% reduced slug egg hatching rates. High concentrations of cinnamamide and ureaformaldehyde killed 100% and 90% of slugs respectively, within 14 hours. Croptex Fungex is already approved as a fungicide and there may be potential for development and registration of copper fungicides as molluscicides. Ureaformaldehyde is used as a slow-release nitrogen fertiliser but its potential formulation and registration as a molluscicide is questionable due to the environmental issue of nitrogen in run-off water. Cinnamamide, an experimental repellent and antifeedant is currently unavailable for commercial use.

Copper-impregnated ground-cover mattings used as root retardants and weed suppressants i.e. Tex-R, Landscape Pro and Supercover Plus, were shown to have significant harmful effects on both slugs and snails in laboratory tests. All reduced slug and snail activity. When slugs and snails were forced to move onto Tex-R, 90% of slugs died and snail activity was reduced by 98%. When given a choice of Tex-R and compost or Mypex, both slugs and snails showed a strong avoidance of Tex-R. In a field trial with *D. panormitanum*, Tex-R greatly reduced slug immigration from the surrounding area. In this trial there were 93% fewer slugs, 90% fewer slug eggs and 68% fewer damaged leaves on plants on Tex-R matting than on those on Mypex matting. In a trial with *O. pfeifferi* on a commercial nursery, there were 36% fewer

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snails on Tex-R plots than on Mypex plots. Here, Tex-R efficacy was reduced by growth of algae on the surface.



Slug infestation of plots in ground-cover matting field trial. A= number of slugs per plot at week 8. B= Accumulated slug eggs per plot until week 8. \*\*\* treatments were significantly different at P < 0.001.

# **Financial benefits**

- During year 1, case studies of slug and snail damage on ten selected HNS nurseries showed that plant losses and downgrading of individual plant species was variable, with estimated plant losses ranging from less than 1 to 100%.
- The average value of containerised HNS plants at point of sale is £6,000 per 100 m<sup>2</sup>. At a conservative estimate of a minimum of 1% losses due to slug and snail damage, these losses represent £60 per 100 m<sup>2</sup>. Losses can be much higher on individual crops. Further losses are incurred in time selecting undamaged plants or trimming off damaged leaves before lifting, and in disposing of unsaleable plants.
- The standard Tex-R ground cover matting costs £128 per 100 m<sup>2</sup> and is likely to be effective in weed and root suppression for ten and five years respectively. HNS growers using Tex-R for the past five years are still observing incidental prevention of slug and snail problems. Assuming Tex-R to be effective for five years, the investment cost per year would be £25.60 per 100 m<sup>2</sup>. Assuming minimum plant losses of 1% due to slug and snail damage, use of Tex-R represents a saving of at least £34.40 per 100 m<sup>2</sup> per crop.
- Until further trials are done on nursery stock species with 'Nemaslug', the cost/benefits of using nematodes for control of existing infestations cannot be confirmed. However, if one application of the current recommended rate of 'Nemaslug' (£19 per 100 m<sup>2</sup>) was shown to give effective control of slugs and snails on HNS, use would lead to a minimum saving of £41 per 100 m<sup>2</sup> per crop.

If Tex-R and 'Nemaslug' were used together in an integrated control strategy, the minimum savings would be  $\pm 15.40$  per 100 m<sup>2</sup>. Should lower rates of 'Nemaslug' prove to give reliable control on commercial nurseries, these would be even more cost-effective.

• Using sustainable, non-chemical methods for slug and snail control would meet growing customer demands for plants produced with minimal use of pesticides.

# Action points for growers

- Maintain good nursery hygiene to reduce shelter and food for slugs and snails. Control algae, mosses, weeds and liverworts as thoroughly and frequently as possible and dispose of unmarketable plants and trimmed plant material promptly.
- Avoid over-watering as wet conditions favour both slug and snail activity and also the growth of algae, mosses and liverworts, on which they feed.
- Remove any slugs and snails on bought-in plants. In addition to checking for plant damage, look under pots and trays where slugs often hide during the day.
- Encourage natural predation of slugs and snails by providing habitats for birds, hedgehogs and beetles, and by minimising the use of pesticides by adopting IPM.
- Tex-R mattings, when used as ground-cover materials for root control and weed suppression, can give protection against slugs or snails as long as the plants are uninfested when stood on the matting, and can give long-term reductions of slug and snail populations on the nursery. Choice of specific matting and methods for keeping it clean to maintain long-term efficacy should be discussed with the supplier.
- Until more nursery trials have been completed, only preliminary recommendations can be given for the use of 'Nemaslug'. If interested, try on a small scale first on susceptible plants in areas prone to slugs or snails. The optimum time for treatment with 'Nemaslug' is in spring or autumn, when compost temperatures are 5-25°C and when slugs and snails are active. Follow label directions carefully, apply the product to moist compost and keep the compost damp for at least two weeks after application.
- Use slug pellets at the recommended rate if necessary against existing infestations.
- Copper fungicides e.g. Croptex Fungex may give some incidental control of slugs and snails when used for disease control. Follow label recommendations carefully.

#### SCIENCE SECTION

#### INTRODUCTION

Slugs and snails damage a wide range of container-grown HNS plant species and are difficult to control with conventional slug pellets. Populations have become established on many nurseries throughout the country, and are damaging plants in plugs, liners and containers, both under protection and outdoors. Plant species affected include alpines, herbaceous plants, perennial and deciduous shrubs, grasses and herbs. Feeding damage is very obvious and causes plant losses or downgrading.

Preliminary investigations prior to the project confirmed the snail species causing problems on one HNS nursery as *Oxyloma pfeifferi*, a small semi-aquatic species. Several slug species have been found on HNS nurseries, but the relative importance of the different species of both slugs and snails as pests of HNS was not known. Methiocarb and metaldehyde pellets are used extensively on susceptible plants on most HNS nurseries, but seem to give only partial control of slugs and little control if any, of the snail species.

The development of alternative, integrated, more effective control measures for slugs and snails would reduce plant losses and use of chemical molluscicides, improve plant quality and offer a sustainable strategy for controlling the pests on HNS. As more growers of HNS are now adopting Integrated Pest Management (IPM) techniques, the development of non-chemical snail and slug control methods will be compatible with IPM strategies and satisfy increasing market demands for plants grown with environmentally-responsible production methods.

The parasitic nematode, *Phasmarhabditis hermaphrodita* (Nemaslug®) has been available for the control of slugs since 1997, but has been promoted mainly for the home/garden market, due to cost implications for large-scale commercial use. Research funded by DEFRA (previously MAFF) and MicroBio Ltd (now Becker Underwood) has been done on the efficacy of the nematodes against both slugs and snails (Glen *et al*, 1996). The nematodes have been shown to be effective against the field slug, *Deroceras reticulatum* and other pest species in wheat and lettuce. The snail species tested were mainly those found in hedgerows around arable fields and these did not include *O. pfeifferi* (Wilson, 2001). However, preliminary laboratory tests with the nematodes before this project began, indicated that 'Nemaslug' had potential against *O. pfeifferi* (Bennison & Maulden, unpublished data).

Copper is known to both kill and repel slugs and snails. Various copper formulations are approved for use on HNS for bacterial disease control and these may have potential against slugs and snails. Copper ammonium carbonate (Croptex Fungex) is

widely used in the HNS industry. 'SpinOut' products containing copper are now commercially available in the UK for use as root retardants and weed suppressants. Products include Tex-R matting for use as a sandbed cover or as an alternative to Mypex, a liquid for application to Mypex, a 'pot-topper' for weed suppression in container plants and an impregnated paper pot. Tex-R matting may have potential for use as a barrier against snails and slugs (Sopp, personal communication).

A grit-like mineral product, 'Snail-Ban'® is available on the amateur market for use as an environmentally-friendly physical barrier to deter snails and slugs. When dry, the product is claimed to prevent snails and slugs moving across the material by absorbing their mucus and when wet, the product acts as an irritant. The product does not dissolve during wet conditions and is claimed to act as a persistent barrier.

Research has been done in the UK on various novel slug and snail repellents. A LINK project part-funded by the HDC Field Vegetables Panel and led by the collaborator in project HNS 105, is investigating improved methods for controlling slugs on field-grown brassicas and lettuce, including the potential role of novel slug repellents.

The main objective of project HNS 105 is to identify the snail and slug species currently causing damage to HNS on commercial nurseries, and to develop an integrated management strategy, including both control of established populations and prevention of infestation of new plants brought onto the nursery.

Detailed objectives of the project are:-

1. To identify the species of slugs and snails currently causing damage and difficulties in control on HNS nurseries.

2. To identify the plant species damaged on a range of nurseries, the time of year infestation and damage occurs, and to estimate the extent of damage both under protection and outdoors.

3. To investigate the biology and likely sources of the predominant snail and slug species in order to plan the optimum timing and components of potential control strategies.

4. To evaluate the efficacy of 'Nemaslug' at various dose rates against the selected predominant snail and slug species, compared with that of chemical molluscicides, both in the laboratory and under commercial conditions.

5. To evaluate the efficacy of physical barrier or repellent techniques against the predominant snail and slug species, to prevent infestation of new plants brought onto the nursery. Techniques to be evaluated will include a copper fungicide spray, Tex-R matting, 'Snail-Ban' and novel repellents such as cinnamamide or those based on wood mulches.

6. To produce a factsheet for growers, with details of the snail and slug species and the results of the research.

#### **OBJECTIVES 1 AND 2**

Work to meet these objectives was completed during year 1. The conclusions were:

# **OBJECTIVE 1: IDENTIFICATION OF THE SPECIES OF SNAILS AND SLUGS CURRENTLY CAUSING DAMAGE AND DIFFICULTIES IN CONTROL ON HNS NURSERIES**

- Oxyloma pfeifferi is the main species of snail occurring on HNS nurseries
- Deroceras panormitanum is the main species of slug occurring on HNS nurseries
- Other snail species identified were *Oxychilus draparnaudi*, a small black spiral species which is semi-predatory on other molluscs, *Zonitoides nitidus, Cepaea nemoralis* and *Discus rotundus*. Other slug species identified were the field slug, *Deroceras reticulatum*, *D. laeve*, *Arion ater*, *Arion* sp. 'Durham', *A. circumscriptus*, *A. distinctus*, *A. fasciatus*, *Limax marginatus* and *Milax budapestensis*.

# **OBJECTIVE 2: IDENTIFICATION OF PLANT SPECIES DAMAGED ON A RANGE OF NURSERIES, TIME OF YEAR OF DAMAGE AND EXTENT OF DAMAGE UNDER PROTECTION AND OUTDOORS**

- A wide range of herbaceous plants, alpines, shrubs, grasses and herbs in plugs, liners and containers are damaged on commercial nurseries by snails and slugs.
- Damage by snails and slugs occurs both under protection and outdoors.
- *D. panormitanum* seems to damage mainly soft-leaved plants whereas *O. pfeifferi* seems to damage both soft and fleshy-leaved plants (later studies during years 2 and 3 showed that both the slug and snail species can cause primary damage to both herbaceous and perennial plants, although not all plant species are damaged by both the slugs and the snails).

- It is possible that on some nurseries, *O. pfeifferi* feeds on algae or mosses on the substrate or polythene tunnel coverings, or on rotting vegetation rather than on HNS species. However, *O. pfeifferi* is of major concern to growers.
- Plant losses and downgrading of individual plant species ranged from less than 1 to 100%.
- Both *D. panormitanum* and *O. pfeifferi* were considered to be endemic on most nurseries visited, although the original source of *O. pfeifferi* in particular was thought to be bought-in liners.
- The age of plants and availability of palatable plant material, time of year, use of molluscicides and nursery hygiene were contributory factors affecting the extent of damage.
- Damage by slugs seems to occur all year round, whereas that by *O. pfeifferi* seems to be mainly between late spring and early winter, due to its apparent hibernation during the winter and early spring (this was later demonstrated in year 2). Worst damage by both snails and slugs occurred in late spring, early summer and autumn.
- Chemical molluscicides, particularly methiocarb, were used extensively on most nurseries visited, although they were considered to be largely ineffective against *O*. *pfeifferi* and only partially effective against slugs. Copper fungicides were thought to give incidental control of snails and slugs at two sites.
- The widespread use of overhead irrigation was thought to encourage snail and slug activity. The use of Tex-R matting and capillary irrigation at one site was considered to be the main factor responsible for reducing the previously high numbers of snails and slugs.
- 'Nemaslug' had been used experimentally at three sites against *O. pfeifferi* and at one site against *D. panormitanum* and was considered to have given some useful control.

# **OBJECTIVE 3: BIOLOGY OF THE PREDOMINANT SLUG AND SNAIL SPECIES**

During year 1, the most common slug and snail species found on HNS nurseries were *Deroceras panormitanum* and *Oxyloma pfeifferi* respectively. During year 2, relevant gaps in knowledge of the biology and behaviour of these two species were addressed by experimental work at both ADAS Boxworth and the University of Newcastle, and by observations on a commercial HNS nursery. During year 3, some aspects of the biology of the slugs and snails were completed and are reported in full in this report.

#### **Materials and Methods**

#### Biology of O. pfeifferi

#### Feeding studies with O. pfeifferi

During year 2, feeding studies were completed with the snails, offering them young leaves of either Choisya or Hosta as a food source. The snails caused damage to Hosta but not to Choisya, and it was concluded that they could sometimes be mistakenly held responsible for slug damage on some HNS species. During year 3, young leaves from a range of HNS species were offered to *O. pfeifferi*, using the same methods as used for Choisya and Hosta in year 2, and any damage was recorded.

As *O. pfeifferi* are often associated with algae on the surface of compost, gravel, Mypex or polythene coverings of tunnels on commercial HNS nurseries, an experiment was set up during year 2 to identify whether immature snails could grow and complete their development on algae alone. The results of this experiment showed that although the snails could survive for two to three weeks on algae on either compost or gravel, body lengths increased only marginally and mortality rates were high after three weeks. It was concluded that the snails must need an additional source of nutrients to thrive and complete their development. As *O. pfeifferi* are often observed on decaying leaves left on the surface of the compost or substrate after trimming HNS plants on commercial nurseries, a laboratory experiment was completed in year 3, to identify whether the snails can thrive as well on algae and decaying leaves, as they do when also offered fresh plant material.

The two food sources offered to the snails were:

- Algae-covered compost and gravel, and a decaying *Hedera* leaf collected from the surface of the compost in pots on the commercial monitoring site.
- Algae-covered compost and gravel, a decaying *Hedera* leaf collected from the commercial monitoring site, and a portion of Chinese cabbage leaf.

Each food source was added to 20 replicate liner pots containing damp compost. *O. pfeifferi* measuring 8-9.8 mm long were collected from the commercial monitoring site. One snail was added to the surface of each pot and the pot was covered with a square of mesh and a square of perforated polythene, secured with an elastic band.

The pots were incubated at 20°C at a 12:12 hrs light:dark cycle for six weeks. The pots were checked weekly, misted with water and the length of each snail was measured. Snails were recorded as live, dead or 'attached', i.e. when the snail sealed its operculum with a layer of mucus and became inactive, often prior to death unless the snail is entering hibernation.

### Biology of D. panormitanum

The study on the biology of the slugs started in Year 2, and preliminary results were given in the Year 2 report. The studies were completed during Year 3 and are reported in full in this report.

#### Feeding studies with D. panormitanum

The feeding studies started in year 2 were completed during year 3. Experiments were done in autumn 2002 and spring 2003, to determine the following:

- The effect of selected food sources in a no-choice situation on slug mortality, weight change and egg production.
- Whether the slugs act as both primary or secondary feeders on the selected HNS species, Choisya and Hosta.

Slugs weighing 150-300mg were collected in the field and then kept separately in 500ml transparent plastic tubs lined with damp blue tissue paper in incubators at 15° C and a 16:8 light:dark cycle. In the autumn 2002 experiments the slugs were fed with one of the following food sources:

- Chinese cabbage leaf (optimum food source as a control)
- *Choisya ternata* two undamaged leaves, one old and one young light green
- *Hosta cv. elegans* two leaves, one green, one partly decaying but both showing some physical damage.
- A suspension of algae, collected from a local nursery and kept on an algal growth medium

Plants were offered as whole leaves, which were kept fresh by keeping their base immersed in water in 5ml glass tubes. The boxes were cleaned once a week, and plant damage, slug mortality and egg production were recorded. The slugs were weighed every 7 days. The experiment was continued until all slugs were dead.

A similar series of experiments was done in spring 2003 to examine the effects of prior damage on the feeding success of *D. panormitanum*. The method was identical to that above except that the food sources were:

• A whole fresh undamaged Hosta cv. elegans leaf

- A fresh Hosta leaf with four holes punched into it
- Two whole undamaged Choisya leaves (one old and one fresh light green)
- Two whole Choisya leaves with one whole punched in each leaf (one old and one fresh light green)

Holes were punched into the leaves using a paper puncher approximately 5 mm in diameter.

#### Development of D. panormitanum at different temperatures

Slugs were collected in the field and kept in incubators at 15° C and a 16:8 light:dark cycle and fed with Chinese cabbage and carrots. Egg batches with 15–30 eggs were removed on a daily basis, transferred into Petri dishes with damp filter paper and placed into incubators with temperatures of 12, 15 and 20° C (a total of 20 egg batches per temperature). Egg development time and hatching rate were recorded. Newly hatched slugs were kept in batches and reared to adult size over a 30-week period. The slugs were reared in 500ml transparent sandwich boxes and provided with damp filter paper, Chinese cabbage, chicken food "Layers Mash" (mainly wheat seeds) and a piece of cuttle fish bone (calcium source). Cultures were cleaned once a week. Mortality and egg production of maturing adults was recorded weekly; the slugs were weighed every 14 days for 30 weeks.

*Performance of field-collected D. panormitanum at different temperatures* Slugs (155-375 mg) were collected in the field in early April and kept separately in 3 litre transparent sandwich boxes in incubators at 12, 15, 20 and 22° C and a 16:8 light:dark cycle. There were two boxes with 20 slugs each per temperature. Slugs were fed with Chinese cabbage and carrots and provided with a piece of cuttle fish bone. The boxes were cleaned once a week and slug mortality and egg production were recorded. The slugs were weighed every 14 days. The experiment continued until all slugs were dead.

#### Population dynamics of D. panormitanum in the field

Thirty slugs were collected from the field (clover patch at the University of Newcastle Close House field station near Heddon-on-the–wall, Northumberland) each month over a 16-month period. Wooden boards, approximately 40x40 cm were used as "traps", using the slugs' tendency to seek shelter under material at the end of the night. Slugs were weighed and then kept separately in Petri dishes, for the first four months in a polythene tunnel, therafter in a shaded area of an unheated greenhouse (long-term temperature range 4-40° C). Slugs were fed with Chinese cabbage and carrots and provided with a piece of cuttle fish bone. The Petri dishes were cleaned once a week and slug mortality and egg production were recorded for a period of two weeks. This experiment started in November 2001 and ran until February 2003. Temperatures were measured with a Tinytalk® data logger at 4-hour intervals.

#### Population dynamics of D. panormitanum in the glasshouse

One hundred and fifty slugs were collected from the field at the University of Newcastle field station. The slugs were weighed and then transferred into plant propagators (40x60x20 cm), 30 slugs in each of five propagators. The propagators were filled with approximately 1cm of coarse sand, covered with Mypex matting and contained four Chinese cabbage plants each. The Chinese cabbage plants were replaced twice a month. Each month six slugs were randomly sampled from each of the five propagators, weighed and then transferred into Petri dishes and treated as described above under "population dynamics in the field". After 14 days all surviving slugs and all the eggs laid during this period were returned to their respective propagator.

One set of five propagators was started in late October 2001 and kept in a polythene tunnel at the University field station until January 2002, when two of the five propagator populations were lost due to severe frost and the remaining three propagators were transferred to an unheated glasshouse. A second set of five propagators was set up in late February 2002 and was kept in the glasshouse all of the time. This experiment continued until March 2003.

#### Diurnal behaviour of D. panormitanum and O. pfeifferi

The study on diurnal behaviour of the slugs and snails started in Year 2, and preliminary results were given in the Year 2 report. The study was completed during Year 3 and are reported in full in this report.

Slugs were collected in the field and acclimatised to the laboratory conditions (15°C and a 16:8 light:dark cycle) for at least four weeks before the experiments were done. Snails were reared in the laboratory and acclimatised to the same laboratory conditions as the slugs for at least one week prior to the experiments. The weight range of the slugs was 300-400mg (mean 355 mg) and that of the snails was 100-160 mg (mean 125 mg). The diurnal behaviour of the snails and slugs was identified using low-light, time-lapse video, with infra-red illumination during the dark cycle. One slug or snail was placed in each of 20 replicate, 16 cm-diameter white plastic dishes containing damp compost. The dishes contained a 9cm<sup>2</sup> piece of Chinese cabbage leaf and a 9 cm<sup>2</sup> shelter, made of '2H' wool-based black horticultural matting. The compost was kept damp at all times and the walls of the dishes were painted with Fluon<sup>®</sup> to prevent the slugs or snails from escaping. The snails were marked with a small spot of Tipp-Ex ® on their shells to produce a good contrast with the background. The slugs or snails were kept in the arena 24h prior to the start of the video to allow them to explore the new environment and were then recorded for 24 hours. The video was then digitised and analysed using the software package

EthoVision ®, at first analysing the differences between dark and light periods and then analysing the 30 minute intervals in more detail. This included fully automated analysis of movement activity (track position and derived track length). In addition to movement activity, activities such as resting, sheltering and feeding were analysed using a keystroke-activated event recorder. For the slugs the 24 hour period was divided into three equal periods of "Dark", "Light am" and "Light pm" as there were obvious differences between the activity in the morning and the afternoon.

#### Statistical analysis

For the slugs, the data were analysed using analysis of variance (ANOVA) and some of the observed activity parameters were combined, e.g. "under leaf" and "under shelter" were combined as "sheltering". For the snails, the "Dark" period (eight hours) and "Light" period (16 hours) were compared using angular-transformed data of the proportions in paired t-tests or the non-parametric equivalent, the Wilcox test.

#### **Results and Discussion**

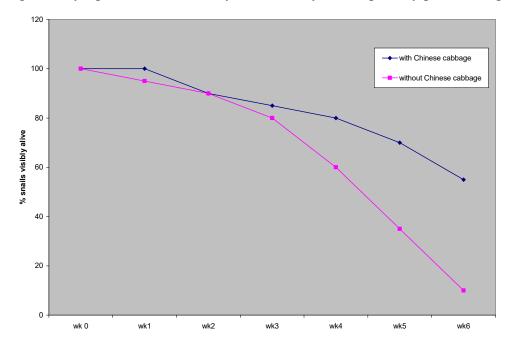
#### Biology of O. pfeifferi

#### Feeding studies with O. pfeifferi

In the feeding bioassays, damage was recorded to *Ajuga*, *Euonymus*, lupin, *Phormium*, *Campanula* and *Viola*. No damage was recorded on Japanese anenome, *Hemerocallus*, *Saxifrage* and *Aubretia*, although all these species have been recorded as susceptible plants to damage by *O. pfeifferi* on commercial nurseries.

When offered algae on compost and gravel, and a decaying *Hedera* leaf, with or without a portion of Chinese cabbage leaf as food sources, the snails grew larger and survived longer when Chinese cabbage was offered in addition to the algae and decaying leaf (Figs 1 & 2) After a 6-wk period, 55% of the snails fed on the diet including Chinese cabbage were still alive and the mean body length had increased by 1.6mm. Only 10% of the snails fed on the diet without the Chinese cabbage were alive after six weeks and the mean body length was less than when the experiment was set up, due to the larger snails having died. The snails laid eggs when fed on both diets, and young snails were recorded in the pots with the Chinese cabbage by week 2. By week 4, 45% and 35% of the pots with or without the Chinese cabbage respectively contained young snails.

It was concluded that snail survival, development and egg-laying is more successful when a source of fresh plant material is available. However, large numbers of snails of all sizes were seen on the commercial monitoring site during years 2 and 3, on pots of *Hedera* and *Euonymus*, where very little plant damage was seen. This demonstrates that *O. pfeifferi* can reach large numbers when feeding secondarily on



algae decaying leaves and that they do not always cause primary plant damage.

Fig.1. Mean % live *O. pfeifferi* over a 6-wk period when offered algae on compost and on gravel, and a decaying *Hedera* leaf with or without a portion of Chinese cabbage leaf as a food source.

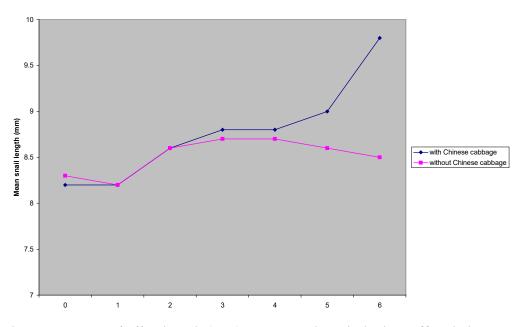


Fig.2. Mean *O. pfeifferi* length (mm) over a 6-wk period when offered algae on compost and on gravel, and a decaying *Hedera* leaf with or without a portion of Chinese cabbage leaf as a food source.

**Biology of D. panormitanum** 

#### Feeding studies with D. panormitanum – Autumn 2002 experiment

When the slugs were fed on the highly palatable food source Chinese cabbage most individuals showed a large increase in weight (Fig.3). Weight nearly doubled within four weeks and remained at a high level thereafter. Slugs fed on damaged *Hosta* leaves gained little weight in the first week and kept their weight a little below the initial weight for the remaining time. Slugs fed on the algae suspension or *Choisya* leaves continuously lost weight from the start of the experiment.

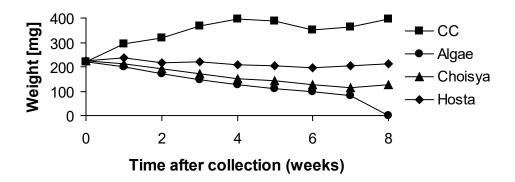


Fig.3. Weight development of *D. panormitanum* with different food sources in autumn feeding trial.

The damage caused to the highly palatable Chinese cabbage leaf was significantly higher than on the other food sources (week 8: N =20, Kruskal-Wallis test: P < 0.001, Fig. 4). Damage to the Chinese cabbage was more than 10 times higher than the algae-soaked paper tissue that was eaten. The damage on Choisya was marginal, just 1 % in comparison with the Chinese cabbage. No data is available for Hosta as the leaves already had some physical damage when offered as food and it was not possible to record slug damage to the decaying leaves. With all four food sources damage occurred on the blue paper tissue lining the plastic containers. Damage to the paper was highest in the algal food source as it was applied onto the tissue. Damage to the paper was also high when leaf damage was low or absent (especially for Choisya).

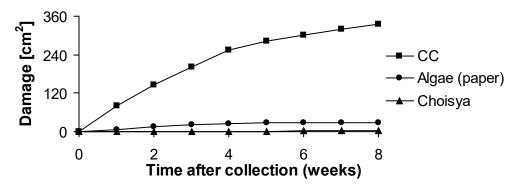


Fig. 4. Accumulated damage of *D. panormitanum* on different food sources over a period of eight weeks in autumn feeding trial.

As in previous experiments at 15 °C, egg production of field-collected slugs was low during the first week after collection. Overall egg production was highest when slugs were fed with Chinese cabbage (Fig.5). However, until four weeks after collection, slugs fed on the algal suspension or Hosta laid more or as many eggs as those fed on Chinese cabbage, and the accumulated egg production per slug did not significantly differ between food sources after eight weeks (N = 20, Kruskal-Wallis test: P > 0.05). The mean egg batch size was low, six eggs or less (Fig.6). Generally slugs fed on Chinese cabbage had the largest batch sizes, while with the other food sources batch sizes were fairly small. With algae as a food source, the high total egg production was due to a high frequency of slugs that laid eggs (Fig.7).

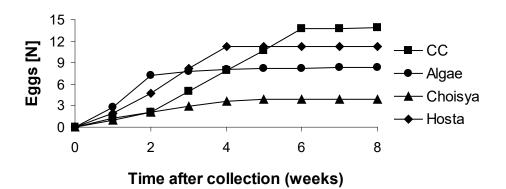


Fig.5. Accumulation of egg production of *D. panormitanum* (eggs per initial number of slugs) with different food sources over a period of eight weeks in autumn 2002 feeding experiment.

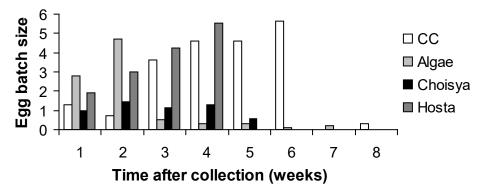


Fig. 6. Egg production (batch size) of *D. panormitanum* with different food sources in autumn 2002 feeding experiment.

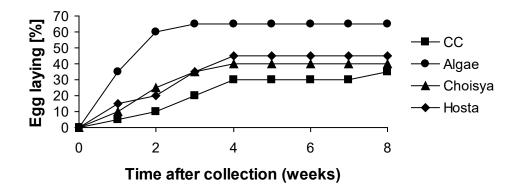


Fig.7. Frequency of *D. panormitanum* egg production with different food sources over a period of eight weeks in autumn 2002 feeding trial.

Survival of the slugs was not significantly influenced by the food source offered (N = 80, Kaplan-Meier survival analysis: P > 0.05, Fig.8).

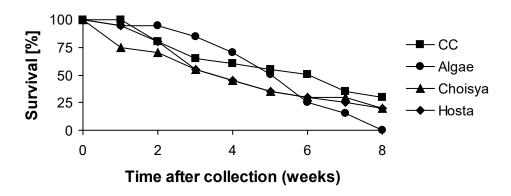


Fig.8. Survival of *D. panormitanum* with different food sources over a period of eight weeks in autumn feeding trial.

#### Feeding studies with D. panormitanum – Spring 2003 experiment

In the first week of the feeding experiment assessing the effects of prior damage, further damage occurred on three of the four food sources at frequencies above 80 %. Punched Choisya leaves were only damaged by 50 % of all slugs in the first week which was significantly less frequent than undamaged Choisya leaves (N = 20, Chi-Square test: P < 0.05, Fig.9). In the following weeks punched leaves were always more frequently damaged than undamaged leaves and this was significant for Hosta leaves in the fourth week (N =20, Chi-Square test: P < 0.05, Fig.9). When the position of damage to punched leaves was compared, positions away from the punched hole were preferred in the first week and the punched hole was preferred in all the following weeks (Fig.10).

Generally, Choisya was damaged more frequently than Hosta. Damaged areas were typically very small (Fig. A, Appendix I), only occasionally Choisya leaves were "shredded". The harder, older Choisya leaves were never damaged. The maximum

number of individual holes caused by one slug in one week was 14 on a Hosta leaf and 41 on a Choisya leaf. With all four food sources damage occurred on the blue paper tissue.

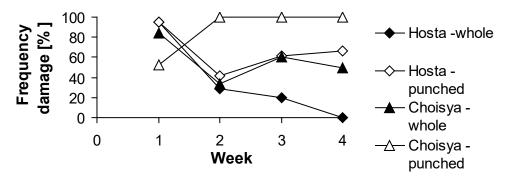


Fig.9. Frequency at which live slugs were causing damage over a period of four weeks with different food sources in spring 2003 feeding experiment.

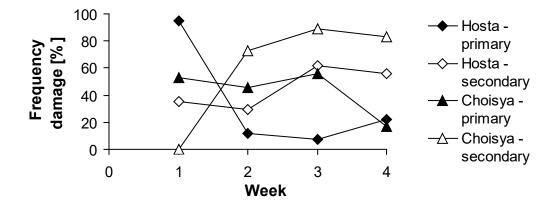


Fig.10. Frequency at which live slugs were causing secondary (at existing punched hole) and primary (anywhere else) damage to punched leaves over a period of four weeks in the spring 2003 feeding experiment.

There was little egg production by slugs feeding on Choisya or Hosta (Fig.11). Slugs feeding on Choisya produced a slightly higher number of eggs until the fourth week compared with those in the previous feeding experiment. Slugs feeding on whole Hosta leaves had approximately half the accumulated egg production than those feeding on decaying Hosta leaves in the earlier experiment, but still more than those feeding on Choisya. After four weeks egg production of slugs feeding on punched and whole leaves of the same plant were very similar. None of the four food sources had significantly more egg production than another (N = 20, Kruskal-Wallis test: P > 0.05).

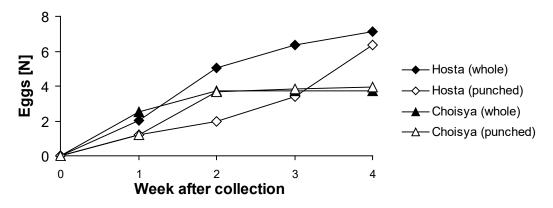


Fig.11. Accumulation of egg production of *D. panormitanum* (eggs per initial number of slugs) with different food sources over a period of four weeks in spring 2003 feeding experiment.

Of those slugs that laid eggs, the batch size was much larger than in the previous feeding experiment (Fig.12). In the first two weeks egg production was very similar with different food sources. In the second two weeks batch size was larger when slugs were feeding on punched Hosta leaves than on unpunched leaves. No eggs were laid by slugs feeding on whole Choisya leaves.

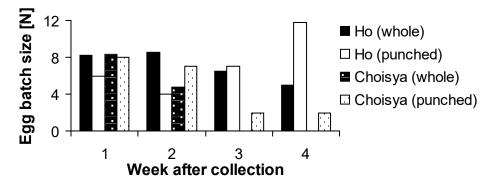


Fig.12. Egg production (mean batch size) of live *D. panormitanum* with different food sources in the spring 2003 feeding experiment.

The rate of survival of slugs was much lower in the spring (30 - 45 %) than in the autumn (55 -85 % after four weeks). This result is likely to have been due to the natural seasonal mortality patterns i.e. older slugs dying over the winter or in the early spring, before the slugs were collected for the spring experiment. In the spring experiment there was no significant difference in survival between slugs fed on different food sources (N =80, Kaplan-Meier survival analysis: P > 0.05, Fig.13).

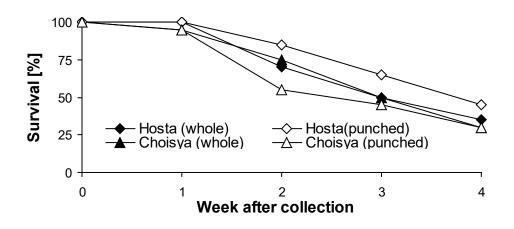


Fig.13. Survival of *D. panormitanum* with different food sources over a period of four weeks in the spring 2003 feeding experiment.

#### Development of D. panormitanum at different temperatures

When *D. panormitanum* eggs were kept at constant temperatures there were significant differences in the development time (n= 1416, Anova, P<0.001, Table 1). However, the hatching rate was not affected by the temperature (n= 1416, Anova, P=0.60, Table 1). According to the egg development times a threshold for egg development could be calculated as being 5.4 °C, with approximately 210 day degrees above this threshold needed to complete egg development.

Table 1. Egg batch size, development time and hatching rate of *D. panormitanum* at constant temperatures. Different letters show significant differences between treatments (P<0.001).

Т [°С]	batch size [eggs]	dev. time [d]	hatch rate [%]
12	22.8 ± 1.4	32.4 ± 0.5 A	84.4 ± 3.0
15	24.4 ± 1.7	21.9 ± 0.3 B	89.1 ± 3.4
20	21.9 ± 0.3	14.6 ± 0.1 C	86.7 ± 3.4
Р	n.s.	***	n.s.

A total of 1229 slugs hatched and were reared to adult size. Mortality of juvenile slugs was particularly high in the first 4-5 weeks (Fig. 14). In the first ten weeks mortality was highest at 20 °C (N=1229, G-Test, P<0.001, Table 2). Later, significant differences in mortality rates between different temperatures disappeared (G-Test, for 20 and 30 weeks P>0.05,) and finally after 30 weeks mortality rates were very similar (Fig.14).

Table 2. Mortality rate and weight of *D. panormitanum* juveniles at constant temperatures. Different letters show significant differences between treatments

(D < 0.001)	
(P < 0.001)	).

	mo	ortality rate	weight [mg]					
Т [°С]	week 10	week 20	week30	week 10	week 20	week30		
total [N]	604	392	218	604	392	218		
12	51 A	65	82	27 ± 1 A	67 ± 4	88 ± 7 A		
15	44 A	73	83	30 ± 1 A	71 ± 7	146 ± 19 B		
20	63 B	69	81	62 ± 5 B	110 ± 11	104 ± 16 A		
Р	*	n.s.	n.s.	***	n.s.	***		

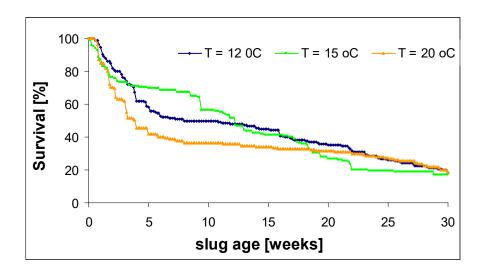


Fig.14. Survival of juvenile *D. panormitanum* kept at constant temperatures.

The surviving slugs grew very slowly in the first four weeks. Growth rates were more or less the same for 12 and 15 °C until week 20, when slugs at 15 °C started growing much faster than at 12 °C. The increase in weight was much faster at 20 °C where the average weight reached 100 mg 14 weeks after hatching. From that time onwards the average weight levelled out, partly because large animals were dying. While slugs kept at 20 °C were significantly heavier than those at 12 and 15 °C after 10 weeks (N=604, Anova, P<0.001, Table 2), slugs which were kept at 15 °C continued to grow until the end of the experiment (Fig.15) and were therefore significantly heavier than slugs at 12 and 20 °C at the end of the experiment (N=218, Anova, P<0.001, Table 2).

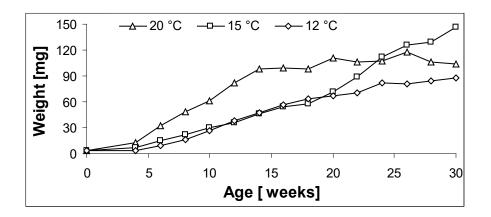


Fig.15. Development of juvenile *D. panormitanum* weight when kept at constant temperatures.

The egg production of maturing *D. panormitanum* was generally very low, even though individual slugs reached weights above 150 mg in week 12, 14 and 6 for 12, 15 and 20 °C respectively. At 20 °C egg laying started 63 days after hatching, at 15 °C after 118 days. Slugs kept at 12 °C did not lay any eggs during the 30-week period, but one individual laid a few eggs just after the trial finished. Taking these observations into account the temperature threshold for sexual maturation of slugs could be calculated as 8.5 °C, with approximately 750 day degrees above this threshold needed for sexual maturation. At 20 °C the slugs laid a total of 2309 eggs, which represents five eggs per hatched slug or 30 eggs per slug alive at the end of the experiment. At 15 °C the slugs laid only a total of 28 eggs.

#### Performance of field collected D. panormitanum at different temperatures

In the first two weeks after transferring the adult slugs into laboratory conditions there was little mortality in the treatments. After that the rate of mortality increased in all temperatures except 12 °C where animals survived for up to 15 weeks. At the higher temperatures most animals died within eight weeks (Fig. 16). Overall the survival of slugs was significantly higher at 12 °C than in all other temperatures (N=160, Kaplan-Meier survival analysis, Breslow procedure, P < 0.001).

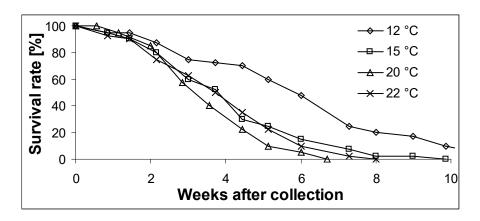


Fig.16. Survival of field-collected D. panormitanum at constant temperatures.

Adults generally gained a lot of weight in the first two weeks. At 15 and 22.5 °C weight gain continued until week 4, at other temperatures a decrease in average weight started already in the third week (Fig.17). At no time was there a significant difference in weight between the cultures kept at different temperatures (Anova, P>0.05, Table 3).

Table 3. Weight development of field-collected *D. panormitanum* at constant temperatures.

	weight [mg]										
Т [°С]	start	week 2	week 4	week 6	week 8						
12	232 ± 8	332 ± 15	321 ± 19	298 ± 33	310 ± 37						
15	227 ± 7	309 ± 12	316 ± 24	221 ± 35	168 ± 75						
20	221 ± 5	292 ± 10	247 ± 55	218 ± 123	na						
22.5	231 ± 7	290 ± 11	312 ± 18	240 ± 57	na						
Р	0.94	0.57	0.55	0.55	na						

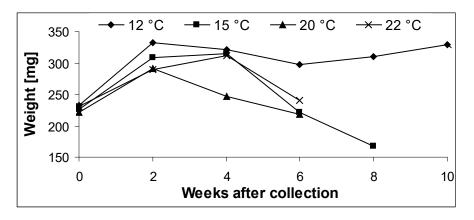


Fig. 17. Weight development of adult field-collected *D. panormitanum* at constant temperatures.

There was a delay before the onset of egg production that increased with temperature (N=6, regression analysis with logarithmic model P<0.05, Fig. 18). The higher the temperature, the sooner the animals stopped producing eggs; usually a week or more before they had all died. 15 °C can be regarded as the long-term optimum temperature for egg laying.

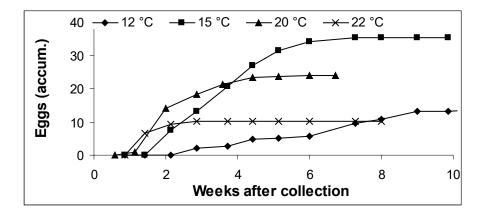


Fig. 18. Egg production of adult field-collected *D. panormitanum* at constant temperatures.

#### Population dynamics of D. panormitanum in the field

A total of 480 slugs were sampled over 16 months. The weight of the collected slugs ranged from 18- 420 mg. However, the majority of slugs weighed between 150 and 300 mg (75%) and there were only very few months when average weight was significantly higher or lower than in other months (N=480, Anova, P<0.001, Table 4). For example in spring and autumn 2002 (March, April and November) slugs were significantly lighter than in winter 2002 (Feb) and 2003 (Jan). Within similar average weights, there were still some differences in the population structure (Fig.19).

Table 4. Temperature conditions during field sample period and weights of *D*. *panormitanum*. Different letters represent a significant difference between the months.

Month	Temperat AVG (14 days)		Weight [mg]	eggs laying ind. [%]	mortality rate [%]
Nov-01* <sup>1</sup>	7.0	6.7	203 ± 14 AB	3 AB	0 A
Dec-01	3.1	3.3	194 ± 11 AB	0 A	0 A
Jan-02	4.7	6.3	195 ± 13 AB	0 A	0 A
Feb-02	7.2	8.4	222 ± 11 B	0 A	10 AB
Mar-03* <sup>2</sup>	13.5	13.3	179 ± 11 A	7 ABC	13 AB
Apr-02	15.3	15.0	183 ± 10 A	17 ABC	10 AB
May-02	16.4	16.8	177 ± 16 AB	47 DE	13 AB
Jun-02	16.6	16.6	174 ± 12 A	33 BCDE	30 B
Jul-02	17.6	17.2	195 ± 17 AB	60 E	10 AB
Aug-02	16.8	16.7	182 ± 14 AB	47 DE	7 AB
Sep-02	13.9	12.9	221 ± 15 AB	40 CDE	13 AB
Oct-02	13.3	12.8	187 ± 15 AB	53 DE	7 AB
Nov-02	12.1	12.2	169 ± 11 A	30 BCDE	7 AB
Dec-02	11.8	11.7	190 ± 10 AB	40 CDE	3 AB
Jan-03	12.0	11.9	231 ± 10 B	43 DE	13 AB
Feb-03	14.0	11.7	193 ± 8 AB	20 ABCD	10 AB
Р			***	*	*

\*<sup>1</sup> Temperature estimated based on Min/Max-readings (data logger failure)

\*<sup>2</sup> At the beginning of March 2003 propagators and petri dishes were moved from unheated polythene tunnel to unheated greenhouse

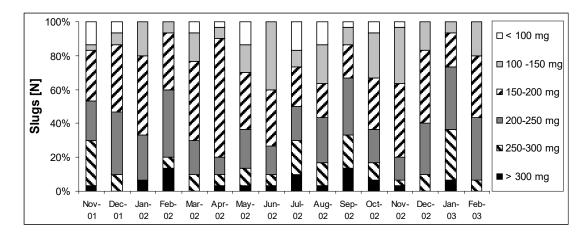


Fig. 19. Size class distribution of *D. panormitanum* when collected in the field.

During the very cold conditions in winter 2001/2002 there was no egg production of slugs kept in the unheated polythene tunnel. When slugs were transferred and kept in an unheated greenhouse, where the temperature never dropped below 8.6 °C, egg production started and reached a maximum of 2 eggs per slug in two weeks in July 2002 (Fig. 20). In July the average egg production was significantly higher than in the months November 2001- March 2002 (N=480, Kruskal-Wallis-test, P<0.001, Fig. 20). Only 28% of all slugs laid eggs; a total of 3677 (Table 4 and Table 5). The highest frequency of egg laying appeared in the size classes of individuals between 200 and 300 mg, with more than one third of all slugs laying eggs. In these two size classes the frequency of egg laying was significantly higher than in size class 6 (<100 mg) (N=480, G-test P<0.05, Table 5). Animals heavier than 250 mg laid most eggs, about 15 in two weeks. However, there were no significant differences in the average number of eggs laid per slug between the size classes (N=480, Kruskal-Wallis-test, P>0.05, Table 5). For those slugs that laid eggs a significant effect both of slug weight and average temperature on egg production could be found (N=132, multiple regression, R<sup>2</sup>=0.363, P<0.001).

The mortality during the 14-day observation was generally low, on average only 9%. During the very cold conditions from November 2001 to January 2002 there was significantly less mortality occurring during the observation period than in the warm month of June 2002 (N=480, G-test, P<0.05, Table 4), when mortality reached 30%. Individuals of the heaviest and lightest size classes had the highest mortality rates, but were not significantly different from any other size class (N=480, G-test: P>0.05, Table 5).

we class	ight [mg]	Total ind. [N]	total [N]	eggs laid (AVG ± SE)	ind. [%]	mortality rate [%]
1	> 300	23	314	14 ± 5	30 AB	13
2	250-300	57	911	16 ± 3	40 A	4
3	200-250	124	1273	10 ± 1	35 A	8
4	150-200	171	936	5 ± 1	23 AB	10
5	100-150	79	206	3 ± 1	22 AB	11
6	<101	26	37	1 ± 1	8 B	12
to	total		3677	7.7	28	9
	Р			n.s.	*	n.s.

Table 5. Egg production and mortality rate of field-collected *D. panormitanum* at ambient temperature, separated into size classes. Different letters show a significant difference between size classes.

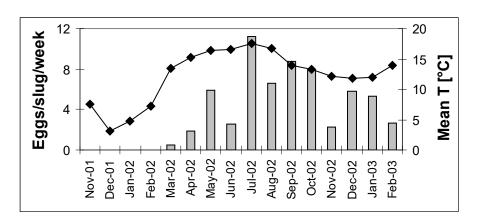


Fig. 20. Egg production of adult field-collected *D. panormitanum* (columns) at ambient average temperatures (line); polythene tunnel & glasshouse).

#### Population dynamics of D. panormitanum in the glasshouse

A total of 467 slugs were sampled from the ten propagators. Slugs between 1 and 552 mg were sampled from these glasshouse populations. In December 2001 there were 48 hours when temperatures stayed below 0 °C during the 14-day sampling period. At all other times during the 14-day sampling period the temperature was above 0 °C. The slug populations in the plant propagators were affected by two periods of frost during December 2001 and January 2002 which caused the extinction of two of the five populations from the first set in late January 2002. Two populations from the second set were extinct due to the death of all adult slugs which left no eggs behind before dying.

The size class distribution varied strongly over the period of a year. Both sets of propagators had an increase in large slugs during the first months (Fig. 21 and Fig. 22). Then nearly all of the adults died in spring to leave a very young population behind. Populations recovered within a couple of months to rebuild a population with all six size classes. Consequently the mean weight of the sampled individuals

changed significantly over time. In both sets the slugs were heaviest in March and lightest later in spring (April and May respectively, Table 6).

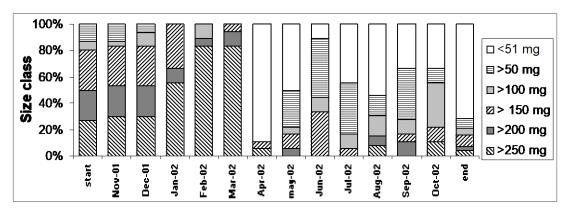


Fig. 21. Size classes of *D. panormitanum* greenhouse populations (set 1).

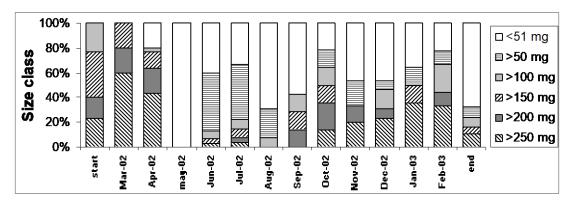


Fig. 22. Size classes of *D. panormitanum* greenhouse populations (set 2).

The slugs sampled from the first set laid a total of 236 eggs, the slugs from the second set 519 eggs (Table 6). There were significant differences between months in the frequency of egg laying. In the first set, significantly more slugs laid eggs in the autumn (October) than in other seasons (N=235, G-Test, P<0.05, Table 6). In the second set, peak egg laving occurred in spring (April, N=232, G-Test, P<0.05, Table 6), with smaller peaks in October 2002 and January 2003. However, there was no significant difference in the number of eggs laid per individual (Kruskal-Wallis-test, P>0.05, Table 6). In the first set, small adults between 100 and 150 mg laid most eggs (29% of individuals, mean of three eggs per slug), but this trend was not significant (N=235, G-test, P>0.05, Kruskal-Wallis-test, P>0.05, Table 6). In the second set, heavier slugs laid eggs significantly more often than slugs below 100 mg (N=232, G-test, P<0.05, Table 7). The maximum egg production (11 eggs per slug) occurred in the heaviest size class (N=232, Kruskal-Wallis-test, P<0.001, Table 7). When investigating the egg production of those slugs which had laid eggs, a significant effect of the slug weight was found (multiple regression, R<sup>2</sup>=0.41 and 0.23 respectively, average temperature rejected as factor, P<0.001).

The mortality rates were slightly higher than in the field-collected slugs, particularly in propagator set 2, where the overall mortality rate was 26%. In the first set, mortality was significantly higher in June than in three other months (N=235, G-test, P<0.05, Table 6). In the second set, mortality was very high in August and November 2002, but there were no significant differences between the months (N=232, G-test, P>0.05, Table 6). Individuals between 150 and 200 mg had the highest mortality rate (G-test, P>0.05, Table 7).

	set 1 (Nov 01-Oct 02)							set 2 (Mar02-Feb03)							
weight month	sample slugs [N]	weigł [mg]		total [N]	eggs la ind. [%]	(AVG ± SE)	[	morta N]	ality [%]	sampl slugs [	•		eggs la ind. [%]	id (AVG ± SE)	morta [N]
Nov-01	30	203 ± 1	4	0	0 A	0.0 ± 0.0		0	0 A	х	х	х	х	х	х
Dec-01	30	218 ± 1	8	0	0 A	$0.0 \pm 0.0$		3	10 AB	х	х	х	х	х	х
Jan-02	18	251 ± 1	8	0	0 A	$0.0 \pm 0.0$		1	6 AB	х	х	х	х	х	х
Feb-02	18	314 ± 2	23	0	0 A	$0.0 \pm 0.0$		2	11 AB	х	х	х	х	х	х
Mar-02	18	331 ± 2	20	0	0 A	$0.0 \pm 0.0$		4	22 AB		30 279 ± 17	246 BC	33 BC	8.2 ± 2.7	1
Apr-02	18	50 ± 29	)	0	0 A	$0.0 \pm 0.0$		2	11 AB		30 224 ± 29	287	40 C	9.6 ± 2.8	0
May-02	18	68 ± 15	5	41	11 AB	2.3 ± 1.8		0	0 A		30 17 ± 2	0	0 A	0.0 ± 0.0	3
Jun-02	18	108 ± 1	2	23	17 AB	1.3 ± 0.9		7	39 B		30 67 ± 11	7	3 AB	0.2 ± 0.2	1
Jul-02	18	58 ± 11		12	11 AB	0.7 ± 0.5		2	11 AB		27 84 ± 15	22	15 ABC	0.8 ± 0.4	2
Aug-02	13	79 ± 20	)	7	15 AB	$0.5 \pm 0.5$		1	8 AB		13 34 ± 9	2	8 ABC	0.2 ± 0.2	4
Sep-02	18	82 ± 14	ŀ	34	22 AB	1.9 ± 1.0		1	6 AB		7 97 ± 19	17	14 ABC	2.4 ± 2.4	2
Oct-02	18	109 ± 1	9	119	50 B	6.6 ± 2.1		0	0 A		14 152 ± 23	57	36 BC	4.1 ± 2.1	0
Nov-02 x	(	х	2	x	х	х	х	х			15 134 ± 36	4	13 ABC	0.3 ± 0.2	7
Dec-02 x	(	х	2	x	х	х	х	х			13 141 ± 38	31	15 ABC	2.4 ± 2.3	2
Jan-03 x	(	х	2	x	х	х	х	х			14 165 ± 34	60	36 BC	4.3 ± 2.0	1
Feb-03 x	(	х	2	x	х	х	х	х			9 193 ± 36	32	11 ABC	$3.6 \pm 3.6$	1
1 year	235			236	9	1.0		10	9	:	232	519	19	2.2	24
P					*	n.s.		*					*	n.s.	n.s

Table 6. Monthly egg production and mortality rates of propagator-collected *D. panormitanum* at ambient temperatures, between November 2001 and February 2003.

Table 7. Egg production and mortality rates of propagator-collected *D*. *panormitanum* at ambient temperatures, separated into size classes.

we	set 1 (Nov 01-Oct 0 veight Total eggs laid				,		mortality Total					set 2 (Mar02-Feb03) eggs laid					
class	[mg]	slugs [N]	[N]	ind. [%]	(AVG ± SE)	[N]	[%]		slugs [N]	[N]	ind. [%	$[AVG \pm SE)$	[N]				
1	>250	62	53	5	0.9 ± 0.6	4	4 6		49	526	43 A	10.7 ± 2.3 A	11				
2	200-250	23	31	4	1.3 ± 1.3	2	2 9		21	114	43 A	5.4 ± 2.0 AB	5				
3	150-200	38	44	11	1.2 ± 0.6	ç	9 24		18	41	28 A	2.3 ± 1.0 ABC	8				
4	100-150	21	65	29	3.1 ± 1.2	4	l 19		13	73	46 A	5.6 ± 2.6 AB	4				
5	50-100	37	43	22	1.2 ± 0.4	4	11		38	11	8 B	0.3 ± 0.2 BC	8				
6	<50	54	0	0	0	n.a.	n.a.		93	0	0 C	0 C	n.a. ı				
to	otal	235	236	9	1.0	23	3 13		232	765	1	9 3.3	36				
G-	test			n.s.	n.s	n	.s.				*	***	n.:				

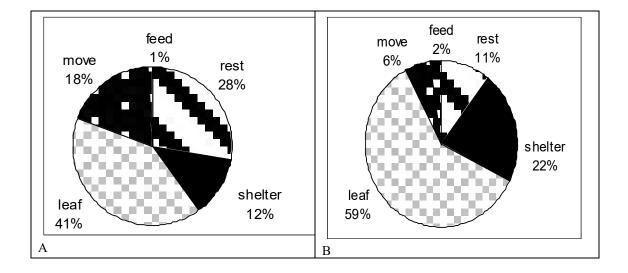
Diurnal behaviour of D. panormitanum and O. pfeifferi

The size and circular shape of the dishes used in the laboratory experiments may have led to some unnatural behaviour patterns. However, the arena diameter was eight

times the length of a slug and approximately 25 times the length of a snail. Providing shelters and food, and acclimatising the slugs and snails to the arena 24 hours before the recording in order to reduce exploration behaviour also encouraged a more natural behaviour during the recording time.

#### D. panormitanum – behaviour patterns

The proportion that each activity contributed to the total behaviour pattern over 24 hours is shown in four separate charts; for the three eight-hour periods "Dark", Light am" and "Light pm" and for the entire 24-hour period (Fig. 21, A-D). The slugs were more active during the dark period and this tendency towards nocturnal behaviour confirms earlier work on *D. panormitanum* (Morton, 1979). The slugs were only active i.e. moving or feeding for approximately 20% of the 24 hour period. For the rest of their time, the slugs were passive, with approximately 30% of their inactive time spent resting and exposed on the surface and 70% spent sheltering. These behaviour patterns have important implications for both monitoring and control on HNS nurseries. The slugs are difficult to monitor during the day as they are hiding under pots, trays or leaves, and thus their numbers may easily be under-estimated. As much of their time is spent sheltering, they may only be exposed to control measures e.g. parasitic nematodes or slug pellets, for a small proportion of their time.



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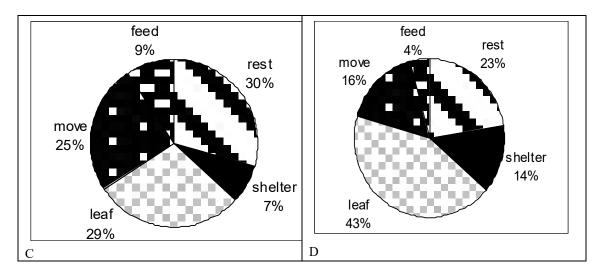


Fig. 21. A-D. Activity pattern of *D. panormitanum* A.) during 8-hour Dark, B.) during 8-hour Light am, C.) during 8-hour light pm, D.) 24-hour period

#### D. panormitanum – movement activity

The slugs travelled long distances during the dark phase and in the early hours of the morning (Fig. 22). The mean distance travelled in 24 hours was 6.1 m. The behaviour was predominantly nocturnal, but the movement activity peaked immediately after the light was switched on at 06:00 (Fig. 23). During the first 30-minute interval of the light period the slugs travelled a mean distance of 44 cm. However, overall distance travelled was significantly higher during the eight hour dark period than in the second eight hour light period ("light pm") and approximately 50 % higher than in the first eight hour light period ("light am") (See 'Track' columns in Fig. 24, n = 20, P < 0.001).

The pattern for the mean time spent moving (Fig. 23) was similar to that of the distance travelled. The total length of time taken up with moving in 24 hours was 234 minutes (3.9 hours). The peak time for moving activity was from 06:30 to 07:00 with a mean moving activity of 13.5 minutes per 30 minute interval. During the dark period, moving activity was significantly higher than during the second eight hour light period and 36 % higher than during the first eight hour light period (See 'Move' columns, Fig. 24, n = 20, Anova, P < 0.001). The most active individual moved for nearly eight hours out of 24, while another slug with a higher than average velocity moved nearly 13 m in 24 hours.

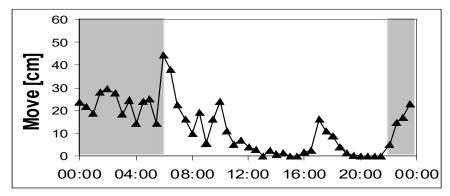


Fig. 22. Distance travelled by *D. panormitanum* in 30-minute intervals over a 24-hour period.

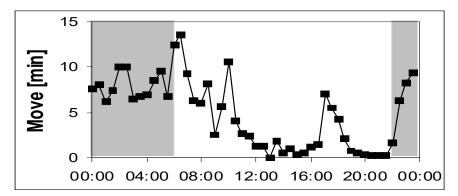


Fig. 23. Movement activity of *D. panormitanum* in 30-minute intervals over a 24-hour period.

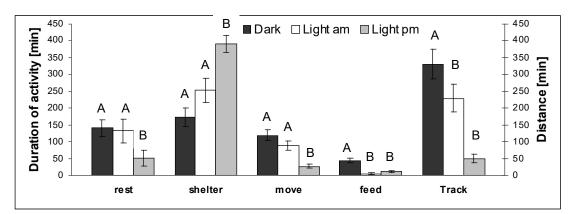


Fig. 24. Activity of *D. panormitanum* during three 8-hour periods, different letters represent a significant difference between periods, P < 0.01 for resting and P < 0.001 for other parameters.

#### D. panormitanum – feeding activity

Feeding activity could be clearly distinguished from sheltering under the leaf or moving over the leaf as all the slug feeding activity caused clearly visible holes in the Chinese cabbage leaf. There was a very clear nocturnal behaviour pattern with 75 % of all feeding taking place during the dark (Fig. 25). Feeding activity was significantly higher during the dark period than during the two light periods, between which the feeding activity did not significantly differ (Fig. 24, n = 20, Anova, P <0.001). The peak of feeding activity was reached in the interval from 23:00 to 23:30, (60-90 minutes after the start of the dark phase) when seven minutes out of 30 were spent on feeding. During the 24 hour period one hour was spent on feeding. The maximum total duration an individual slug spent feeding was nearly three hours.

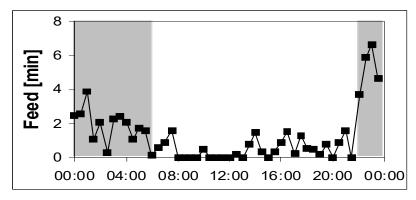


Fig. 25. Feeding activity of *D. panormitanum* in 30-minute intervals over a 24-hour period.

#### *D. panormitanum – sheltering behaviour*

During the entire 24 hour period the slugs preferred to shelter under the leaf rather than under the shelter made of horticultural matting. When the recording started in the afternoon, four individuals (of the 20 observed) were hiding under the matting shelter, and nine were hiding under the leaf. One slug sheltered under the leaf for nearly the entire light period, from 6:03 to 16:58, without feeding on the leaf. The highest proportion of sheltering in a 30-minute interval was in the interval starting at 19:00 with a mean of 27 minutes (Fig. 26). During the second light period (light pm) the slugs sheltered for more than 80 % of their time (Fig. 21) which was significantly higher than in the two other eight hour periods (Fig. 24, n = 20, P < 0.001). It was difficult to analyse the 'homing' behaviour of the slugs as there were only two shelters of differing quality. However, when comparing those slugs sheltering under the leaf with those under the matting at the beginning of recording, both groups significantly preferred their initial choice for the remainder of the experiment (t-test for 24-hour period, df = 11, total n = 13, P < 0.001).

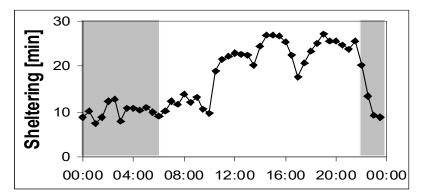
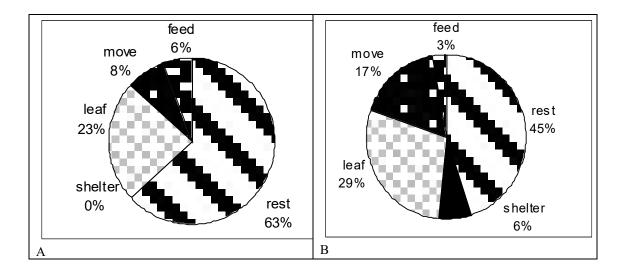


Fig. 26. Time *D. panormitanum* spent sheltering in 30-minute intervals over a 24-hour period.

#### O. pfeifferi – behaviour patterns

The proportion that each activity contributed to the behaviour pattern of the snails is presented in three separate charts for the two periods "Dark" and "Light" and for the entire 24-hour period (Fig. 27). The activity pattern of the snails was quite different from that of the slugs. The snails were more active during the day, showing two peaks of moving activity, in the early morning and towards the end of the day. As with the slugs, the snails were only active i.e. moving or feeding for 20% of the 24-hour period. However, unlike the slugs which spent most of their inactive time sheltering, the snails are active and visible during the day on HNS nurseries, they are more easily noticed than the slugs which shelter for most of the day, thus the snails could be mistakenly held responsible for slug damage on some plant species e.g. Choisya. However, even if not causing primary damage, if present in large numbers the snails may lead to quality problems as contaminants.



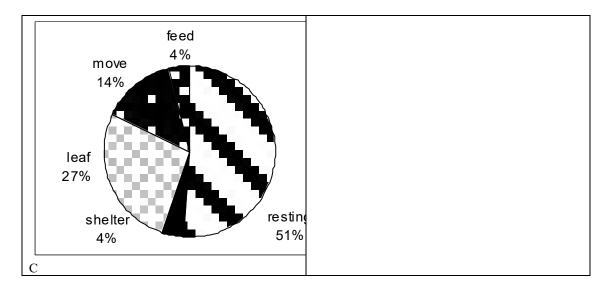


Fig. 27. Activity pattern of *O. pfeifferi* A) during eight-hour dark B) during 16-hour light C) during 24-hour period.

#### O. pfeifferi – movement activity

The snails had two peaks of moving long distances during the light phase, one in the early morning, the other one in the late afternoon (Fig. 28). The maximum distance covered in a 30-minute interval was 7 cm between 17:30 and 18:00. The mean distance travelled in 24 hours was 93 cm. The distance moved was significantly higher during the light than during the dark phase. During the light period the snails covered a significantly greater distance than during the dark period (Fig. 29, paired t-test, n = 20, P < 0.05).

The pattern for the mean time spent moving was similar to that of the distance moved (Fig. 30). Moving activity lasted for a total of 200 minutes (3.3 hours) in 24 hours. As with the peak distance travelled, the peak time of moving activity was also from 17:30 to 18:00, with a mean movement activity of 10.6 minutes per 30 minute interval. The moving activity of individual snails was observed to fluctuate, with great changes from one 30 minute period to another. Unlike with the slugs, during the dark period the proportion of the snails' time spent moving was significantly lower than during the light period (Fig. 29, paired t-test, n = 20, P < 0.01). The most active individual moved for more than seven out of 24 hours, while another snail with a higher than average velocity moved nearly 2.6 m in the same time period.

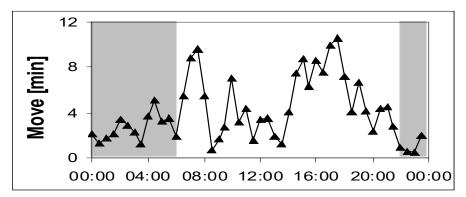


Fig. 28. Movement activity of O. pfeifferi in 30-minute intervals over 24 hours.

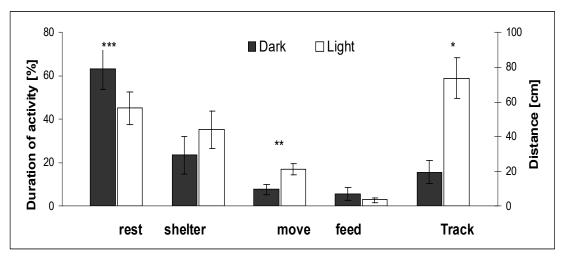


Fig. 29. Activity of O. pfeifferi during dark and light period. \* P<0.05, \*\* P<0.01.

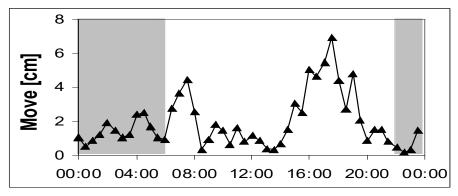


Fig. 30. Distance moved by O. pfeifferi in 30-minute intervals over 24 hours.

#### *O. pfeifferi – feeding activity*

Snail feeding activity on the Chinese cabbage leaf was low compared with that of the slugs. As the feeding damage during the experiments was often only superficial or at the edge of the leaf, it was not always possible to distinguish completely between 'feeding' and 'being in contact with the leaf'. In addition, there was a great variation between the feeding activity of individual snails. As *O. pfeifferi* can also feed on algal films on the surface of compost, substrates and polythene tunnels, some of the movement activity could also be regarded as 'feeding'.

Overall, the snails fed for a mean of 55 minutes in the entire 24 hour period. There were three periods of high feeding activity, one in the early morning, and two during the dark period (Fig. 31). The longest mean time spent feeding in one 30 minute interval was 4.2 minutes between 07:00 and 07:30. As a consequence there was no significant difference between feeding activity during the dark and light periods (Fig. 29, Wilcoxon test, n = 20, P = 0.51) even though the snails fed for proportionately nearly twice as much time during the dark as during the light. The individual snail spending the most time feeding fed for nearly seven hours in the 24-hour period.

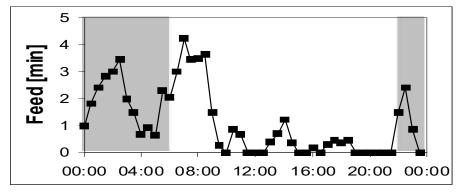


Fig. 31. Feeding activity of O. pfeifferi in 30-minute intervals over a 24-hour period.

#### *O. pfeifferi* – *sheltering behaviour*

The snails rarely used the horticultural matting for shelter, with only two snails sheltering under it, between 07:30 and 21:30, but in those two cases for several hours by each snail. Sheltering under the leaf made up 87 % of the entire sheltering activity. In some cases the snails sheltered inside a rolled-up leaf. The snails spent long periods on the lower side of the leaf without any sign of causing even superficial leaf damage. The longest period a snail spent under a leaf was 14.5 hours. Sheltering peaked in the early evening when it occupied 50% of the snails' time (Fig. 32). The snails spent a mean of 7.5 hours sheltering and 12 hours resting, exposed on the compost surface. The mean proportion of time spent sheltering was 33% higher during the light period than during the dark, but this difference was not significant (Wilcoxon rank test , n = 20, P = 0.18). However, the proportion of time taken up with being exposed (resting on the surface and moving) was significantly higher in the dark period (Fig. 29, Wilcoxon test, n = 20, P < 0.05).

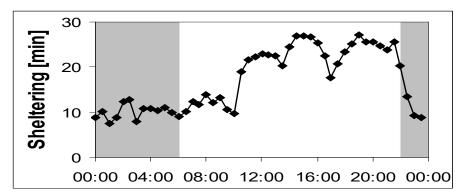


Fig. 32. Time *O. pfeifferi* spent sheltering in 30-minute intervals over a 24-hour period.

# Conclusions on biology of *D. panormitanum* and *O. pfeifferi* over the three project years

#### O. pfeifferi

- In studies between September 2001 and March 2002, *O. pfeifferi* hibernated between September/October and February/March on the sides of pots, on the structure of polythene tunnels and on plants. All hibernating snails were juveniles (less than 8 mm long). Over the winter period, 20-50% natural mortalities occurred. Snail activity began again during February and most were active by late March.
- In studies during 2002, *O. pfeifferi* laid eggs between late March and August, with mixed ages and overlapping generations occurring at any one time during this period. Damp or wet conditions stimulated activity and egg-laying and favoured survival. Most eggs took two weeks to hatch but development time was dependent on temperature. The life cycle from egg to egg was 14 weeks at 20-22°C.
- *O. pfeifferi* fed on soft-leaved herbaceous plants including Hosta, Campanula and Viola, and also on some tougher-leaved perennials e.g. Phormium and Euonymus. However, the snails did not cause primary damage to certain perennials such as Choisya and Hedera, although they are often found on these plant species on commercial nurseries.
- The snails also fed on algae on the surface of compost and gravel, on liverwort and on decaying leaves, which indicates the importance of strict nursery hygiene to help to reduce infestations. However, they could not survive on a diet of algae alone, and their survival and development on algae plus decaying leaves was favoured by the additional availability of palatable fresh plant material.
- Studies on diurnal behaviour of the snails showed that unlike *D. panormitanum*, peak moving activity occurred during the day, and even when resting they are

often exposed on the surface of plants, compost or pots. As the snails are active and visible during the day, they could be mistakenly held responsible for slug damage on some HNS species e.g. Choisya. However, even if not causing primary damage, if present in large numbers they may lead to quality problems as contaminants.

• The optimum times for control of *O. pfeifferi* could be late March/April and again in late August/early September, i.e. when snails are fully active, just after and just before the hibernation period. However, if using parasitic nematodes for control, compost temperatures need to be within the optimum range (5-25°C) and this will affect when applications should best be timed, e.g. temperatures under protection may be too warm for effective use of the nematodes in August/early September.

#### D. panormitanum

- *D. panormitanum* was active all year round, with peaks of activity occurring in spring and autumn. Large adult slugs of this species die in late winter to early spring.
- Environmental conditions have a strong influence on *D. panormitanum* biology. As with *O. pfeifferi* the slugs need high relative humidity for continuous growth and egg production. Irrigation methods will thus be an important factor affecting slug abundance, with generous overhead watering favouring egg production, population growth and overlapping generations.
- Temperature is another important factor affecting *D. panormitanum* biology e.g. growth rates, onset and rate of egg production and egg development. In studies between November 2001 and February 2002, older, potentially sexually mature *D. panormitanum* layed very few eggs in temperatures below 5.4°C, which was considered the threshold for egg production during these studies. The minimum temperature threshold for sexual maturation of juveniles was established as (8.5°C). Thus, even in unheated glasshouses or polythene tunnels, temperatures will exceed this threshold more frequently and for longer than outdoors. Consequently, egg-laying and hatching is likely to start earlier in the year under protection than outdoors.
- Under favourable conditions *D. panormitanum* adults increase in size over the winter and egg production starts in early spring (or any overwintering eggs resume development in early spring). Slug size is not necessarily related to slug age and/or sexual maturity, as great weight variation of same-age juveniles can occur, and very large and heavy laboratory-reared individuals did not lay eggs at 12 °C).
- Many juvenile *D. panormitanum* appear in the spring while the previous generation adults are dying. During studies in year 2 of the project, egg laying started in March and large numbers of juveniles were present in April. Under favourable conditions egg laying can continue all summer.

- The optimum times for control of *D. panormitanum* are likely to be similar to those for *O. pfeifferi* i.e. in spring, to break the life cycle and prevent a new generation and if necessary, again in the early autumn (September and October). Timing will depend on the control strategies used e.g. any products with activity against both slugs and snails and on egg-laying and development would be appropriate to use in the early spring, to prevent a new generation. Products with good molluscicide activity would be suitable for use in the autumn, to reduce the size of the overwintering population and to prevent damage to evergreen plants, on which any damage remains evident until the spring when leaf grazing is covered up by new growth.
- Under optimum conditions the life cycle can be as short as 11 weeks. Laboratoryreared young slugs showed a great variation in growth rates and onset of egg production, which indicates that generations will probably overlap. In protected HNS where favourable mild, damp conditions are present between spring and autumn, two or even three generations may develop each year, leading to heavy infestations. Outdoors, in very dry summers and cold winters, slug activity may be very low.
- 15°C was shown to be the optimum temperature for the long-term development of *D. panormitanum* juveniles and for sustainable egg production. However, their survival, growth and egg-laying rates were also high at 20°C, and this species seems to be well-adapted to the high temperatures present in glasshouses and polythene tunnels. During the trials evaluating control with nematodes in a commercial glasshouse reported under Objective 4 in this report, *D. panormitanum* numbers increased between mid-July and mid-August, and egg masses and young juveniles were frequently seen during assessments on Choisya liners. During this period, temperatures reached 40°C and the high 30's°C on many dates (see Appendix II).
- *D. panormitanum* was shown to feed on the selected perennial and herbacous HNS hosts, Choisya and Hosta, both as a primary and secondary feeder on fresh leaves, and on decaying plant material (Hosta). However, in comparison with the highly palatable Chinese cabbage used as the control, the extent of the visible damage remained relatively low with small circular holes (0.1-0.3mm diameter) representing the majority of the observed damage, and leaf edge feeding and shredding representing only 10-20% of the damage.
- As with *O. pfeifferi*, *D. panormitanum* was also shown to feed on algal films, although survival rates on algae alone were low. Any empty production areas should be kept as dry as possible and free from potential food sources including decaying organic matter, moss and algae. This would make conditions for slugs as unfavourable as possible and help to prevent the appearance of a new generation.
- Studies on diurnal behaviour showed that *D. panormitanum* have a nocturnal behaviour pattern typical of slugs, with both moving and feeding activity being

mainly during the dark. The slugs were only active for 20% of the 24-hour period and the inactive time was spent sheltering. However, in favourable conditions they can also show some activity during the day, and this was confirmed by obervations both in the field and under protection on commercial HNS nurseries, e.g. if misty or raining outdoors, or in overcast conditions or under shade screens under protection.

- The tendency for nocturnal activity and the large amount of time spent sheltering means that the abundance of *D. panormitanum* can often be underestimated by growers, and the slugs are only exposed to control measures e.g. parasitic nematodes for a small proportion of their time if hiding in untreated refuges.
- Time-lapse video recording showed that *D. panormitanum* moved approximately seven times further than *O.pfeifferi*, with means of 6.1 and 0.9 m travelled over 24 hours respectively. This difference can be partly explained by the larger size of the slugs. The distance travelled indicates the relative risk of infestation of previously slug and snail-free production areas. The slugs were also shown to be able to survive for several weeks without food, thus absence of plant damage or visible slugs does not guarantee that slugs or eggs are not present.

#### **OBJECTIVE 4: EVALUATION OF PARASITIC NEMATODES** ('NEMASLUG') AGAINST THE PREDOMINANT SNAIL AND SLUG SPECIES

During years 1 and 2, the parasitic nematodes, *Phasmarhabditis hermaphrodita* 'Nemaslug' were found to be effective against both *D. panormitanum* and *O. pfeifferi* in laboratory bioassays, when applied under ideal conditions at recommended and lower dose rates. During year 3, the efficacy of the nematodes was tested on HNS plants on a commercial nursery.

#### **Materials and Methods**

#### First trial

A trial was set up on a commercial HNS nursery on 18 July, to evaluate the efficacy of 'Nemaslug' at four dose rates against *D. panormitanum* and *O. pfeifferi*, compared with that of methiocarb (Draza) slug pellets. The trial was set up in mid-July, so that

assessments would be completed by mid-August, before the snails were expected to enter hibernation from September onwards.

#### Experimental plants

The trial was done with Choisya plants in liners (12 pots per tray), stood on Mypex in a glasshouse. The liners were biodegradable pots stood in plastic trays. Plants selected for assessments avoided any liners which were disintegrating, and those around the outer edges of the trays, which may have dried out in between irrigations. Assessment plants were labelled to aid selection on subsequent dates.

#### Treatments

- 1. Water control
- 2. Additional water control
- 3. Methiocarb (Draza, 4% w/w) at the recommended rate of 5.5 kg/ha (0.6g per m<sup>2</sup>).
- 4. 'Nemaslug' at the recommended rate of 12 million per  $40m^2$  (300,000 per m<sup>2</sup>)
- 5. 'Nemaslug' at half the recommended rate  $(150,000 \text{ per m}^2)$
- 6. 'Nemaslug' at quarter the recommended rate  $(75,000 \text{ per m}^2)$
- 7. 'Nemaslug' at eighth the recommended rate  $(37,500 \text{ per m}^2)$

All pots in all treatments were thoroughly watered until the compost was moist, before treatments were applied. Treatments 1,2 and 4-7 were applied as a drench in 10,000 l water/ha (1 litre water per  $m^2$ ).

Treatments 1, 2 and 4-7 were applied with a knapsack sprayer, using a coarse nozzle (Lurmark FCX04) without a filter. Sub-samples of the nematode pack supplied for the trial were taken to check numbers of viable nematodes per g of product, and numbers per ml of made-up suspension in the spray tank and in replicate test sprayed amounts. Numbers of viable nematodes were checked using standard nematological techniques in the laboratory the day before the experiment was set up. Nematodes were made up in suspension as recommended on the label. The spray tank was kept agitated during application, to avoid nematodes settling to the bottom.

#### Irrigation of plots

The grower was asked to check the plants daily and to irrigate when necessary, to avoid the compost drying out. The aim was to keep the compost damp but to avoid waterlogging of the plots.

*Experiment design* Number of trays per plot: three Plot size: Three trays side by side, measuring  $1 \ge 0.53 \text{ m} (0.53 \text{ m}^2)$ .

Number of replicates: Four per treatment (total 28 plots), in a randomised block design (see trial plan, Appendix II).

Barriers: Strips of Tex-R matting (40 cm wide) were used to separate each plot, to prevent migration of slugs and snails between plots. A 10 cm gap of Mypex was left around each plot of three trays, i.e. between the block of trays and the Tex R barrier, to avoid spraying nematodes onto the Tex R. Thus the total plot size was  $1.20 \times 0.73m$ , but the area treated was  $1 \times 0.53m$ .

The area used for the experiment was dampened with water before the Tex R strips and trays were put in position. This helped the Tex R strips to lie flat on the Mypex and provided a similar humid environment under the trays, as in the rest of the commercial crop.

#### Slug and snail infestation

The plants were naturally infested with slugs, predominantly *Deroceras panormitanum*, but also there were low numbers of *Arion ater*.

The plants had a low infestation of snails (*Oxyloma pfeifferi*) but in order to provide enough snails to adequately test the treatments, three additional *O. pfeifferi* per pot were added, after watering but before application of either 'Nemaslug' or slug pellets. The snails were collected from polythene tunnels at the nursery's container unit.

#### Slug and snail assessments

Assessments were carried out just before treatment application, and two and four weeks after treatment. On each assessment date, the following records were made on ten plants per plot:

- Number of live slugs per plant (first 20 slugs recorded to be identified to species).
- Number of live snails and empty shells per plant.
- Number of live slugs and snails per tray (the underneath of each of the three trays per plot was assessed).

#### Temperature assessments

The following temperature assessments were made:

- Temperature of water used to make up 'Nemaslug' solution on the nursery.
- Maximum, minimum and mean daily temperatures of compost, using a 'Tiny talk' temperature logger, left in position throughout the duration of the trial.

#### Statistical analysis

The mean number of live slugs and snails per plot were subjected to analysis of variance (ANOVA).

#### Second trial

As the first trial gave inconclusive results, thought to be due to the exceptionally hot weather during the trial period adversely affecting nematode viability and efficacy, a second trial was set up at the same site on 3 October. The same experimental plants, treatments and assessments were used as in the first trial, with the following modifications:

- *O. pfeifferi* were not added to the plants, thus the trial was done on slugs only.
- Number of damaged leaves per assessment plant were recorded on each assessment date.

#### **Results and Discussion**

#### First trial

#### 'Nemaslug' viability checks

Sub-samples of the 'Nemaslug' pack assessed in the laboratory before application to the trial plants indicated that only 74% of the nematodes were viable. Thus, proportionately more product was used to make up the suspension applied to the trial plants, to make up the numbers of viable nematodes to the required dose rates. Numbers of nematodes per ml of suspension in the dose-adjusted sprayed sub-samples were confirmed to be accurate, indicating that the required numbers of live nematodes were being applied through the spray nozzle.

#### Numbers of live slugs and snails per plot

- On the day the trial was set up, there were similar numbers of slugs per plot of ten plants in all treatments, with an overall mean of 13 slugs per plot (Table 8). The majority of slugs, including the first 20 identified during the assessments, were *D. panormitanum*, although there were also a few *Arion ater*. Only numbers of *D. panormitanum* were included in the analysis.
- 14 days after treatment, numbers of slugs and snails were higher in all treatments than on the day the trial was set up, as the slugs had been breeding (egg masses and young juveniles were frequently seen on the liners during assessments) and extra snails had been added after the pre-treatment assessment had been done.

- There were no significant differences (at the 95% probability level) between any of the treatments in numbers of live slugs or snails, on either of the post-treatment assessment dates. The lack of control given by the nematodes is likely to have been due to the exceptionally hot conditions during the trial period, when maximum temperatures exceeded those suitable for nematode survival, and compost moisture levels are likely to have been very low between irrigations (see temperature assessments section below). The lack of control given by the Draza may have been partly due to the slugs spending less time on the surface of the compost when the pots were dry between irrigations, and thus reducing their chances of encountering the pellets.
- The snails suffered high mortalities between 14 and 28 days after treatment, including in the control plots. This is likely to have been due to the hot, dry conditions during the trial period, but also to some possible natural mortalities of adult snails during August, as had been observed during the hibernation studies during year 2.

Table 8. Mean numbers of live *D. panormitanum* and *O. pfeifferi* per plot, 0, 14 and 28 days after treatment with either 'Nemaslug' at various dose rates or methiocarb (Draza).

Treatment	Days after treatment	Mean live slugs per plot (10 plants)	Mean live snails per plot (10 plants)		
1. Control	0	12.3	4.5		
	14	27.5	18.5		
	28	31	3.3		
2. Control	0	15.8	4.5		
	14	24.3	23.5		
	28	27.5	3.8		

3. methiocarb	0	11.3	2.5
(Draza)	14	15.3	24
	28	20.8	1.8
4. Nemaslug	0	16.5	5
(rec. rate)	14	24.8	14.5
	28	22.8	3.5
5. Nemaslug	0	14	3.8
(half rate)	14	25.8	24.5
	28	26.3	2.3
6. Nemaslug	0	11.3	4.8
(quarter rate)	14	24.3	25.8
	28	24.5	5.3
7. Nemaslug	0	11.3	2.3
(1/8  rate)	14	24.5	20
	28	17	2.3
		N.S.	N.S.

#### *Temperature assessments*

- The temperature of the nursery water supply used to make up the nematode suspension for application to the trial plots was 23°C, which was within the optimum temperature range (5-25°C) for this nematode species.
- Temperatures during the trial period after treatments had been applied were unusually high due to a prolonged hot, sunny period. Maximum temperatures reached 40°C and the high 30's on many dates (see Appendix II), which led to the liners drying out between irrigations, even though the grower applied extra water by a hose to the trial plots during the day, in addition to the automatic overhead irrigation every evening.
- The drying of the liners and the high temperatures are likely to have led to poor nematode survival and efficacy.

#### Second trial

#### 'Nemaslug' viability checks

Sub-samples of the fresh 'Nemaslug' pack assessed in the laboratory before application to the trial plants indicated that only 40% of the nematodes were viable. It was not possible to leave application to the trial until the following week, or to order a different pack of 'Nemaslug', as the grower was holding the trial plants and could not delay marketing any longer than the 4-week period agreed for the trial. Thus, with the agreement of the nematode supplier and the HDC project co-ordinator, proportionately more product was used to make up the suspension applied to the trial plants, to make up the numbers of viable nematodes to the required dose rates. The nematode batch used for the trial also showed a sudden failure in the supplier's QC tests. The cause for this QC failure was noted and dealt with in-house at the supplier's. Although the nematode numbers used in the trial were adjusted for the application, the supplier advised retrospectively that it is probable that the nematodes were also poorly infective.

#### Numbers of live slugs per plot

- On the day the trial was set up, there were similar numbers of slugs per plot of ten plants in all treatments, with an overall mean of 12 slugs per plot (Table....).
- There were no significant differences (at the 95% probability level) between any of the treatments in numbers of live slugs, on either of the post-treatment assessment dates (Table 9).
- Draza failed to give significant control of the slugs. It is possible that the single application at the recommended rate which was used in the trial is too low for effective control on heavily-infested HNS plants (many growers use much higher than recommended rates). In the laboratory bioassays in year 1, Draza gave 60% kill of *D. panormitanum*, but the single pellet per pot applied was equivalent to three times the recommended rate, and the Draza was re-applied after two weeks as the pellets had become mouldy. However, as none of the nematode treatments gave significant control of slugs in the trials on the commercial nursery either, Draza should be re-evaluated under commercial conditions in comparison with 'Nemaslug' in any future trials.

Table 9. Mean numbers of live <i>D. panormitanum</i> and <i>O. pfeifferi</i> per plot, 0, 14 and
28 days after treatment with either 'Nemaslug' at various dose rates or methiocarb
(Draza). No significant differences between treatments on either post-treatment date.

Treatment	Days after treatment	Mean live slugs per plot (10 plants)
1. and 2. (mean of	0	15.9
both control	14	14.6
treatments)	28	11.8
3. (Draza)	0	12.3
	14	9.3
	28	7.8
4. Nemaslug	0	14.5
(rec. rate)	14	12.5
	28	12.0

5. Nemaslug	0	13.8
(half rate)	14	11.5
	28	11.3
6. Nemaslug	0	15.5
(quarter rate)	14	10.3
	28	8.8
7. Nemaslug	0	14.8
(1/8 rate)	14	10.8
, , ,	28	9.8
	N.S.	N.S.

- The questionable quality of the 'Nemaslug' could have been the major factor affecting the efficacy of the nematodes, despite numbers being adjusted to allow for the 60% non-viable nematodes in the pack.
- As agreed with the HDC project co-ordinators, 'Nemaslug' was put to the most severe test on HNS i.e. on liners, which present very little treated compost area to the slugs or snails. Thus the slugs may not have come into contact with the nematodes for long enough for infection on their way up to the foliage, during their nocturnal foraging trips.
- Results in laboratory tests in years 1 and 2 of the project showed that the slugs avoid contact with nematode-treated compost. This reaction may have caused the slugs to accelerate moving over the compost at night, or to move back down the sides of the liners or under the trays, in preference to spending much time on the compost, thus avoiding infection.
- Results in the final year of this project have also shown that some *D*. *panormitanum* individuals can survive for four or more weeks without feeding on higher plants (see Results section in Objective 3). This indicates that if the slugs avoided moving onto nematode-treated compost, many could have survived the trial period without feeding on the plants.
- Compost temperatures were within the optimal range for the nematodes (5-25°C) for the majority of the trial period (see Temperature assessments section below).
- The grower made a special effort to check the plants daily for compost moisture and the liners were always damp on the assessment dates.

#### Numbers of damaged leaves per plant

Numbers of damaged leaves per plant were recorded before treatments were applied and at each of the post-treatment assessment dates, and the data were subjected to analysis of variance. The data is not presented in this report as no meaningful conclusions could be drawn on the comparative efficacy of the treatments. There were significant differences in slug damage between treatments, (although none between any of the treatments and the controls) even before treatments had been applied, with an overall mean of 6.7 damaged leaves per plant (range of 4.4-10.1). When the relative increase in numbers of damaged leaves at 14 days after treatment were compared with the number of damaged leaves at day 0, there were no significant differences between treatments. When the relative increase in numbers of damaged leaves at 28 days after treatment were compared with the number of damaged leaves at day 0, there were significantly less damaged leaves per plant with the three reduced rates of nematodes when compared with the controls (P<0.05). However, this comparison was invalid as there were less damaged leaves on plots treated with these three treatments than at the pre-treatment assessment. Some of the top leaves on the plants 28 days after treatment had been severed between day 14 and day 28, possibly by slug damage to the stem or petioles, thus leaf counts dropped on some plants between these two dates.

For any future trials on efficacy of 'Nemaslug' on HNS, a leaf damage scoring system should be used as in the laboratory tests, rather than recording numbers of damaged leaves per plant, particularly as work in this project has now shown that *D. panormitanum* tend to prefer to feed on previously damaged Choisya leaves rather than intact ones. Thus assessments of number of damaged leaves on sequential dates does not accurately reflect comparative damage to the plants.

#### Temperature assessments

- Compost temperatures were within the optimal range for the nematodes (5-25°C) for the majority of the trial period (see Appendix III).
- The maximum temperature was 24.1°C on 3 October and the minimum temperature was 3.5°C on 20 October.
- Temperatures fell below 5°C on only three dates: 3.9-4.6°C for five hours on 19 October, 3.5- 4.6°C for eight hours on 20 October and 4.6°C for two hours on 24 October. This should not have affected nematode viability. Although the nematodes would not have been active below 5°C, they would have remained alive and infective at any temperature above freezing, and would have become active again as soon as temperatures rose above 5°C.

#### Conclusions over the three project years

- In laboratory tests, 'Nemaslug' at half-rate was as effective as recommended and double rates in killing *D. panormitanum*, with a mean of 80% mortality four weeks after treatment.
- In laboratory tests, 'Nemaslug' at 1/8 rate was as effective as recommended or 1/4 rates in killing *O. pfeifferi*, with 100% mortality three weeks after treatment.
- In laboratory tests, half, recommended and double rates of 'Nemaslug' all significantly reduced mean % leaf feeding by *D. panormitanum* over a 5-wk

period after treatment, with double rate giving significantly better reduction (70%) than recommended (45%) and half-rates (48%).

- In laboratory tests, half, recommended and double rates of 'Nemaslug' all significantly reduced the mean % *D. panormitanum* remaining in contact with treated compost during the first week after treatment, i.e. the nematodes had a repellent effect on the slugs.
- Time-lapse video recording confirmed that when given the choice, the slugs preferred untreated compost rather to that treated with 'Nemaslug'.
- Time-lapse video recording showed that activity of *O. pfeifferi* was significantly reduced on compost treated with 'Nemaslug' when no choice of untreated compost was given, ie. an irritant effect was demonstrated.
- When 'Nemaslug' was applied at recommended, half, quarter and one eighth rates to replicated plots of Choisya plants in liners on a commercial nursery in two consecutive trials, none of the treatments led to a significant increase in kill of *D. panormitanum* or *O. pfeifferi* when compared with water-treated control plots. In the first trial during late July and early August, lack of control was likely to have been due to exceptionally high temperatures leading to poor nematode survival and efficacy. In the second trial during October, which did not include the snails, lack of slug control is likely to have been due to the questionable quality of the nematodes supplied for the trial, although the known avoidance response of the slugs to nematode-treated compost may also have contributed.
- Assessments of numbers of damaged leaves per pot on the second trial on the commercial nursery did not give conclusive results. In any future efficacy trials, a leaf damage scoring system should be used.
- In both the trials on the commercial nursery, Draza at the recommended rate gave no control of the slugs or snails.
- Further research is needed to evaluate the efficacy of 'Nemaslug' in an integrated control strategy against slugs and snails on a commercial HNS nursery. Recent research by the 'Nemaslug' supplier in Europe has demonstrated the improved efficacy and cost-effectiveness of a 'little and often' approach to using nematodes against slugs on field vegetable crops. This approach should be investigated on HNS, where regular low-dose applications could be made using the overhead irrigation system.

### **OBJECTIVE 5: EVALUATION OF BARRIER OR REPELLENT TECHNIQUES AGAINST THE PREDOMINANT SNAIL AND SLUG SPECIES**

Barriers or repellents could prevent infestation of new plants brought onto the nursery and also reduce the abundance of resident snail and slug populations.

#### **Materials and Methods**

In the first two years of the project, the potential repellent, irritant, antifeedant or barrier effects of fourteen products were investigated against *D. panormitanum* and *O. pfeifferi*, using replicated laboratory bioassays. During year 3, some additional tests on these products were conducted, using similar approaches to those in years 1 and 2, in conditions conducive to slug and snail activity i.e. 15°C, high RH% and 16:8 Light:Dark. The methods and results are reported below.

### Behavioural response of slugs and snails (low concentrations of soluble products, nochoice experiments)

In the second year report only preliminary data were presented. The results of the full ten replicates are reported here. The behavioural response of the snails and slugs to low concentrations of three treatments were identified using low-light, time-lapse video. The treatments were cinnamamide and ureaformaldehyde at concentrations between 0.1 and 1% and Croptex-Fungex ® at concentrations between 0.1 and 0.625% (the current maximum concentration when applied as a fungicide). If proved effective, these low concentrations would be more cost-effective than higher dose rates.

One slug or snail was placed in each of 10 replicate, 16 cm-diameter white plastic containers containing damp compost, onto which the treatment was added, and slug behaviour was recorded for one night (14h). Individuals of *D. panormitanum* weighed between 300 and 400 mg and those of *O. pfeifferi* weighed between 100 and 160 mg. The video was then digitised and analysed using the software package EthoVision  $\mathbb{R}$ .

# Suppression of D. panormitanum egg development (low concentrations of soluble products)

In an extension of work reported in Year 2, cinnamamide, Croptex-Fungex ® and ureaformaldehyde were evaluated at 0.005 -0.05%. Egg batches with 15-30 eggs were collected from *D. panormitanum* laboratory cultures on a daily basis. Egg batches were transferred into Petri dishes containing filter paper slightly dampened with 1 ml water per dish. Four ml of each test solution were poured onto the filter paper around the egg batch. Hatching rate and egg development was recorded every day for four weeks after egg laying commenced.

Behavioural response of slugs and snails (mattings, no-choice experiments) The data for adult *D. panormitanum* were presented fully in the year 2 report. In this report, data for adult *O. pfeifferi* (for which only preliminary data was given in the year 2 report) and juvenile *D. panormitanum* are presented. The behavioural response of *O. pfeifferi* to six ground-cover mattings (Table 10) were identified using low-light, time-lapse video. One snail was placed in each of 10 replicate, 16 cm-diameter white plastic containers containing damp compost, onto which the relevant matting was added, and snail behaviour was recorded for one night (14h). All mattings were soaked before adding to the compost and water as sprayed onto the surface immediately before the start of the experiment. Individuals of *O. pfeifferi* weighed between 100 and 160 mg. The video was then digitised and analysed using the software package EthoVision  $\mathbb{R}$ .

Trade Name	Description of matting	Abbreviation
Mypex ®	plastic ground cover (woven)	MYP
Florimat 3®	synthetic capillary matting	FM
GeoBond®	thick capillary matting (synthetic/wool blend)	GB
Tex-R ®	copper-impregnated non-woven matting I	TEX-R
Tex-R Landscape Pro®	copper-impregnated non-woven matting II	TEX LP
Tex-R Supercover Plus ®	copper-impregnated plastic woven ground cover	TEX SCP

Table 10. Ground-cover mattings used in laboratory and field trials.

Very similar experiments were done with juvenile *D. panormitanum*. Recording was done for only three hours and experiments started either in the morning or early afternoon under artificial light. Data was analysed separately for three one-hour intervals. Juvenile slugs weighed between 12 and 15 mg and were approximately four weeks old. There were 20 replicate dishes.

#### Field trials with mattings

The preliminary results for the four-week assessment of these two trials were presented in the year 2 report. The full results are reported below.

An area at the University field station planted with clover was used for the trial with *D. panormitanum*. This area had a very large slug population and was regularly used for slug collection during the project. Square pieces of ground-cover matting (1.4 m<sup>2</sup>) of Mypex (a) and Tex-R (b) Landscape Pro (b) were placed on the ground. Thirty-six potted plants, six plants each of Hosta, Choisya, Impatiens, Geranium, Marigold and Chinese cabbage were placed on the matting. All plants were re-potted immediately before the trial and were presumed to be "slug free". Plants were arranged in six rows with a random row arrangement (see Fig. 4a, Appendix I, year 2 report). A 15-20 cm-wide strip at the edge of the matting was left free of pots to act as a barrier against slugs. The number of slugs on each plot was assessed by sampling all pots, including presence under pots. The plant damage was assessed by counting damaged leaves on 12 specific plants on each plot. The assessments took place every 14 days for eight weeks. There were 10 replicates.

One polythene tunnel on a commercial nursery was used for tests against snails. The floor of the tunnel was covered with gravel. Square pieces of horticultural matting  $(1.4 \text{ m}^2)$  of Mypex and Tex-R Landscape Pro ® were placed on the ground. Sixteen potted plants, four plants each of *Choisya* and *Euonymus* and eight plants of *Elaeagnus* were placed on the matting in four rows (4x4 square). A 15-20 cm wide strip at the edge of the matting was left free of pots to act as a barrier against slugs and snails. Each plot was surrounded by 60-80 *Elaeagnus* plants with a natural infestation of *O. pfeifferi* of approximately 15-30 snails per plant. The number of slugs and snails on the plot and plants and the damage caused to the plants were assessed for eight specific plants per plot after four and eight weeks respectively. There were six replicates of each treatment.

#### **Results and Discussion**

#### Behavioural response of slugs and snails (low concentrations of soluble products, nochoice experiments)

The slugs in the untreated arena were very active (Table 11). They moved a mean distance of 12.6m in 14 hours. They were moving for 58 % of the time with a mean velocity of 2.6 cm/min. Their weight loss over 14 hours was marginal and a few slugs even increased in weight during the experiment. No mortality occurred in the control. The behaviour of the slugs when forced to move onto treated surfaces was completely different to that in the control. The slugs moved for significantly less time in all treatments (N = 100, ANOVA: P < 0.001, Table 11). At higher concentrations they often moved at a significantly lower speed (N = 100, ANOVA: P < 0.001, Table 11). In all treatments except 0.1 % copper ammonium carbonate the weight loss during the 14 hours was significantly higher than in the control (N = 100, ANOVA: P < 0.001, Table 11). The mortality rate was significantly increased with 0.5 and 1 % of cinnamamide and ureaformaldehyde (N = 100, G-test: P < 0.05, Table 11). The concentration of the product, the proportional weight loss of the slug and the absence or presence of any product were found to be factors significantly influencing the track length (N = 100, multiple regression:  $P < 0.001 \text{ R}^2 = 0.69$ ). The distance travelled by the slug was reduced by at least 31 % in comparison with the control (Table 11). This reduction was significant in all treatment-dose combinations except the lowest concentration of copper ammonium carbonate and ureaformaldehyde (N = 100, ANOVA: P < 0.001, Fig. 33). A visual output of the track D. panormitanum

moved in an eight-hour period at different concentrations of ureaformaldehyde can be seen in Fig. 34.

Table 11. Behavioural and physiological response and mortality of *D. panormitanum* to different concentrations of molluscicides over a 14 hour period. Asterisks indicates where means are significantly different from control, \*\*\* = P < 0.001, \* = P < 0.05. CT= control, CF= Croptex Fungex®, CIN=cinnamamide, UF=ureaformaldehyde.

treat-	reduction of	move	velocity	weight	weight	mortality
ment	track length [%]	[min]	[cm/min]	loss [mg]	loss [%]	rate [%]
СТ	x	484 ± 23	2.6 ± 0.1	6 ± 8	1 ± 2	0
CF 0.1%	31	338 ± 26***	2.6 ± 0.2	53 ± 15	15 ± 4	0
CF 0.5%	51	296 ± 34***	2.2 ± 0.1	129 ± 12***	37 ± 3***	10
CF 0.6%	58	265 ± 20***	2.0 ± 0.1***	124 ± 14***	36 ± 4***	20
CIN 0.1%		337 ± 18***	2.1 ± 0.1	113 ± 17***	34 ± 6***	0
CIN 0.5%		208 ± 19***	1.2 ± 0.1***	175 ± 11***	50 ± 3***	60*
CIN 1%		107 ± 15***	1.0 ± 0.1***	200 ± 17***	57 ± 5***	100*
UF 0.1%	<b>39</b>	313 ± 36***	2.5 ± 0.1	82 ± 16***	24 ± 5	20
UF 0.5%	78	161 ± 20***	1.8 ± 0.2***	210 ± 10***	60 ± 3***	90*
UF 1%	80	123 ± 11***	2.1 ± 0.2	191 ± 23***	54 ± 6***	50*
Ρ	n.a.	***	***	***	***	*

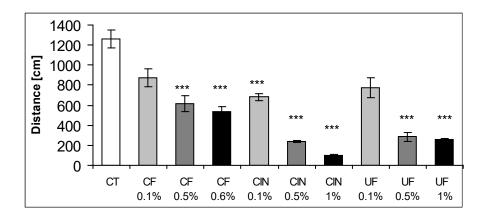


Fig. 33. Distance moved by *D. panormitanum* in 14 hours when forced to move onto treated surfaces. \*\*\* indicates where means are significantly different from control, P < 0.001. Abbreviations as in Table 11.

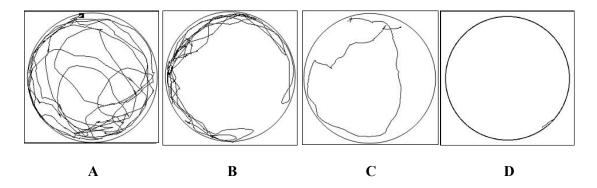


Fig. 34. Representative tracks from *D. panormitanum* for an eight-hour dark period: A= Untreated compost (control), B –D Ureaformaldehyde: B= 0.1 %, C= 0.5 %, D= 1 %.

## Suppression of egg development (low concentrations of soluble products) The three potential molluscicides investigated had significant effects on egg development at much lower concentrations than those at which effects on feeding and movement were observed. At the previously investigated concentrations of 0.1, 0.5 and 1 % no egg development was observed. Hatching rates lower than in the control were observed in concentrations as low as 0.005 % active ingredient. Hatching rates were significantly lower than in the control at 0.5 % (all treatments), but also at a tenth of that concentration i.e. 0.05% (cinnamamide, N = 200, Kruskal-Wallis test: *P* < 0.001, Table 12). An additional effect was the delay in the individual egg development which was observed to be significant for all treatment-dose combinations except 0.01 % copper ammonium carbonate (N = 2618, Kruskal-Wallis test: *P* < 0.001, Table 12).

Table 12. Egg development of *D. panormitanum* at different concentrations of molluscicides. \*\*\* indicates where means are significantly different from control, P < 0.001. Abbreviations as in Table 11.

Treatment	batch	hatching	egg
	size [eggs]	rate [%]	dev [d]
total N	200	200	2618
СТ	23.1	92 ± 2	15.0 ± 0.1
CF 0.005%	22.3	61 ± 5***	15.6 ± 0.1***
CF 0.01%	21.2	78 ± 6	16.6 ± 0.4***
CF 0.05%	21.1	0 ± 0***	n.a.
CIN 0.005%	20.7	82 ± 4	15.5 ± 0.2***
CIN 0.01%	19.9	65 ± 6***	14.6 ± 0.2
CIN 0.05%	19.4	0 ± 0***	n.a.
UF 0.005%	21.4	92 ± 2	15.5 ± 0.1***
UF 0.01%	23.7	86 ± 4	15.4 ± 0.1***
UF 0.05%	22.8	43 ± 9***	15.9 ± 0.2***
<i>P</i>	n.s.	***	***

#### Behavioural response of slugs and snails (mattings, no-choice experiments)

When snails were forced to move onto ground-cover mattings in the laboratory nochoice experiment, all types of copper-impregnated mattings significantly reduced the activity and the resulting track length of the snails (N = 10, Anova, P < 0.001, Table 13 and Fig. 35) in comparison with the standard plastic woven matting (Mypex®). The highest reduction in track length was 91 % with Tex-R Landscape Pro. Capillary mattings also reduced snail activity by up to 33 % in comparison with the plastic woven matting, but these trends were not significant. The main effect was irritation causing dehydration and severe weight loss. Some representative tracks for the different mattings can be seen in Fig. 36, A-F. On some of the mattings the velocity of the snails was higher than on Mypex and on others it was lower, but none of these differences were significant (N =10, Anova: P > 0.05, Table 13).

Table 13. Movement activity of *O. pfeifferi* on different ground-cover mattings in a 14-hour period. Different letters represent significant differences between treatments (P < 0.001).

	Treatment								
-	MYP FM GB TEX-R TEX LP TEX SCP P								
Distance [cm]	283 ± 60 a	267 ± 54 a	190 ± 22 a	59 ± 6 b	27 ± 22 b	35 ± 7 b	***		
Move [min]	360 ± 71 a	351 ± 53 a	240 ± 25 a	91 ± 25 b	42 ± 7 b	44 ± 10 b	***		
Velocity [cm/min]	0.8 ± 0.1	0.7 ± 0.1	0.8 ± 0.1	0.5 ± 0.1	0.7 ± 0.1	0.9 ± 0.2	ns		

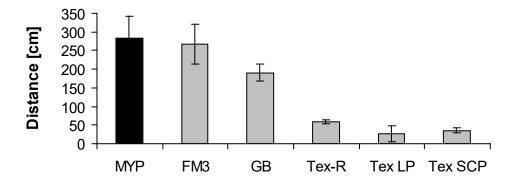


Fig. 35. Track length of *O. pfeifferi* on different horticultural mattings in 14 hours in no-choice experiment.

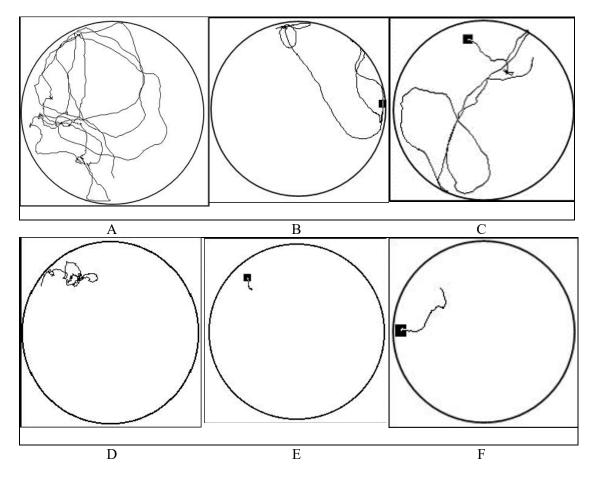


Fig. 36. A-F: Representative tracks of *O. pfeifferi* on different horticultural mattings during an eight-hour dark period:

A: Mypex	B: Florimat3	C: Gabon
D: Tex-R	E: Tex-R Landscape Pro	F: Tex-R Supercover Plus

Unfortunately, weight loss of the snails at the end of the experiment was not recorded for all treatments. However, for those recorded there was a clear difference in weight loss between copper-impregnated mattings (Tex-R Landscape Pro and Tex-R Supercover Plus; above 30 %) and other mattings (Florimat3 and Geobond; 8 %). Little mortality occurred until the end of the experiment (10 % with Tex-R Landscape Pro and 20 % with Tex-R Supercover Plus). No statistical analysis was done on snail weight loss and death, due to the lack of a full data set for all treatments.

When juvenile slugs were forced to move onto Tex-R matting, their activity was very strongly reduced. On average slugs on Tex-R stopped moving after only 28 minutes and in most cases, died shortly after that. The irritant effect of the matting was so strong that high levels of mortality were reached within a few hours. With 95 % of the slugs, mortality after three hours was significantly higher on Tex-R than on Mypex (N = 20, Fisher test: P < 0.001). All slugs on Tex-R matting stopped moving within the second hour of the experiment. This result was probably caused by severe and irreversible dehydration of the slugs, despite the Tex-R being damp. Whereas slugs moving on Mypex lost only 8 % of their weight, those on Tex-R lost significantly more (62 %; N = 20, t-test: P < 0.001, Table 14). Only 15 % of all slugs on Tex-R managed to crawl the linear distance of 4 cm from the centre of the arena to its edge, which was significantly lower than on the Mypex (where all slugs reached the edge; N = 20, Fisher test: P < 0.001). Comparing Tex-R with Mypex, the time spent moving was reduced by 80 % and the distance travelled was reduced by 93 %, in both cases this was a significant reduction (N = 20, t-test: P < 0.001, Table 14). A detailed summary of all the recorded parameters can be found in Table 14.

Observed nerometer	Matt	ing	Р	Change [%] TEX	
Observed parameter	MYP TEX		F	in comparison with MYP	
Frequency of activity	2.5 ± 0.5	1.3 ± 0.1	*	-49	
Total duration of activity [min]	124 ± 7	25 ± 2	***	-80	
Mean duration per activity [min]	81 ± 11	21 ± 1	***	-74	
Total Distance [cm]	233 ± 21	17 ± 1	***	-93	
Mean distance per activity [cm]	159 ± 26	15 ± 1	***	-91	
Velocity [cm/min]	1.8 ± 0.1	0.7 ± 0.0	***	-62	
Time until immobilisation [min]	129 ± 7	28 ± 3	***	-79	
Weight loss [mg]	1.7 ± 0.2	8.4 ± 0.4	***	405	
Weight loss [%]	12 ± 2	62 ± 3	***	50	
Mortality rate [%]	0	95	***	95	

Table 14. Performance of juvenile *D. panormitanum* on Mypex and Tex-R matting during a three-hour period.

On Mypex the juvenile slugs moved a distance of more than 2 m in three hours (Table 14 and Fig. 37) which is quite remarkable considering their body size and weight. In the first hour alone they moved more than a metre (Fig. 38), but this distance continuously decreased over time, which is a common behaviour pattern in this type of experiment, after initial exploration of the arena. As already stated earlier in this section, slugs on Tex-R only moved only for the first and second hour, but they moved for a shorter distance than those on Mypex, approximately 15 cm in the first hour and less than 2 cm in the second hour.

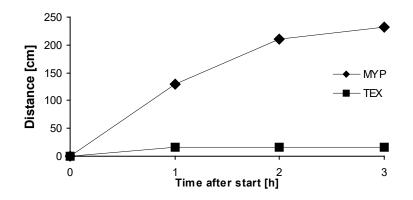


Fig. 37. Accumulated distance travelled by juvenile slugs on Mypex and Tex-R matting over a period of three hours.

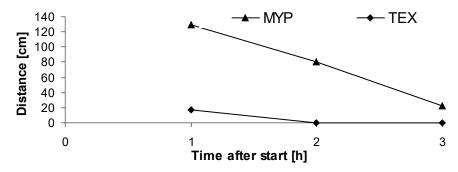


Fig. 38. Distance travelled by juvenile slugs on Mypex and Tex-R matting over a period of three hours.

During the first hour, slugs on Mypex moved for nearly all of the time (Fig. 39). This high proportion of the time spent moving gradually fell to less than one third of the time in the third hour. On Tex-R, the slugs moved for approximately 25 minutes during the first hour and for less than a minute in the second hour.

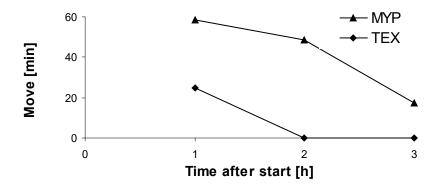


Fig. 39. Time spent moving by juvenile slugs on Mypex and Tex-R mattings in three periods of one hour.

The speed at which juvenile slugs moved on Mypex during the first hour was extremely high in relation to their body size. They moved at 2.2 cm/min (Fig. 40), i.e. more than half the speed of adult slugs for a comparable period. Their speed continuously decreased but not as quickly as the distance travelled or the time spent moving decreased over time. Mean slug speed always remained higher on the Mypex than on the Tex-R during the first hour.

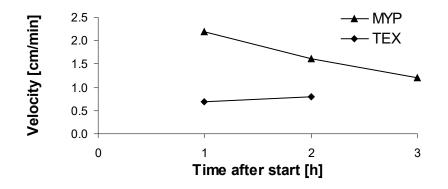


Fig. 40. Change in velocity of juvenile slugs on Mypex and Tex-R mattings over a period of three hours.

#### Field trials with mattings - slugs

Slug species found on the plots included *D. panormitanum*, *D. reticulatum*, *Limax maximus* and several species of Arionidae. After two weeks approximately 10 % of plant pots placed on the Mypex matting were found to be infested and this increased to 35 % after 8 weeks (Fig. 41). There was a lag in the egg infestation of pots placed on Mypex, but in the last two weeks levels of infestation with eggs were similar to those with adults. The infestation with slugs and eggs of plant pots placed on Tex-R matting remained under 4.5 % until the end of the trial. After eight weeks the frequency of both slugs and eggs found under pots was significantly lower on Tex-R than on Mypex matting (N = 360, G-test: P < 0.05).

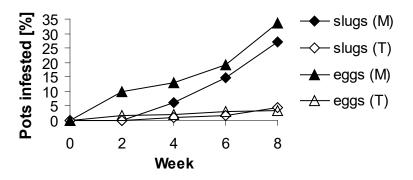


Fig. 41. Frequency of infestation of plant pots with slugs and slug eggs in field trial with mattings over a period of eight weeks (M = Mypex; T = Tex-R).

At the final assessment, a mean of 22 slugs were found per Mypex plot. On Tex-R plots, less than two slugs were found per plot, which was significantly lower, a

reduction by 93 % (N = 10, paired t-test: P <0.001, Fig. 42A). The accumulated number of eggs found under and in plant pots was 134 on Mypex and only 13 on Tex-R plots, a significant reduction in eggs by 90 % (N = 10, paired t-test: P <0.001, Fig. 42B).

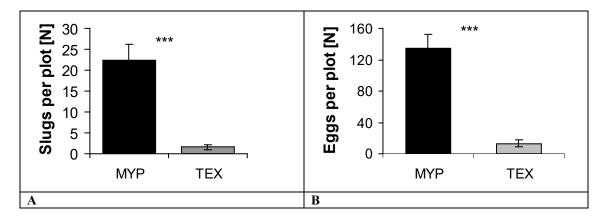


Fig. 42. Slug infestation of plots in matting field trial. A: number of slugs per plot at wk 8. B: Accumulated slug eggs per plot until wk 8.

On Mypex at the start of the trial, the number of slugs found under pots at the edge of the plot was not higher than under those in the centre of the plot. In contrast, in the second half of the field trial, there was a clear trend of more slugs towards the centre of each plot.

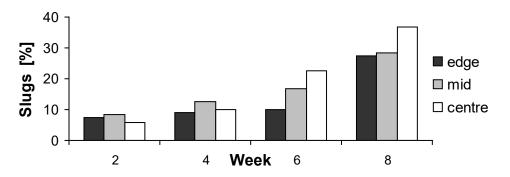


Fig. 43. Frequency of slugs found under plant pots on Mypex at edge, mid and centre-position over a period of eight weeks.

On Tex-R, slugs were only found under pots at the edge of the plot at the first assessment, but only 2.5 % of all assessed plant pots were infested. The later assessments had no clear trends regarding the presence of slugs, the frequency of presence under pots at a certain position seemed to vary (Fig. 44). Observations for the frequency of position of eggs under pots were similar for Mypex plots. On Tex-R plots more eggs were laid under the central pots.

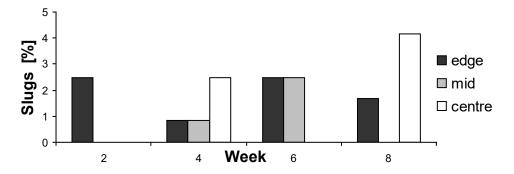


Fig. 44. Frequency of slugs found under plant pots on Tex-R at edge, mid and centreposition over a period of eight weeks.

On Mypex plots, the frequency of damaged plants was already 73 % after two weeks and this increased to levels above 85 % in the following assessments (Fig. 45). On Tex-R matting the frequency of damage was lower, especially at the first assessment when 62 % of all plants had no damage at all. Later, approximately 70 % of all plants had some damage. At the final assessment the proportion of plants with damage was significantly (more than 20 %) lower on the Tex-R matting than on the Mypex (N= 360, G-test: P < 0.05).

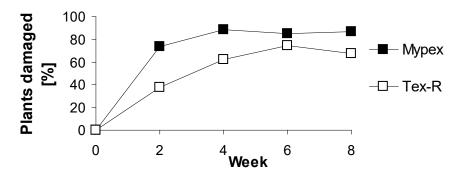


Fig. 45. Frequency of damaged plants in matting field trial over eight weeks.

The extent of the damage on plants was significantly lower on plants placed on Tex-R than on those placed on Mypex (N = 10, paired t-test: P < 0.001, Fig. 46). After eight weeks the accumulated number of damaged leaves for the Mypex plots reached more than 300 (based on 12 plants, extrapolated per plot: 915 leaves), for the Tex-R plots this was less than 100 leaves (extrapolated: 290), a reduction by 68 %.

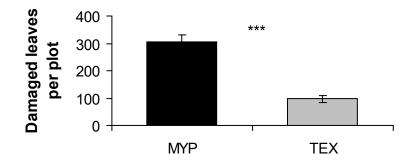


Fig. 46. Accumulated number of damaged leaves per plot (based on 12 plants sampled) during an eight-week field trial comparing Tex-R and Mypex mattings.

On the Mypex plots, nearly all plants were damaged at the end of the trial. Only some Geranium and Hosta plants remained completely undamaged (Fig. 47). For all plant species the proportion of damaged plants was much higher than the proportion of plant pots found to be infested with slugs or slug eggs. The frequency of plant damage and the intensity of the damage were not as much reduced by Tex-R matting as the level of plant infestation and there are two possible reasons for this :

- (1) Slugs are nocturnal, but assessments were done during the day. This means that only the population which had established itself on the plots was recorded (with limitations of finding all slugs during the day due to sampling technique). Slugs which may have been invading the plots at night from the surrounding clover patch were not recorded.
- (2) There was some "background noise" in the data due to some insect plant damage which was difficult to distinguish from slug damage.

On the Mypex plots, the Choisya and Impatiens plant pots had the highest rate of infestation with slugs (65%), while the Hosta and Impatiens plant pots had the highest rate of infestation with slug eggs (Fig. 47). The frequency of infestation of the Chinese cabbage plant pots with both slugs and slug eggs was remarkably low (only 10 and 35 % respectively, despite the 100 % plants damaged and a high number of damaged leaves per plant). In the Choisya, Impatiens and Marigold plant pots the frequency of infestation with slugs was higher than that of slug eggs. For the Chinese cabbage it was the opposite and for the Geranium and Hosta the frequency of slugs and slug eggs on each respective plant species was the same.

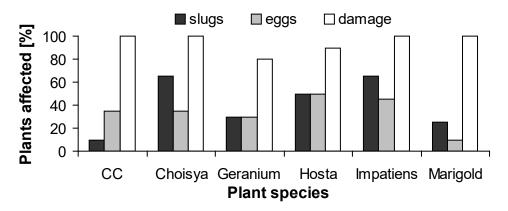


Fig. 47. Percentage of plants placed on Mypex plots infested with slugs or slug eggs and % plants damaged for different plant species at the final assessment in the matting field trial (CC = Chinese cabbage).

#### Field trials with mattings - snails

In the commercial nursery trial many more snails (*O. pfeifferi*, 84 %) were found than slugs (*D. panormitanum*, *D. reticulatum* and Arionidae; 16 %). Mean slug numbers on Tex-R plots were reduced by 20 % in comparison with the Mypex plots. However, this difference was not significant (N = 6, paired t-test, P > 0.05, Table 15, Fig. 48). In comparison with the Mypex plots the mean number of snails found on the matting or associated with the plant pot was 36 % lower on the Tex-R plots, a significant reduction (N = 6, paired t-test, P < 0.01, Table 15, Fig. 48). Overall mollusc infestation was 34 % lower on Tex-R than on Mypex (N = 6, paired t-test, P < 0.05, Table 15, Fig. 48).

Of the molluscs found on the plots, the proportion found on the matting itself was low (between 1.5 and 14 % of the total). Most slugs or snails were found on the pots, on the compost or on the plant. The proportions of slugs or snails found on the matting were lower on the Tex-R plots than on the Mypex plots. There were 78 % less slugs and 60 % less snails on the Tex-R matting in comparison with the Mypex matting. This reduction was significant for the snails (P<0.01, Table 15) but not for the slugs, as there were much fewer slugs than snails on the matting and the variation in slug numbers was high.

The efficacy of Tex-R matting in this trial was probably reduced due to several factors:

- Plants were already infested with snails when the trial started (adults were removed when found, but some adults and probably nearly all eggs were overlooked).
- Plants grew more quickly than anticipated and the surrounding plants grew to touch the plants on the matting and functioned as "bridges" for the slugs and snails to move onto the plants on the plots.

Tex-R Landscape Pro was used, which is a thick matting intended for use on sandbeds with capillary irrigation. The plants in the trial were watered by overhead irrigation and the gravel floor was always wet and the relative humidity very high. Thus the mattings were probably always very damp and algae growth developed on the Tex-R matting, reducing its efficacy.

Table 15. Mean number of slug, snail and total molluscs ( $\pm$ SE) found per plot at the final assessment of an eight-week matting trial on a commercial nursery.

	slugs [N]				total		
Matting	pots	matting	total	pots	matting	total	molluscs
MYP TEX	-	$\begin{array}{c} 1.5\pm0.8\\ 0.3\pm0.3\end{array}$	-		$\begin{array}{c} 14\pm 4\\ 6\pm 2\end{array}$	$\begin{array}{c} 83\pm13\\ 53\pm7\end{array}$	$\begin{array}{c} 97\pm17\\ 64\pm8 \end{array}$
Ρ	ns	ns	ns	*	**	**	*
120 100 80 60	) - ) -	■ MYPE □ TEX-F		**		Ī	*

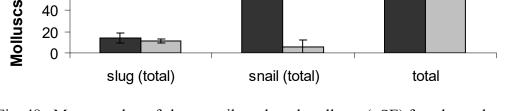


Fig. 48. Mean number of slugs, snails and total molluscs ( $\pm$ SE) found per plot at the final assessment of an eight-week matting trial on a commercial nursery.

#### **Conclusions over the three project years**

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The potential repellent, irritant, antifeedant or barrier effects of fourteen products • were investigated against D. panormitanum and O. pfeifferi, using replicated laboratory tests. During year 1, several of these products showed good repellent and mortality effects and all barrier treatments tested significantly reduced leaf damage by the slugs and/or the snails. The barrier treatments tested in year 1 included aluminium and copper foil, the experimental seed dressing/ repellent / antifeedant cinnamamide, the fungicide copper ammonium carbonate (Croptex Fungex), garlic concentrate, a mulch based on chipboard waste, a grit-like mineral product (SnailBan) and the copper-impregnated ground-cover matting Tex-R, used on sandbeds as a root retardant and weed suppressant. The width of the barrier treatment needed to be in an appropriate proportion to the slug or snail body size, to prevent access to the plant material.

- During year 2, additional products were tested in the laboratory, including liquid ureaformaldehyde, the active ingredient in the mulch and also used in pellets as a slow-release nitrogen fertiliser, and a range of other ground-cover mattings including additional copper-impregnated ('Spin Out'-treated) mattings. The products were confirmed to have seven different modes of action against slugs and snails:
  - > Repellency, causing a chemical barrier effect
  - Physical barrier effect (e.g. foils and some mattings)
  - Irritation
  - Mortality of adults and juveniles
  - Antifeedant effect
  - Suppression of egg development (egg mortality)
  - Reduction in egg production
- The most promising of the novel liquid molluscicides against both slugs and snails were considered to be cinnamamide, Croptex Fungex and ureaformaldehyde. These products were further evaluated against the slugs in no-choice laboratory tests at a range of reduced concentrations during years 2 and 3. These tests showed that even at very low concentrations, the three products had long-term harmful effects on the slugs. Slug movement and speed on damp compost were reduced and extreme weight loss occurred within 14 hours, which is likely to reduce egg-laying and survival in the long-term. Both cinnamamide and ureaformaldehyde led to up to 100% and 90% slug death respectively (depending on concentration) within the 14 hours. All three products significantly reduced slug egg hatching rates at concentrations as low as 0.005% i.e. 50-100 times lower than those effective against adult slugs (0.5-1%). The low concentrations of the three products also delayed slug egg development time, which are likely to lead to reduced population growth.
- With *O. pfeifferi*, 0.1-1% concentrations of all three novel liquid molluscicides significantly reduced snail damage, and 1% cinnamamide and 0.6% Croptex Fungex reduced snail activity by 99% and 93% respectively.
- Croptex Fungex is approved for use as a fungicide at concentrations of up to 0.6% and this and other copper-based fungicides may give incidental control of slugs and snails when applied for disease control. The work in this project has shown that there may be the commercial potential for the development and registration of copper fungicides as molluscicides, particularly if used at low rates.

- Ureaformaldehyde is already used as a slow-release nitrogen fertiliser, but its potential registration as a molluscicide would be questionable due to the current environmentally-sensitive issue of the potential run-off of nitrogen, particularly in NVZs (nitrate vulnerable zones) and in areas with overhead irrigation. However, if used at very low rates or as a combined, controlled-release nitrogen fertiliser and molluscicide, it may have potential for registration for this use.
- Cinnamamide has also been used as an experimental repellent against both vertebrate and invertebrate pests, e.g. as a seed dressing against slugs, but it may be too expensive for commercial use. However, as work in this project has demonstrated its potential at low concentrations as a molluscicide and against slug eggs, with careful timing, it may prove cost-effective should a product ever be registered as a molluscicide.
- Copper-impregnated mattings were shown to have significant harmful effects on both slugs and snails in laboratory tests. In year 1, slugs and snails forced to move onto Tex-R matting in laboratory tests led to 90% slug death, 91% reduction in slug activity and 98% reduction in snail activity. When given a choice of compost or Tex-R, both slugs and snails showed a strong preference for the compost. In similar experiments in year 2, when given a choice between Tex-R and Mypex, both slugs and snails showed a strong preference for the Mypex, with 88% and 93% slug and snail movement being on the Mypex. In year 3, three types of Tex-R mattings (Tex-R, Landscape Pro and Supercover Plus were compared in no-choice experiments and all of them significantly reduced snail and slug activity. Although the slugs moved further on these surfaces than the snails, weight loss and mortality rates of the slugs were higher. Juvenile slugs forced to move onto Tex-R stopped moving within two hours and 95% died within four hours.
- Tex-R matting was selected for testing in field trials during years 2 and 3, due to its strong adverse effects on both slugs and snails in laboratory tests and to its current commercial availability and practicality. In a slug trial at Newcastle University field station in an area planted with clover, and heavily infested with slugs including *D. panormitanum*, damage to a range of plant species including Choisya and Hosta was significantly lower on plants stood on Tex-R plots (over 20% less damaged plants and 68% less damaged leaves) than on those stood on Mypex. Slug numbers and egg production were also significantly lower (93% and 90% reduction respectively) on Tex-R plots than on Mypex ones. However, the ability of a few slugs to overcome the plant-free border of the Tex-R plots (15-20 cm border on a square of matting 1.4m<sup>2</sup>) and to spread into the centre of the plots demonstrated the likely need to cover the entire production area of susceptible plant species with the matting, i.e. not just the edge as a repellent physical-

chemical barrier. If an established slug infestation is already present or if slugs move onto the Tex-R in a heavy infestation, use of a suitable molluscicide (biological or chemical) in an integrated control strategy within the production area would be necessary. The work in this project focussed on *D. panormitanum* and *O.pfeifferi* but larger slugs e.g. *Arion* species may be less effectively repelled than the smaller *Deroceras* species.

- In a snail trial with Choisya, Euonymus and Elaeagnus plants in a polythene tunnel on a commercial HNS nursery in years 2 and 3, which was heavily infested with *O. pfeifferi*, numbers of snails per plot were significantly reduced (by 36%) on Tex-R Landscape Pro plots rather than Mypex plots. In this trial, the efficacy of Tex-R is likely to have been reduced by the layer of algae which grew on the surface in the very wet conditions in the tunnel, and by other experimental problems encountered.
- All the copper-impregnated mattings tested in this project significantly repelled slugs and snails, reduced their activity and with slugs, led to high mortalities. Several commercial growers have commented that where they have used Tex R for its root control properties, slug and snail problems have been considerably reduced. However, the surface of the matting may need to be swept, scraped or sprayed with an algicide between crops, to avoid accumulation of compost, plant debris, algae and moss, which would reduce the matting efficacy. The observed effect on all nurseries where Tex R mattings are being used is the prevention of slug and snail movement into the covered area. However, any slugs and snails on infested plants are not controlled, so unless the plants are completely uninfested when placed on the matting, a molluscicide is needed to prevent plant damage. There has been some commercial success using copper fungicide sprays which are reported to have an incidental effect on slugs and snails by causing them to leave the plants.
- Choice of specific mattings will depend on different production and irrigation systems e.g. if using overhead irrigation, a woven matting allowing rapid water drainage and thus preventing algal and moss growth would be more effective against slugs and snails. The mattings should be viewed as a preventive measure for slugs and snails; on any heavily-infested plants, control with a molluscicide will be necessary. The mattings need to be tested further against both slugs and snails within an integrated control strategy on a commercial HNS nursery, together with cultural control measures and biological control with parasitic nematodes.

#### **OBJECTIVE 6: PRODUCTION OF FACTSHEET FOR GROWERS**

The text and photographs for Factsheet 07/02 were provided by the research partners, summarising the year 1 results of the project, and the Factsheet was distributed to all HNS levy payers by HDC. At the end of year 3, it was agreed with the HDC Technical Manager that production of the final Factsheet should be delayed until the completion of a further year's research to identify and validate an effective and reliable integrated control strategy for slugs and snails on HNS, should further HDC funding be agreed.

#### **TECHNOLOGY TRANSFER**

#### Presentations

- Jude Bennison presented the results of the project to date to growers at the HDC / Efford Open Day on 25 September 2001.
- Ingo Schüder presented the results of the project at the IOBC conference 'Slug and Snail Pests' in Lyon, France in March 2001.
- Jude Bennison presented the key results of the project to date in a paper at the IOBC conference 'Integrated Control in Protected Crops', Victoria, Canada, in May 2002 (see below for publication in the conference proceedings).
- Ingo Schüder presented a poster at the BCPC conference, Brighton in October 2002, on novel pesticides for slug and snail control in horticulture (see publication from this conference below).
- Jude Bennison presented a summary of key results of the project to growers during five Defra-funded IPM workshops for growers of ornamentals and herbs around the country during January 2003.
- Ingo Schüder presented a joint paper on behalf of the research partners on 'Sublethal effects of the parasitic nematode *Phasmarhabditis hermaphrodita* on the slug *Deroceras panormitanum* and the snail *Oxyloma pfeifferi*' at the IOBC conference on using parasitic nematodes against soil pests in Salzau, Germany in May 2003.
- Ingo Schüder will present a joint paper on behalf of the research partners on 'Integrated Management of slug and snail pests in Hardy Nursery Stock' at the BCPC conference 'Slugs and snails' on 8-9 September 2003 and a paper will be published in the conference proceedings.
- Ingo Schüder will present a poster on 'Automated analysis of slug and snail behaviour' at the BCPC conference 'Slugs and snails' on 8-9 September 2003 and a paper will be published in the conference proceedings.

#### Publications

- Bennison, J.& Schüder, I. (2001). Slugs and snails in the firing line. *HDC News* No. 77.
- Bennison, J., Umpelby, R. & Buxton, J. (2002). IPM on protected hardy ornamental nursery stock in the UK. *IOBC/wprs Bulletin* **25(1)**, 13-16.
- Schüder, I., Port, G. & Bennison, J. (2002). Screening novel pesticides for slug and snail control in horticulture. *BCPC Conference Proceedings 2002*, 873-878.
- Schüder, I., Port, G. & Bennison, J. (2003). Barriers, repellents and antifeedants for slug and snail control. *Crop Protection*, in press.
- Schüder, I., Port, G. & Bennison, J. A paper 'The diurnal behaviour of *Deroceras panormitanum* and *Oxyloma pfeifferi*' has been submitted to the Journal of Molluscan Studies for consideration of publication.
- Schüder, I., Port, G. & Bennison, J. A paper 'The behavioural response of slugs and snails to novel molluscicides' has been submitted to the Journal of Zoology for consideration of publication.
- An article summarising the results of the project is planned for publication in the 'Grower' in late 2003.

#### **Industry communications**

- During 2002, Jude Bennison discussed the results of the project to date and the practicality of using parasitic nematodes and Tex-R matting with selected growers who were visited during year 1 of the research.
- During the project, Jude Bennison and Heather Maher discussed laboratory results and demonstrated the commercial nursery trials with the parasitic nematodes 'Nemaslug' with the supplier, Becker Underwood.
- Ingo Schüder and Jude Bennison provided information on the results with Tex-R matting, to the suppliers, Fargro Ltd, to include in their technical newsletter.
- During the life of the project, Jude Bennison discussed the results of the project with individual HNS growers during consultancy visits and with ADAS Horticultural Consultants.
- Jude Bennison discussed the key results of the project during a session 'Reducing pesticides with confidence' at the HNS conference 'CONTACT' on 10 January 2003.

#### ACKNOWLEDGEMENTS

Thanks to Becker Underwood for supplying the Nemaslug, to Fargro for the Tex-R mattings, to Certis for the Croptex Fungex and to commercial nursery stock growers for providing experimental sites and plants for the research.

#### REFERENCES

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## Appendix 1



**Figure A**: Typical damage caused by an individual *D. panormitanum* in one week on punched (left) and whole (right) Choisya leaves

#### **Appendix II:**

# HNS 105 Hardy nursery stock: Integrated control of slugs and snails

#### Commercial nursery trial layout and randomisation

				G	L	A	S	S	н	0	U	s	Е		W	A	L	L				
	Р	В	т			Р	В	т					Р	В	т			Р	В	т		
					-					_											_	
	1	1	6			8	2	3					15	3	5			22	4	1		
	2	1	5		]	9	2	7		]	P A		16	3	3			23	4	2	]	
	3	1	4		]	10	2	2		]	T H B		17	3	6			24	4	4	]	
	4	1	2		]	11	2	4		]	E T W E		18	3	1			25	4	7	]	
	5	1	1		]	12	2	5		]	E N B		19	3	4			26	4	5	]	
	6	1	3		]	13	2	6		]	A Y S		20	3	7			27	4	3	]	
	7	1	7		]	14	2	1		]			21	3	2			28	4	6	]	
End door	End door				С	Е	Ν	т	R	Е		Ρ	A	т	н				_	Centr	e aisle	

#### Treatments

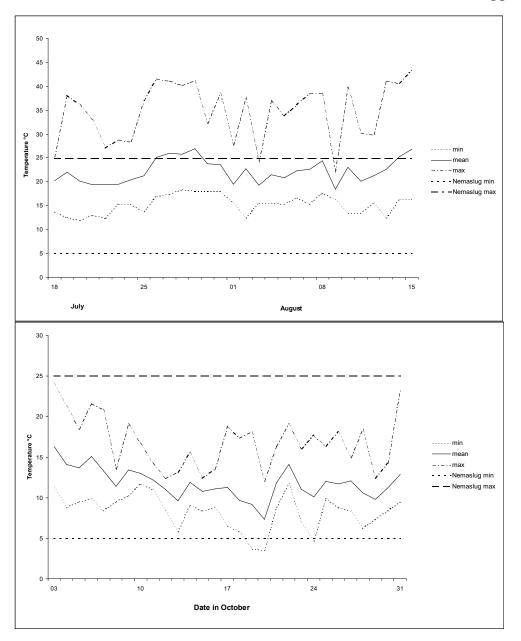
- 1 Untreated
- 2 Untreated
- Draza @ 5.5 kg/ha (0.3g / plot) 3
- 4 Nemaslug @ 300,000 / sqm
- Nemaslug @ 150,000 / sqm 5
- 6 Nemaslug @ 75,000 / sqm
- 7 Nemaslug @ 37,500 / sqm

Plot size : 0.5 sqm

Spray volume (T4-T7) 10,000 l/ha (0.5 l/plot)

- P plot B block **T treatment**

#### **Appendix III**



Compost temperatures (°C) in 'Nemaslug' trials on commercial nursery in first trial (18 July - 15 August and in second trial (3 - 31 October)