European Journal of Plant Pathology **105:** 783–790, 1999.

The influence of fungicide sprays on infection of apple cv. Bramley's seedling by *Nectria galligena*

Louise R. Cooke
Applied Plant Science, Department of Agriculture for Northern Ireland and The Queen's University of Belfast, Newforge Lane, Belfast, BT9 5PX, UK (Fax: +44 1232 668375)

Accepted 30 August 1999

Key words: apple canker, fungicides, disease control

Abstract

In 1990, a long-term trial was initiated by planting young apple trees, cv. Bramley's Seedling, inoculated, at single sites in the leader shoots, with *Nectria galligena*. The effect of spring–summer and autumn fungicide spray programmes, applied during 1991–1993, on the development of new cankers was assessed up to May 1994. Spring–summer fungicide programmes, applied as for the control of apple scab, reduced numbers of new cankers by between 65% and 76% compared with the untreated control. Sterol demethylation inhibiting fungicides (hexaconazole, myclobutanil, penconazole) had a similar effect on canker to dithianon. Autumn application of copper oxychloride at 5% and 50% leaf-fall further reduced numbers of new cankers. The combination of a spring–summer benzimidazole (carbendazim or thiophanate-methyl) with a scab fungicide (dithianon) and autumn copper oxychloride did not improve canker control compared with applying copper oxychloride after spring–summer myclobutanil+mancozeb. The percentage of fruit which developed rots in long-term storage was decreased by spring–summer benzimidazole application, but not by other fungicide programmes even those which achieved similar levels of canker control.

Introduction

European canker, caused by *Nectria galligena*, is an important disease of apples in the UK and is particularly prevalent in wetter regions such as Northern Ireland, where the culinary cultivar Bramley's Seedling is the main cultivar grown commercially.

Until the last few years, it has been generally accepted that *N. galligena* spreads from tree to tree via air-borne and splash-dispersed spores. It was considered to require some form of wound to facilitate its entry to the tree, infection occurring during the spring–summer period through bud-scale scars and other damage points, and in the autumn through leaf scars (Swinburne, 1971). However, it has been suggested recently that symptomless, systemic infection may occur, particularly in some dessert apple cultivars (Lovelidge, 1995). Nonetheless, the greater incidence of canker in areas of high rainfall and the increased infection which develops following wet years provide circumstantial evidence in favour of a role for rain in the dispersal of *N. galligena*.

Fungicide sprays have proved effective in preventing canker. Application of copper- or phenylmercury-based formulations at leaf-fall reduced leaf-scar infections (e.g. Byrde et al., 1965; Bennett, 1971). In Northern Ireland, Swinburne et al. (1975) showed that, in Bramley's Seedling, spring–summer fungicide programmes caused a greater reduction in canker numbers than autumn fungicides alone. This was further investigated in two trials in Northern Ireland between 1983 and 1990 (Cooke et al., 1993). These were initiated in order to find alternative treatments to replace autumn application of phenylmercury nitrate (PMN), then permitted under an EC derogation (although subsequently banned), and to evaluate the effect on canker of the newer spring–summer fungicides, particularly those of the sterol demethylation inhibiting (DMI) group.

These trials showed that copper oxychloride was a reasonable alternative to PMN as an autumn treatment. Trees treated with dodine pre-blossom and dithianon post-blossom tended to develop less canker than those receiving a programme of spring–summer DMI fungicides (myclobutanil, penconazole), but fewest cankers occurred when the benzimidazole fungicide carbendazim was used throughout the spring–summer programme. However, it was not possible to assess the overall impact of these programmes on canker development as no untreated controls were included in these trials.

A long-term trial was therefore run from 1991 to 1994 to allow the absolute effect of a range of DMI programmes to be assessed by comparing canker development on treated trees with that on trees receiving no fungicide sprays. The leader shoot of each tree was inoculated with *N. galligena* to provide an initial inoculum source. The impact of combining such programmes with autumn copper oxychloride was also evaluated. A preliminary report of the effect of fungicides on the total number of wood cankers, compared with those of the earlier trials, has been published (Cooke and Watters, 1994).

Materials and methods

Trees

Apple trees, cv. Bramley's Seedling on M26 rootstock, were grown at the Northern Ireland Horticultural and Plant Breeding Station, Loughgall, Co. Armagh. They were planted in January 1990 in four fully randomised blocks with ten treatments per block in five-tree plots. Nylon mesh screens 2 m high were erected between plots to limit dispersal of sprays and inoculum. The leader shoot on each tree was inoculated (Swinburne et al., 1975) using a 1989 Northern Ireland isolate (N1/89) of *N. galligena*. Two months later, all trees were checked for the presence of active lesions around inoculation points.

Treatments

Fungicide sprays were applied at high volume in 1000 l ha^{-1} using a portable Fox Motori Wagon sprayer. During 1990, all trees received a routine dithianon spray programme. Treatments were applied from spring 1991 until autumn 1993. During the spring–summer, treatments were applied as for the control of apple scab at ca. 10-day intervals (9 applications April–July 1991, 11 applications April–July 1992, 12 applications April–August 1993); in the autumn, where appropriate, two sprays were applied at 5% and 50% leaf-fall (22 November, 6 December 1991; 3, 16 November 1992; 2, 9 November 1993). Details of fungicide formulations are given in Table 1 and spray programmes in Table 2.

Assessments

Numbers of new cankers were recorded in May 1991 (pre-treatment assessment), March and May 1992, March and May 1993 and May 1994. For each canker, the infection site was recorded using the designations of Swinburne et al. (1975) viz. crotch – in the crotch between branches, basal – in the basal five leaf scars, distal – in the leaf scars above the basal five. Wood age was also recorded for basal and distal cankers. Each canker was marked with waterproof paint when assessed, so that on each occasion only new cankers were recorded.

Table 1. Details of fungicide formulations and application rates

Fungicide	Proprietary name	Manufacturer	Application rate (g ha^{-1})
Carbendazim	Bavistin	BASF	500
Copper oxychloride	Cuprokylt	Universal	5000
Dithianon	Delan-Col	Zeneca	840
Hexaconazole	Anvil	Zeneca	50
Mancozeb	Karamate	Rohm & Haas	1680
Myclobutanil	Systhane	Rohm & Haas	66
Penconazole	Topas	Novartis	50
Penconazole + dithianon	Topas D	Novartis	50 + 500
Thiophanate-methyl	Cercobin	Rhône-Poulenc	1000

Table 2. Details of fungicide programmes

Treatment number	Spring–summer		Autumn
	Pre-blossom	Post-blossom	
1	None	None	None
2	Dithianon	Dithianon	None
3	Hexaconazole	Hexaconazole	None
4	Penconazole	Penconazole	None
5	Penconazole + dithianon	Penconazole + dithianon	None
6	Myclobutanil	Myclobutanil	None
7	Myclobutanil	Myclobutanil + mancozeb	None
8	Myclobutanil	Myclobutanil + mancozeb	Copper oxychloride
9	Dithianon + thiophanate-methyl	Dithianon + thiophanate-methyl	Copper oxychloride
10	Dithianon + carbendazim	Dithianon + carbendazim	Copper oxychloride

In 1992 and 1993, the fruit from each plot was harvested and stored in a controlled atmosphere store. The following February (1993) or March (1994), the total numbers of rotted fruit from each plot and the number of fruit with symptoms of *Nectria* eyerot were recorded.

Analyses

All results were subjected to analyses of variance. The assessments of rotted fruit were expressed as percentages and analysed after arcsine transformation.

Results

Development of new cankers over time

All inoculation sites developed canker lesions. Analysis of the May 1991 assessment of cankers which had resulted from infection before application of treatments showed a lack of any significant difference between treatment plots. No benefit was found from using these initial counts as a co-variate in the analyses.

New cankers were counted twice a year in 1992 and 1993. In the untreated trees, the numbers of new cankers at each of these assessments varied between 36 and 60 per plot (Figure 1). It was not possible to make an early spring assessment in 1994, but in May 1994, it was found that a very large number of new cankers had developed, ca. 400 per plot in the untreated control. The year 1993 had a greater total rainfall (899 mm) than 1991 or 1992 (711 and 807 mm, respectively) and the period December 1993–March 1994 was unusually wet (rainfall 165% of 30-year mean), which would have encouraged infection.

More canker developed in this trial than in two similar trials at Loughgall during the 1980s. In these earlier trials (which did not include untreated controls), the cumulative totals of new cankers which developed on six-tree plots receiving the standard programmes (dodine and dithianon) were 58 (1983–1986 trial) and 62 (1988–1990 trial) compared with 148 per five-tree dithianon-treated plot in the present trial (1991–1994). The annual rainfall totals during the early 1990s were no greater than in the 1980s, so it is unlikely that the development of canker was influenced by the macroclimate. However, the present trial was planted in a more sheltered and lower-lying area so that a more humid microclimate may have encouraged spread.

Effect of fungicide treatment on canker development

Compared with the untreated control, fungicide treatments reduced canker by between 65% and 90% (Figure 1). The standard spring–summer dithianon programme reduced canker by 75%. There was a significant effect of treatment on total numbers of new cankers and cumulative totals at all assessments except March 1992.

The three programmes containing DMI fungicides alone with no autumn treatment (treatments 3, 4 and 6) reduced canker numbers to a level similar to that with dithianon. Myclobutanil tended to be slightly

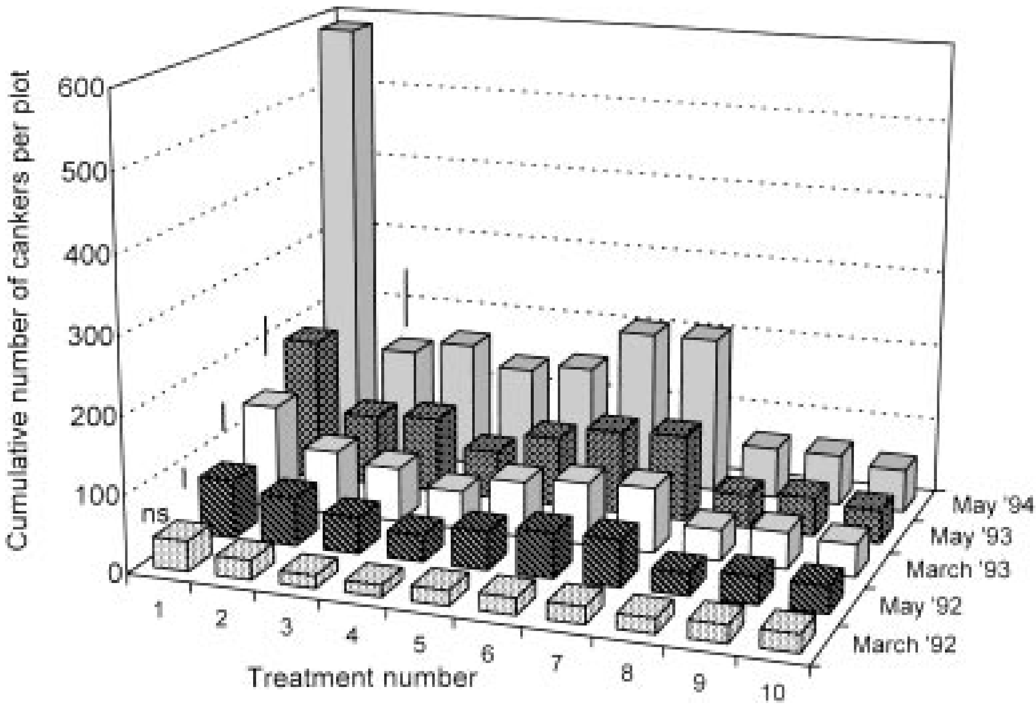

Figure 1. Total numbers of cankers at each assessment. Details of treatments are given in Tables 1 and 2. Vertical bars indicate LSD values ($P < 0.05$).

less effective and penconazole slightly more effective than hexaconazole in reducing canker numbers, but the differences were not significant ($P > 0.05$). Addition of dithianon to penconazole (treatment 5) or mancozeb to myclobutanil (treatment 7) did not affect canker incidence (Figure 1) as compared with the DMI fungicides alone.

The greatest reductions in canker numbers were achieved by the three programmes which included two autumn applications of copper oxychloride (treatments 8, 9 and 10). The addition of autumn copper oxychloride to the myclobutanil + mancozeb programme (treatment 8) decreased canker incidence by 68% compared with the same programme without an autumn fungicide (treatment 7). Trees treated with the two programmes including a spring–summer benzimidazole fungicide and autumn copper oxychloride (treatments 9 and 10) developed similar numbers of new cankers to those which received the myclobutanil/myclobutanil + mancozeb/copper oxychloride programme (treatment 8).

Effect of wood age on canker development

Over the whole period of the trial, the majority of new cankers were located on one-year-old wood, about half as many on two-year-old wood and relatively few on older wood (Figure 2). The predominance of cankers on one-year-old wood was particularly marked when assessment was made in May 1994 and, as these were associated with leaf scars, it seemed likely that they had been initiated at leaf-fall in autumn 1993. The ranking order of fungicide treatments with respect to canker numbers was very similar on wood of different ages. However, as wood age increased, the effect of treatment decreased from very highly significant on one-year-old wood ($P < 0.001$), to highly significant on two-year-old wood ($P < 0.006$), significant on three-year-old wood ($P < 0.014$) and non-significant on four-year-old wood ($P > 0.05$).

Approximately equal numbers of cankers developed in basal and distal positions on branches, with a smaller

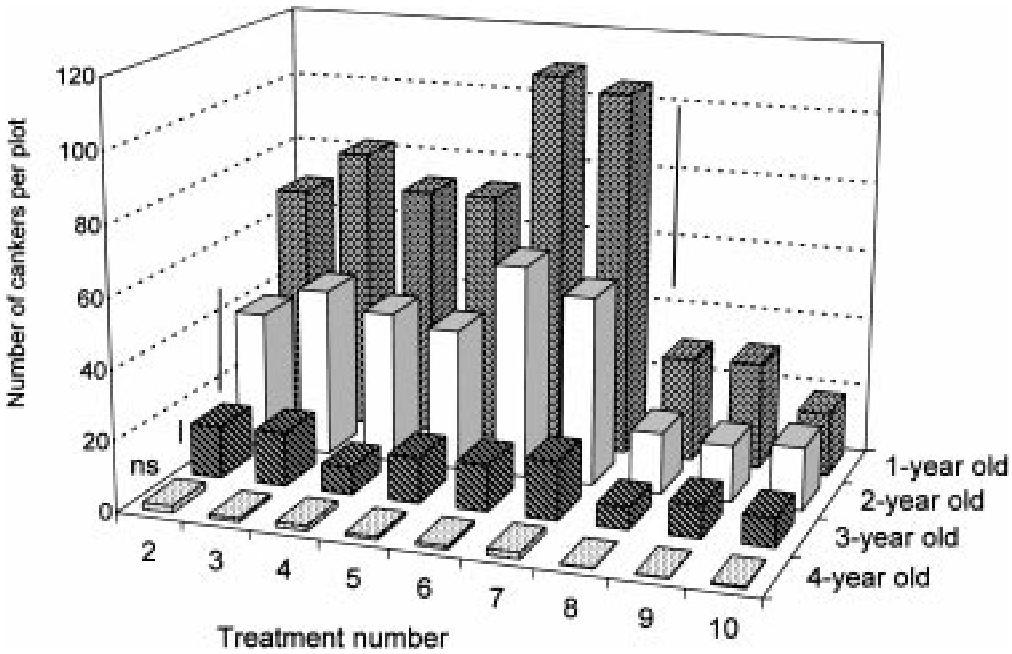

Figure 2. Canker development on wood of different ages (excluding the untreated control). Details of treatments are given in Tables 1 and 2. Vertical bars indicate LSD values ($P < 0.05$).

proportion in crotches (Figure 3). Although the ranking order of treatments was the same for basal, distal and crotch cankers, treatment had less effect on the incidence of crotch cankers ($P < 0.002$ cf. $P < 0.001$ for basal and distal canker totals).

Effect of treatment on fruit rotting

When the apples were removed from storage in March 1993, there were significant differences between treatments in the percentage of *Nectria* rotted fruit, but not in the percentage of all rotted fruit (Table 3). The apples from the plots which received a spring–summer benzimidazole (carbendazim or thiophanate-methyl; treatments 9 and 10) had no *Nectria* rots, this was significantly less than those from the myclobutanil/myclobutanil + mancozeb/copper oxychloride programme (treatment 8), although the canker incidence was similar for these three programmes.

In February 1994, treatment significantly affected the percentage of all rotted fruit, but very few *Nectria*-rotted fruit were identified (Table 3). The apples from plots which received the two spring–summer benzimidazole programmes (treatments 9 and 10) developed significantly fewer rots than those from any other treatment except hexaconazole (treatment 3).

Discussion

The results of this trial confirmed the conclusions of Swinburne et al. (1975) that spring–summer fungicide applications, intended to control apple scab, also cause substantial reductions in canker. There was no evidence that the DMI fungicides used for apple scab control were less effective in preventing canker than older protectant fungicides. Programmes including autumn application of copper oxychloride achieved the greatest reductions in canker numbers. This is in agreement with the results of two trials at Loughgall during the 1980s (Cooke et al., 1993) where copper oxychloride was found to be the best available alternative to autumn phenylmercury nitrate.

In a previous trial (Cooke et al., 1993), a spring–summer benzimidazole programme proved much more effective than a spring–summer myclobutanil/myclobutanil + mancozeb programme in preventing new cankers when no autumn treatments were applied.

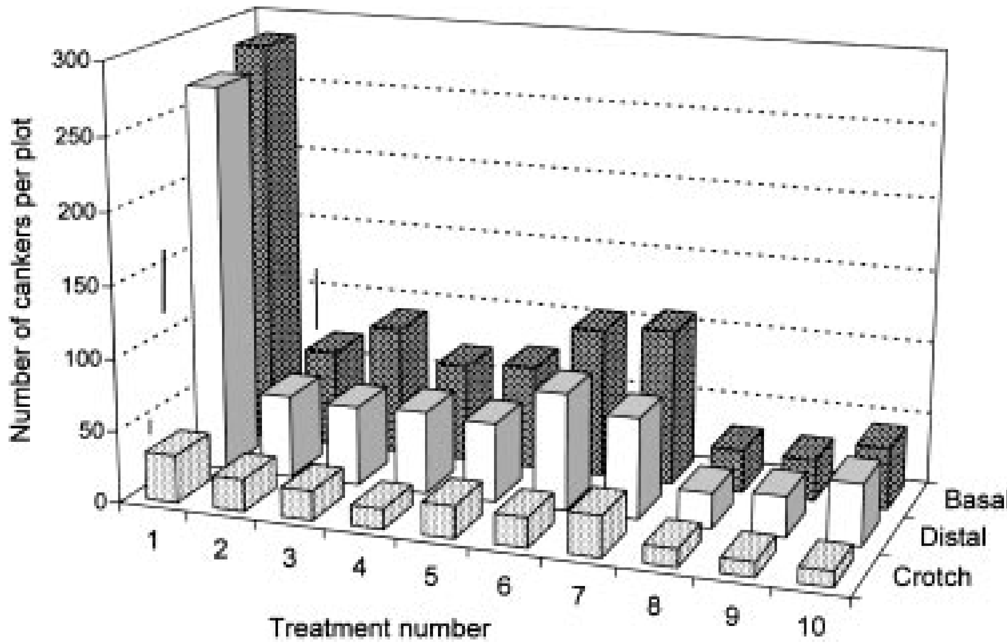

Figure 3. Canker development at different infection sites on trees (for definitions see Materials and methods.) Details at treatments are given in Tables 1 and 2. Vertical bars indicate LSD values ($P < 0.05$).

Table 3. Assessment of rotting of apples after storage, 1993 and 1994

Treatment number[b]	Rotted fruit (arcsin %)[a]			
	March 1993		February 1994	
	Total	*Nectria*	Total	*Nectria*
1	23.2a	8.6ab	22.1a	10.9a
2	30.1a	5.9ab	25.3a	2.9a
3	15.8a	11.3a	19.1ab	2.3a
4	27.1a	12.4a	26.1a	3.2a
5	20.6a	14.3a	23.8a	3.8a
6	24.7a	7.7ab	23.5a	5.2a
7	19.8a	5.1ab	24.2a	0.0a
8	20.2a	9.4a	22.7a	4.9a
9	13.2a	0.0b	6.1c	0.0a
10	14.7a	0.0b	12.3bc	2.3a
SE (27 DF)	3.86	3.19	2.87	2.35
Significance[c]	n.s.	*	***	n.s.

[a]Letters in columns followed by the same letter do not differ significantly ($P > 0.05$).
[b]For details of treatments, see Tables 1 and 2.
[c]n.s. = $P > 0.05$, $^{*}P < 0.05$, $^{***}P < 0.001$.

In the present trial, the two programmes including spring–summer benzimidazole fungicides with autumn copper oxychloride were no more effective than the myclobutanil/myclobutanil + mancozeb/copper oxychloride programme. This would suggest that the effect of using benzimidazole fungicides in the spring–summer is not additive to the use of an autumn fungicide in preventing formation of new cankers. The spring–summer benzimidazole programmes without an autumn fungicide, which would have been required to test this possibility, were not included in the present trial. Benzimidazoles are known to suppress sporulation of the pathogen for prolonged periods (Swinburne et al., 1975) and may thus prevent both spring and autumn infection by *N. galligena* without the need for an additional autumn treatment.

The antisporulant effect of the benzimidazole fungicides may also have been responsible for the reduction in fruit rotting which occurred with the carbendazim and thiophanate-methyl programmes. This was not simply due to decreased inoculum resulting from control of canker, since trees treated with myclobutanil + mancozeb + autumn copper oxychloride had similar numbers of cankers, but more of the harvested fruit developed rots. The benzimidazoles also reduced the incidence of fungal rots other than those caused by *N. galligena*. This was not unexpected since they are active against most higher fungi and the majority of non-*Nectria* rots were caused by the Ascomycete pathogens

Penicillium expansum, *Sclerotinia fructigena* and *Gloeosporium* spp.

Benzimidazole and DMI fungicides are systemic, however, up-take into woody tissue from foliar fungicide sprays is extremely limited and insufficient to produce a fungicidal dose, since their translocation is almost exclusively acropetal (Crowdy, 1977). Fungicide sprays are thus unlikely to kill the pathogen within established cankers or to inhibit possible systemic spread via the xylem. Recently, Xu and Butt (1996) reported that when curative fungicide sprays were applied to potted apple trees, they were relatively ineffective in preventing canker development at pruning cuts 48 or 36 h after inoculation with *N. galligena*. Hence, the dramatic decrease in canker numbers on one-year-old wood following fungicide treatments, compared with those on untreated trees, would imply an external source of inoculum rather than the development of cankers from systemic infections. Since the cankers on the one-year-old wood were associated with leaf scars, it seems likely that they had been initiated by dispersal of conidia from existing cankers at leaf-fall in autumn 1993. If, however, symptomless systemic infection by *N. galligena* has a role initiating canker in Bramley's Seedling, as it appears to do in some dessert cultivars, then cankers originating in this way would not be prevented by fungicide sprays.

Thus use of an effective scab control programme based on DMI and/or non-systemic fungicides such as dithianon should largely prevent canker from becoming established in an orchard as long as the major source of inoculum is external. Supplementing this with two autumn applications of copper oxychloride is worthwhile in wet areas such as Northern Ireland, where it can substantially reduce leaf-scar infection. Fungicide spray treatments cannot, however, eradicate existing infections, so if trees are already visibly infected, the programme must be supplemented by cutting out and removing cankers, treating wounds with an effective canker paint (Cooke and McCracken, 1988.). In addition, spring–summer application of benzimidazole fungicides may be worthwhile for one or two seasons where canker is a major problem within an orchard, to reduce new infections and, particularly, fruit rotting. However, benzimidazoles alone cannot be relied on to control scab, because of the risk of selecting benzimidazole-resistant strains of *Venturia inaequalis* (Gilpatrick, 1982). They must therefore be supplemented by other scab fungicides, making this an expensive treatment. Prolonged use of benzimidazole fungicides in orchards is also undesirable because they have adverse effects on earthworms (Stringer and Wright, 1976).

Two new groups of fungicides are now available for the control of apple scab. The anilinopyrimidine pyrimethanil and the strobilurin kresoxim-methyl have recently been approved for application to top fruit in the UK and several other European countries. There have been no published reports of their effects on apple canker, but in view of their spectra of activity, it seems likely that they will reduce infection in a similar way to other scab fungicides.

Acknowledgements

I am very grateful to Mr. B.S. Watters, formerly head of the Top Fruit Section of the Northern Ireland Horticultural and Plant Breeding Station (NIHPBS), Loughgall for his help and co-operation throughout this work and to the staff of NIHPBS who provided invaluable technical assistance. I also thank Dr. S. Watson, Biometrics Division, for her advice on the trial design and analyses and Dr. Averil Brown, The Queen's University of Belfast, for her helpful and constructive comments on the manuscript. The following companies kindly donated some of the fungicides used in the trials; Novartis, Rohm & Haas, Universal Crop Protection and Zeneca.

References

Bennett M (1971) Comparison of a copper and mercury spray programme against apple canker (*Nectria galligena* Bres). Plant Pathology 20: 99–105

Byrde RJW, Evans SG and Rennison RW (1965) The control of apple canker in two Somerset orchards by a copper-spray programme. Plant Pathology 14: 143–149

Cooke LR and McCracken AR (1988) Control of apple canker using fungicidal paints and gels. Annals of Applied Biology 113: 279–285

Cooke LR and Watters BS (1994) The effect of fungicide sprays on the incidence of apple canker (*Nectria galligena*) in cv. Bramley's Seedling. Brighton Crop Protection Conference – Pests and Diseases 2: 779–784

Cooke LR, Watters, BS and Brown AE (1993) The effect of fungicide sprays on the incidence of apple canker (*Nectria galligena*) in Bramley's Seedling. Plant Pathology 42: 432–442

Crowdy SH (1977) Translocation. In: Marsh RW (ed) Systemic Fungicides. 2nd edition. (pp 92–114) Longman, London

Gilpatrick JD (1982) Case study 2: Venturia of pome fruits and Monilinia of stone fruits. In: Dekker J, Georgopoulos SG (eds) Fungicide Resistance in Crop Protection. (pp 195–206) Pudoc, Wageningen

Lovelidge, B (1995) Solving the apple canker mystery. Grower 123: 23–25

Stringer A and Wright MA (1976) The toxicity of benomyl and some related 2-substituted benzimidazoles to the earthworm *Lumbricus terrestris*. Pesticide Science 7: 459–464

Swinburne TR (1971) The seasonal release of spores of *Nectria galligena* from apple canker in Northern Ireland. Annals of Applied Biology 69: 97–104

Swinburne TR, Cartwright J, Flack NJ and Brown AE (1975) The control of apple canker (*Nectria galligena*) in a young orchard with established infections. Annals of Applied Biology 81: 61–73

Xu X and Butt DJ (1996) Tests of fungicides for post-germination activity against *Nectria galligena*, causal agent of canker and fruit rot of apple. Crop Protection 15: 513–519