

Grower Summary

CP 124

Managing ornamental plants
sustainably (MOPS):

Evaluation of disinfectants
against *Fusarium* sp. ex stocks
and *Pythium* sp.

Annual 2014

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The results and conclusions in this report may be based on an investigation conducted over one year. Therefore, care must be taken with the interpretation of the results.

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Read the label before use: use pesticides safely.

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AHDB Horticulture,
AHDB
Stoneleigh Park
Kenilworth
Warwickshire
CV8 2TL

Tel – 0247 669 2051

AHDB Horticulture is a Division of the Agriculture and Horticulture Development Board.

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Work package title: Evaluation of disinfectants against *Fusarium* sp. ex stocks and *Pythium* sp.

Work package leader: Dr Erika Wedgwood

Contractor: ADAS

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GROWER SUMMARY

Headline

Three out of nine disinfectant products tested, Disolite, Unifect G and Domestos Extended Germ Kill, gave complete control of *Fusarium* even when immersion was for only five minutes at half rate, with Disolite and Unifect G also preventing *Pythium* survival

Background and expected deliverables

Factsheet 03/14 collated information from AHDB Horticulture projects on the efficacy of various disinfectants. Several actives listed in the factsheet are no longer available or permitted (e.g. dichlorophen, formaldehyde, high boiling point tar phenols). Actives currently marketed for use in other areas (e.g. amines and chlorhexidine used in veterinary hygiene and designated as product-type 3) will no longer be permitted for use in horticulture unless a successful application under the BPCR for their use as a product-type 2 active is made.

Nine disinfectants currently used or likely to be available for use in commercial horticulture will be evaluated for their effectiveness against two common ornamental plant pathogens a *Fusarium* sp. (e.g. *Fusarium oxysporum* f. sp. *mathiolae*) and an oospore producing *Pythium* sp.. Work on *Pythium* sp. is expected to be relevant to other oomycetes such as root and foliar infecting *Phytophthora* spp. and downy mildews. Products selected will be from across a range of biocide types, representing the major different chemical modes of action (e.g. hydrogen peroxide, benzoic acid, iodophor, quaternary ammonium compound).

A series of laboratory tests will be done with *Pythium* and *Fusarium* mycelium containing resting spores to determine the effect of i) disinfectant concentration ii) exposure time iii) the presence of organic matter and iv) surface type on product activity. The effect of the concentration and contact duration of the disinfectants on *Fusarium* spores (conidia) will also be examined. Methods devised to test against *Mycosphaerella melonis* and reported in AHDB Horticulture project report PE 001a will be used.

The objectives of this work are:

1. To determine the efficacy of a range of chemical disinfectants, from different active ingredient groups, against spores of *Fusarium*
2. To determine the efficacy of a range of chemical disinfectants, from different active ingredient groups, against mycelium of *Fusarium* and *Pythium*

3. To determine the efficacy of selected disinfectants for reduction of *Fusarium* and *Pythium* on five surfaces (glass, aluminium, rigid plastic, concrete and woven ground cover material).
4. To determine the effect of peat contamination of selected disinfectants against mycelium of *Fusarium* and *Pythium*

Summary of the work and main conclusions

Nine disinfectant products were selected for efficacy testing against two pathogens of ornamental plants, *Fusarium* and *Pythium*, whose resting spores can contaminate nursery beds, benches and re-used containers and lead to root infestation. Products were selected to include disinfectants with different active ingredients, some having been tested by ADAS using the same methods in previous AHDB Horticulture projects (as summarised in Factsheet 03/14) and thus included to aid comparison with the current work. The products selected for this project are shown in Table 1. All products were tested at both full rate and half rate, with contact times of both five and 30 minutes (four treatments).

Table 1. Details of products tested

Product	Active ingredient(s)	Full rate on product label
1. Jet 5	Hydrogen peroxide + peroxyacetic acid	1:125
2. Citrox P	Plant extract	1:150
3. Disolite	Phenolics	2%
4. FAM 30	Iodophor	1:125
5. Menno Florades	Benzoic acid	1%
6. Hydrocare	Hydrogen peroxide + silver	3%
7. Unifect G	Quaternary ammonium compounds + aldehyde	4%
8. Virkon S	Peroxygen compounds + organic acids	1:100
9. Domestos Extended Germ Kill	Chlorine-based bleaching agents (sodium hypochlorite)	120 ml into 5 L

The pathogens used in the work were *Fusarium oxysporum* f. sp. *matthiolae* isolated from wilted stems of column stocks (*Matthiola incana*) and *Pythium irregulare* from yew (*Taxus baccata*) roots.

Spores were collected from three-week old agar plates of *Fusarium* sp. Disinfectant was added to the spore suspension to obtain either a full or half rate product dilution before centrifuging after five or 30 minutes so that the disinfectant could be pipetted off and replaced with sterile distilled water (SDW). Spores from each of the four treatments per disinfectant were pipetted onto agar, giving ten replications. The control treatment was SDW. The agar plates were incubated and *Fusarium* growth recorded at intervals up to seven days.

In order to treat resting spores of *Pythium* sp. (oospores) and *Fusarium* sp. (chlamydospores), mycelium was grown for three weeks on 7 mm diameter filter paper discs on agar plate that could be lifted off for immersion in the disinfectants. The same four treatments and ten replicates were used per disinfectant as for the spore testing, together with a SDW control. The discs were removed from the disinfectants after the required interval, rinsed in SDW and allowed to drip dry before incubating on agar plates for seven days to allow recording of colony growth.

The mycelium disc testing was repeated at full rate for five and 30 minutes immersion of *Fusarium* and *Pythium* with the addition of 0.1% w/v John Innes No1 with peat to the disinfectant. Organic matter can affect the efficacy of some disinfectants (AHDB Horticulture Factsheet 03/14).

For both spore and mycelium tests, disinfectant efficacy was generally the same regardless of the concentration or contact time except for the control of *Fusarium* spores by Hydrocare, where there was no control at half rate for five minutes, but complete control at full rate. Jet 5, Citrox P, Disolite FAM 30, Unifect G and Domestos Extended Germ Kill gave complete control of *Fusarium* spores. Efficacy of most products was, however, poor against *Fusarium* mycelium with only Disolite, Unifect G and Domestos Extended Germ Kill giving complete control, and these were still effective when organic matter was added. *Pythium* mycelium control was more variable, with Disolite and Unifect G giving complete control, but with Jet 5, Citrox P, FAM 30 giving 50% to 80% control at half rate, but some complete control at full rate. After the addition of organic matter to full rate disinfectants Jet 5, FAM 30 and Unifect G gave complete control after 5 minutes contact.

Table 2 shows the percentage control given by each disinfectant used at full rate for 30 minutes (the number of replicates out of the ten without any surviving pathogen). The most effective disinfectants against *Fusarium* and *Pythium* were Disolite, Unifect G and Domestos Extended Germ Kill.

Table 2. Summary of the control given by disinfectants used at full rate for 30 minutes contact time

Table 2. The proportion of replicates out of ten without pathogen growth after full rate for 30 minutes					
No survival	80% - 90% control	50% - 70% control	30% - 40% control	10 – 20% control	Zero control (alive)
+++++	++++	+++	++	+	-

Product name	<i>Fusarium</i> spores	<i>Fusarium</i> mycelium	<i>Pythium</i> mycelium	<i>Fusarium</i> mycelium + peat	<i>Pythium</i> mycelium + peat
1. Jet 5	+++++	+++	+++++	-	+++++
2. Citrox P	+++++	-	++++	+	++++
3. Disolite	+++++	+++++	+++++	+++++	+++++
4. FAM 30	+++++	-	+++++	-	+++++
5. Hydrocare	+++++	-	++++	+	-
6. Virkon S	++++	-	+	-	-
7. Unifect G	+++++	+++++	+++++	+++++	+++++
8. Menno Florades	+++	-	+++	-	+
9. Domestos Extended Germ Kill	+++++	+++++	+++++	+++++	+++++

The type of surface being treated can affect the efficacy of some disinfectants (AHDB Horticulture PE 001a) and so materials were brought into the laboratory for testing. The selected surfaces were a glass sheet, a polypropylene grow-bag tray, an aluminium sheet, a concrete slab and woven ground cover material. These were cleaned down with running water and marked into sections for inoculation. The mycelium and spores were scraped off three-week old agar plates of *Fusarium* sp. and of *Pythium* sp. used to produce an even suspension of unfiltered inoculum. Droplets of the inoculum of one or other pathogen were dispensed over each surface and spread out over each 100 mm x 100 mm area and left to dry. Each of the nine disinfectants and a SDW control was then sprayed to the point of run-off onto the square areas of each pathogen and left for 30 minutes. Five cotton wool bud

swabs were then used in turn to wipe across each surface per disinfectant and each swab streaked over a different agar plate. The plates were incubated and examined after seven days for the presence or absence of mycelial growth.

The failure of Citrox P, FAM 30, Hydrocare, Virkon S and Menno Florades to control *Fusarium* was again shown in the surface tests (Table 3). Disolite and Unifect G still gave very good control, although sometimes incomplete. Domestos Extended Germ Kill performed much poorer than in the immersion tests, only working on glass, but this problem was not seen with *Pythium* (Table 4). Relative product performance at full rate for 30 minutes across the surface types was similar to that seen for the same rate and duration in the *Pythium* mycelium dip tests, with Virkon S giving poor control. However, the previously complete control by Disolite was incomplete on all of the surfaces. No adverse effects on the surfaces were seen from any of the products.

In conclusion, Disolite (a phenolic currently used by the mushroom industry) and Unifect G (a quarternary ammonium compound + aldehyde) performed consistently well against both *Fusarium* and *Pythium*, giving complete control in immersion tests even with short contact time or using a half rate. Domestos Extended Germ Kill (sodium hypochlorite) gave complete control of both with a 30 minute contact period. *Fusarium* resting spores survived treatments that controlled *Pythium*. Where *Pythium* is the main concern on a nursery then Jet 5 (hydrogen peroxide + peroxyacetic acid) or Citrox P (plant extract) at full rate should be effective. Where other bacterial, fungal or virus pathogens are to be controlled then information should be sought on the likely spectrum of activity of the disinfectant types and consideration given to, for example, odour, corrosiveness and biodegradability.

Table 3. Effect of treatments on five surfaces contaminated with *Fusarium* after 7 days incubation, showing % control 30 minutes after the application of disinfectant sprays at full rate

Tables 3&4. The proportion of five replicates without pathogen growth after full rate for 30 minutes					
No survival	80% control	60% control	40% control	20% control	Zero control (alive)
+++++	++++	+++	++	+	-

Product name	Surfaces contaminated with <i>Fusarium</i> prior to disinfectant spray				
	Glass	Plastic	Aluminium	Concrete	Woven ground-cover
1. Water (untreated)	-	-	-	-	-
2. Jet 5	-	++++	-	-	-
3. Citrox P	-	-	-	-	-
4. Disolite	+++++	+++++	+++++	++++	++
5. FAM 30	-	-	-	-	-
6. Hydrocare	-	-	-	-	-
7. Virkon S	-	-	-	-	-
8. Unifect G	+++++	+++++	++++	+++++	+++++
9. Menno Florades	-	-	-	-	-
10. Domestos Extended Germ Kill	+++++	-	-	-	-

Table 4. Effect of treatments on five surfaces contaminated with *Pythium* after 7 days incubation, showing the % control 30 minutes after the application of disinfectant sprays at full rate

Product name	Surfaces contaminated with <i>Pythium</i> prior to disinfectant spray				
	Glass	Plastic	Aluminium	Concrete	Woven ground-cover
1. Water (untreated)	-	-	-	-	+++
2. Jet 5	+++++	+++++	+++++	++++	+++++
3. Citrox P	+	+++++	-	+++	+++++
4. Disolite	+++	+++	++++	++++	+++
5. FAM 30	+++++	+++++	+++++	++++	+++++
6. Hydrocare	++++	++++	+++++	+++++	++++
7. Virkon S	-	-	+	++++	++++
8. Unifect G	+++++	+++++	+++++	+++++	+++++
9. Menno Florades	+	+++++	++++	+++++	++++
10. Domestos Extended Germ Kill	+++++	+++++	+++++	+++++	+++++

Action Points

- Ensure thorough disinfection of surfaces and containers between crops, particularly if there has been a root disease problem
- Before disinfecting a surface remove as much debris as possible to remove sources of resting spores that can resist treatment and to prevent any loss of product efficacy
- Refer to product labels and information in AHDB Horticulture Factsheet 03/14 together with the results from this research to select the most effective disinfectant product for the situation
- Do not use disinfectants on plants, soil or growing substrates
- Ensure that the recommended personal protective clothing is worn when handling the product and ensure safe disposal of any waste product