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The results and conclusions in this report are based on an investigation conducted over one year. The conditions under which the experiments were carried out and the results obtained have been reported with detail and accuracy. However, because of the biological nature of the work it must be borne in mind that different circumstances and conditions could produce different results. Therefore, care must be taken with interpretation of the results especially if they are used as the basis for commercial product recommendations.

## 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 M Scrace Senior Research Scientist ADAS Arthur Rickwood

..... Date

Report authorised by ..... Dr M Heath Team Manager ADAS Arthur Rickwood

..... Date

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## PRACTICAL SECTION FOR GROWERS

#### **Background and objectives**

Several of the most popular species of bedding plants are susceptible to rust diseases. Rust-affected plants suffer from reduced quality and/or a lack of vigour, and if severely affected may even die. Because there can be a considerable period from the time a plant is first infected to the production of symptoms, there is great potential for the spread of rust diseases between nurseries on infected but symptomless plants and cuttings.

The symptoms and life-cycles of the rust diseases of some of the most widely-grown bedding plant hosts, together with details of previous work into cultural and chemical control measures, are described in the literature review and grower survey for this project, published in April 2000. See also HDC factsheet 23/00.

At the time this project commenced, the number of fungicides that could be used on protected bedding plants for rust control was very limited. Recent changes to the offlabel arrangements for the use of pesticides have meant that more products are now available (although in most cases these are used at the grower's risk).

The objective of this part of Project PC 175 was to assess a range of fungicides, most of which can be used by growers of protected bedding plants, for their crop safety and ability to control rust diseases. The assessment of disease control ability included separate measurements of protectant and eradicant activity.

At this stage only the more 'modern' fungicides were evaluated, as single-product treatments (rather than forming part of a programme). The most effective products from these experiments will go forward to the next stage of the project, where they will be evaluated as the components of fungicide programmes including some of the older broad-spectrum fungicides (e.g. mancozeb, chlorothalonil, etc.). In this way it is hoped that fungicide programmes will be devised which, in addition to giving effective disease control, minimise the risk of the rust fungi developing fungicide resistance.

#### Summary of results

#### Crop safety

The crop safety of ten fungicides was assessed on seven different bedding plant subjects (Antirrhinum, Bellis, Chrysanthemum, Cineraria, Dianthus, Fuchsia and Pelargonium). Two sprays of each of the fungicides were applied, with a seven day interval between them. The first spray was applied during hot, sunny conditions, the second during cooler, cloudy conditions. See Table 1 below for a list of fungicides applied.

Trade Name	Active Ingredient	Application Rate (single)	Application Rate (double)	Approval Status*
Control	water only			
Alto 240EC	cyproconazole	0.33 ml/litre	0.66 ml/litre	4
Amistar	azoxystrobin	1.00 ml/litre	2.00 ml/litre	3
Dorado	pyrifenox	0.25 ml/litre	0.50 ml/litre	\$
Nimrod-T	bupirimate +	3.20 ml/litre	6.40 ml/litre	1
	triforine			
Plantvax 75	oxycarboxin	0.50 g/litre#	1.00 g/litre#	2
Plover	difenoconazole	1.00 ml/litre	2.00 ml/litre	3
Systhane 20EW	myclobutanil	0.45ml/litre	0.90 ml/litre	3
Tilt 250EC	propiconazole	0.40 ml/litre	0.80 ml/litre	3
Twist	trifloxystrobin	1.00 ml/litre	2.00 ml/litre	4
Experimental	mepanipyrim	0.80 g/litre	1.60 g/litre	4

 Table 1:
 Fungicides Used in Phytotoxicity Experiment

# plus wetter.

\* Approval status:

1 - On-label use for protected ornamentals.

- 2 On-label use for specific protected ornamental crop(s). Can be used on other protected ornamentals at grower's risk under the Revised Long-Term Arrangements for Extension of Use (2000).
- 3 Specific Off-Label Approval (SOLA) for use on a protected crop. Can be used on any protected ornamental crop at grower's risk under the Revised Long-Term Arrangements for Extension of Use (2000).
- 4 No approval for use on protected ornamentals.
- \$ Approval for use on protected ornamentals has been recently revoked. There is a two year use-up period for existing stocks.

Most of the fungicides caused little or no damage to the plants, even when applied at double the recommended rate. However, scorch symptoms were produced on some of the plants from the first application of Amistar (azoxystrobin), Twist (trifloxystrobin),

Nimrod-T (bupirimate + triforine) or Plantvax 75 (oxycarboxin). No damage was caused by the second applications of these products.

The second application of Alto 240EC (cyproconazole) resulted in severe damage symptoms on a range of plant types. A specific effect of fungicides from the triazole group (Alto 240EC (cyproconazole), Tilt 250EC (propiconazole), Plover (difenoconazole) and Systhane 20EW (myclobutanil)) was noted on Fuchsia, consisting of distortion and thickening of young leaves. Application of Twist (trifloxystrobin) to Fuchsia resulted in a change in flower shape (later experiments showed that this may have been a specific effect on the variety used in the crop safety experiment).

There were few significant effects of the fungicides on either plant height or dry weight.

In an observation trial to assess the effect of compost drenches of Plantvax 75 (oxycarboxin), damage symptoms were produced on most of the plant types.

#### Disease control ability

This was assessed using the rusts of Bellis (*Puccinia distincta*), Fuchsia (*Pucciniastrum epilobii*) and Pelargonium (*Puccinia pelargonii-zonalis*).

The strobilurin fungicides Amistar (azoxystrobin) and Twist (trifloxystrobin) performed well as protectants against all three of the rusts. They were less effective at eradicating established infections, although Amistar in particular did show some eradicant activity when applied shortly after the plants had been inoculated with the rusts.

The fungicides from the triazole group (Plover, Systhane 20EW, Tilt 250EC) showed excellent activity against Bellis and Fuchsia rusts, as both protectants and eradicants. Their performance against Pelargonium rust, however, was poor, particularly when they were used as protectants. This poor performance contrasts with the results of work carried out in the USA, and further work will be done to try and determine

whether this discrepancy is due to fungicide resistance in the rust population used in this project.

Nimrod-T (bupirimate + triforine) and Plantvax 75 (oxycarboxin) also gave useful reductions in disease levels, but Dorado (pyrifenox) did not perform well.

### Action points for growers

- Of the fungicides used in these experiments, seven can be used legally on protected bedding plants. These are Amistar (azoxystrobin), Dorado (pyrifenox), Nimrod-T (bupirimate + triforine), Plantvax 75 (oxycarboxin), Plover (difenoconazole), Systhane 20EW (myclobutanil) and Tilt 250EC (propiconazole). Approval for use of Dorado on ornamentals has been recently revoked, but there is a two-year use-up period for existing stocks. It has recently become known that Plantvax is not being supported in the EU pesticide review programme and the registration approval will lapse in 2003.
- The results of the crop safety experiment showed that the majority of the fungicides were safe to the majority of plant subjects when used at recommended rates. However, care should be taken to apply the fungicides during appropriate environmental conditions (i.e. avoid hot, sunny or very bright conditions). Products should also be evaluated on a small scale if they have not been used previously on a particular plant species or variety.
- Amistar works extremely well as a protectant product. It is recommended that it is used as a protectant fungicide and not to control existing rust infections.
- Plover and Systhane can be used as both protectants and eradicants against Bellis and Fuchsia rust. As they belong to the same chemical group it is not recommended to use both products in a spray programme. There is some potential for crop damage from these products on Fuchsia, so they should be used with care. Tilt 250EC (again from the same chemical group) gave no advantages over Plover

or Systhane in terms of disease control, and shows greater potential for crop damage. It is therefore recommended that Tilt is not used on Fuchsia.

- Plantvax 75 and Nimrod-T also gave useful reductions in rust disease, and should be considered as part of a rust control programme (particularly protectant programmes). Plantvax 75 is particularly useful, as it belongs to a different chemical group from both Amistar and Plover/Systhane.
- Plantvax 75 should not be used as a drench treatment on bedding plants. It caused obvious crop damage to most of the plant species on which it was tested.
- Differences in the performance of the fungicides against the different rusts were sometimes noted. This should be remembered when using the products against rusts of other bedding or pot plant species.
- The most successful products from these experiments will now be incorporated into programmes with broad-spectrum protectant materials (e.g. chlorothalonil, mancozeb) to try and enhance the anti-resistance component of the programmes.

## Practical and financial benefits

The value of the UK bedding plant industry in 1998 was approximately £150 million (MAFF Basic Horticultural Statistics 1988-1998). The value of Pelargonium crops alone was £11.5 million, and that of Fuchsias £5.5 million.

Losses due to rust diseases are variable, but probably average from two to five per cent overall of those crops susceptible to the disease. However, some Bellis crops have suffered severe losses due to rust, and this work has shown that total crop loss could occur where plants remain untreated. Heavy losses have also been reported in some Fuchsia crops.

These initial experiments have identified fungicides which give effective control of rusts, and which can be used legally by growers of protected bedding and pot plants.

Effective fungicides have been identified from a number of different chemical groups, which should help prevent the development of fungicide resistance in the rusts. It is hoped that the next stage of the work will increase the range and choice of fungicides still further, and provide growers with effective fungicide programmes.

## SCIENCE SECTION

#### **Introduction**

Many different species of rust are known to attack bedding plants, most of which are specific to one plant genus only. The symptoms produced are variable according to the host plant, but commonly pustules are produced containing large numbers of spores, which may be yellow, orange, brown or black in colour. The pustules are invariably produced on the leaves, but other parts of the plant (e.g. stems, flower parts) may be affected on some hosts. The disease affects not only the aesthetic quality of the plants, but can also reduce vigour and, occasionally, result in death.

The spores are usually splash-dispersed and/or carried on air currents, and in this way spread the disease through the growing house. Research has shown that the spores of most of the bedding plant rusts can survive for weeks or sometimes even months on detached leaves. The spores can be carried long distances on the wind and cause new outbreaks. However, with most of the rusts a more common cause of long-distance spread is the transport of infected plants or cuttings. These may be carrying the disease in a latent (symptomless) state. Depending on the environmental conditions, symptoms may not be produced until several weeks after infection has taken place.

Environmental conditions which are conducive to infection vary somewhat between the different rust species, but a critical factor is the presence of free water on the leaf surfaces. If this is present, and other criteria such as a suitable temperature are satisfied, rust spores on the leaf surface can germinate and infect the plant, usually through the stomata. Germination and infection can occur in just a few hours.

The symptoms and life-cycles of the rust diseases of the following host plants, together with details of previous research work into cultural and chemical control measures, are described in the literature review and grower survey for this project, published in April 2000:

Pelargonium (Puccinia pelargonii-zonalis)

Fuchsia (Pucciniastrum epilobii)
Bellis (Puccinia distincta, Puccinia obscura, Puccinia lagenophorae)
Antirrhinum (Puccinia antirrhini)
Dianthus (Uromyces dianthi (carnations/pinks), Puccinia arenariae (sweet william))
Chrysanthemum (Puccinia horiana (white rust), Puccinia chrysanthemi (brown rust))
Cineraria (Puccinia lagenophorae, Coleosporium tussilaginis)

Control of rust diseases on bedding plants grown under protection can be difficult for a number of reasons:

- 1) the disease is usually only noticed when the level of infection is fairly advanced and sporulation is actually noted on plant material.
- 2) there are only a limited number of fungicides which possess approval for use under protection (although the number of products with activity against rusts which can be used on protected ornamentals has increased recently due to changes in the off-label arrangements).
- the efficacy of the fungicide used appears to vary depending upon the rust disease present.

The findings of the literature review revealed that there had been fairly extensive research into the chemical control of rusts on crops which are also widely grown for the cut flower market, such as carnation and chrysanthemum. However, for many of the other crops the amount of previous research was either very limited, had been carried out many years ago (and therefore did not include many of the currently-available fungicides), or was conducted overseas (mainly in the USA).

There had also been only limited research into the true eradicant effect of fungicides against rusts, i.e. control of an infection that has already taken place, rather than a gradual reduction in the overall level of disease resulting from the protection of newly-emerging leaves. Whilst occasionally there seemed to be quite extensive eradicant activity (Dickens, 1990, 1991; Weber & Tilston, 1999) other work seemed

to indicate eradicant activity lasting for just one or two days after infection had taken place (Spencer, 1976,1979).

The objectives of the experiments described in this report were to evaluate a range of fungicides for both crop safety and disease control ability. The fungicides would include:

- those which have been available for some time for use on a range of protected ornamental crops, e.g. Plantvax 75, Nimrod-T.
- those which have recently become available for use on protected ornamentals, or which can be used on a greater range of ornamentals, due to changes in the off-label regulations, e.g. Amistar, Systhane 20EW, Plover, Tilt 250EC.
- those which do not have approval for use on protected ornamentals, but which give good control of rusts on other crops, e.g. Alto 240EC, Twist.

The experiment to check for crop safety would include all of the plant types covered in the literature review. The disease control trials would be conducted using the rusts of Bellis (*Puccinia distincta*), Fuchsia (*Pucciniastrum epilobii*), and Pelargonium (*Puccinia pelargonii-zonalis*).

It was originally intended that Dianthus rust (*Uromyces dianthi*) would be used. However, preliminary work indicated that some bedding Dianthus types could be affected by both *Uromyces dianthi* (carnation rust) and *Puccinia arenariae* (sweet william rust), indicating that these plants had a mixed carnation/sweet william parentage. It could not be guaranteed that a fungicide with activity against one of these rusts would be equally effective against the other, which could lead to problems in the commercial exploitation of the results.

It was therefore decided to substitute bedding Dianthus with Bellis, and to use the 'new' Bellis rust, *Puccinia distincta*. This rust has caused widespread and severe problems on Bellis over the last 3-4 years (Weber *et al*, 1998: Weber & Webster,

1999). Whilst Bellis is known to be affected by two other *Puccinia* species in the UK, all of the outbreaks examined recently in the ADAS Plant Clinic have been caused by *P. distincta*.

A small observation trial was also set up to look into the crop safety of applying compost drenches of Plantvax 75 (oxycarboxin), as this fungicide has approval for drench treatment of carnations grown in peat bags.

#### Materials and methods

#### Location of work

All of the work described in this report was carried out at ADAS Wolverhampton. Disease inoculum for application to the plants was prepared in the laboratories of the ADAS Plant Clinic. The experiments themselves were located in compartments of a multi-compartment glasshouse with computerised control of irrigation and environmental conditions.

#### Isolate collection and maintenance

#### Collection of rust isolates

Infected plant material was received from commercial nurseries in the UK, from ADAS consultants and through the ADAS Plant Clinic.

#### Maintenance of rust isolates

Because rust fungi cannot be grown successfully away from their host plant, the three rust fungi used in the efficacy experiment (Bellis rust, *Puccinia distincta*; Fuchsia rust, *Pucciniastrum* epilobii and Pelargonium rust, *Puccinia pelargonii-zonalis*) were maintained on potted specimens of their respective host plants (known as 'rust stock plants'). Spore preparations were made from the various rust isolates collected and used to inoculate these plants. As the same plants were inoculated (and later reinoculated to maintain the rust in a 'fresh' condition) with each of the various isolates

of their respective rust, a mixed population was built up on these plants for use in the disease control experiments.

For details of the preparation of spore suspensions and the inoculation of plants, please consult the methods section for the disease control experiments.

# Experiment 1: Evaluation of the phytotoxicity of candidate fungicides towards a range of bedding plant species

The potential phytotoxicity of the candidate fungicides was evaluated on a range of the bedding plant species commonly affected by rust diseases. The experiment was a split-plot design with four replicate blocks. Each plot consisted of six plants of each of the bedding plant types.

#### Plant species

The following bedding plant species were used in this experiment: Antirrhinum 'Chimes Light Pink', Bellis 'unknown', Chrysanthemum (garden mum) 'Bravo', Cineraria maritima 'Silverdust', Dianthus 'Strawberry Parfait', Fuchsia 'Beacon Rose' and Pelargonium 'Maverick Red'. All were bought-in as plug plants.

## Crop husbandry

The plugs of each plant type were potted into six-cell packs using Sinclair SHL compost. The exception to this was the Bellis which, due to their larger plug size, were potted individually into seven centimetre square plastic pots.

The plants were grown on raised aluminium mesh benches. Each plot consisted of a separate, isolated piece of capillary matting, onto which the seven plant types were placed. In this way any cross-contamination by fungicides between plots, via contamination of the capillary matting, was avoided. Each plot was irrigated individually via its own trickle pipe. Irrigation was computer-controlled on a timed cycle, with additional hand-watering as and when required. Some protection from direct sunlight was given by spraying 'Summer Cloud' shading onto the glass.

No fungicide applications in addition to the experimental treatments were necessary. However, recurring outbreaks of aphids, particularly on the Bellis plants, required control. Due to the need to avoid the development of any phytotoxicity symptoms which might complicate later assessments, nicotine shreds were used, as these are generally regarded as being safe to use on a wide range of plants, provided the foliage is dry. The shreds were used at the recommended rate of 225g per 560 cubic metres. Five treatments were required during the course of the experiment, at intervals ranging from seven to twenty-one days.

Interestingly, whilst six of the seven plant types showed no symptoms of phytotoxicity from the nicotine, the Dianthus 'Strawberry Parfait' plants began to develop damage symptoms five days after the first application, despite having completely dry foliage at the time of application. The symptom consisted of necrosis of the leaf tips (most noticeable on younger leaves), initially a grey-green colour but progressing to a light brown or almost bleached lesion with a purple margin (see Appendix 1). These symptoms developed following each further application of nicotine, on any leaves that had emerged since the previous application.

## Fungicide application

The candidate fungicides were applied to the point of run-off using an Oxford Precision Sprayer pattern with a carbon dioxide-generated pressure of 1.5 bar, giving a coarse spray output. Details of the fungicides and the rates used are given in Table 1.

In order to carry out a rigorous assessment of phytotoxicity, each fungicide was applied at :

1) The recommended rate (from the label or Specific Off-Label Approval (SOLA)).

2) Double the recommended rate.

Two sprays of each treatment were applied, with a seven-day interval between them. The first sprays were applied on a hot, sunny day - this would be expected to increase the risk of phytotoxicity symptoms developing. The second sprays were applied under cooler, cloudy conditions.

Trade Name	Active Ingredient	Application Rate (single)	Application Rate (double)	Approval Status*
Control	water only			
Alto 240EC	cyproconazole	0.33 ml/litre	0.66 ml/litre	4
Amistar	azoxystrobin	1.00 ml/litre	2.00 ml/litre	3
Dorado	pyrifenox	0.25 ml/litre	0.50 ml/litre	\$
Nimrod-T	bupirimate +	3.20 ml/litre	6.40 ml/litre	1
	triforine			
Plantvax 75	oxycarboxin	0.50 g/litre#	1.00 g/litre#	2
Plover	difenoconazole	1.00 ml/litre	2.00 ml/litre	3
Systhane 20EW	myclobutanil	0.45ml/litre	0.90 ml/litre	3
Tilt 250EC	propiconazole	0.40 ml/litre	0.80 ml/litre	3
Twist	trifloxystrobin	1.00 ml/litre	2.00 ml/litre	4
Experimental	mepanipyrim	0.80 g/litre	1.60 g/litre	4

Fungicides Used in Phytotoxicity Experiment Table 1:

# plus wetter.

\* Approval status: 1 - On-label use for protected ornamentals.

- 2 On-label use for specific protected ornamental crop(s). Can be used on other protected ornamentals at grower's risk under the Revised Long-Term Arrangements for Extension of Use (2000).
- 3 Specific Off-Label Approval (SOLA) for use on a protected crop. Can be used on any protected ornamental crop at grower's risk under the Revised Long-Term Arrangements for Extension of Use (2000).
- 4 No approval for use on protected ornamentals.
- \$ Approval for use on protected ornamentals has been recently revoked. There is a two year use-up period for existing stocks.

#### Assessments

The plants were assessed prior to the first application of the fungicides, and any symptoms noted that could be mistaken for phytotoxicity in later assessments. After the application of the fungicide treatments the plants were checked on a regular basis for the development of symptoms. Detailed assessments were made seven days after each application of fungicides, and immediately prior to the end of the experiment. An assessment was made of any effects on flowering (quantity, quality). At the end of the experiment plant heights and dry weights were measured.

#### Statistical analysis

Plant height and dry weight data was analysed by an ADAS statistician. Assessments of phytotoxicity symptoms were descriptive rather than numerical, and therefore not analysed statistically.

#### Crop diary

Plants bought in as plugs and potted up into six-cell packs or pots:	17-21 April 2000
First fungicide application:	9 May 2000
Second fungicide application:	16 May 2000
Nicotine shreds used for aphid control:	28 April,
	5,12,19 May,
	10 June 2000
Pre-spray assessment:	8 May 2000
Detailed phytotoxicity assessments:	12,15,23,31 May,
	20 June 2000
Flowering assessment:	6 June 2000
Measurement of plant heights and dry weights:	21-23 June 2000
Photographs taken of phytotoxicity symptoms:	24,31 May,
	16 June 2000

## Experiment 2: Evaluation of the protectant activity of the candidate

#### fungicides against rust diseases

The objective of this experiment was to measure the effectiveness of the various fungicides in protecting plants against infection by rust. To do this the plants were first sprayed with the fungicides, and then inoculated with their respective rusts. Inoculations were carried out on two occasions, two and twenty-two days after application of the fungicides. The rust diseases used in the experiment were Bellis rust (*Puccinia distincta*), Fuchsia rust (*Pucciniastrum epilobii*) and Pelargonium rust (*Puccinia pelargonii-zonalis*).

The experiment was a split-plot design with four replicate blocks. Each plot consisted of six plants of each of the bedding plant types.

#### Plant species

The plant species used in the experiment were Bellis 'Medici White', Fuchsia 'Sir Matt Busby' and Pelargonium 'Pink Satisfaction'.

#### General crop husbandry

The plot layout and irrigation technique was the same as used in Experiment 1, except that all plants were potted into seven centimetre square plastic pots. Because fogging was used following the inoculation of the plants with rust, it was found that little irrigation via the trickle lines was necessary, as the fogging provided enough water to keep the capillary matting wet. The temperature in the glasshouse was maintained at approximately 16 °C.

Due to the time of year (autumn) when this experiment was conducted, and the fact that humid conditions were created to favour the development of the rusts, Botrytis was a constant threat. This was counteracted where possible by the removal of senescent flowers and leaves, but it was necessary on one occasion to spray the entire experiment with Rovral (iprodione), a fungicide which has negligible activity against rusts.

As in Experiment 1, aphid infestation occurred, but because of the time of year buildup was slower, and this time was confined to the Bellis plants. The Bellis plants received a single spray of Rapid (pirimicarb) to control the aphids.

### Fungicide application

See Experiment 1 for application method. Details of the fungicides used are given in Table 2. Because it produced severe crop damage on a range of plant types in Experiment 1 (see results section for details), Alto 240EC was not used in Experiments 2 and 3.

Trade Name	Active Ingredient	Rate
Control	water only	
Amistar	azoxystrobin	1.00 ml/litre
Dorado	pyrifenox	0.25ml/litre
Nimrod-T	bupirimate +	3.20 ml/litre
	triforine	
Plantvax 75	oxycarboxin	0.50 g/l*
Plover	difenoconazole	1.00 ml/litre
Systhane 20EW	myclobutanil	0.45 ml/litre
Tilt 250EC	propiconazole	0.40 ml/litre
Twist	trifloxystrobin	1.00 ml/litre
Experimental	mepanipyrim	0.80 g/litre

 Table 2: Fungicides used in protectant activity experiment (Experiment 2)

For details of the approval status of the products, see Table 1.

## Preparation of rust inoculum

The same method of preparation was used for each of the three rusts. Rust-affected leaves were detached from the 'rust stock plants' and washed in deionised water. The leaves were held under the water and the rust pustules brushed with a fine paint brush to dislodge the spores. The number of leaves used for each rust type varied according to the severity of the rust infection on the stock plants. The spore suspension was adjusted to the final volume required to inoculate the experimental plants, and the spore concentration within the suspension was then calculated using a haemocytometer.

## Inoculation of plants

A separate inoculation was carried out for each host plant/rust combination. Taking Pelargonium rust as an example, the rust spore suspension was tipped into a Cooper-Pegler Minipro hand-held pressure sprayer. A timed burst of spray (enough to thoroughly wet the foliage) was given to the six Pelargonium plants in each plot, manipulating the sprayer to try and obtain good coverage of both the upper and lower leaf surfaces.

The Fuchsia and Bellis rusts were applied in the same way. It took approximately two hours in total to apply all three rusts to the trial plots. In order to prevent the spore suspensions drying out during this time, each plot was covered with black polythene when it was not being inoculated.

The concentrations of the spore suspensions used were as follows:

First inoculation (18 October 2000):	Pelargonium rust	-	10,000 spores/ml
	Fuchsia rust	-	5,000 spores/ml
	Bellis rust	-	5,000 spores/ml

Second inoculation (9 November 2000):	Pelargonium rust	-	16,000 spores/ml
	Fuchsia rust	-	9,000 spores/ml
	Bellis rust	-	7,000 spores/ml

### Post-inoculation husbandry

## 1) First inoculation

As soon as the inoculations were complete, the black polythene was removed from each of the plots, and a computer-controlled fogging programme began in order to maintain leaf wetness and provide appropriate conditions for infection by the rusts. The plants received a ten minute burst of fogging once every hour for a total of eighteen hours.

After the initial eighteen hours fogging, the plants received seven hours of fogging in each twenty-four hour period. The fogging again consisted of a ten minute burst every hour, commencing at nine o'clock at night.

## 2) Second inoculation

Instead of removing the black polythene from the plants immediately the inoculations were complete, it was left in place for eighteen hours. After this time, the polythene was removed and the daily seven hours of fogging commenced.

#### Assessments

The plants were monitored throughout the course of the experiment for the development of any phytotoxicity symptoms.

Each plant was assessed for presence or absence of rust infection. Counts were made of rust pustules per plant on the Pelargoniums and Fuchsias. Pustule counts were also made for the Bellis plants in the early stages of the experiment. In the latter stages, when the Bellis in some treatments were severely affected by rust, assessments were made of the percentage leaf area affected.

#### Statistical analysis

Pustule counts and leaf area data were analysed by an ADAS statistician. Data of percentage plants affected are presented without analysis.

#### Crop diary

Plants bought in as plugs and potted up:	4-10 October 2000
Fungicide application:	16 October 2000
First inoculation with rusts:	18 October 2000
First rust symptoms (on all three plant types):	3 November 2000
Rapid (pirimicarb) applied to Bellis for aphid control:	8 November 2000
Second inoculation with rusts:	9 November 2000
First development of symptoms from second inoculation:	20 November 2000
Rovral (iprodione) applied for Botrytis control:	23 November 2000
Disease assessments:	8 November,
	1, 5 December 2000

## Experiment 3: Evaluation of the eradicant activity of the candidate fungicides against rust diseases

The objective of this experiment was to measure the effectiveness of the various fungicides in eradicating an existing rust infection. To do this the plants were inoculated with the rusts and left for a short period, so that the rust spores would germinate and infect them. The plants were then sprayed with the fungicides, and disease development monitored. A second application of fungicides was made later in the experiment, to see whether any of the treaments that had initially given poor disease control could 'clean up' the problem.

A gap of only three days was left between the inoculation of the plants with the rusts and the first application of fungicides, as whilst previous work on the eradicant activity of rust fungicides has given varying results, in some cases (e.g. Spencer 1976, 1979) the eradicant activity was found to be very limited (only one to two days).

The experimental design, plant species, general crop husbandry, fungicides used, fungicide application technique, preparation of rust inoculum, inoculation of plants, assessments and statistical analyses were all identical to those for Experiment 2. The post-inoculation husbandry was as used for the first inoculation in Experiment 2.

The concentrations of the spore suspensions were as follows:

Pelargonium	-	5,000 spores/ml
Fuchsia	-	7,000 spores/ml
Bellis	-	5,000 spores/ml

## Crop diary

Plants bought in as plugs and potted up:	4-10 October 2000
Plants inoculated with rusts:	17 October 2000
First application of fungicides:	20 October 2000
Rapid (pirimicarb) applied to Bellis for aphid control:	7 November 2000
First rust symptoms:	3 November 2000
Second application of fungicides:	28 November 2000
Rovral (iprodione) applied for Botrytis control:	11 December 2000
Assessments:	7, 27 November,
	22 December 2000

## Observation trial: Evaluation of the potential phytotoxicity of Plantvax 75 (oxycarboxin) applied as a compost drench

Plantvax 75 has approval for use as a drench treatment on carnation crops grown in peat bags, for protection against rust. Spencer (1979) found that this application method gave long-lasting protection (complete protection for up to three months, compared to just a couple of weeks for spray applications).

It was felt that there would be merit in assessing the efficacy of this application method for rust control on other bedding plants. However, before this was done, an assessment of the potential phytotoxicity to other bedding plant types from compost drenches of Plantvax 75 would be required. It is known, for example, that root uptake of the fungicide by chrysanthemums can lead to leaf scorch.

A single compost drench of oxycarboxin was given to each of ten well-established potted plants of each of the plant types used in Experiment 1. The rate used corresponded to the recommended rate of 2g Plantvax 75/4l water/peat module (consisting of 42.5 litres of compost) for carnations. The plants were subsequently monitored daily for the development of phytotoxicity symptoms.

#### **Results**

# Experiment 1: Evaluation of the phytotoxicity of candidate fungicides towards a range of bedding plant species

The main symptoms noticed during the pre-spray assessments were the nicotine damage to the Dianthus (described in the materials and methods section), red leaf spots on some of the Fuchsia plants (thought to be physiological in origin), small necrotic spots on the leaves of some Antirrhinum plants (due to the bacterial pathogen *Pseudomonas syringae*) and distortion of some leaves of the Bellis plants due to aphid attack. These, and any other symptoms affecting individual plants, were accurately recorded so that they could not be confused at a later date with phytotoxicity caused by a fungicide treatment.

#### First fungicide application

Both the experimental compound and Dorado left white deposits on the leaves (at both single and double rates) that were still visible some weeks after treatment. It should be noted that the plants were sub-irrigated throughout the experiment (even when hand-watering was required the water was applied directly to the capillary matting) and there was thus no opportunity for any deposits to be washed from the foliage.

As mentioned previously, the first fungicide applications were made during hot and very bright conditions, which would be expected to give a severe test of crop safety.

The first symptoms of phytotoxicity appeared three days after the fungicides were applied, and were all typical of a direct scorch effect (either obvious circular marks typical of droplet scorch, or larger areas of scorching). The effects were seen predominantly on the young leaves. The fungicides which caused damage symptoms were Amistar, Nimrod-T, Plantvax 75 and Twist. Droplet-scorch symptoms were usually produced by Nimrod-T and Plantvax 75, and larger but still localised areas of scorching (often leading to twisting of the leaves) by Amistar and Twist (both of which are strobilurin fungicides). See Appendix 1 for photographs of these symptoms. The plants affected were as follows:

Amistar	-	Antirrhinum (S/D), Dianthus (S/D)
Nimrod-T	-	Antirrhinum (S/D), Dianthus (S/D)
Plantvax 75	-	Antirrhinum (D)
Twist	-	Antirrhinum (D), Chrysanthemum (D), Dianthus (S/D)

S = single rate D = double rate

Six days after the fungicides were applied, a further symptom was noticed on the Fuchsia plants. The symptom again affected the youngest growth, and consisted of the production of smaller, dark green, 'hard', down-curled or distorted leaves (see Appendix 1). It was clear that this symptom was being produced to a greater or lesser

extent by all of the fungicides belonging to the 'triazole' group, namely Alto 240EC, Plover, Systhane 20EW and Tilt 250EC. The effect was most noticeable from Tilt, followed by Alto, Systhane and Plover. Both single and double rates of all of these fungicides produced the symptoms, with the effect generally more pronounced at double rates. The symptoms produced by single rate Plover and Systhane were very mild; in fact it would have been difficult to have seen any effect without being able to make a direct comparison with untreated plants.

The effects of each of the fungicide treatments on the test plants are shown in Table 3.

Table 3: Symptoms produce	d following first	t fungicide application
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Fungicide	Symptoms	Plant Types	
Alto 240EC	None	A,B,Ch,Ci,Di,P	
	Young leaves small, dark, distorted	F (S/ <b>D</b> )	
Amistar	Localised 'bleached' necrosis leading to twisting	A (S/ <b>D</b> ), Di (S/ <b>D</b> )	
	None	B,Ch,Ci,F,P	
Dorado	None	All types	
Nimrod-T	Droplet scorch - sunken areas with 'bleached' margins	A (S/ <b>D</b> )	
	Localised 'bleached' necrosis leading to twisting	Di (S/D)	
	None	B,Ch,Ci,F,P	
Plantvax 75	Droplet scorch - sunken areas with 'bleached' margins	A (D)	
	None	B,Ch,Ci,Di,F,P	
Plover	Young leaves small, dark, distorted	F (S/ <b>D</b> )	
	None	A,B,Ch,Ci,Di,P	
Systhane 20EW	Young leaves small, dark, distorted	F (S/ <b>D</b> )	
	None	A,B,Ch,Ci,Di,P	
Tilt 250EC	Young leaves small, dark, distorted	F (S/ <b>D</b> )	
	None	A,B,Ch,Ci,Di,P	
Twist	Localised 'bleached' necrosis leading to twisting	A (D), Di (S/ <b>D</b> )	
	Localised brown necrosis	Ch (D)	
	None	B,Ci,F,P	
Experimental	None	All types	

Key to plant types: A = Antirrhinum B = Bellis Ch = Chrysanthemum Ci = Cineraria Di = Dianthus F = FuchsiaP = Pelargonium

Application rates in brackets (where more severe symptoms were produced by a particular rate, this is shown in bold):

S = single label/SOLA rate D = double label/SOLA rate

#### Second fungicide application

This was made under cooler, cloudier conditions than the first application. The scorch symptoms resulting from the first applications of Amistar, Nimrod-T, Plantvax 75 and Twist were not reproduced by the second applications.

The symptoms produced by the triazole fungicides on the Fuchsias continued after the second applications. The severity of the symptoms on the fuchsias was scored on a 0-5 scale, with 0 representing unaffected plants and 5 representing the most severely affected (generally in the double rate Tilt treatments). The scores for each plot were added together and averaged to give an index. The results are shown in Table 4:

Table 4: Assessment of the severity of phytotoxicity symptoms produced by the
triazole fungicides on Fuchsia.

Fungicide	Phytotoxicity Index		
	Single Rate	<b>Double Rate</b>	
Alto 240EC	2.25	3.00	
Plover	0.25	1.00	
Systhane 20EW	1.00	1.50	
Tilt 250EC	4.00	4.75	

Two fungicides, Plantvax 75 and Alto 240EC, produced symptoms from the second application that were not seen from the first. Plantvax 75 produced necrosis at or towards the leaf margins of the Bellis, Chrysanthemum and Cineraria. These symptoms are likely to have been the result of uptake of the fungicide through the roots of the plants (see the results of the observation trial with Plantvax drenches). Sprays were applied to the point of run-off, and it is possible that some of the fungicide dripped down onto the compost. Alternatively, the plants may have taken up fungicide from the capillary matting. It is not clear whether the symptoms were a direct result of the second application, or the result of an accumulation of active ingredient from both the first and second applications.

The second application of Alto resulted in leaf necrosis symptoms in Antirrhinum, Bellis and Chrysanthemum, and pink discolouration of the leaf margins in Pelargonium (as well as the symptoms already described in Fuchsia). The symptoms were usually more severe at double rate. In the case of the Antirrhinums the necrosis was sometimes so severe (particularly at double rate) as to cause loss of the growing point and subsequent production of side-shoots (see Appendix 1). It is interesting that these symptoms should have been produced only following the second application, when the conditions at the time of the first spray application should have provided a sterner test of crop safety. It is known that cyproconazole is one of the most rapidly translocated of the triazole fungicides, accumulating in the leaf tips. It is therefore possible that this rapid translocation, coupled with the accumulation of two doses of active ingredient within a short timescale (seven days) was enough to cause symptom production.

The effects of each of the fungicide treatments on the test plants are shown in Table 5.

#### Flowering assessment

There was no indication that any of the fungicides had delayed flowering or increased/decreased flower numbers (obviously, detailed counts of flower numbers per plant were not made). The Cineraria and Pelargonium plants had not flowered at all (including in the untreated plots) at the termination of the experiment, and only limited numbers of flowers had been produced by the Bellis.

Effects of some of the fungicides on the shape and size of the Fuschia flowers were observed. Tilt, and to a lesser extent Alto, caused a reduction in flower size with thickening of the petals and a 'gaping' corolla tube (it will be recalled that these fungicides also caused the most severe leaf symptoms of the triazoles).

Whilst producing no symptoms at all on vegetative growth, the applications of Twist led to fuchsia flowers with elongated, thinner outer petals and an elongated corolla tube. The reasons for this effect are not known.

Table 5: Symptoms	produced	following s	second fur	igicide ar	plication
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Fungicide	Symptoms	Plant Types	
Alto 240EC	Young leaves small, dark, distorted	F (S/ <b>D</b> )	
	Marginal chlorosis/light brown necrosis,	A (S/ <b>D</b> )	
	loss of growing point		
	Light brown necrotic leaf spotting	B (S/ <b>D</b> )	
	Dark brown necrosis on margins &	Ch (S/ <b>D</b> )	
	interveinal areas		
	Pink discolouration of margins	P (S/D)	
	None	Ci, Di	
Amistar	None	All types	
Dorado	None	All types	
Nimrod-T	None	All types	
Plantvax 75	'Bleached' necrosis of leaf margins	B (S/D)	
	Blotchy dark brown necrosis towards	Ch (S/ <b>D</b> )	
	margins, all leaf ages affected		
	Light brown necrotic spots towards	Ci (S/D)	
	margins of older leaves		
	None	A,Di,F,P	
Plover	Young leaves small, dark, distorted	F (S/ <b>D</b> )	
	None	A,B,Ch,Ci,Di,P	
Systhane 20EW	Young leaves small, dark, distorted	F (S/ <b>D</b> )	
	None	A,B,Ch,Ci,Di,P	
Tilt 250EC	Young leaves small, dark, distorted	F (S/ <b>D</b> )	
	Loss of apical dominance	A (D)	
	None	B,Ch,Ci,Di,P	
Experimental	None	All types	

Key to plant types: A = Antirrhinum B = Bellis Ch = Chrysanthemum Ci = Cineraria Di = Dianthus F = FuchsiaP = Pelargonium

Application rates in brackets (where more severe symptoms were produced by a particular rate, this is shown in bold):

S = single label/SOLA rate D = double label/SOLA rate

Dry weight assessments (see Appendix 2 for relevant results tables)

The dry weight data was analysed for any significant differences between the individual fungicides, between the single and double rates, and for any interactions between fungicides and rates.

The only significant differences recorded were between the individual fungicides on Antirrhinum and Dianthus.

Antirrhinum plants treated with Alto 240EC, Dorado and Twist weighed significantly less than the untreated controls. It was also noticeable that with some of the fungicides (e.g Alto 240EC, Twist and Tilt 250EC) the plants treated with double-rate product weighed quite a lot less than those receiving single rates, although the overall analysis for single versus double rates (taking all fungicides into account) was not significant.

Dianthus plants treated with Tilt 250EC weighed significantly less than the untreated controls.

#### Height assessments (see Appendix 2 for relevant results tables)

As with the dry weight assessments, few significant differences were recorded.

Fuchsia plants treated with Alto 240EC and Tilt 250EC were significantly shorter than the untreated controls (see also the photograph of Tilt-treated plants in Appendix 1). Overall, plants treated with double-rate fungicides were significantly shorter than the controls, although the effect varied with the individual product (in a couple of cases the double-rate treated plants were tallest).

In the case of Antirrhinum, there was a significant interaction between rate and product, so that it is not possible to make detailed, separate comparisons between either the individual fungicides or the single versus double rates. However, it can be seen that plants treated with double-rate Alto 240EC were much shorter than the untreated control plants. This is because, as discussed earlier, the growing points of

the plants were often killed, resulting in loss of apical dominance and the production of side-shoots.

# **Experiment 2: Evaluation of the protectant activity of the candidate fungicides against rust diseases**

Rust was first seen on the plants (particularly on the Bellis) sixteen days after the plants were first inoculated. An assessment made twenty-one days after the first inoculation (and just prior to the second inoculation) revealed the following amounts of disease. The data obtained were not suitable for Analysis of Variance (Anova), but were analysed using another statistical test known as a Friedman Test.

Treatment	Mean no. of rust pustules - 8 November 20			
-	Bellis*	Fuchsia#	Pelargonium#	
Control	4.12	0.38	4.06	
Amistar	0.00	0.00	0.00	
Dorado	4.08	0.42	0.63	
Nimrod-T	0.04	0.00	0.83	
Plantvax 75	0.63	0.00	0.54	
Plover	0.00	0.00	0.00	
Systhane 20EW	0.00	0.00	0.63	
Tilt 250EC	0.17	0.00	0.58	
Twist	0.00	0.00	0.00	
Experimental	3.54	0.42	1.25	
P=	< 0.001	0.002	0.007	

 Table 6: Protectant activity of fungicides against rust diseases (21 days after first inoculation).

\* = mean number of pustules on the youngest, fully expanded leaf.

# = mean number of pustules per plant.

These results, and the percentage of plants affected for each plant species are also presented as histograms in Appendix 3.

Bellis

Disease levels on the plants treated with Dorado or the experimental compound were similar to those on the untreated plants. The other treatments all reduced disease levels, with no rust at all visible on the plants treated with Amistar, Plover, Systhane 20EW or Twist.

#### **Fuchsia**

Again, the levels of rust on plants treated with Dorado or the experimental compound were similar to those on the untreated plants. At this stage no rust at all was seen on the other treatments.

#### **Pelargonium**

All of the fungicide treatments reduced disease levels. There was no rust at all on the plants treated with Amistar, Plover or Twist.

The second disease assessment was made seventeen days after the second rust inoculation. The technique used for this inoculation (leaving the plants covered with polythene for eighteen hours, before commencing the fogging programme) led to increased development of disease compared to the first inoculation. It is possible that commencing an eighteen hour period of fogging immediately after applying the rust spores, as occured with the first inoculation, resulted in some spores being washed from the leaves. However, the higher spore concentrations in the suspensions used for the second inoculations are also likely to have played a role in the increased disease levels.

The disease levels recorded in the second assessment are shown in Table 7. The data produced in this assessment were suitable for Analysis of Variance (in the case of the Bellis and Fuchsia, after transformation):

## Table 7: Protectant activity of fungicides against rust diseases (17 days after second inoculation).

Treatment	Mean Disease Severity - 5 December 2000			
	Bellis*	Fuchsia#	Pelargonium#	
Control	15.5 (1.12)	20.2 (1.21)	23.2	
Amistar	1.2 (0.20)	0.4 (0.11)	0.6	
Dorado	14.2 (1.09)	17.1 (1.22)	17.3	
Nimrod-T	3.2 (0.61)	13.1 (1.02)	22.4	
Plantvax 75	5.5 (0.80)	8.0 (0.93)	14.6	
Plover	0.3 (0.09)	0.1 (0.03)	11.1	
Systhane 20EW	0.3 (0.10)	0.2 (0.08)	29.3	
Tilt 250EC	3.9 (0.63)	0.1 (0.03)	27.3	
Twist	1.6 (0.40)	6.3 (0.82)	2.3	
Experimental	12.9 (1.14)	10.4 (1.04)	17.8	
P=	<.001	<.001	<.001	
SED	0.194	0.148	4.38	

\* = mean percentage leaf area affected on the second and third youngest fullyexpanded leaves.

# = mean number of pustules per plant.

These results, and the percentage of plants affected for each plant species are also presented as histograms in Appendix 3.

#### Bellis

All of the fungicide treatments except Dorado and the experimental compound significantly reduced disease levels compared with the untreated control. Levels of rust were significantly lower in plants treated with Amistar, Plover or Systhane 20EW compared with the other fungicide treatments.

It was noted (both in this experiment and in Experiment 3) that the Bellis rust was more severe on the plants situated in plots in the middle of the glasshouse bay. These plants were furthest away from the fogging nozzles, which were situated at the sides of the bay. The foliage of the plants nearer to the nozzles tended to be wetter than those in the middle, whilst the latter dried out more quickly on completion of a fogging cycle. It would therefore seem that *Puccinia distincta* differs somewhat from Fuchsia and Pelargonium rusts in requiring less in the way of leaf wetness to produce damaging levels of disease (although at least some leaf wetness is probably still necessary in order for spore germination and infection to occur). This effect was also noticed whilst maintaining the 'rust stock plants' from which spores were harvested for use in the experiments.

Most of the untreated Bellis plants went on to develop such high levels of disease that they eventually died. In many cases this seemed to be due to an attack on the severely weakened plants by other pests or diseases. It was noted that the roots and stem bases of many of the plants were invaded by sciarid fly larvae.

#### Fuchsia

Amistar, Plover, Systhane 20EW, Tilt 250EC and Twist all significantly reduced disease levels compared to the untreated control. Notable reductions were also obtained from Plantvax 75 and Nimrod-T, although these were not statistically significant.

#### Pelargonium

Only Amistar, Twist and Plover significantly reduced disease levels compared to the untreated control. Amistar also gave a significant reduction compared to all of the other fungicides except Twist. Plantvax 75 reduced disease levels by 40-50%, but this reduction was not statistically significant.

#### Phytotoxicity symptoms noted during Experiment 2

The triazole fungicides Tilt 250EC, Systhane 20EW and Plover produced symptoms on the Fuchsia plants that were identical to those produced in the phytotoxicity experiment (Experiment 1). As in that trial, symptoms were most severe in the plants receiving Tilt 250EC. The symptoms in the plants treated with Systhane 20EW and Plover were once again much milder, and in many cases might not have been noticed at all had there not been untreated plants to compare them with.

The Bellis plants treated with Plantvax 75 again showed some evidence of necrotic spotting at or towards the leaf margin.

The changes in flower habit produced by Twist on the Fuchsia variety (Beacon Rose) used in Experiment 1 did not develop in the variety (Sir Matt Busby) used in Experiments 2 & 3. This effect would therefore appear to be variety-specific, and emphasises the need for caution in the use of pesticides on new or previously untreated varieties, even if they have been shown to be safe to different varieties of the same plant type. In contrast, the effects of the triazole fungicides on Fuchsia and of Plantvax 75 on Bellis were seen on the varieties used in all of the experiments.

## **Experiment 3:** Evaluation of the eradicant activity of the candidate fungicides against rust diseases

Because the plants in this experiment were inoculated with the rusts just once, levels of disease were generally lower than in Experiment 2 (although some secondary spread of disease occurred due to the fogging and occasional overhead watering).

A disease assessment made on 27 November (prior to the second application of fungicides) gave the following results (Table 8). The data obtained were not suitable for Analysis of Variance (Anova), but were analysed using Friedman's Analysis.

Treatment	Mean disease severity - 27 November 2000		
-	Bellis*	Fuchsia#	Pelargonium#
Control	13.20	1.10	6.02
Amistar	0.04	0.21	0.00
Dorado	8.38	0.42	6.54
Nimrod-T	0.23	0.21	3.79
Plantvax 75	2.15	0.08	2.08
Plover	0.08	0.00	0.21
Systhane 20EW	0.02	0.00	0.54
Tilt 250EC	0.21	0.00	1.00
Twist	0.10	0.46	3.04
Experimental	3.71	0.25	2.08
P=	0.002	0.008	0.001

 Table 8: Eradicant activity of fungicides against rust diseases (41 days after inoculation).

\* = mean percentage leaf area affected on the fourth and fifth youngest fullyexpanded leaves.

# = mean number of pustules per plant.

These results, and the percentage of plants affected for each plant species are also presented as histograms in Appendix 3.

#### Bellis

All of the fungicide treatments reduced disease levels to below one per cent leaf area affected (compared to over thirteen per cent in the untreated plants), with the exception of Dorado, Plantvax 75 and the experimental compound.

#### Fuchsia

All of the fungicide treatments reduced the levels of disease by fifty per cent or more. No rust at all was seen on the plants treated with Plover, Systhane 20EW and Tilt 250EC.

#### Pelargonium

There was no reduction in disease from the Dorado treatment. Amistar, Plover and Systhane 20EW all reduced disease levels to below one pustule per plant (compared to over six for the untreated). No disease at all was seen on the Amistar-treated plants.

The assessment on 22 December was made twenty-five days after the second application of fungicides. Once again, data were analysed using Friedman's Analysis.

# Table 9: Eradicant activity of fungicides against rust diseases (25 days aftersecond fungicide application).

Treatment	Mean disease severity - 22 December 2000		
	Bellis*	Fuchsia#	Pelargonium#
Control	44.16	2.08	8.44
Amistar	0.15	0.13	0.00
Dorado	31.40	0.25	5.46
Nimrod-T	0.73	0.04	2.08
Plantvax 75	5.17	0.08	1.67
Plover	0.10	0.00	0.00
Systhane 20EW	0.02	0.00	0.71
Tilt 250EC	1.08	0.00	0.96
Twist	1.44	0.33	2.50
Experimental	22.00	0.25	4.17
P=	<0.001	0.01	< 0.001

\* = mean percentage leaf area affected on second and third youngest fully-expanded leaves.

# = mean number of pustules per plant.

These results, and the percentage of plants affected for each plant species are also presented as histograms in Appendix 3.

#### <u>Bellis</u>

Levels of disease on the untreated plants had increased nearly three-fold since the 27 November assessment. The plants treated with Dorado and the experimental compound were also severely affected, although the experimental compound had reduced the disease levels by approximately fifty per cent. Amistar, Dorado, Nimrod-T, Plover, Systhane 20EW, Tilt 250EC and Twist had all reduced the amount of rust to less than two per cent leaf area affected, compared to more than forty-four per cent on the untreated plants.

Once again, most of the untreated plants died before the experiment was terminated.

#### <u>Fuchsia</u>

Disease levels remained low, with the untreated plants affected by an average of just over two pustules per plant. However, whilst the levels of rust on the untreated plants had increased, those on the fungicide-treated plants had remained constant or declined. This is likely to be due to the protectant activity of the second application of fungicides, preventing secondary spread of the disease from the initial lesions. The disease was assessed as the number of pustules per plant, and the combination of the protection of new growth with the senescence and loss of some of the older leaves (on which the original pustules had been produced) lead to the overall decline in disease levels. This is in contrast to the first assessment, where any reduction in disease levels compared to the untreated plants would have been a true eradicant effect.

#### Pelargonium

Similar results were obtained to the Fuchsias. Whilst the levels of disease on the untreated plants increased slightly compared to the 27 November assessment, those on the majority of the fungicide-treated plants remained constant or declined. Once again, the combination of protection of new growth with the senescence and loss of older, disease-affected leaves is the likely cause of this phenomenon.

#### Phytotoxicity symptoms noted during Experiment 3

Identical symptoms were produced to those noted in Experiment 2, namely the effect of the triazole fungicides on Fuchsia and of Plantvax 75 on Bellis. This is not surprising given that the same plant varieties were used for Experiments 2 and 3.

In the five weeks between the two fungicide applications, the Fuchsia plants gradually reverted to producing symptomless new leaves, even in the case of the plants receiving Tilt 250EC. However, affected leaves were produced again immediately following the second application.

## **Observation trial: Evaluation of the potential phytotoxicity of Plantvax 75** (oxycarboxin) applied as a compost drench.

Phytotoxicity symptoms were first seen thirteen days after the application of the drenches, in the form of marginal necrosis on leaves of the Bellis and Chrysanthemum. Symptoms also developed on most of the other plant types over the following days. The various symptom types are summarised below:

Antirrhinum:	'Bleached' necrosis at leaf tips. Older and middle leaves		
	affected.		
Bellis:	'Bleached' marginal necrosis, plus 'bleached' necrotic spots (2-		
	3mm) near to margin. Leaves of all ages affected. See Appendix		
	1.		
Chrysanthemum:	Marginal necrosis, initially dark brown, progressing to light		
	brown/'bleached'. Leaves of all ages affected.		
Cineraria:	Light brown necrotic spotting on or towards margins. Older		
	leaves only.		
Dianthus:	Blotchy greyish discolouration at leaf tips. Leaves of all ages		
	affected.		
Fuchsia:	No symptoms.		
Pelargonium:	Narrow marginal chlorosis (bright yellow), progressing to		
	necrosis. Older leaves only. See Appendix 1.		

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#### DISCUSSION

The experiments described in this report have identified a number of fungicides which gave good control of the rust diseases studied, whilst causing few or no problems in terms of crop safety. Some products were effective against all the rusts used in the experiments, whilst the efficacy of others varied according to the rust against which they were targeted.

The crop safety experiment (Experiment 1) showed that, encouragingly, most of the fungicides evaluated were safe to most of the plant subjects to which they were applied (in some cases even at double the recommended rate). An exception was Alto 240EC, the second application of which caused extensive damage to range of subjects. This fungicide was selected because it gives effective rust control on a range of arable crops, but it cannot be used legally on protected bedding plants. The experiments showed that there are other, related fungicides (e.g. Plover, Systhane 20EW) which are safer to the range of plant subjects used.

The first application of fungicides was made under hot, sunny conditions, and this resulted in scorch from Amistar, Twist, Nimrod-T and Plantvax 75 on one or two subjects. However, no damage was recorded from the second application of Amistar, Twist or Nimrod-T fungicides, made under cooler, cloudy conditions. This emphasises the need for caution when assessing environmental conditions prior to applying pesticides.

Phytotoxicity symptoms were produced on the Fuchsia plants from both applications of all of the fungicides from the 'triazole' group (Tilt 250EC, Alto 240EC, Plover and Systhane 20EW). Rather than a direct scorch effect, this took the form of irregularities in the leaves produced following fungicide application. Affected leaves were reduced in size, distorted, thickened and darker green. The severity of these symptoms varied according to the fungicide, being particularly prominent in plants treated with Tilt 250EC.

The same symptoms were produced on a different Fuchsia variety used in the disease control experiments, although the plants did slowly recover to produce normal leaves in the weeks following fungicide application. Nonetheless, it is recommended that Tilt 250EC should not be used on Fuchsias.

The case for using Plover and Systhane 20EW in Fuchsias in less clear-cut. Both of these products, when applied at single rate, produced mild phytotoxicity symptoms that may not have been noticed without untreated plants to compare them to. The disease control experiments (discussed below) revealed that they are very effective in controlling fuchsia rust. Given that the plants should also 'grow out' of any symptoms produced by these fungicides, they should not be dismissed when evaluating control strategies for Fuchsia rust, particularly when the plants are under high disease pressure, or there is rust already in the crop. However, it would be prudent to evaluate them on a small number of plants of any variety to which they have not previously been applied, in case a particular variety produces a more severe reaction. In fact, this advice would apply to any pesticide being used on a plant type or variety for the first time.

Compost drenches of Plantvax 75 have been reported to give good control of rust on carnations (Spencer, 1979). However, in the observation trial it was found that applying the fungicide in this way resulted in symptoms of phytotoxicity in most of the subjects to which it was applied (including a bedding Dianthus variety). Care should also be taken when using this fungicide as a foliar spray (particularly on subjects such as Bellis and Chrysanthemum) to ensure that large quantities of the spray do not run off the leaves and into the growing medium.

A number of fungicides performed consistently well in the disease control trials. Amistar and Twist, both fungicides from the strobilurin group, were effective as protectants against all three of the rusts used in the experiments. Amistar usually performed slightly better than Twist, and can be used at the grower's risk on protected bedding plants, whereas Twist only has approval for cereals, and can therefore be used only on outdoor bedding (again at the grower's risk). These fungicides were somewhat less effective when used as eradicants. These results support those obtained from work on a range of diseases on other crops, which has shown that whilst strobilurins have prolonged, effective protectant activity, their effectiveness as eradicants is strictly limited. Interestingly, Amistar in particular did show eradicant activity against Bellis and Pelargonium rusts which was comparable to the triazole fungicides such as Plover and Systhane 20EW. However, it should be remembered that the fungicides were applied only three days after the plants were inoculated with the rusts. It is possible that if this period were increased (allowing the rust to colonise the leaf more extensively) the eradicant activity of the strobilurin fungicides could decline rapidly. For this reason, the strobilurin products are best used as part of a protectant programme, rather than after rust has become established in a crop.

The triazole fungicides Plover, Systhane 20EW and Tilt 250EC all performed well against Bellis and Fuchsia rust, both as protectants and eradicants. The results for Systhane against Bellis rust support those obtained by Weber and Tilston (1999), who used an amateur formulation of the fungicide. Tilt 250EC performed no better than the other two triazole products, and as this fungicide is more likely to cause phytotoxicity problems (see above) it would be preferable to use either Plover or Systhane.

Systhane 20EW and Tilt performed very poorly against Pelargonium rust in the protectant experiment, giving no reduction in disease levels over those on the untreated plants. This is in direct contrast to work reported from the USA, where Raabe (1991) and Chase (1999) both obtained excellent control of the disease using one or both of these fungicides. The fact that the products gave no control at all raises the possibility of fungicide resistance. Resistance of Chrysanthemum White Rust to both of these products has recently been discovered in the UK (Cook, pers comm.). However, there are a number of factors about the results in these experiments which do not fit in with a 'classic' case of fungicide resistance.

Firstly, the closely related fungicide Plover gave reasonable control of the disease in the protectant experiment (although not as good as with the other rusts). Often, a fungus resistant to a fungicide from a particular chemical group (in this case the triazoles) also shows resistance to all other products in that group (cross-resistance) - this should include Plover. Secondly, all of the triazoles gave good reductions in the levels of Pelargonium rust in the eradicant experiment (Experiment 2). Further work will therefore be required to try and ascertain whether this is a true case of fungicide resistance.

Plantvax 75 and Nimrod-T, whilst in general being less effective than the strobilurin (particularly in the protectant experiments) or triazole fungicides, often gave useful reductions in disease levels, and should certainly not be dismissed when devising fungicide control programmes. Plantvax 75 in particular, which belongs to a different chemical group (the carboxamides), would be useful as part of an anti-resistance strategy.

Dorado performed poorly in most cases. This fungicide has carried a label recommendation for the control of Fuchsia rust, but its use on ornamentals (excluding roses) has recently been revoked although there is a two-year period during which existing stocks can be used up.

The next stage of Project PC 175 will evaluate fungicide programmes for crop safety and disease control ability. The programmes will include the better-performing products from this first series of experiments. They will also include some of the older, broad-spectrum protectants such as chlorothalonil, mancozeb, etc. in a bid to increase the diversity of fungicides and so enhance the anti-resistance strategy. The programmes will be compared with the most effective single-product treatments from these first trials. The opportunity will also be taken to evaluate any new fungicides or novel treatments.

#### CONCLUSIONS

- Techniques have been successfully developed for the large-scale inoculation of plants with rusts.
- A range of fungicides have been assessed for crop safety (on seven different bedding plant types) and their ability to control disease (using three different rust species).
- The crop safety experiment showed that most of the fungicides were safe to most of the plant types used. However, some products (Amistar, Twist, Plantvax 75 and Nimrod-T) caused scorch on some subjects when applied under hot, sunny conditions.
- Alto 240EC caused severe damage to a range of plant types, which appeared to be due to the cumulative effect of two applications at a short interval.
- Fungicides from the triazole group caused phytotoxicity symptoms on the new growth of Fuchsias. The amount of damage varied according to the fungicide and the rate.
- Plantvax 75, when applied as a compost drench, caused damage to most of the plant subjects.
- Excellent protectant activity was obtained from the strobilurin fungicides Amistar and Twist against the three rust fungi used in the experiments. Some eradicant activity was also seen, particularly with Amistar, when these fungicides were applied shortly after plants had been inoculated with the rusts.
- The triazole fungicides Plover and Systhane 20EW showed excellent protectant and eradicant activity against Bellis and Fuchsia rusts. Protectant activity of Systhane 20EW in particular against Pelargonium rust was very poor. Further

work needs to be done to assess whether this is due to fungicide resistance in the population of Pelargonium rust used in the experiments.

• The fungicides that have performed well in this series of experiments will now be evaluated in programmes including broad-spectrum protectant materials. It is hoped to develop programmes giving excellent disease control, whilst minimising the chance of the rusts developing fungicide resistance.

## **TECHNOLOGY TRANSFER**

HDC Grower Factsheet 23/00, summarising the results of the literature review, was published in September 2000.

The project was discussed at an HDC Seminar on Bedding Plants R&D at HRI Wellesbourne on 26 October 2000.

Results of the project to date were presented at a meeting of the Midlands Bedding and Pot Plant Group on 30 January 2001.

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## ACKNOWLEDGEMENTS

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The Pelargonium plants used in Experiments 2 and 3 were kindly donated by Young Plants Ltd. Thanks are also due to the various growers and consultants who supplied rust-affected plant material for use in the project.

## **Appendix 1: Phytotoxicity Symptoms**

1. Scorch on leaf tips of Dianthus, caused by nicotine smoke applied for aphid control.

2. Stunting of Fuchsia caused by single-rate Tilt 250EC. Untreated on left, Tilt-treated on right.

3. Damage to young leaves of Fuchsia from single-rate Tilt 250EC. Leaves are dark green, reduced in size, distorted and thickened.

4. Effect on single-rate Systhane 20EW on Fuchsia. Slight distortion of young leaves and reduction in leaf size.

5. Effect of second application of double-rate Alto 240EC on Antirrhinum. Severe leaf scorch, death of growing point and production of side-shoots.

6. Effect of first application of single-rate Plantvax 75 on Antirrhinum. Centre leaf in photograph shows droplet scorch - slightly sunken areas with bleached necrotic margins.

7. Effect of first application of double-rate Twist on Antirrhinum. Bleached necrotic areas of scorch leading in some cases to leaf twisting.

8. Effect of double-rate Twist on flowers of Fuchsia. Narrower, elongated outer petals plus longer corolla tube.

9. Effect on Bellis of Plantvax 75 applied as a compost drench. Necrotic areas on and around leaf margins.

10. Effect on Pelargonium of Plantvax 75 applied as a compost drench. Chlorosis at leaf margins (later progressing to necrosis).

## Appendix 2: Experiment 1 - Assessments of Dry Weight (grams).

## 1. Antirrhinum

Fungicide	Rate		Fungicide Mean
	Single	Double	
Control	-	-	12.78
Alto 240EC	10.55	9.18	9.86
Amistar	12.45	11.93	12.19
Dorado	11.10	10.80	10.95
Nimrod-T	12.00	12.43	12.21
Plantvax 75	12.68	11.78	12.23
Plover	12.35	12.28	12.31
Systhane 20EW	12.23	11.98	12.10
Tilt 250EC	12.73	10.93	11.83
Twist	11.08	9.98	10.53
Experimental	11.18	11.55	11.36
Rate Mean	11.83	11.28	-
Fungicides: $P = 0.00$	5  SED = 0.699		
Rates: NS			
Fungicides x Rates:	NS		

NS = not significant (P<0.05)

## 2. Dianthus

Fungicide	Rate		Fungicide Mean
	Single	Double	
Control	-	-	8.19
Alto 240EC	7.55	8.25	7.90
Amistar	8.63	9.05	8.84
Dorado	7.90	6.57	7.24
Nimrod-T	9.42	8.50	8.96
Plantvax 75	9.00	9.70	9.35
Plover	8.72	6.80	7.76
Systhane 20EW	8.60	7.10	7.85
Tilt 250EC	6.87	6.50	6.69
Twist	7.98	8.63	8.30
Experimental	7.78	9.30	8.54
Rate Mean	8.24	8.04	-
Fungicides: $P = 0.00$	1  SED = 0.608		
Rates: NS			
Fungicides x Rates:	NS		
NS = not significant	$(D_{1}, 0, 0.5)$		

NS = not significant (P<0.05)

## Assessment of Height (millimetres)

## 1. Antirrhinum

Fungicide	Rate		Fungicide Mean
	Single	Double	
Control	-	-	401.6
Alto 240EC	343.6	230.6	287.1
Amistar	389.3	385.0	387.1
Dorado	347.1	334.3	340.7
Nimrod-T	368.4	337.8	353.1
Plantvax 75	391.4	381.5	386.5
Plover	373.8	392.1	382.9
Systhane 20EW	363.4	368.0	365.7
Tilt 250EC	408.9	335.3	372.1
Twist	357.7	308.4	333.0
Experimental	345.4	385.6	365.5
Rate Mean	368.9	345.9	-
Fungicides: $P = \langle 0.0 \rangle$	3001  SED = 21.41		
Rates: $P = 0.019$ S	SED = 16.58		
Fungicides x Rates: $P = 0.030$ SED = 26.22			

## 2. Fuchsia

Fungicide	Rate		Fungicide Mean
	Single	Double	
Control	-	-	227.6
Alto 240EC	181.5	164.0	172.7
Amistar	234.2	238.8	236.5
Dorado	215.8	196.0	205.9
Nimrod-T	232.1	198.3	215.2
Plantvax 75	239.8	242.3	241.1
Plover	224.9	193.3	209.1
Systhane 20EW	226.4	220.5	223.4
Tilt 250EC	167.6	156.6	162.1
Twist	239.9	210.2	225.0
Experimental	223.1	238.7	230.9
Rate Mean	218.5	205.9	-
Fungicides: $P = <0.001$ SED = 12.78			
Rates: $P = 0.030$ SED = 9.90			
Fungicides x Rates: NS			

NS = not significant (P<0.05)

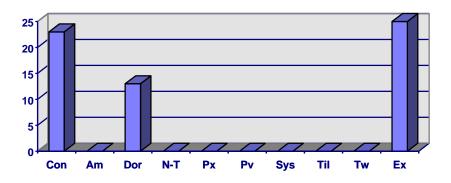
## Appendix 3.

**Experiment 2: Percentage plants affected on 8 November 2000.** 

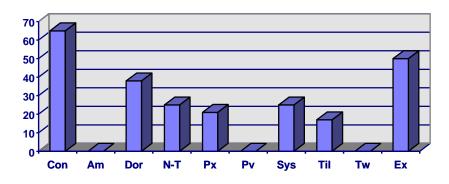
80 70 60 50 40 30 20 10 0 Con Am Dor N-T Px P٧ Sys Til Tw Ex

<u>Bellis</u>

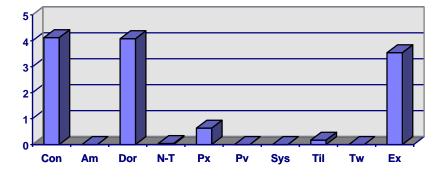
## Fuchsia



## <u>Pelargonium</u>

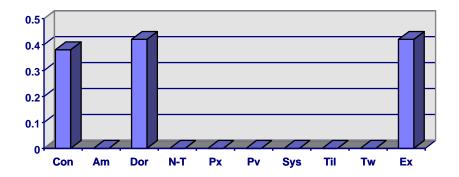


## Experiment 2: Assessments of the Number of Rust Pustules on 8 November 2000

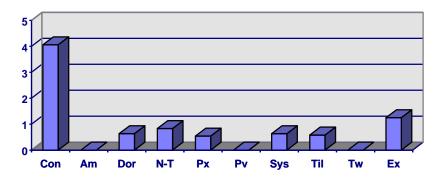


<u>Bellis</u>

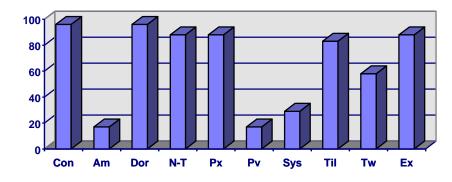
## Fuchsia



Pelargonium

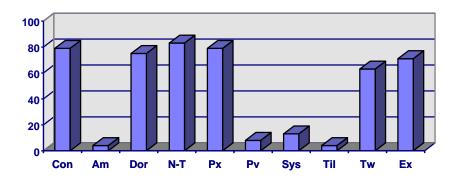


## **Experiment 2: Percentage Plants Affected on 5 December 2000**

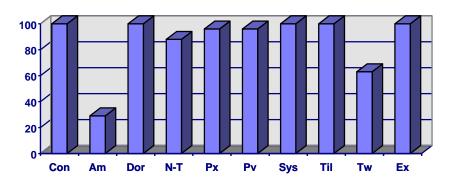


<u>Bellis</u>

#### **Fuchsia**

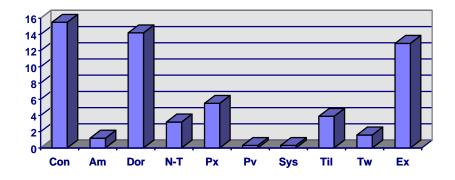


Pelargonium



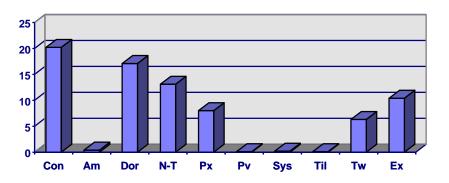
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### **Experiment 2: Disease Assessment on 5 December 2000**

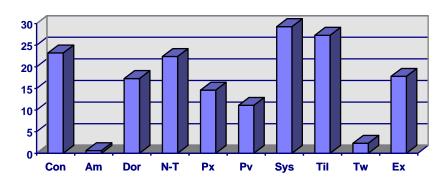


Bellis (mean % leaf area affected on second and third fully-expanded leaves).

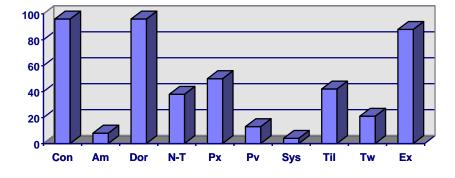
Fuchsia (mean number of pustules per plant).



Pelargonium (mean number of pustules per plant).

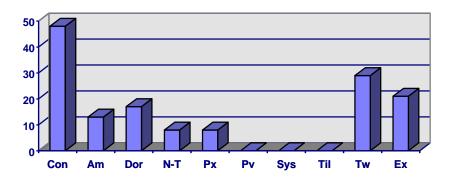


#### **Experiment 3: Percentage Plants Affected on 27 November 2000**

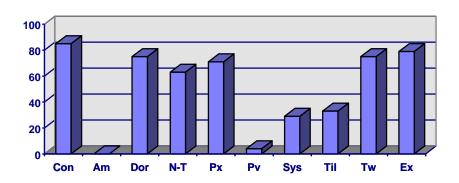


<u>Bellis</u>

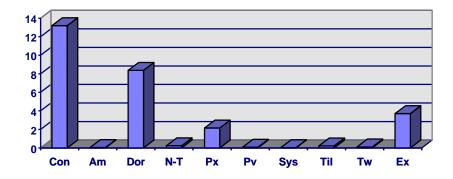
## **Fuchsia**



## Pelargonium

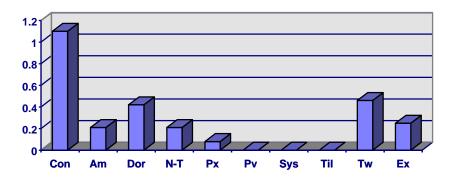


## **Experiment 3: Disease Assessment on 27 November 2000**

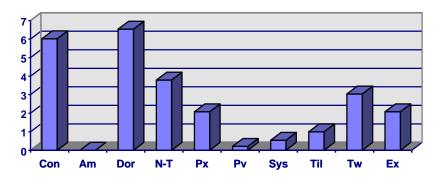


Bellis (mean % leaf area on the fourth and fifth fully-expanded leaves)

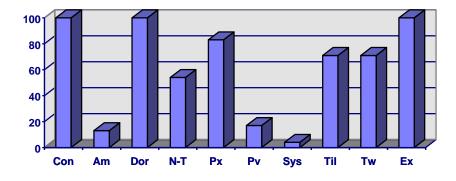
Fuchsia (mean number of pustules per plant).



Pelargonium (mean number of pustules per plant).

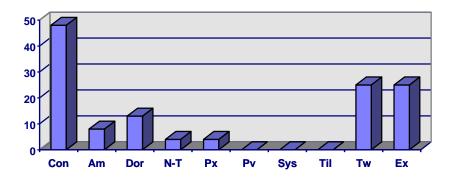


#### **Experiment 3: Percentage Plants Affected on 22 December 2000**

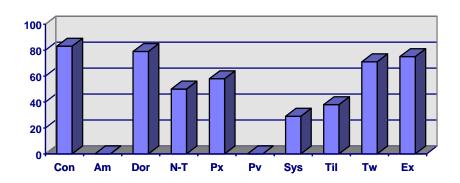


<u>Bellis</u>

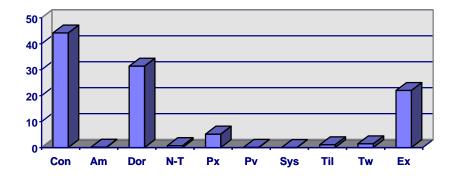
## **Fuchsia**



Pelargonium

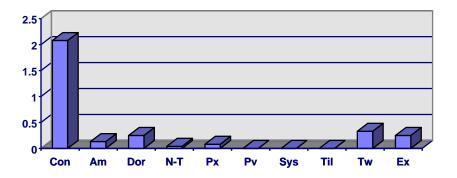


## **Experiment 3: Disease Assessment on 22 December 2000**



Bellis (mean % leaf area on the second and third fully-expanded leaves).

Fuchsia (mean number of pustules per plant).



Pelargonium (mean number of pustules per plant).

