Grower Summary

PE 018

Efficacy of conventional fungicides, biofungicides and disinfectants against tomato leaf mould (Passalora fulva)

Final 2015

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AHDB Horticulture,
AHDB
Stoneleigh Park
Kenilworth
Warwickshire
CV8 2TL

Tel – 0247 669 2051

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

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Project Leader:	Dr Tim O'Neill, ADAS
Contractor:	ADAS UK Ltd
Industry Representative:	Brian Moralee, Vitacress Tomatoes, Arreton, Isle of Wight
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GROWER SUMMARY

Headline

- Reliable development of tomato leaf mould in experimental systems required high levels of inoculum and prolonged periods (96 h) of high humidity
- Effective conventional fungicides for control of tomato leaf mould include Amistar, Switch, Signum and Teldor, with treatment timing key to reducing disease
- The biological fungicide Serenade ASO also had good efficacy when used preventatively and under low disease pressure
- A range of disinfectant products were effective against both the spores and mycelium of *Passalora fulva*

Background

Tomato leaf mould caused by *Passalora fulva* (previously *Cladosporium fulvum*) is a destructive foliar disease of increasing importance in the UK. Outbreaks have occurred most years over the last decade and affected a range of varieties. Previously well controlled by genetic resistance, the new outbreaks appear to be caused by the cultivation of varieties with no claimed resistance and the emergence of strains capable of overcoming the resistance genes deployed in current varieties. Amistar (azoxystrobin) has given good control in some crops, but grower reports indicate resistant strains can develop within a few years. The disease has also affected organic crops, where use of Amistar and other conventional fungicides is not permitted by the Soil Association. No plant protection products currently permitted on tomato carry a label recommendation for leaf mould control. Spores of *P.fulva* appear to be very resistant to dryness and low temperatures and are believed to survive in a dormant state from one crop to the next. The fungus can also survive saprophytically in dried leaf debris. There is little information on the relative effectiveness of different disinfectants in reducing inoculum of *P.fulva*. The aim of this study was to provide tools for improved management of tomato leaf mould in both conventional and organic crops through identification of effective conventional fungicide and biofungicide treatments for use in crops, and of disinfectant treatments for use between crops.

Specific objectives of the project were:

1. To develop a controlled infection technique on tomato seedlings with *P.fulva*
2. To determine the efficacy of selected conventional fungicides and biofungicides applied as protectant and curative spray treatments for control of tomato leaf mould.
3. To determine the effectiveness of selected disinfectants for reduction of *P.fulva*

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inoculum on surfaces and in debris.

Summary

<u>Objective 1 – Development of controlled infection technique</u>

In Experiment 1, a range of spore concentrations and periods of high humidity were applied to tomato plants cv. Gardener's Delight, a variety with no claimed genetic resistance to *P. fulva*. *P. fulva*, isolated from a crop in early 2014, was cultured on agar, and a spore suspension was used to inoculate the lower surface of tomato plant leaflets. The high concentration was 1 x 10^5 spores/ml, and the low concentration was an order of magnitude lower at 1 x 10^4. Results showed that disease development was greatest with the high level of inoculum, and an extended period (96 hours) of high humidity (Table 1 and Figure 1).

Table 1. Effect of inoculation rate and humidity period on the development of leaf mould in glasshouse tomato cv. Gardener's Delight – ADAS Boxworth, 2014 (Experiment 1)

Treatment	Inoculation rate	Humidity period (h)	% leaf area affected at intervals after inoculation		Crop vigour (0-5 index)
			15 days	18 days	18 days
1	Uninoculated	24	0.0	0.2	4.8
2	Uninoculated	48	0.0	0.0	5.0
3	Uninoculated	96	0.0	0.0	5.0
4	Low rate	24	0.3	5.0	5.0
5	Low rate	48	1.0	4.5	4.8
6	Low rate	96	1.8	**13.8**	5.0
7	High rate	24	1.8	8.8	5.0
8	High rate	48	3.3	**15.0**	5.0
9	High rate	96	**15.0**	**35.0**	**3.5**
Probability (F value) Inoculation x humidity			<0.001	0.005	<0.001
LSD (24 d.f.)			3.55	9.31	0.62

*Values in bold are significantly different from uninoculated plants.

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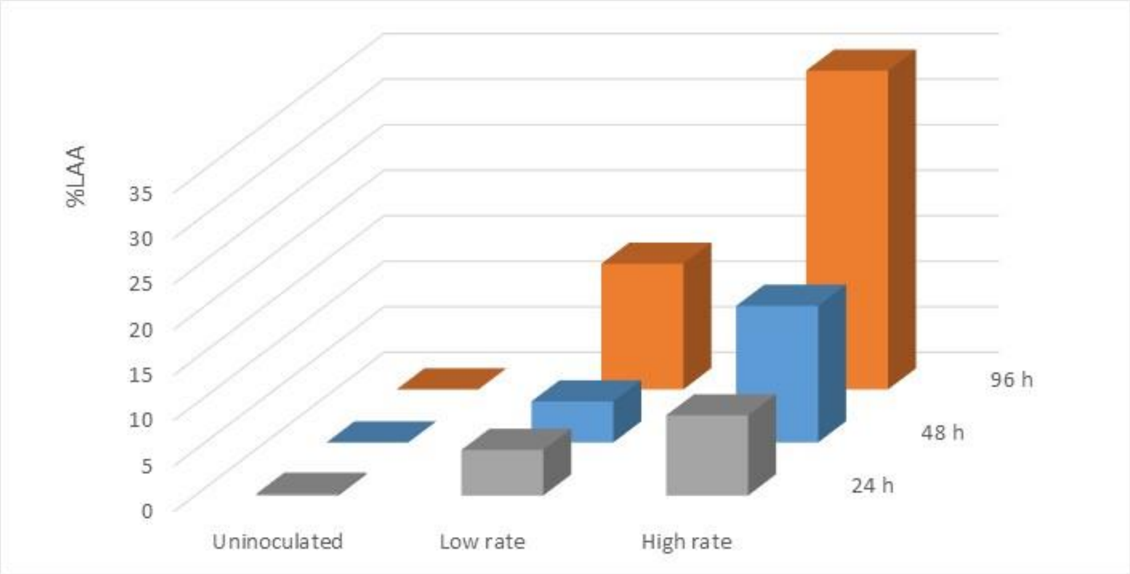

Figure 1. Effect of inoculum level and duration of high relative humidity on percentage tomato leaf area affected by tomato leaf mould as assessed 15 May, 2014 – ADAS Boxworth.

Objective 2 – Evaluation of the efficacy of conventional fungicide and biofungicide products

A total of eight products (four fungicides and four biofungicides) were tested against a water control; all of the products were approved for use on protected tomato except the coded biofungicides F185 and F186. Each product was sprayed only once, but at five different timings with reference to inoculation. These timings ranged from 5 days before to 5 days after inoculation, also including 1 day before, the day of inoculation and 1 day after. This was to establish any curative action of products against *P. fulva*, or to determine if products acted only preventatively.

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Table 2. Overall effects of plant protection product and spray timing on tomato leaf mould – 2014 (Experiment 2)

Factor	Mean % plants affected		Mean % leaf area affected (Severity)	
	11 Jun	19 Jun	11 Jun	19 Jun
<u>Product</u>				
1. Water control	93.8 (8.4)	100 (<0.1)	1.8	4.8
2. Amistar	24.7 (6.0)	**47.3** (6.4)	**0.2**	0.9
3. Signum	46.8 (6.0)	**65.4** (5.0)	1.0	2.4
4. Switch	40.1 (7.1)	**70.4** (5.5)	**0.7**	2.7
5. Teldor	46.8 (6.9)	**73.8** (5.9)	**0.7**	1.5
6. Prestop	58.4 (6.7)	**86.4** (4.6)	1.2	2.6
7. Serenade ASO	42.7 (5.1)	**56.3** (3.8)	0.9	2.3
8. HDC F185	46.3 (6.4)	**76.1** (5.5)	0.9	1.9
9. HDC F186	42.8 (6.8)	**72.6** (5.8)	**0.7**	2.5
P value (120 d.f.)	0.143	0.002	0.033	0.142
LSD	-	-	0.9425	2.356
<u>Timing</u>				
-5	40.6 (5.2)	67.2 (4.0)	0.6	1.7
-1	32.9 (4.8)	58.6 (4.9)	0.4	1.3
0	36.8 (5.1)	65.6 (4.2)	0.4	2.1
1	50.0 (5.3)	72.7 (4.2)	1.0	2.0
5	58.6 (4.9)	79.0 (3.8)	**1.4**	3.4
P value (120 d.f.)	0.016	0.051	<0.001	0.005
LSD	-	-	0.9126	2.281

*Values in bold are significantly different from the water control (upper columns) or the day 0 timing (lower columns); ( ) – standard error. See Tables 11 and 12 for results of individual treatments.

Results show that whilst all treatments reduced incidence of leaf mould compared to the untreated on 19 June (2 weeks after inoculation), Amistar treated plots contained fewest infected plants (Table 2). At the earlier 11 June assessment, Amistar, Switch and Teldor had significantly reduced disease severity, as had the biological HDC F186, whereas at the 19 June assessment, no treatments resulted in a severity significantly different to the untreated. Amistar, Signum and Teldor did show a trend for a reduction in severity at this assessment. The biological products Serenade ASO and HDC F185 also showed a trend towards a reduction in disease severity at the later assessment (Table 2). In terms of spray timing, the effect of spray timing was found to be significant. On 11 June there was significantly more disease (% leaf area affected) in the plots sprayed at +5 days than in plots sprayed at -1 or 0 days. There was also an observable trend for the efficacy of most products to decrease when used after inoculation at both assessment dates (Table 2). Therefore, we can conclude that the tested products are most effective when applied as protectants.

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Amistar and Prestop were seen to cause phytotoxicity when applied at the time plants were bagged to create conditions conducive to infection (Fig 1), causing yellowing and leaf distortion of the younger leaves at the head of the plant. Amistar is known to cause such a reaction in warm temperatures and high humidity, but this reaction to Prestop has not been observed before. Neither Amistar nor Prestop caused phytotoxicity when the treatment was applied before or after bagging. No differences in crop vigour that were not attributable to phytotoxicity were observed during the trial.

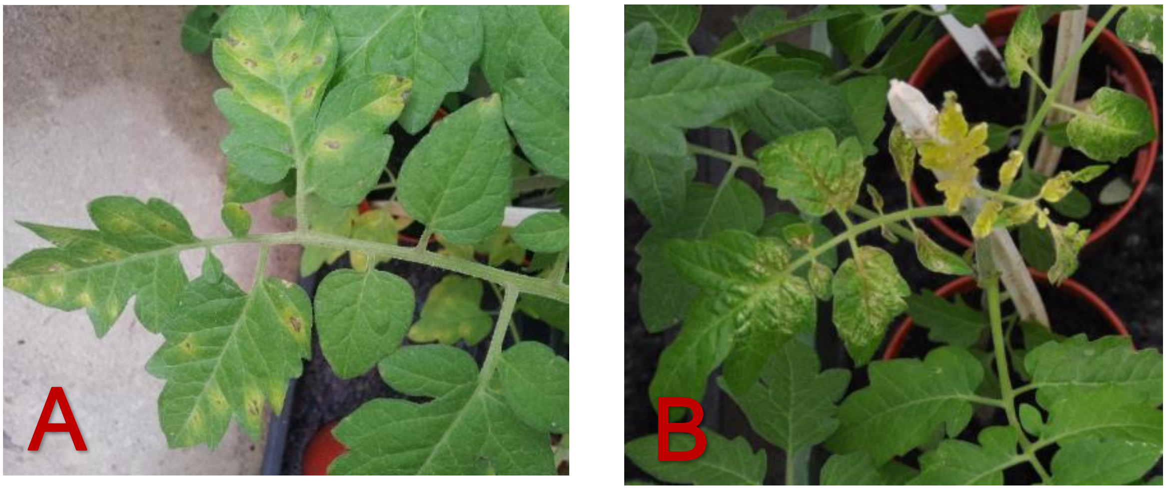

Figure 2. A – Symptoms of leaf mould 2 weeks after inoculation. B – Phytotoxicity following application of Amistar and imposed high humidity.

Products were further evaluated in another experiment on cv. Gardener's Delight. Fungicides were applied once at 3 days before inoculation or 3 days after inoculation; biofungicides were applied twice at -7 and 0 days before inoculation (Table 3).

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Table 3. The effect of the best performing plant protection products on tomato leaf mould at 20 days after inoculation (Experiment 3) – 2014

Treatment	Application timing (days relative to inoculation)				% leaves affected	% leaf area affected	% area of inoculated layer affected
	-7	-3	0	3	24 July	31 July	31 July
1. Untreated					30	26.0	55.6
2. Serenade ASO	✓		✓		30	20.0	**31.2**
3. HDC F185	✓		✓		30	20.0	**28.7**
4. HDC F186	✓		✓		30	17.5	43.8
5. Amistar		✓			**10**	**8.5**	**6.9**
6. Switch		✓			**0**	**2.9**	**2.1**
7. Teldor		✓			30	23.8	37.5
8. Amistar				✓	**10**	**2.8**	**3.8**
9. Switch				✓	**10**	**0.5**	**0.4**
10. Teldor				✓	30	**10.5**	**17.5**
P value					<0.001	<0.001	<0.001
LSD between treatments					0.14	10.17	22.98
LSD vs. untreated					0.12	8.80	19.90

Values in bold are significantly different from untreated plants.

This experiment can be viewed as a more difficult test of the plant protection products than previously, as the severity achieved in untreated plots reached 26% leaf area affected. Perhaps because of this increased disease pressure, the biological products tested did not perform well against *P. fulva*. Of the three biological fungicides tested, only Serenade ASO and HDC F185 resulted in a significant reduction in leaf mould at the final assessment, and only when the inoculated leaf layer was assessed alone. HDC F186 did not give any observable reduction in leaf mould at any of the assessments. Of the conventional fungicide products, both Switch and Amistar significantly reduced disease incidence and severity at every assessment when applied at 3 days before inoculation (Table 3).

It should be noted that products were applied only once (Experiment 2), or once for fungicides and twice for biofungicides (Experiment 3) in line with experiment objectives. All of the registered products can be applied several times in a commercial setting (see Table 5) and it is possible that greater levels of control may be achieved where multiple sprays are used.

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<u>Objective 3 – Efficacy of disinfectants for reduction of *P. fulva*</u>

Six commercially available disinfectant products were tested for efficacy against *P. fulva* in a series of laboratory experiments.

Initially, using specialised agar plates, disinfectants were tested against the spores and the mycelium of *P. fulva*. *P. fulva* spores and mycelium were challenged with the disinfectant at their recommended and half rates, and for 5 or 30 minutes at each rate. All of the disinfectants tested were effective when used at their recommended rate with 30 minutes of exposure.

Products were generally more effective against spores than against mycelium, and the products were not statistically different from one another in their efficacy. Those that maintained good efficacy when used at half rate or for only 5 minutes against mycelium (Jet 5, Unifect G, Menno Florades and Hortisept Pro) were used in two further experiments. In a comparison of disinfectants on four surfaces (aluminium, concrete, glass and plastic) Unifect G appeared the most effective against total fungal and bacterial growth; and aluminium, glass and plastic appeared easier to disinfect than concrete. However, no results were obtained specifically for *P. fulva* as the fungus was not recovered, not even from the untreated controls. It is likely growth on agar plates was swamped by more rapidly growing bacteria and fungi recovered concomitantly from the test surfaces. In a final experiment, the best performing products were tested against infected crop debris, which was used to inoculate tomato plants cv. Gardener's Delight, to determine if the debris was still infective (Table 4).

Table 4. Effect of disinfectant treatment on infectivity of tomato leaf debris affected by *P. fulva* – ADAS Boxworth, 2014

Treatment	% leaf area affected		% area affected inoculated leaf layer	
	12 Sep	19 Sep	12 Sep	19 Sep
1. Untreated	3.8	13.0	14.3	26.3
2. Jet 5	**0.6**	**6.8**	**5.4**	**16.3**
3. Unifect-G	**0.2**	**2.0**	**0.7**	**4.3**
4. Menno Florades	**0.1**	**0.8**	**0.3**	**2.6**
5. Hortisept Pro	**0.1**	**0.4**	**0.2**	**2.0**
P value	<0.001	<0.001	0.001	<0.001
LSD	1.114	3.405	6.117	5.240

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At the first and second assessments, all disinfectant treatments succeeded in reducing disease transmission by infected leaf debris when compared to inoculation with untreated leaf debris (Table 4). There were initially no statistical differences between disinfection products. At the final assessment, however, differences between treatments became clearer. When Jet 5 was used to treat leaf debris, significantly higher disease levels were observed than when debris was treated with the other disinfectant products. There were no statistical differences in efficacy between Hortisept Pro, Menno Florades and Unifect-G.

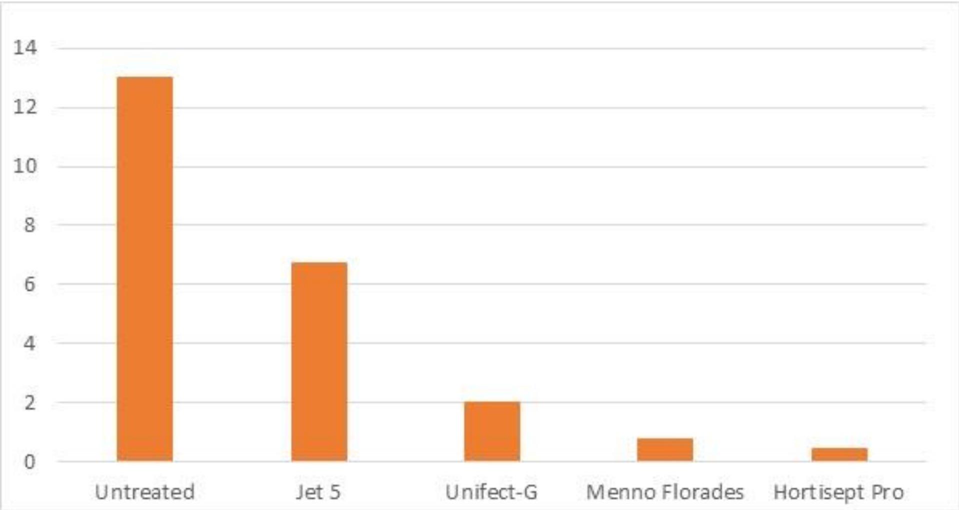

Figure 2. % leaf area affected by leaf mould after inoculation with disinfectant-treated leaf debris, 19 September 2014 – ADAS Boxworth

Financial Benefits

Good management of tomato leaf mould is likely to depend on both effective management of glasshouse humidity and use of effective plant protection products and disinfectants. Keeping relative humidity low in tomato glasshouses is already implemented as far as is allowed by other cropping factors. Similarly, large scale disinfection of glasshouses between crops is performed, but knowing which products will be most effective against *Passalora fulva* will allow this investment and labour to be more cost-effective. The disinfectant products found to be most effective in these trials are of comparable cost to others on the market. The plant protection products Amistar and Switch are already used in spray programmes against Botrytis, and a single spray application is estimated to cost £250 per hectare per season. There will also be the added benefit of providing some control of both diseases when sprays are applied, rather than having to add a new product to any spray programmes in place. However, spray timing may need to be adjusted to obtain the most effective control of leaf mould.

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Action Points

- Minimise prolonged periods of high humidity (over 80% RH) as far as possible within the glasshouse
- Fungicides most effective against *P. fulva* include Amistar, Switch, and to a lesser extent, Teldor
- Although less effective than Amistar in this study, consider use of Signum when broad-spectrum disease control is required (eg Botrytis, leaf mould and powdery mildew)
- Use fungicides from two or more fungicide groups to reduce the risk of leaf mould developing resistance on your nursery; grower experience indicates that leaf mould can become resistant to azoxystrobin (Amistar) and it is likely that the related fungicide pyraclostrobin (in Signum) would also be ineffective in this situation.
- Biofungicides effective against *P. fulva* include Serenade ASO and a coded product (not currently registered for use on tomato)
- Generally, using plant protection products preventatively for control of leaf mould (i.e. at the very early stage in the disease epidemic) is more effective than using curatively
- The scientific literature indicates *P. fulva* can easily persist on a nursery between crops; carefully clean up all crop debris after crop removal, especially in cases where leaf mould has occurred
- After crop removal and clean-up, treat the glasshouse structure and floor with a suitable disinfectant so that any remaining very small leaf fragments are disinfected
- Disinfectants found most effective against *P. fulva* in this work include Hortisept Pro, Unifect-G and Menno Florades
- Regularly washing hands with soap and water or alcohol gel may help to prevent spread of leaf mould
- Disinfectant products are most effective when used at their full recommended rates, for as long a time as possible
- On more uneven, porous surfaces, such as concrete, it is likely that a disinfectant product will require a longer time to be fully effective

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