



Agriculture & Horticulture
DEVELOPMENT BOARD



Grower Summary

SF 98

Sustainable management of
Mucor and Rhizopus in
strawberry

Final 2011

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Project Leader:	Dr Angela M Berrie, EMR
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Headline

The fungicides Switch (cyprodonil + fludioxonil), Signum (pyraclostrobin + boscalid) and Thianosan (thiram) were shown in laboratory tests to be active at relatively low concentrations against *Mucor* and *Rhizopus* soft rots

Background and expected deliverables

Mucor and *Rhizopus* both cause soft rots (also called leak disease) in raspberries and strawberries and losses can be significant when conditions are favourable resulting in rapid spread of the fungi in harvested fruit. They are primarily post-harvest rots but also occur on ripe fruit in the field. Both fungi cause similar symptoms on fruit. Infected fruit are initially slightly discoloured, gradually turning pale brown. The tissue rapidly softens and collapses and, under humid conditions, the fruit is soon covered with a dense, fluffy white mycelium which bears stiff, black-headed sporangia (pin mould). Both fungi survive in soil and crop debris between seasons and produce thick walled resting bodies (zygospores) which serve as long-term resting spores. Some circumstantial evidence suggests that the rots may be encouraged by green manures.

Mucor and *Rhizopus* are both wound pathogens and favoured by wet, moderately warm conditions (around 18°C). The black spores are easily spread by wind, rain and pickers hands. *Rhizopus* is inhibited by temperatures below 6°C whereas *Mucor* can grow and infect fruit at 0°C as well as it can at higher temperatures. This means that the former can be controlled in soft fruit by cool chain management whereas *Mucor* will continue to develop during cold storage and marketing.

Neither fungus is well controlled by fungicides and there is evidence that the occurrence of these rots may be increased by the use of certain fungicides such as Rovral (iprodione) to control botrytis rot. Currently control of these soft rots is based on cultural measures including removal of all ripe fruit at harvest.

In order to develop improved methods of managing these rots to minimise losses it is important first to establish the relative incidence of *Rhizopus* and *Mucor* in soft fruit crops. Although fungicides appear to be relatively ineffective against these fungi there may be alternative chemicals such as those used in the food industry that suppress *Mucor* or *Rhizopus* and could be used near harvest.

Over two years, this project aimed to identify the relative incidence of *Mucor* and *Rhizopus* in soft fruit plantations. This will enable management systems for minimising losses to be developed. In addition, laboratory studies were done to identify potential alternative chemicals active against these rots.

Summary of the project and main conclusions

In 2009, 131 samples of soft rotted berries were received from fruit farms in England and Scotland. *Mucor* spp. was the more frequently isolated from strawberry and raspberry soft rots than *Rhizopus*. 74% of isolates from strawberry and 88% from raspberry were identified as *Mucor* spp. All samples except for 6 (open field Elsanta) were obtained from tunneled crops whether raspberry or strawberry. All isolates from the open field crop were *Mucor* spp. From this data it is not possible to draw any definitive conclusions on the effect of tunnels on the incidence of *Mucor* and *Rhizopus*. Very little information on management practices was collected from the growers that provided the rot samples so it was not possible to investigate the relation between practice and rot incidence. Management of fruit and rots at harvest did vary on the farms visited. Most removed all rots and non-marketable fruit from the plantation at harvest which would tend to reduce the soft rot inoculum and hence the risk of infection by *Mucor* and *Rhizopus*.

In 2010, 16 lots of fruit samples (13 strawberry samples, 3 raspberry samples) were received from growers and consultants. These were mainly from Kent (9 samples) but also from Staffordshire, Yorkshire and Cambridgeshire. None were received from Scotland.

Isolations were made from 164 berries, yielding 115 fungal isolates with characteristics typical of members of the Mucorales. Of these, 80 were positively identified as *Mucor* spp. and 35 as *Rhizopus* spp.

Thirteen of the *Mucor* spp. isolates and 15 of the *Rhizopus* isolates were recovered from raspberry. Similarly, 67 of the strawberry soft rot associated fungi were *Mucor* spp. and 20 were *Rhizopus*. That is, 77% of the strawberries with symptoms of soft rot had *Mucor* spp. as the causal agent, which are similar results to 2009 (74%). In contrast to 2009 (88%), 46% of the soft-rotted raspberries were infected by *Mucor* spp. The raspberry samples came from two farms in Kent of cvs. Maravilla and Octavia, and so the sample size was rather small.

The efficacy of a range of chemicals against *Mucor* and *Rhizopus* was tested in the laboratory using PDA (Potato Dextrose Agar) amended with the chemical under test at

concentrations of 0, 1, 10, 100, 1000, 5000 ppm (*in vitro* tests). Growth of mycelial plugs of the test fungus was measured after 1 or 2 days. Standard isolates of *Mucor piriformis*, *Mucor mucedo* and *Rhizopus stolonifera* were used as well as isolates of *Mucor* and *Rhizopus* obtained during the survey. In the *in vitro* plate tests Signum (pyraclostrobin + boscalid), Switch (cyprodonil + fludioxonil), HDC F 13 (experimental) and Thianosan (thiram) were most effective in inhibiting mycelial growth of *Mucor* and *Rhizopus*. Sodium bicarbonate, potassium bicarbonate, potassium sorbate and sodium benzoate all inhibited growth at high concentrations, below that at which the products are used, and so may be worth further evaluation. Sulphur, Amistar (azoxystrobin), Frupica (mepanipyram) and ascorbic acid were ineffective.

Chemicals and biocontrol agents (BCAs) were also tested on tomatoes inoculated with either *Mucor* or *Rhizopus* spp. (*in vivo* tests). Fungal rots spread more slowly in tomatoes which makes it easier for scientists to assess the effect of fungicides. In these tests, Thianosan significantly reduced rotting on tomato fruit inoculated with *Mucor* spp. Switch, Signum, HDC F 13, Thianosan and the BCA Boni Protect forte all significantly reduced rotting on tomatoes inoculated with *Rhizopus* spp.

The results are summarised in Table 1a. The most effective products will be evaluated in field trials in strawberry in 2011 as part of the SCEPTRE Horticulture LINK project (HL 01109, CP 77).

In laboratory studies to investigate whether fruit flies were involved in the spread of *Mucor* in strawberries, fruit flies obtained from a laboratory supply company were placed in plastic boxes containing strawberries previously inoculated with *Mucor*. After two days the flies were transferred to healthy strawberries and plated on to PDA. The strawberry fruit in the plastic boxes where the flies were transferred rapidly developed soft rots with *Mucor* sporulating on the affected fruit. Similarly flies plated out onto PDA rapidly developed colonies of *Mucor*. This study shows that fruit flies are able to spread *Mucor*.

Financial benefits

Currently soft rots caused by *Mucor* and *Rhizopus* are managed by cultural methods only or not at all and are often overlooked as problems. Recently the incidence of these rots has increased, particularly after the last two seasons when the weather has been wet and favourable for spread. The incidence is high, especially after harvest during marketing when rot spread in punnets can be unacceptable to the retailers. Once the disease is present in

harvested fruit, spread can be very rapid especially *Mucor*, which continues to grow at the temperatures used in cold storage and cool chain marketing. For developing strategies to control these rots it is important to know the relative incidence of *Mucor* and *Rhizopus* in soft fruit crops. If *Mucor* were the predominant species then additional control measures would need to be considered pre-harvest to control the problem. Such measures would not be needed for *Rhizopus* which would be suppressed by the temperatures used in cool chain marketing. Failure to recognise soft rots as a potential problem and take steps to manage them could result in the problem arising during marketing leading to rejection of the consignment with financial loss. This project has established the relative incidence of the two rots in soft fruit plantations, and identified chemicals or biocontrol agents that could be used to control the problem.

Action points for growers

- Most of the soft rots were due to *Mucor* spp. which can grow at the low temperatures at which raspberries and strawberries are held after picking and during marketing. Therefore it is important to minimise the risk of *Mucor* infection of fruit during harvest.
- The fungicides Switch, Signum and Thianosan which are approved for use on strawberry showed activity at low concentrations against *Mucor* and *Rhizopus* in laboratory tests and could be used to help reduce the soft rots in commercial crops.
- Signum and Switch could similarly be used in raspberries.
- Fruit flies were shown to spread *Mucor* in strawberry fruit. Measures to minimise fruit fly infestations in plantations and in the packhouse must be considered as part of the management strategy for control of soft rots.

Table 1a. Chemicals and BCAs evaluated in the laboratory *in vitro* (Plate tests) or *in vivo* (Tomato tests) for efficacy against *Mucor* spp. or *Rhizopus* spp. in 2010

Test Product	Active ingredient	Rate/ha	Rate/ml	<i>In vitro</i> testing	Efficacy	<i>In vivo</i> testing	Efficacy
Amistar	azoxystrobin	1 L	1 ml	X	NE		
Frupica	mepanipyram	0.9 L	0.9 ml	X	NE		
Scala	pyrimethanil	2.0 L	2 ml	X	*		
Signum	Pyraclostrobin (6.7%) + boscalid (26.7%)	1.8 kg	1.8 mg	X	***	X	*** (R)
Switch	cyprodonil (37.5%)+ fludioxonil (25%)	1.0 kg	1.0 mg	X	***	X	*** (R)
Headland Sulphur	sulphur	600 ml/100 L	0.006 ml	X	NE		
Teldor	fenhexamid	1.5 kg	1.5 g	X	*		
Thianosan	thiram	3 kg/1000L	3 g	X	***	X	*** (M + R)
HDC F 13	experimental	0.8 L	0.8 ml	X	***		*** (R)
Vitamin C	ascorbic acid	-	-	X	NE		
Chitoplant	chitosan	1 g/L	1 mg			X	NE
Potassium bicarbonate	potassium bicarbonate	5 kg	5.0 mg	X	**	X	NE
Potassium sorbate	potassium sorbate	10 g/L	10.0 mg	X	**		NE
Sodium benzoate	sodium benzoate	10 g / L	10 mg	X	**		
Sodium bicarbonate	sodium bicarbonate	20 g/L	20 mg	X	**		
Wetcit	alcohol ethoxylate	2 ml/L	0.002 ml			X	*
Boni Protect forte	<i>Aureobasidium pullulans</i>	1.2 kg (at 2000 L/ha)	0.6 g			X	*** (R)
Prestop	<i>Gliocladium catenulatum</i>	100 g/20 L	5 g			X	NE
Serenade	<i>Bacillus subtilis</i>	10 L	10 ml			X	NE

X = tested * Some efficacy, *** Very effective, NE not effective R= Effective on *Rhizopus* only, M + R = Effective on *Mucor* and *Rhizopus*