CONTRACT REPORT FOR THE
HORTICULTURAL DEVELOPMENT COUNCIL

(PC39)

EVALUATION OF FUNGICIDES AND A BIOLOGICAL CONTROL AGENT FOR THE CONTROL OF WILT (*Verticillium dahliae*) AND BLACK ROOT ROT (*Phomopsis sclerotioides*) IN ROCKWOOL GROWN CUCUMBERS

HORTICULTURE RESEARCH INTERNATIONAL

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AUTHENTICATION

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

Signed .....

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Summary

A replicated cucumber trial using the cv. Corona grown to a good commercial standard in rockwool evaluated a range of products (carbendazim as Bavistin FL, prochloraz-manganese as Sporgon 50WP, myclobutanil as Systhane 6W and a coded biocontrol agent as BCA 1001) for the control of both black root rot (*Phomopsis sclerotioides*) and wilt (*Verticillium dahliae*).

Following artificial inoculation *P. sclerotioides* developed slowly though by the end of the investigation had caused severe root damage in inoculated control plots. Inoculation with *V. dahliae* appeared to have little effect on the plants and could not be recovered from the root zone at any stage in the investigation. However, in an interim assessment of root discoloration in early September and in a final assessment at crop termination a significantly higher level of root discoloration was observed in the verticillium inoculated plots. Of particular interest was the consistent recovery of *V. dahliae* from nodes taken approximately 1 ft (30 cm) up the stem, including from nodes from the uninoculated control plants, in destructive tests at crop termination. A satisfactory explanation for these results has not been found.

Both Bavistin FL and Sporgon 50WP significantly reduced the severity of root infection and discoloration caused by both *P. sclerotioides* and *V. dahliae*. In addition both Systhane 6W and the biocontrol agent, BCA 1001, significantly reduced root discoloration in the verticillium inoculated plots.

Stem disease caused by *Mycosphaerella melonis* (*Didymella bryoniae*) and *Botrytis cinerea* was prevelant in this trial and plant losses, due in part to these diseases, increased over the trial period until by September over 50% of the plants were dead in the inoculated control plots. The high level of plant losses were attributed to a combination of root infection by the

pathogens introduced and poor control of stem disease, brought about by the limited fungicide programme available. Certain fungicides, eg benzimidazoles, could not be used because of their potential interaction with the experimental fungicide programmes applied to the root zone.

A marked reduction in the incidence of both stem diseases was observed following fungicide treatment, particularly with Bavistin FL applied at the higher rate. This fungicide applied at the higher rate reduced plant losses significantly though surprisingly this did not affect the final crop yield.

Introduction

It was generally assumed that the move to hydroponic systems of crop culture would eliminate the problem of soil-borne root infecting fungi. Initially, disease levels in hydroponic crops were alleviated by isolation from the soil. Some diseases persisted, however, particularly those caused by water-moulds eg, phycomycete fungi (*Pythium* and *Phytophthora* spp). A few fungicides are now available with label recommendations for the control of these diseases in hydroponic crops eg, Filex, Aaterra. Whilst both *Pythium* and *Phytophthora* remain the most frequent root pathogens of hydroponic crops other diseases occur sporadically and can cause both yield and quality losses in some crops.

Recently cucumber growers have experienced increasing problems with the soil-borne fungi *Phomopsis sclerotioides*, cause of black root rot and *Verticillium dahliae* cause of vascular wilt. It is assumed that these diseases are the result of earlier soil-borne contamination. Whilst fungicides are available to control these pathogens in soil or straw bale grown crops recommendations for their use does not extend to hydroponic crops.

The purpose of this investigation was to evaluate the efficacy of carbendazim as Bavistin FL alongside a range of other potential products for the control of both *P. sclerotioides* and *V. dahliae* following artificial inoculation in rockwool cucumbers. A secondary aim was to generate residue data following carbendazim use in support of a Specific Off-Label Approval for the use of Bavistin FL in hydroponic crops, assuming efficacy could be demonstrated.

Materials and Methods

Sites

The replicated efficacy trial was located in a 0.1 ha (1/4 acre) multispan polycarbonate house (No. 6) at Horticulture Research International, Stockbridge House, Cawood, Selby, North Yorkshire, YO8 0TZ.

Two further glasshouse sites were used (House 9 and Fairfield House 1) for non-replicated (residue) trials using carbendazim (Bavistin FL).

Cultivars

The cultivar Corona was used in the replicated trial in House 6 and also in Fairfield House 1. The cultivar Rebella was used in House 9.

Trial Design

The replicated efficacy trial was a randomised block, split plot design with 3 replicates. Plots consisted of a double row, each row containing 5 rockwool slabs (10 plants) with a total of 20 plants/plot. One row in each plot was inoculated with *P. sclerotioides*, the other with *V. dahliae*.

The unreplicated plots for residue studies in House 9 and Fairfield House 1 consisted of 6 rockwool slabs (12 plants). Pathogen inoculation was not carried out in these areas.

Crop Husbandry

All three trial crops were maintained at ADAS blueprint, ie 21°C day temperature and 19°C night temperature with the vent setpoint at 24°C. Crop nutrition was maintained at ADAS blueprint controlled by timer but varying due to the demand by the crop from the levels of solar radiation. The aim was to achieve 20% run-off at all times.

Pest and disease control was maintained using an IPM programme whereas foliar diseases were suppressed with carefully selected pesticides. With the exception of the experimental treatments, benzimidazole fungicides were not applied to the crops during these trials.

Fungaflor (imazalil) was used routinely as a HV spray to the foliage for powdery mildew control with 4 spray applications during the season. Protectant HV sprays of Rovral (iprodione) and Bravo 500 (chlorothalonil), used in an alternating programme with 5 sprays in total, were applied for the suppression of stem disease (*Botrytis* and *Mycosphaerella*). Filex (propamocarb hydrochloride) was applied to the root zone immediately post-planting and again 2-3 weeks later for the control of *Pythium* spp. In addition a 3 spray programme of Torque (fenbutatin oxide) was used for the control of red spider mite followed by a final spray of Childion (dicofol+tetradifon). All treatments were applied according to manufacturers recommendations.

Treatments

Treatments in the trial were as follows:

1. Uninoculated control.

2. Inoculated control.

3. Carbendazim (Bavistin FL) applied manually as a drench at monthly intervals to the rockwool block/slab at a rate of 500 ml/plant of a solution containing 4 ml product/100 litres water.

4. Carbendazim (Bavistin FL) applied manually as a drench at monthly intervals to the rockwool block/slab at a rate of 500 ml/plant of a solution containing 10 ml product/100 litres water.

5. Prochloraz-manganese (Sporgon 50WP) applied manually as a drench at monthly intervals to the rockwool block/slab at a rate of 500 ml/plant of a solution containing 6.8 g product/100 litres water.

6. A coded biocontrol agent (BCA 1001) from Kemira Oy, Helsinki applied manually as a drench at monthly intervals to the rockwool block/slab at a rate of 60 ml/plant of a solution containing 1 g product/10 litres water.

7. Myclobutanil (Systhane 6W) applied manually as a drench at monthly intervals to the rockwool block/slab at a rate of 500 ml/plant of a solution containing 10 g product/100 litres water.

Fungicide and BCA Application

All treatments were applied manually to individual plants by drenching each rockwool block with the fungicide or BCA and allowing the product to infiltrate the rockwool slab. The first application was made 7 days prior to pathogen inoculation in the replicated trial. At each treatment date drenches were applied in 500 ml solution/plant. The only exception to this was with the biocontrol agent where 60 ml solution/plant was applied.

Inoculation

V. dahliae and *P. sclerotioides* were introduced into individual rockwool slabs in the sub-plots by aseptically removing a 1 cm diameter rockwool core from the centre of each slab using a cork-borer and inserting a 1 cm diameter disc of agar containing *V. dahliae* or 0.1 g crushed root debris containing micro-sclerotia of *P. sclerotioides*. Rockwool cores were re-inserted and the slabs resealed using sticky tape.

Pathogen Monitoring

Various techniques including a cucumber seedling bioassay were used in an attempt to monitor pathogen progess in this trial, though none were found to be totally satisfactory. Visual observation and microscopic examination was finally used as a method for determining the success of inoculation procedures and subsequent pathogen establishment.

Sample Collection for Residue Analysis

In the replicated trial (House 6) a minimum of 4 cucumber fruit/plot were picked by hand 24, 48 and 72 hours after 4, 6, 7 and 8 carbendazim applications in Treatments 1 (untreated), 3 (4 ml Bavistin FL/100 l water) and 4 (10 ml Bavistin FL/100 l water). Fruit from the 3 replicates were bulked for analysis. In House 9 and Fairfield House 1 a minimum of 4 fruit/plot were collected for analysis at each sampling date. The fruit harvested from each plot were immediately placed in labelled, sealed, polythene bags. Remaining fruit harvested from the trial areas were discarded.

Samples from House 9 after eight applications of carbendazim could not be obtained as the crop was terminated prematurely.

Storage of Residue Samples

All harvested fruit awaiting analysis was immediately placed in a deep freeze maintained at -20°C.

Despatch and Transportation of Residue Samples

Fruit samples taken after the last application of carbendazim (eighth application in House 6 and Fairfield House 1 and seventh application in House 9) were transported to G C Laboratories Ltd, Faldo Road, Barton-Le-Clay, Bedfordshire, MK25 4RL for residue analysis. Samples were transported in insulated crates and upon receipt were immediately returned to deep freeze facilities maintained at -20°C.

Analysis of Residue Samples

All analyses were conducted to GLP standards by G C Laboratories, Faldo Road, Barton-Le-Clay, Bedfordshire. The results of the analyses are presented in a separate report (Annex I "Determination of carbendazim residues in cucumbers").

Assessments

Assessments of crop vigour and disease development were made as the trial progressed. These included:

1. Incidence of stem disease caused by *M. melonis* and *B. cinerea*.

2. Incidence of leaf disease caused by powdery mildew.

3. Incidence of wilting and dead plants.

4. Severity of root discoloration in rockwool cores and presence of microsclerotia of *P. sclerotioides*.

Rockwool cores were either examined microscopically for the presence of microsclerotia of *P. sclerotioides* or root discoloration attributed to *V. dahliae* This latter assessment was carried out using a 0-3 scale where:

0 = No root discoloration
1 = Slight root discoloration
2 = Moderate root discoloration
3 = Severe root discoloration

At termination of the trial an assessment of root development in the slab and root discoloration in the block and slab was made on the roots visible after removal of the polythene sleeves. Assessments were based on a 0-3 scale where:

Root Development

0 = No root visible
1 = Slight root development
2 = Moderate root development
3 = Extensive root development

Root Discoloration

0 = No root discoloration
1 = Slight root discoloration
2 = Moderate root discoloration
3 = Severe root discoloration

Where interim and final disease assessments were made using a 0-3 scale an index was calculated using the formula:

$$\frac{1\,(1) + 2\,(2) + 3\,(3)}{\text{Number Assessed}} \times \frac{100}{3}$$

At crop termination stem sections were taken from 5 plants/sub-plot in an attempt to recover *V. dahliae* from the vascular tissues. 1 cm sections of node tissue was placed on moist sterile filter papers in Petri dishes and incubated for 72 hours. Microscopical examination was used as a means of confirming pathogen presence.

All fruit was harvested, counted, weighed, and graded according to size and quality.

Statisical Analysis

Data was subjected to a split plot analysis of variance where appropriate. Treatment means were separated using Duncan's Multiple Range Test. The uninoculated control treatment was omitted from the analyses, with the exception of the yield data.

Crop Diary

The cucumber seed for House 6, Fairfield House 1 and House 9 were sown in perlite on 24 December, 13 December 1990 and 22 January 1991 respectively. Seedlings were pricked out into propagation blocks when the radicle had emerged and subsequently transferred to the growing houses on 21 January, 9 January and 14 February respectively. The first application of fungicides and biological control agents was applied on 25 February before inoculation with both pathogens on 4 March (House 6 only). Subsequent applications of fungicide and biological control agents were made on 26 March, 23 April, 21 May, 18 June, 15 July, 13 August and 10 September. Repeat inoculation with *V. dahliae* and *P. sclerotioides* was carried out on 22 May and 1 July respectively. Rockwool core samples for assessment were taken on 2 September. Plants were assessed for disease symptoms on 18 June, 9 July, 2 September and at trial termination on 27 May, 15, 16 and 17 July, 13, 14 and 15 August and 10, 11 and 12 September.

Samples were submitted to G C Laboratories for residue analysis on 22 September. A completed report was subsequently received on 1 December 1991.

A residue report was prepared and submitted to the NFU via HDC in support of a Specific Off-Label Approval for Bavistin FL on 24 January 1992. An Off-Label Approval Notice No. 0713/92 was finally issued on 23 June 1992.

Results

Following artificial inoculation an attempt to monitor pathogen establishment and subsequent development was made using a 14 day old cucumber seedling bioassay. However, neither *P. sclerotioides* or *V. dahliae* were recovered consistently using this technique even after several attempts. Instead, an alternative technique was used whereby rockwool cores were removed at intervals, particularly from the blocks and slabs in Treatment 2 (inoculated control). The roots teased out from these cores were examined visually and microscopically for evidence of pathogen establishment and subsequent disease development.

Using the latter technique it became clear that *P. sclerotioides* developed only slowly following inoculation. The first visible sign of root infection was on 18 June when microsclerotia of the pathogen were observed on a few plants only. To ensure an even distribution of *P. sclerotioides* in the trial reinoculation was carried out in the same manner as previously described on 1 July.

Following inoculation with *V. dahliae* slight root discoloration became apparent though attempts to observe the fungus on the roots microscopically were unsuccessful and indeed isolation onto artificial agar media failed to confirm *V. dahliae* as the cause of the discoloration. As *V. dahliae* is a vascular pathogen it would have been preferable to have taken samples from the stem near the plant base but destructive sampling was not possible because of the limited plant numbers in the trial. It was assumed that our inoculation technique had been unsuccessful and plots were reinoculated on 22 May in a further attempt to secure disease establishment. Even after repeat inoculation with *V. dahliae* however, we were left with no evidence of successful pathogen establishment.

On 8 May the first signs of stem disease caused by mycosphaerella, botrytis and penicillium were recorded. A full assessment carried out on 18 June indicated that mycosphaerella was the most prevalent stem disease though botrytis was also common.

The infection by *P. oxalicum* observed earlier on 8 May failed to develop further and the pathogen was not detected again in the trial.

28% plants were recorded with one or more stem lesions of mycosphaerella on 18 June (Table 1) with an indication of increased plant susceptibility in the phomopsis inoculated sub-plots. The same effect was not observed where *V. dahliae* had been introduced into the root zone. A similar effect was recorded with botrytis though disease incidence was much lower. (Table 2).

The experimental products were in some cases, effective in reducing the incidence of stem disease (Table 1). Bavistin FL, particularly at the higher rate of application, (Treatment 4) significantly reduced the incidence of stem infection by mycosphaerella from 47% to 0% plant infection in the phomopsis inoculated sub-plots in this interim assessment on 18 June. A similar effect was recorded in the verticillium inoculated sub-plots though the effect here was not significant at the 5% level of probability. Some of the other experimental treatments particularly the biocontrol agent (BCA 1001) also appeared to suppress stem infection by mycosphaerella, though only following inoculation with *P. sclerotioides*. None of the treatments evaluated in the trial reduced the incidence of stem lesions caused by Botrytis in this interim assessment (Table 2).

By 18 June some plant losses had occurred (Table 3) and this was largely attributed to stem infection of mycosphaerella and botrytis. Treatment with Bavistin FL and Systhane were effective in reducing these plant losses, though the effect was not statistically significant in this interim assessment.

The incidence of stem infection by both mycosphaerella and botrytis had increased by 9 July when a further assessment was made (Tables 4-5). The results appeared fairly variable at this second assessment and perhaps surprisingly none of the treatments significantly reduced the incidence of mycosphaerella and botrytis stem infection (main stem to wire only).

Wilting of individual plants was evident at this stage and plant losses had increased slightly from the previous assessment (Tables 6-7). The incidence of wilting was as great in the uninoculated plots as it was in the inoculated plots and whilst there was an indication of a reduction following fungicide application in some cases the results were too variable to draw conclusions. Similarly where plant losses were recorded, fungicide treatment reduced the losses but the effect was not significant ($P = 0.5$).

A final assessment of stem infection at crop termination was carried out on 27 September (Table 8). This assessment was based on aggressive 'wet' lesions only and limited 'dry' lesions were not assessed. In contrast to earlier results, where the incidence of stem infection was higher following inoculation with phomopsis, at trial termination the incidence of aggressive lesions of mycosphaerella was much lower. A similar effect was observed in the verticillium inoculated plots though here all treatments applied significantly reduced the number of plants with aggressive stem lesions.

Root infection became more pronounced as the trial progressed. Rockwool cores removed on 2 September (Table 9) demonstrated an extremely high incidence of root infection by phomopsis in the inoculated plots. A marked reduced in root infection was secured using Bavistin FL particularly at the higher rate. Interestingly Sporgon containing prochloraz-manganese was the most effective treatment, reducing the incidence of plants with root infection from 86.7% to 33.3%.

A moderate level of root discoloration was recorded in the verticillium inoculated control plots and both Bavistin FL (high rate) and Systhane reduced this significantly. Whether the root infection assessed was a direct result of verticillium infection is uncertain.

At crop termination a large proportion of plants in the cropping house were either wilting (Table 10) or dead (Table 11) Bavistin FL significantly reduced the plant losses, particularly at the higher rate used (Treatment 4) and this was in both the phomopsis and the verticillium inoculated plots. Sporgon was the only other treatment to significantly reduce plant losses though only in the phomopsis inoculated plots. Where the data for wilting and dead plants was combined (Table 12) Bavistin FL (high rate), Sporgon 50WP and Systhane (verticillium inoculated plots only) were effective in significantly reducing losses as compared with the inoculated control. Plant losses (wilting and dead) were reduced from 58% (uninoculated control) to 15% (Bavistin 10 ml/100 l).

A full assessment of the roots in the blocks and slabs was only possible at the end of the trial when the polythene sleeves could be removed. Bavistin FL, Sporgon 50WP and Systhane 6W significantly increased root development in both the phomopsis and the verticillium inoculated slabs (Table 13).

Bavistin FL at both the low and high rate and Sporgon 50WP significantly reduced the incidence and severity of root discoloration caused by phomopsis in the rockwool blocks (Table 14). Only BCA 1001 and Systhane 6W significantly reduced the root discoloration in the verticillium inoculated plots, though again it is unclear whether the symptom observed was a direct result of the verticillium inoculum introduced.

In the slabs Bavistin FL applied at the higher rate and Sporgon significantly reduced root discoloration caused by *P. sclerotioides* (Table 15). All treatments applied reduced root discoloration in the verticillium inoculated plots, though again whether this was caused by the inoculated pathogen remains unclear.

At crop termination there was an opportunity to carry out destructive tests using the vascular tissues of the main stem to determine whether the verticillium inoculation had been successful. Stem slices were taken across the nodes and incubated under high humidity conditions in damp chambers at 95° RH for 48-72 hours to encourage aerial development of the fungus on the stem slices. Microscopic observations after this incubation period revealed verticillium in almost every plant including the uninoculated control plants (Table 16).

Throughout the trial duration fruit yield was assessed (Tables 17-18). No significant differences were recorded in fruit number, or fruit weight, though surprisingly, fruit quality was significantly increased following pathogen inoculation.

The encouraging results obtained using Bavistin FL highlighted the need to pursue residue data generation for carbendazim, the active ingredient of Bavistin FL. Samples collected during the season were submitted to G C Laboratories, Bedfordshire who conducted the analyses to Good Laboratory Practice (GLP)

standards and subsequently issued a residues report (Annex I). The results indicated that residue levels in all samples taken within 24-72 hours of the seventh or eighth application of Bavistin FL at either 4 ml or 10 ml per 100 l water (500 ml/plant) were very low.

A residue report was subsequently prepared and submitted to the Pesticide Safety Division of MAFF, via the HDC and NFU, in support of an emergency Specific Off-Label Approval application for Bavistin FL.

The report was evaluated by PSD and on 23 June 1992 a Specific Off-Label Approval for Bavistin FL was issued (Annex II) which allows growers to routinely use Bavistin FL on hydroponic cucumbers. Unfortunately the SOLA only provisionally Approves use of Bavistin FL at the lower rate tested in the trial and additional data requirements have been set for both the lower and higher rates. Data generation in support of Specific Off-Label Approvals is being coordinated as part of a separate HDC funded project. It is anticipated that additional data will be collected following use of Bavistin FL to allow full Off-Label Approval, hopefully at the higher rate of use.

Unfortunately, as the SOLA application was submitted BASF plc announced their intention to rationalise the various formulations of Bavistin. They therefore announced their intention to withdraw Bavistin WP and Bavistin FL, and to concentrate their label recommendations on the DF formulation.

Subsequent correspondence (Annex III) confirmed that PSD will be prepared to consider an application for Off-Label Approval for Bavistin DF. Outstanding data requirements would naturally still apply.

Table 1: Interim assessment of Mycosphaerella stem lesions in the trial on 18 June 1991.

Treatment		% Plants with Stem Lesions of Mycosphaerella		
		Phompsis Inoculation	Verticillium Inoculation	Mean
1.	Uninoculated+	33.3	23.3	28.3
2.	Inoculated*	46.7 b	23.3 ab	35.0 b
3.	Bavistin FL* (4 ml/100 l)	20.0 ab	30.0 ab	25.0 ab
4.	Bavistin FL* (10 ml/100 l)	0.0 a	6.7 a	3.4 a
5.	Sporgon 50WP* (6.8 g/100 l)	26.7 ab	20.0 ab	23.4 ab
6.	BCA 1001* (1 g/10 l)	16.7 a	40.0 b	28.4 b
7.	Systhane 6W* (10 g/100 l)	20.0 ab	20.0 ab	20.0 ab
SED		11.34	11.34	9.72
Degrees of Freedom		12	12	10

* One row in each plot inoculated with either *P. sclerotioides* or *V. dahliae*.

\+ Uninoculated control omitted from analysis.

Figures with the same letter in the suffix within each column do not differ significantly at the 5% (P = 0.05) level of probability (Duncan's Multiple Range Test).

Table 2: Interim assessment of Botrytis stem lesions in the trial on 18 June 1991.

Treatment		% Plants with Stem Lesions of Botrytis		
		Phompsis Inoculation	Verticillium Inoculation	Mean
1.	Uninoculated+	6.7	10.0	8.4
2.	Inoculated*	13.3	13.3	13.3
3.	Bavistin FL* (4 ml/100 l)	6.7	16.7	11.7
4.	Bavistin FL* (10 ml/100 l)	13.3	6.7	10.0
5.	Sporgon 50WP* (6.8 g/100 l)	13.3	16.7	15.0
6.	BCA 1001* (1 g/10 l)	10.0	20.0	15.0
7.	Systhane 6W* (10 g/100 l)	20.0	30.0	25.0
SED		NSD	NSD	NSD
		13.30	13.30	11.43
Degrees of Freedom		12	12	10

* One row in each plot inoculated with either *P. sclerotioides* or *V. dahliae*.

\+ Uninoculated control omitted from analysis.

NSD No significant difference at the 5% level of probability (P = 0.05).

Table 3: Interim assessment of plant losses in the trial on 18 June 1991.

Treatment	% Plants Dead		
	Phompsis Inoculation	Verticillium Inoculation	Mean
1. Uninoculated*	13.3	10.0	11.7
2. Inoculated*	13.3	16.7 ab	15.0
3. Bavistin FL* (4 ml/100 l)	3.3	6.7 a	5.0
4. Bavistin FL* (10 ml/100 l)	0.0	13.3 ab	6.2
5. Sporgon 50WP* (6.8 g/100 l)	10.0	16.7 ab	13.4
6. BCA 1001* (1 g/10 l)	6.7	26.7 b	16.7
7. Systhane 6W* (10 g/100 l)	5.0	5.0 a	5.0
SED	NSD 7.79	7.79	NSD 7.37
Degrees of Freedom	12	12	10

* One row in each plot inoculated with either *P. sclerotioides* or *V. dahliae*.

· Uninoculated control omitted from analysis.

Figures with the same letter in the suffix within each column do not differ significantly at the 5% level of probability (Duncan's Multiple Range Test).

NSD No significant difference at the 5% level of probability ($P = 0.05$).

Table 4: Interim assessment of Mycosphaerella stem lesions in the trial on 9 July 1991.

Treatment		% Plants with Stem Lesions of Mycosphaerella		
		Phompsis Inoculation	Verticillium Inoculation	Mean
1.	Uninoculated+	30.0	33.3	31.7
2.	Inoculated*	43.3	23.3	33.3
3.	Bavistin FL* (4 ml/100 l)	56.7	30.0	43.4
4.	Bavistin FL* (10 ml/100 l)	40.0	13.3	26.7
5.	Sporgon 50WP* (6.8 g/100 l)	36.7	33.3	35.0
6.	BCA 1001* (1 g/10 l)	23.3	26.7	25.0
7.	Systhane 6W* (10 g/100 l)	50.0	50.0	50.0
SED		NSD 17.29	NSD 17.29	NSD 13.37
Degrees of Freedom		12	12	10

* One row in each plot inoculated with either *P. sclerotioides* or *V. dahliae.*

\+ Uninoculated control omitted from analysis.

NSD No significant difference at the 5% level of probability (P = 0.05).

Table 5: Interim assessment of Botrytis stem lesions in the trial on 9 July 1991.

Treatment		% Plants with Stem Lesions of Botrytis		
		Phompsis Inoculation	Verticillium Inoculation	Mean
1.	Uninoculated+	20.0	10.0	15.0
2.	Inoculated*	20.0	6.7	13.4
3.	Bavistin FL* (4 ml/100 l)	10.0	16.7	13.4
4.	Bavistin FL* (10 ml/100 l)	3.3	13.3	8.3
5.	Sporgon 50WP* (6.8 g/100 l)	3.3	6.7	5.0
6.	BCA 1001* (1 g/10 l)	16.7	6.7	11.7
7.	Systhane 6W* (10 g/100 l)	0.0	15.0	7.5
		NSD	NSD	NSD
SED		8.39	8.39	6.47
Degrees of Freedom		12	12	10

* One row in each plot inoculated with either *P. sclerotioides* or *V. dahliae*.

\+ Uninoculated control omitted from analysis.

NSD No significant difference at the 5% level of probability (P = 0.05).

Table 6: Interim assessment of wilting plants in the trial on 9 July 1991.

Treatment		% Plants Wilting		
		Phompsis Inoculation	Verticillium Inoculation	Mean
1.	Uninoculated+	43.3	33.3	38.3
2.	Inoculated*	16.7	30.0 abc	23.4 ab
3.	Bavistin FL* (4 ml/100 l)	6.7	20.0 ab	13.4 a
4.	Bavistin FL* (10 ml/100 l)	3.3	56.7 bc	30.0 bc
5.	Sporgon 50WP* (6.8 g/100 l)	23.3	60.0 c	41.7 c
6.	BCA 1001* (1 g/10 l)	6.7	10.0 a	8.4 a
7.	Systhane 6W* (10 g/100 l)	35.0	40.0 abc	37.5 bc
		NSD		
SED		15.49	15.49	6.34
Degrees of Freedom		12	12	10

* One row in each plot inoculated with either *P. sclerotioides* or *V. dahliae*.

\+ Uninoculated control omitted from analysis.

Figures with the same letter in the suffix within each column do not differ significantly at the 5% level of probability (Duncan's Multiple Range Test).

NSD No significant difference at the 5% level of probability (P = 0.05).

Table 7: Interim assessment of dead plants in the trial on 9 July 1991.

Treatment		% Plants Dead		
		Phompsis Inoculation	Verticillium Inoculation	Mean
1.	Uninoculated+	16.7	20.0	18.4
2.	Inoculated*	20.0	23.3	21.7
3.	Bavistin FL* (4 ml/100 l)	6.7	13.3	10.0
4.	Bavistin FL* (10 ml/100 l)	0.0	26.7	13.0
5.	Sporgon 50WP* (6.8 g/100 l)	10.0	16.7	13.4
6.	BCA 1001* (1 g/10 l)	10.0	30.0	20.0
7.	Systhane 6W* (10 g/100 l)	5.0	10.0	7.5
SED		NSD 9.64	NSD 9.64	NSD 8.96
Degrees of Freedom		12	12	10

* One row in each plot inoculated with either *P. sclerotioides* or *V. dahliae*.

\+ Uninoculated control omitted from analysis.

NSD No significant difference at the 5% level of probability (P = 0.05).

Table 8: Assessment of aggressive stem infection by *Mycosphaerella melonis (Didymella bryoniae)* at crop termination on 27 September 1991.

Treatment		% Plants with Stem Lesions Caused by *Mycosphaerella melonis*#		
		Phomopsis Inoculation	Verticillium Inoculation	Mean
1.	Uninoculated+	20.0	40.0	30.0
2.	Inoculated*	3.3	23.3 b	13.3 b
3.	Bavistin FL* (4 ml/100 l)	0.0	6.7 a	3.3 a
4.	Bavistin FL* (10 ml/100 l)	0.0	0.0 a	0.0 a
5.	Sporgon 50WP* (6.8 g/100 l)	0.0	3.3 a	1.7 a
6.	BCA 1001* (1 g/10 l)	0.0	0.0 a	0.0 a
7.	Systhane 6W* (10 g/100 l)	0.0	0.0 a	0.0 a
SED		NSD 3.50	3.50	2.76
Degrees of Freedom		12	12	10

* One row in each plot inoculated with either *P. sclerotioides* or *V. dahliae*.

\+ Uninoculated control omitted from analysis.

\# Assessment of aggressive 'wet' stem lesions only.

Figures with the same letter in the suffix within each column do not differ significantly at the 5% level of probability (Duncan's Multiple Range Test).

NSD No significant difference at the 5% level of probability (P = 0.05).

Table 9: Interim assessment of root disease in 5 cm rockwool cores taken from growing slabs on 2 September 1991.

Treatment		Incidence of *P. sclerotioides* (%)	Index of Root Discoloration for *V. dahliae* (0-3)
1.	Uninoculated+	0.0	0.0
2.	Inoculated	86.7 bc	17.7 c
3.	Bavistin FL (4 ml/100 l)	73.3 abc	11.1 abc
4.	Bavistin FL (10 ml/100 l)	40.0 ab	0.0 a
5.	Sporgon 50WP (6.8 g/100 l)	33.3 a	8.8 abc
6.	BCA 1001 (1 g/10 l)	100.0 c	15.5 bc
7.	Systhane 6W (10 g/100 l)	80.0 abc	3.35 ab
SED		20.79	5.19
Degrees of Freedom		10	10

+ Uninoculated control omitted from analysis.

Figures with the same letter in the suffix within each column do not differ significantly at the 5% level of probability (Duncan's Multiple Range Test).

Table 10: Assessment of wilting and dead plants at crop termination on 27 September 1991.

Treatment		% Plants Wilting		
		Phompsis Inoculation	Verticillium Inoculation	Mean
1.	Uninoculated*	16.7	0.0	8.4
2.	Inoculated*	3.3 ab	6.7	5.0
3.	Bavistin FL* (4 ml/100 l)	6.7 ab	3.3	5.0
4.	Bavistin FL* (10 ml/100 l)	3.3 ab	0.0	1.7
5.	Sporgon 50WP* (6.8 g/100 l)	0.0 a	0.0	0.0
6.	BCA 1001* (1 g/10 l)	10.0 ab	3.3	6.7
7.	Systhane 6W* (10 g/100 l)	13.3 b	0.0	6.7
SED		4.91	NSD 4.91	NSD 3.33
Degrees of Freedom		12	12	10

* One row in each plot inoculated with either *P. sclerotioides* or *V. dahliae*.

* Uninoculated control omitted from analysis.

Figures with the same letter in the suffix within each column do not differ significantly at the 5% level of probability (Duncan's Multiple Range Test).

NSD No significant difference at the 5% level of probability (P = 0.05).

Table 11: Assessment of wilting and dead plants at crop termination on 27 September 1991.

Treatment		% Plants Dead		
		Phompsis Inoculation	Verticillium Inoculation	Mean
1.	Uninoculated·	23.3	56.7	40.0
2.	Inoculated*	56.7 cd	50.0	53.4 bc
3.	Bavistin FL* (4 ml/100 l)	30.0 abc	30.0	30.0 ab
4.	Bavistin FL* (10 ml/100 l)	3.3 a	23.3	13.3 a
5.	Sporgon 50WP* (6.8 g/100 l)	20.0 ab	33.3	26.7 a
6.	BCA 1001* (1 g/10 l)	76.7 d	43.3	60.0 c
7.	Systhane 6W* (10 g/100 l)	45.0 bc	25.0	35.0 ab
			NSD	
SED		12.88	12.88	10.10
Degrees of Freedom		12	12	10

* One row in each plot inoculated with either *P. sclerotioides* or *V. dahliae*.

· Uninoculated control omitted from analysis.

Figures with the same letter in the suffix within each column do not differ significantly at the 5% level of probability (Duncan's Multiple Range Test).

NSD No significant difference at the 5% level of probability (P = 0.05).

Table 12: Mean assessment of plants either wilting or dead at crop termination on 27 September 1991.

Treatment		% Plants Wilting or Dead		
		Phompsis Inoculation	Verticillium Inoculation	Mean
1.	Uninoculated+	40.0	56.7	48.4
2.	Inoculated*	60.0 cd	56.7 b	58.4 cd
3.	Bavistin FL* (4 ml/100 l)	36.7 bc	33.3 ab	35.0 ab
4.	Bavistin FL* (10 ml/100 l)	6.6 a	23.3 a	15.0 a
5.	Sporgon 50WP* (6.8 g/100 l)	20.0 ab	33.3 ab	26.7 ab
6.	BCA 1001* (1 g/10 l)	86.7 d	46.6 ab	66.7 d
7.	Systhane 6W* (10 g/100 l)	58.3 c	25.0 a	41.7 bc
SED		11.92	11.92	9.38
Degrees of Freedom		12	12	10

* One row in each plot inoculated with either *P. sclerotioides* or *V. dahliae*.

\+ Uninoculated control omitted from analysis.

Figures with the same letter in the suffix within each column do not differ significantly at the 5% (P = 0.05) level of probability (Duncan's Multiple Range Test).

Table 13: Assessments of root development at crop termination on 27 September 1991.

Treatment		Index of Root Development in Rockwool Slabs		
		Phompsis Inoculation	Verticillium Inoculation	Mean
1.	Uninoculated+	91.0	84.4	87.7
2.	Inoculated*	75.5 a	79.9 a	77.7 a
3.	Bavistin FL* (4 ml/100 l)	82.1 ab	86.6 ab	84.4 ab
4.	Bavistin FL* (10 ml/100 l)	86.6 bc	93.2 b	89.9 bc
5.	Sporgon 50WP* (6.8 g/100 l)	93.2 c	93.2 b	93.2 c
6.	BCA 1001* (1 g/10 l)	79.9 ab	82.1 a	81.0 a
7.	Systhane 6W* (10 g/100 l)	86.6 bc	93.2 b	89.9 bc
SED		4.52	4.52	3.61
Degrees of Freedom		12	12	10

* One row in each plot inoculated with either *P. sclerotioides* or *V. dahliae*.

\+ Uninoculated control omitted from analysis.

Figures with the same letter in the suffix within each column do not differ significantly at the 5% (P = 0.05) level of probability (Duncan's Multiple Range Test).

Table 14: Assessments of root discoloration at crop termination on 27 September 1991.

Treatment		Index of Root Discoloration in Rockwool Blocks		
		Phompsis Inoculation	Verticillium Inoculation	Mean
1.	Uninoculated+	0.0	16.6	8.3
2.	Inoculated*	78.2 c	45.3 b	61.8 c
3.	Bavistin FL* (4 ml/100 l)	49.9 b	26.6 ab	38.3 b
4.	Bavistin FL* (10 ml/100 l)	22.2 a	33.3 ab	27.8 ab
5.	Sporgon 50WP* (6.8 g/100 l)	14.4 a	21.7 ab	18.1 a
6.	BCA 1001* (1 g/10 l)	64.4 bc	12.3 a	38.4 b
7.	Systhane 6W* (10 g/100 l)	55.0 bc	18.3 a	36.7 b
SED		10.21	10.21	6.67
Degrees of Freedom		12	12	10

* One row in each plot inoculated with either *P. sclerotioides* or *V. dahliae*.

\+ Uninoculated control omitted from analysis.

Figures with the same letter in the suffix within each column do not differ significantly at the 5% (P = 0.05) level of probability (Duncan's Multiple Range Test).

Table 15: Assessments of root discoloration at crop termination on 27 September 1991.

Treatment		Index of Root Discoloration in Rockwool Slabs		
		Phompsis Inoculation	Verticillium Inoculation	Mean
1.	Uninoculated+	33.3	42.2	37.8
2.	Inoculated*	66.6 bc	66.6 b	66.6 c
3.	Bavistin FL* (4 ml/100 l)	53.3 ab	15.6 a	34.5 ab
4.	Bavistin FL* (10 ml/100 l)	35.5 a	22.2 a	28.9 a
5.	Sporgon 50WP* (6.8 g/100 l)	33.3 a	24.4 a	28.9 a
6.	BCA 1001* (1 g/10 l)	88.8 c	26.6 a	57.7 bc
7.	Systhane 6W* (10 g/100 l)	60.0 ab	26.7 a	43.4 abc
SED		11.84	11.84	10.10
Degrees of Freedom		12	12	10

* One row in each plot inoculated with either *P. sclerotioides* or *V. dahliae*.

\+ Uninoculated control omitted from analysis.

Figures with the same letter in the suffix within each column do not differ significantly at the 5% (P = 0.05) level of probability (Duncan's Multiple Range Test).

Table 16: Percentage recovery of *V. dahliae* in destructive tests at crop termination on 27 September 1991.

Treatment		% Recovery of *V. dahliae*
1.	Uninoculated*	96.7
2.	Inoculated*	100.0
3.	Bavistin FL* (4 ml/100 l)	95.2
4.	Bavistin FL* (10 ml/100 l)	88.9
5.	Sporgon 50WP* (6.8 g/100 l)	100.0
6.	BCA 1001* (1 g/10 l)	93.3
7.	Systhane 6W* (10 g/100 l)	91.7
SED		NSD
Degrees of Freedom		9.04
		10

NSD No significant difference at the 5% level of probability (P = 0.05).

Table 17: Marketable yield and mean fruit size during the period of the investigation.

Treatment	No. of Fruit per m^2	Weight of Fruit per m^2 (kg)	Weight per Fruit (g)
1. Uninoculated+	103.6	45.53	438.7
2. Inoculated*	102.8	45.36	439.4
3. Bavistin FL* (4 ml/100 l)	107.1	47.39	442.1
4. Bavistin FL* (10 ml/100 l)	114.2	50.19	438.5
5. Sporgon 50WP* (6.8 g/100 l)	108.4	48.23	444.0
6. BCA 1001* (1 g/10 l)	101.5	45.94	452.2
7. Systhane 6W* (10 g/100 l)	106.5	48.12	451.6
SED	NSD	NSD	NSD
	5.33	2.64	5.6
Degrees of Freedom	10	10	10

NSD No significant difference at the 5% level of probability (P = 0.05).

Table 18: Fruit quality assessments during the period of the investigation.

Treatment		% Cucumbers By Grade	
		Class I	Class II
1.	Uninoculated+	72.65 a	21.42 b
2.	Inoculated*	79.05 c	16.51 a
3.	Bavistin FL* (4 ml/100 l)	77.60 b	16.09 a
4.	Bavistin FL* (10 ml/100 l)	78.91 bc	16.03 a
5.	Sporgon 50WP* (6.8 g/100 l)	77.31 b	17.40 a
6.	BCA 1001* (1 g/10 l)	77.12 b	16.95 a
7.	Systhane 6W* (10 g/100 l)	77.55 b	16.88 a
SED		0.824	0.694
Degrees of Freedom		10	10

Figures with the same letter in the suffix within each column do not differ significantly at the 5% (P = 0.05) level of probability (Duncan's Multiple Range Test).

Discussion

Disease establishment in this trial, whilst slow to develop, appears to have been effective. Certainly, inoculation with *P. sclerotioides* was successful as at crop termination microsclerotia of this pathogen were abundant on the roots of infected plants. 87% of plants exhibited evidence of phomopsis infection in an interim assessment on 2 September. Root discoloration became apparent following the introduction of *V. dahliae* into rockwool slabs though failure to recover the pathogen from the roots made it difficult to draw any firm conclusions. Yet, in destructive tests at crop termination *V. dahliae* was recovered from almost 100% of plants including the uninoculated control plants. It does appear that the pathogen had established successfully in the trial site and spread to infect uninoculated plants. The method of pathogen transfer to the uninoculated plots was not determined.

It proved to be difficult to monitor the progress of root infection in the trial. Many records were based on assessment of stem disease caused by *M. melonis (D. bryoniae)* and *B. cinerea*. Stem infection by these fungi was moderate to high in this trial and indeed high levels of plant losses occurred. Whether the plant losses were directly attributed to these stem diseases or whether they were caused by increasing severity of root infection remains in some doubt, though most likely they resulted from a combination of stem and root disease.

The most encouraging aspect was that the plant losses whatever their cause, were markedly reduced following treatment with Bavistin FL, particularly at the higher rate used in the trial.

The residue data for Bavistin FL generated from this set of trials was used successfully to obtain a Specific Off-Label Approval for Bavistin FL on hydroponic cucumbers, albeit only

Provisionally at this stage and for the lower rate of use tested. Additional data requirements have been set by the regulatory authorities (PSD) and it is hoped that these can be met as part of a further HDC funded project, as yet unregistered.

It is also encouraging that PSD have acknowledged the similarity in the two Bavistin formulations (Bavistin FL and DF) and have agreed to consider a SOLA application for Bavistin DF using the existing data. A new application for Bavistin DF should be pursued (see Annex III).

Conclusions

1. *Phomopsis sclerotioides*, artificially introduced into hydroponic cucumbers, established successfully to cause typical 'black root rot' disease symptoms.

2. Moderate root discoloration occurred in hydroponic cucumbers following artificial inoculation with *Verticillium dahliae*, though it was unclear whether the pathogen was primarily responsible. *V. dahliae* did appear, however, to spread to uninoculated plants readily.

3. Stem disease caused by *Mycosphaerella melonis* and *Botrytis cinerea* was severe in the trial and contributed to the plant losses. Stem disease was reduced significantly following fungicide treatment with Bavistin FL particularly in the early stages of the trial.

4. Treatment with Bavistin FL, particularly at the higher rate used significantly reduced plant losses. The plant losses were attributed to a combination of root infection by *P. sclerotioides* and *V. dahliae* and stem infection by *M. melonis* and *B. cinerea*.

5. Neither of the root pathogens significantly reduced yield and none of the experimental treatments enhanced it.

6. Residue levels of carbendazim taken 24-72 hours after 7 or 8 applications of Bavistin FL at 4 ml and 10 ml/100 l water were below the Codex MRL of 0.5 mg/kg.

7. A Specific Off-Label Approval application submitted by the NFU on behalf of HDC was successful and a Specific Off-Label Approval Number 0713/92 was granted on 23 June 1992.

l is Provisional only and additional data
s have been set. As BASF plc have indicated
tion to withdraw Bavistin FL any further studies
ducted using Bavistin DF.

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ANNEX I

AUTHENTICATION

Sponsors Study Ref. : Cucumbers

Our Study No. : J 7709

I, the undersigned hereby declare that this study was performed under my direction and that this report presents a true and accurate record of the results obtained. I confirm that the study was conducted in accordance with the principles of Good Laboratory Practice as defined by the United Kingdom Compliance Programme.

J.S.C. Smith

(Study Director)

6.12.91

(Date)

Your Ref:- Cucumbers

Our Ref:- J7709

QUALITY ASSURANCE AUDIT STATEMENT

STUDY TITLE:

DETERMINATION OF CARBENDAZIM RESIDUES IN CUCUMBERS

Two audits were carried out by the Quality Assurance Unit of G.C. Laboratories Ltd. The first, part of a random procedure-based inspection and the second, on a critical phase specific to this project.

	1	2
Date of Audit:-	22 October 1991	26 November 1991
Date of Report to Study Director:-	N/A	27 November 1991

The report of this study was audited by the Quality Assurance Unit, in order to assess compliance with the protocol and to confirm that it truly reflected the conduct and findings of the study.

Date of start of Audit:- 3 December 1991

Date of Report to Study Director:- 5 December 1991

Date of reply from Study Director:- 5 December 1991

Date of final agreement of Report of Study:- 5 December 1991

Signed..J.Lamond.....(J K Lamond) Date..5:12:91..

Quality Assurance Officer

GC LABORATORIES LTD
FALDO ROAD
BARTON-LE-CLAY
BEDS. MK45 4RL
TEL: LUTON (0582) 882081

ANALYTICAL REPORT

TO:— Dept. of Plant Pathology,
Government Buildings,
Lawnswood,
LEEDS
W. Yorks.
LS16 5PY

FOR THE ATTENTION OF:— Dr.G.M.McPherson

OUR REF:— J7709

YOUR REF:— Cucumbers

DATE:— 1st December 1991

ENQUIRIES TO:— Mr. J.S.C. Smith

ANALYSIS OF 21 SAMPLE(S) RECEIVED ON 22nd October 1991

Determination of Carbendazim Residues in Cucumbers

1. OBJECTIVES

To measure Carbendazim residues in the submitted samples of cucumbers.

2. SAMPLE RECEIPT

The samples were received at G.C. Laboratories Ltd. on 22nd October 1991 in a frozen condition.

The project was assigned our Project Number J7709 and each sample was individually and uniquely identified with our sample numbers from C397/91/2062 to C324/91/2082 inclusive.

All samples were put into deep freeze storage immediately after labelling and were maintained at a temperature of minus 20°C or less between receipt and analysis. Temperatures of the freezers used to store the samples were read and recorded once per day. The samples have been stored in our freezers Reference Numbers 2 and 5 from receipt until the date of this report.

3. EXPERIMENTAL PROCEDURES

The samples were analysed according to G.C. Laboratories Ltd. Protocol No. RES/91/137.

Our Ref. J7709

3(a) Analytical Method

The method used was G.C. Laboratories Method No. M349 which is based on : "Analytical Methods for Residues of Pesticides", 4th Edition (1985), Multi-Residue Method 3, Submethod 2 (Ministry of Welfare, Health & Cultural Affairs Leidschendam - Netherlands). This was adapted from the procedure described by J.E. Farrow et al (Analyst, 102, 752-758, 1977).

3(b) Analytical Standard

The Analytical Standard employed was 99% pure Carbendazim supplied by Riedel-de-Haen (Our Ref. S-1148).

4. RESULTS OF ANALYSIS

4(a) Recovery Experiments

Recovery Level(mg/kg)	Our Expt.No. J7709/	Recovery %
0.501	2	101
0.251	19	102
0.050	11	94

Our Ref. J7709

4(b) Sample Residue Results

Sample No.	Treatment	Date	Our Expt. No.J7709/	Carbendazim mg/kg
1	1 Untreated	11.9.91	18	0.02
2	1 Untreated	12.9.91	20	0.04
3	1 Untreated	13.9.91	21	0.05
4	3 Bavistin 4ml/100L	11.9.91	9	0.08
5	3 Bavistin 4ml/100L	12.9.91	12	0.19
6	3 Bavistin 4ml/100L	13.9.91	13	0.13
7	4 Bavistin 10ml/100L	11.9.91	3	0.27
8	4 Bavistin 10ml/100L	12.9.91	6	0.19
9	4 Bavistin 10ml/100L	13.9.91	7	0.19
10	Fairfield Bav.4ml/100L	11.9.91	14	0.05
11	Fairfield Bav.4ml/100L	12.9.91	8	0.04
12	Fairfield Bav.4ml/100L	13.9.91	15	0.05
13	Fairfield Untreated	11.9.91	22	0.05
14	Fairfield Untreated	12.9.91	23	0.05
15	Fairfield Untreated	13.9.91	4	0.03
16	House 9 Bav.4ml/100L	14.8.91	16	0.16
17	House 9 Bav.4ml/100L	15.8.91	5	0.30
18	House 9 Bav.4ml/100L	16.8.91	17	0.25
19	House 9 Untreated	14.8.91	10	0.01
20	House 9 Untreated	15.8.91	1	0.03
21	House 9 Untreated	16.8.91	24	0.06

Our Ref. J7709

5. OTHER INFORMATION

5(a) Copies of typical chromatograms from sample and recovery experiments, and also data from a linearity check, are attached in the Appendix.

5(b) All raw data, and a copy of the final report, will be stored in the archives of G.C. Laboratories Ltd., Barton-le-Clay, Beds. MK45 4RL

5(c) The Study was carried out at :-

G.C. Laboratories Ltd.
Faldo Road,
Barton-le-Clay,
Beds. MK45 4RL

The Analysis was commenced on 14th November 1991 and completed on 29th November 1991.

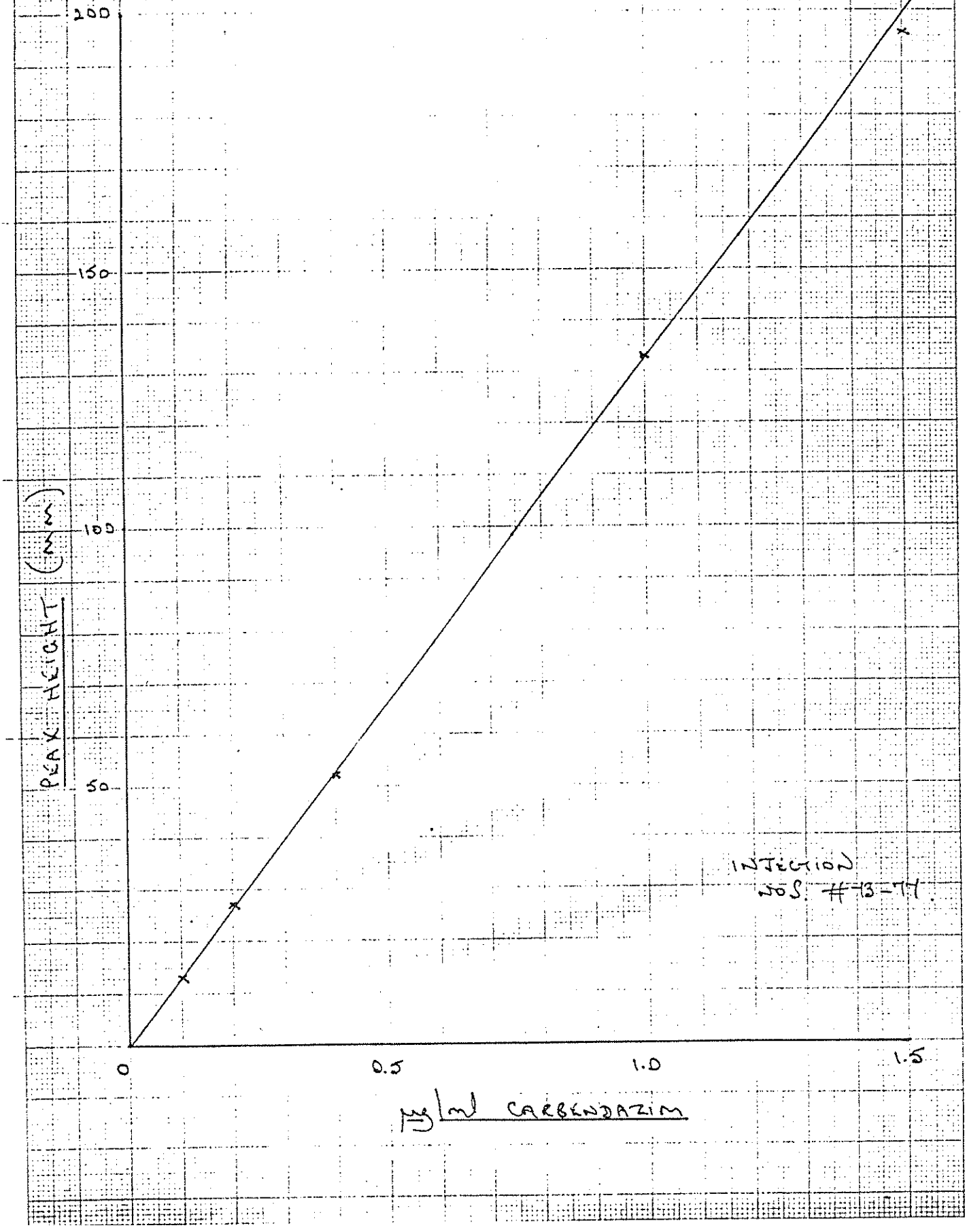
Signed P.C. Smith (J.S.C. Smith) Date 6.12.91

(Study Director)

Chartwell

DETERMINATION OF CARBENDAZIM RESIDUES IN CUCUMBERS

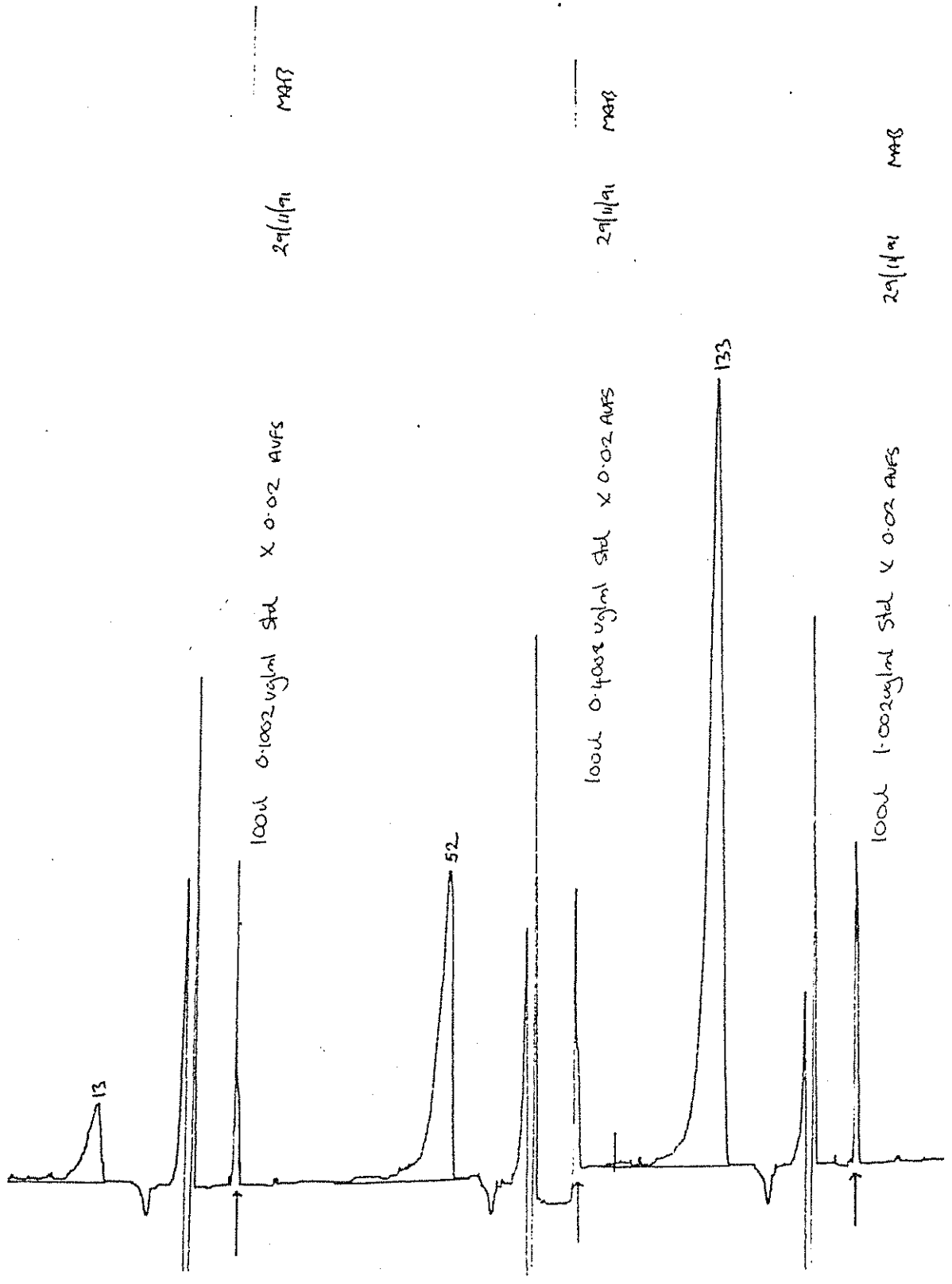
CALIBRATION DATA GRAPH 29.11.91 #4409



DETERMINATION OF CARBENDAZIM RESIDUES IN CUCUMBERS J7709

Linearity Data : 29/11/91

Standard 0.100ug/ml	Standard 0.401ug/ml	Standard 1.00ug/ml
#75	#74	#73

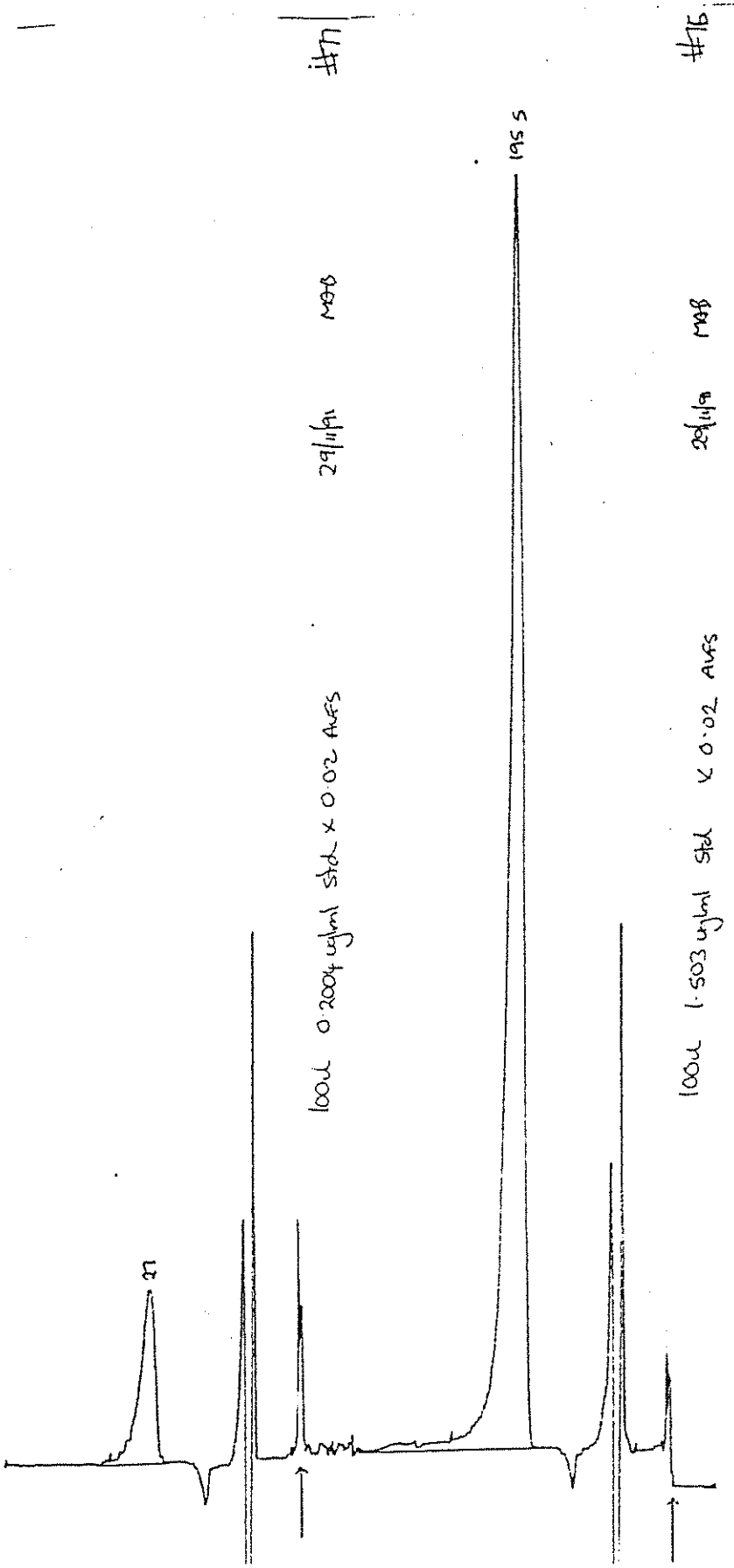


DETERMINATION OF CARBENDAZIM RESIDUES IN CUCUMBERS J7709

Linearity Data : 29/11/91

Standard
0.200ug/ml

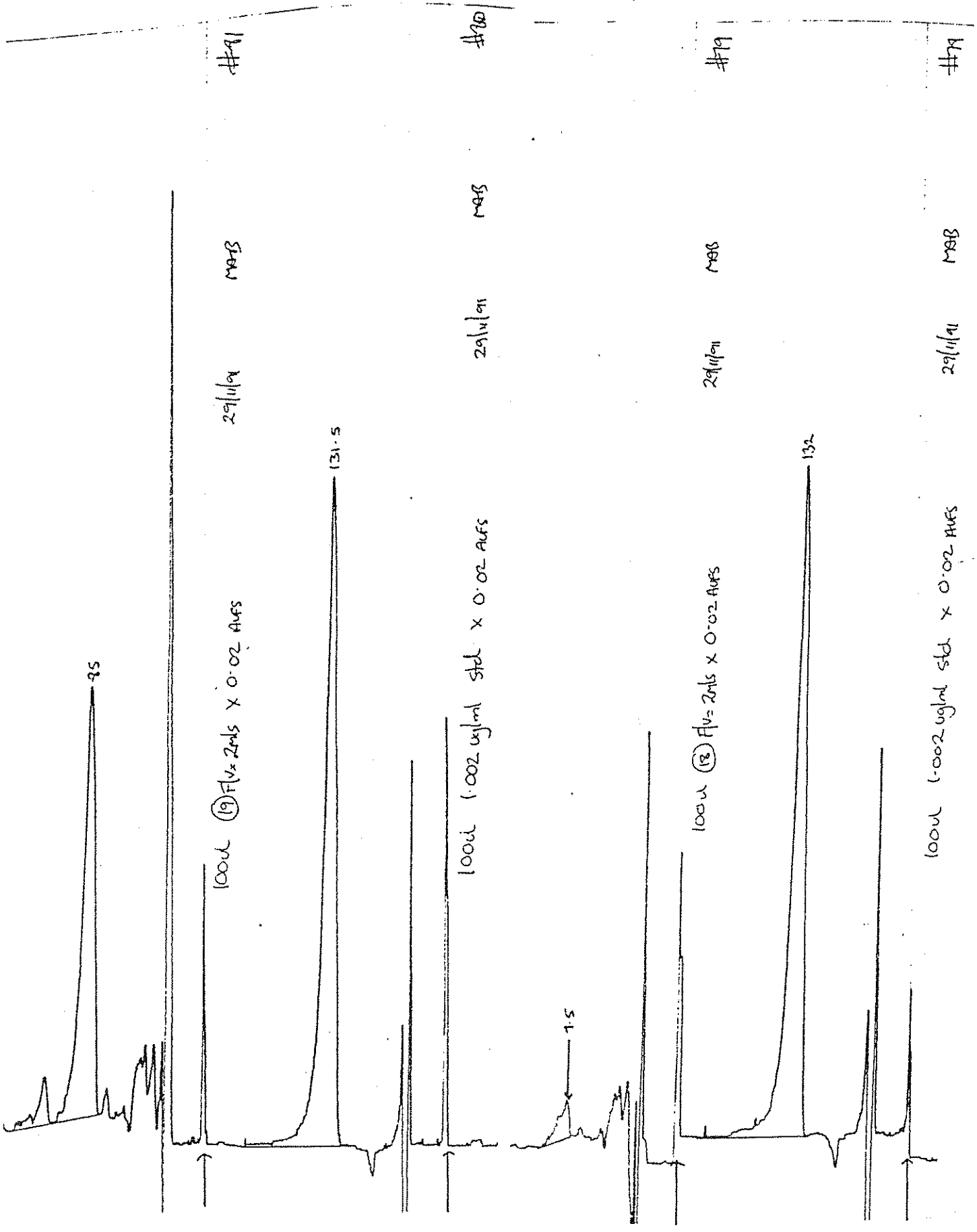
Standard
1.50ug/ml



G. C. LABORATORIES LTD.

DETERMINATION OF CARBENDAZIM RESIDUES IN CUCUMBERS J7709

Recovery Expt. 0.25ImG/kg J7709/19	Standard 1.00ug/ml	Untreated Sample No.1 J7709/18	Standard 1.00ug/ml

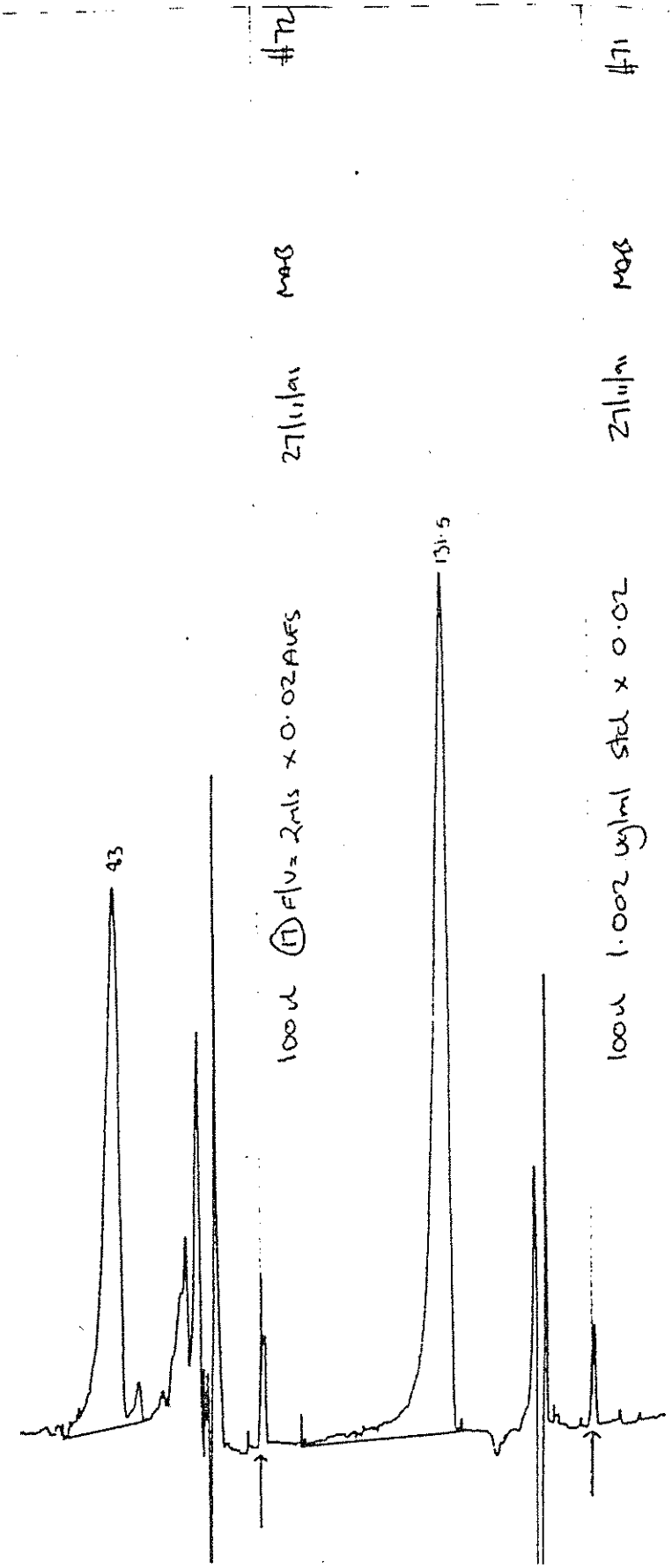


G. C. LABORATORIES LTD.

DETERMINATION OF CARBENDAZIM RESIDUES IN CUCUMBERS J7709

Treated
Sample No.18
J7709/17

Standard
1.00ug/ml



ANNEX II

NOTICE OF APPROVAL NO.0718/92

CONTROL OF PESTICIDES REGULATIONS 1986
(S.I. 1986 NO. 1510):
APPROVAL FOR OFF-LABEL USE OF AN APPROVED PESTICIDE PRODUCT

This approval provides for the use of the product named below in respect of crops and situations, other than those included on the product label. Such "off-label use", as it is known, is at all times done at the user's choosing, and the commercial risk is entirely his or hers.

The conditions below are statutory. They must be complied with when the off-label use occurs. Failure to abide by the conditions of approval may constitute a breach of that approval, and a contravention of the Control of Pesticides Regulations 1986. The conditions shown below supersede any on the label which would otherwise apply.

Level and scope: In exercise of the powers conferred by regulation 5 of the Control of Pesticides Regulations 1986 (SI 1986/1510) and of all other powers enabling them in that behalf, the Minister of Agriculture, Fisheries and Food and the Secretary of State, hereby jointly give provisional approval for the advertisement, sale, supply, storage and use of

Product name: Bavistin FL containing

Active ingredient: 500g/l carbendazim

Marketed by: BASF UK Limited under MAFF NO. 00218 subject to the conditions relating to off-label use set out below:

Date of issue: 23 June 1992

Data submission deadline: 30 June 1994

Date of expiry: 30 June 1996
subject to the continuing approval of MAFF 00218
see note*

C713/92

D-1

Ministry of Agriculture, Fisheries and Food
Pesticides Safety Division
Rothamsted. Harpenden. Herts AL5 2SS
Telephone: 0582-462100 Ext. GTN: 3091 Telex: 827517 Fax: 0582-462919

Mr R Turner
National Farmers Union
Agriculture House
KNIGHTSBRIDGE
London
SW1X 7NJ

Your ref:

Our ref: MAFF 00218

Date: 8 June 1992

Dear Mr Turner

COP 92/00182: PROVISIONAL OFF-LABEL APPROVAL FOR THE USE OF 'BAVISTIN FL' ON CUCUMBERS GROWN HYDROPONICALLY - OFF-LABEL EMERGENCY PROCEDURE.

1. I am writing to inform you that the Data Evaluation Unit of Pesticides Safety Division has now completed its evaluation of the application for approval of the following product:

   Product name: Bavistin FL (contains carbendazim)

   MAFF No: 00218

   Marketing company: BASF UK Limited

2. The evaluation related to your company's request for off-label approval for the use of 'Bavistin FL' on cucumbers grown hydroponically. Application dated 27 February 1992.

3. The Unit has considered the application described in paragraphs 1 and 2 above and by way of this letter has recommended that Ministers give provisional approval for four years as set out in the attached draft Notice of Approval; subject to the provision of the outstanding data listed in paragraph 5 by the data submission deadline.

   This approval is recommended without prejudice to the outcome of the review of carbendazim.

4. THIS LETTER DOES NOT CONSTITUTE AN APPROVAL unless it includes a Notice of Approval duly signed by the authorising Department.

5. To support a future application for full approval for the off-label use of 'Bavistin FL' on cucumbers grown hydroponically at a rate of 4 mls product/100 litres water, all the following data/information MUST BE SUBMITTED BY THE DATA SUBMISSION DEADLINE OF two years from the date of issue of the approval.

   (i) Covering letter
   (ii) Copies of relevant correspondence and the previous Notice(s) of Approval
   (iii) One seasons residue data (3-4 trials) for cucumbers grown hydroponically.

   See 'Guidelines for generating and reporting crop residue data' published in the Pesticides Register, issue 12 December 1991 for guidance..

   Please note

   To support a future application for provisional approval for the off-label use of 'Bavistin FL' on cucumbers grown hydroponically at a rate of 10 mls product/100 litres water we require the following data:

   One seasons residue data (2-3 trials) for cucumbers grown hydroponically.

   nb. to support an application for full off-label approval we require the following data:

   One seasons residue data (5-7 trials) for cucumbers grown hydroponically.

6. PLEASE NOTE that if all the data/information are submitted by the deadline, this approval will continue for a further two years or until the data are evaluated and a decision taken, whichever is the sooner. If the deadline is not met, the approval for the unsupported use(s) will be revoked immediately. All off-label uses are subject to the continuing approval of the product on which the approval for the off-label use was based.

Other specific restrictions:

(1) This product must only be applied if the terms of this approval, the product label and/or leaflet and any additional guidance on off-label approvals have first been read and understood.

(2) The maximum concentration must not exceed 4ml product/100 litres water.

Signed .....
~~(Authorised~~ signatory)

Date 23 June 1992

Application Reference Number: COP 92/00182

THIS NOTICE OF APPROVAL IS NUMBER 0713 of 1992

ADVISORY INFORMATION

This approval relates to the use of 'Bavistin FL' for the control of root diseases of hydroponically grown cucumbers. Application should be made as a drench treatment at a rate of 500ml diluted solution/plant.

Note: Continuing approval of this off-label use is also subject to receipt of data by the data submission deadline. If the required data are not submitted, approval for this use will cease on that date. It is the users' responsibility to ensure that the approval is still extant if they intend to make use of it after that date. Details of the data requirements specific to this approval are as follows:

One seasons residue data (3-4 trials) for cucumbers grown hydroponically.
See 'Guidelines for generating and reporting crop residue data'.

Futher information is given in Appendix 6 and associated Working Documents of 'Data Requirements for Approval under the Control of Pesticides Regulations 1986'.

Advice can be obtained from Branch H, Pesticides Safety Division.

7. If it becomes apparent that any of the required data/information may not be available for submission before the data deadline, the Technical Secretariat should be informed in writing as soon as possible, with a detailed explanation of the problem and assurances as to when the data will be submitted. Where data are not submitted by the deadline or where any other failure to comply with requests for information takes place, no re-submission of application for the relevant specific off-label approval will be accepted until after the expiry of a two year period following the data submission deadline.

8 IMPORTANT NOTES:

(i) All approvals remain subject to immediate revocation, suspension or amendment of the conditions of approval at any time if safety considerations so demand.

(ii) Approval holders must submit immediately any data which show an adverse effect on humans, animals, crops or the environment.

9. Copies of this letter and draft Notice of Approval are being sent to the Department of Employment and the Health and Safety Executive.

Yours sincerely

pp S Miller
Data Evaluation Unit

cc Registration Officer
BASF UK Ltd

ANNEX III

Field of use: ONLY AS AN HORTICULTURAL FUNGICIDE

Crops: Cucumber grown hydroponically under protection

Maximum individual dose: see "Other specific restrictions"

Maximum number of treatments: 8 per crop

Latest time of application: Two days before harvest

Environmental protection: Since this product is harmful to fish or aquatic life, surface waters or ditches must not be contaminated with chemical or used containers.

Ministry of Agriculture, Fisheries and Food
Pesticides Safety Division
Rothamsted, Harpenden, Herts AL5 2SS
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Dr G M McPherson
Horticulture Research International
Stockbridge House
Cawood
Selby
North Yorkshire
YO8 0TZ

Your ref : HRI / 011

Our ref : FEH 1271D / MAFF 00218D

Date : 29 May 1992

Dear Dr McPherson

RE : COP 92 / 00182 - OFF LABEL APPROVAL FOR THE USE OF 'BAVISTIN FL' ON CUCUMBERS GROWN HYDROPONICALLY.

I refer to your letter of 10 April 1992, in which you enquired whether the application for Off - Label approval for the use of 'Bavistin Fl' (MAFF 00218) on cucumbers grown hydroponically can be switched to 'Bavistin DF' (MAFF 03848), and if not what additional residue data would be required for 'Bavistin DF' (MAFF 03848), to ensure the availability of the active ingredient to growers of substrate - grown cucumbers in the UK.

The Unit has considered your request, and decided that if similar active ingredient solubilities can be demonstrated for both products i.e. 'Bavistin FL' (MAFF 00218) and 'Bavistin DF' (MAFF 03848), then the Unit would be prepared to consider an application for Off - Label approval for 'Bavistin DF' (MAFF 03848), (i.e. with the proposed use being the same as that currently proposed for 'Bavistin FL' (MAFF 00218)).

Please note - Any outstanding data requirements set for 'Bavistin FL' (MAFF 00218) would also apply to 'Bavistin DF' (MAFF 03848).

If you have any future questions regarding the data requirements for 'Bavistin FL' (MAFF 00218) / 'Bavistin DF' (MAFF 03848) please contact L. Holmes (Branch K), Pesticides Safety Division.

Yours sincerely

Steven Miller

S. Miller
Data Evaluation Unit

cc Mr R Turner, NFU

Ministry of Agriculture, Fisheries and Food
Pesticides Safety Division
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Horticulture Research International
Stockbridge House
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YO8 0TZ

Your ref : HRI / 011

Our ref : FEH 1271D / MAFF 00218D

Date : 29 September 1992

Dear Dr McPherson

RE : COP 92/00182 - OFF LABEL APPROVAL FOR THE USE OF 'BAVISTIN FL' ON CUCUMBERS GROWN HYDROPONICALLY.

Further to my letter to you of 29 May 1992 (copy enclosed), regarding your request of 10 April 1992, in which you enquired whether the application for Off-Label approval for the use of 'Bavistin Fl' (MAFF 00218) on cucumbers grown hydroponically can be switched to 'Bavistin DF' (MAFF 03848),the Unit has received a letter from BASF plc in which they addressed the question specified in my letter of 29 May 1992, ie. 'if similar active ingredient solubilities can be demonstrated for both products i.e. 'Bavistin FL' (MAFF 00218) and 'Bavistin DF' (MAFF 03848), then the Unit would be prepared to consider an application for Off-Label approval for 'Bavistin DF' (MAFF 03848), (i.e. with the proposed use being the same as that currently approved for 'Bavistin FL' (MAFF 00218)'.

The Unit has considered the information specified in BASF's plc letter and decided to accept their arguement for extrapolation of data from 'Bavistin FL' (MAFF 00218) to 'Bavistin DF' (MAFF 03848) for use on cucumbers grown hydroponically, based on the solubility of the active ingredient.

The Unit would therefore be prepared to consider an application for Off-label approval for 'Bavistin DF' (MAFF 03848), (ie. with the proposed use being the same as that currently approved for 'Bavistin FL' (MAFF 00218), under COP 92/00182, Notice of Approval Number 0713/92), Nb. A copy of this letter should be included with any such application.

Please note - Any outstanding data requirements set for 'Bavistin FL' (MAFF 00218) also apply to 'Bavistin DF' (MAFF 03848).

If you have any future questions regarding the data requirements for 'Bavistin FL' (MAFF 00218) / 'Bavistin DF' (MAFF 03848) please contact L. Holmes (Branch K), Pesticides Safety Division.

Yours sincerely

Steven Miller

S. Miller
Data Evaluation Unit

cc Mr R Turner, NFU
Mr P. R. Mathews, BASF plc