



# Maleic Hydrazide Application Alters Bacterial Wilt Severity in Mechanically Topped and Harvested Tobacco

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# Mechanization & the Spread/Severity of Bacterial Wilt in SC



# Background

- Field observations suggested maleic hydrazide (MH) application could possibly suppress mechanical transmission of bacterial wilt following mechanical topping and leaf removal.
- Mechanical transmission of *R. solanacearum* during flower and leaf removal coincides with the time when MH is applied to arrest axillary shoot growth (suckers).
- Growth regulators have been shown to effect disease development.

## Previous studies with MH

- Evaluated the effect of MH application on *R. solanacearum* establishment and disease development following mechanical transmission of the bacterium using:
  - Growth Chambers
  - Greenhouse

## Previous Results – Growth Chamber

Treatment	Disease Severity	Separation Level	Stem Necrosis	Separation Level
Inoc. with <i>R. solanacearum</i>	5.00	A	5.00	A
Late MH + inoculum	4.00	A	4.00	A
MH + inoculum	2.25	B	2.50	B
Early MH + inoculum	1.50	B	1.75	B

# Previous results with MH

- MH suppresses the severity of bacterial wilt.
- Disease suppression is strongly linked to MH application timing.

## Previous Results with MH



Inoc. with  
*R. solanacearum*  
(inoculum)

Early MH +  
inoculum

MH +  
inoculum

Late MH +  
inoculum

# Objective

- To evaluate the effect of MH application on *R. solanacearum* establishment and disease development in the field following mechanical transmission of the bacterium during topping and harvesting.



# Materials and Methods – Field Tests

- Experiments were conducted at Clemson's Research and Education Center in Florence, SC, summer 2010.
- Plants of K346 were grown under standard agronomic practices for South Carolina.
- Plots consisted of two rows 15.2 m long with a 1.2m row spacing.
- Each row was fumigated with 1,3 D + chloropicrin (10.5 gal/A) 21 days prior to transplanting (Telone C-17).
- Experimental design was a randomized complete block with four replications.
- Experiments were repeated in time for a second complete run.

## Inoculation – Field Tests

- Isolates of *R. solanacearum* were grown and suspended in deionized water at Optical Density<sub>600</sub> = 0.2 =  $10^8$  cells/ml and used as a stock culture.
- The  $10^8$  suspension was diluted to  $2 \times 10^6$  cells/ml for inoculation.
- MH was applied with a Backpack CO<sub>2</sub> pressure system at a rate equivalent to 50 gal/A (15 ml/500 ml).
- Inoculation was performed to simulate mechanical flower and leaf removal
  - Topping test : a steel cutter blade was misted individually with the isolate suspension and used to top plants 15cm below the inflorescence.
  - Harvest test : a steel cutter blade was misted individually with the isolate suspension and used to remove lower leaves at harvest.

# Treatments – Topping Tests

Treatment	Description
<i>R. solanacearum</i>	Inoculated with <i>R. solanacearum</i> (at $2 \times 10^6$ cells/ml) <u>without</u> MH application
Pre-Treatment MH + <i>R. solanacearum</i>	MH application <u>8 days prior</u> to inoculation with <i>R. solanacearum</i>
Pre-Treatment MH + <i>R. solanacearum</i>	MH application <u>4 days prior</u> to inoculation with <i>R. solanacearum</i>
MH + <i>R. solanacearum</i>	MH application <u>immediately prior</u> to inoculation with <i>R. solanacearum</i>

**\*\*Each treatment row was paired with a MH treated/non-inoculated control row.**

# Treatments – Harvesting Tests

Treatment	Description
<i>R. solanacearum</i>	Inoculated with <i>R. solanacearum</i> (at $2 \times 10^6$ cells/ml) <u>without</u> MH application
Pre-Treatment MH + <i>R. solanacearum</i>	MH application <u>14 days prior</u> to inoculation with <i>R. solanacearum</i>
Pre-Treatment MH + <i>R. solanacearum</i>	MH application <u>7 days prior</u> to inoculation with <i>R. solanacearum</i>
Post MH + <i>R. solanacearum</i>	MH application <u>immediately after</u> inoculation with <i>R. solanacearum</i>
Post MH + <i>R. solanacearum</i>	MH application 4 days post inoculation with <i>R. solanacearum</i>

**\*\*Each treatment row was paired with a MH treated/non-inoculated control row.**

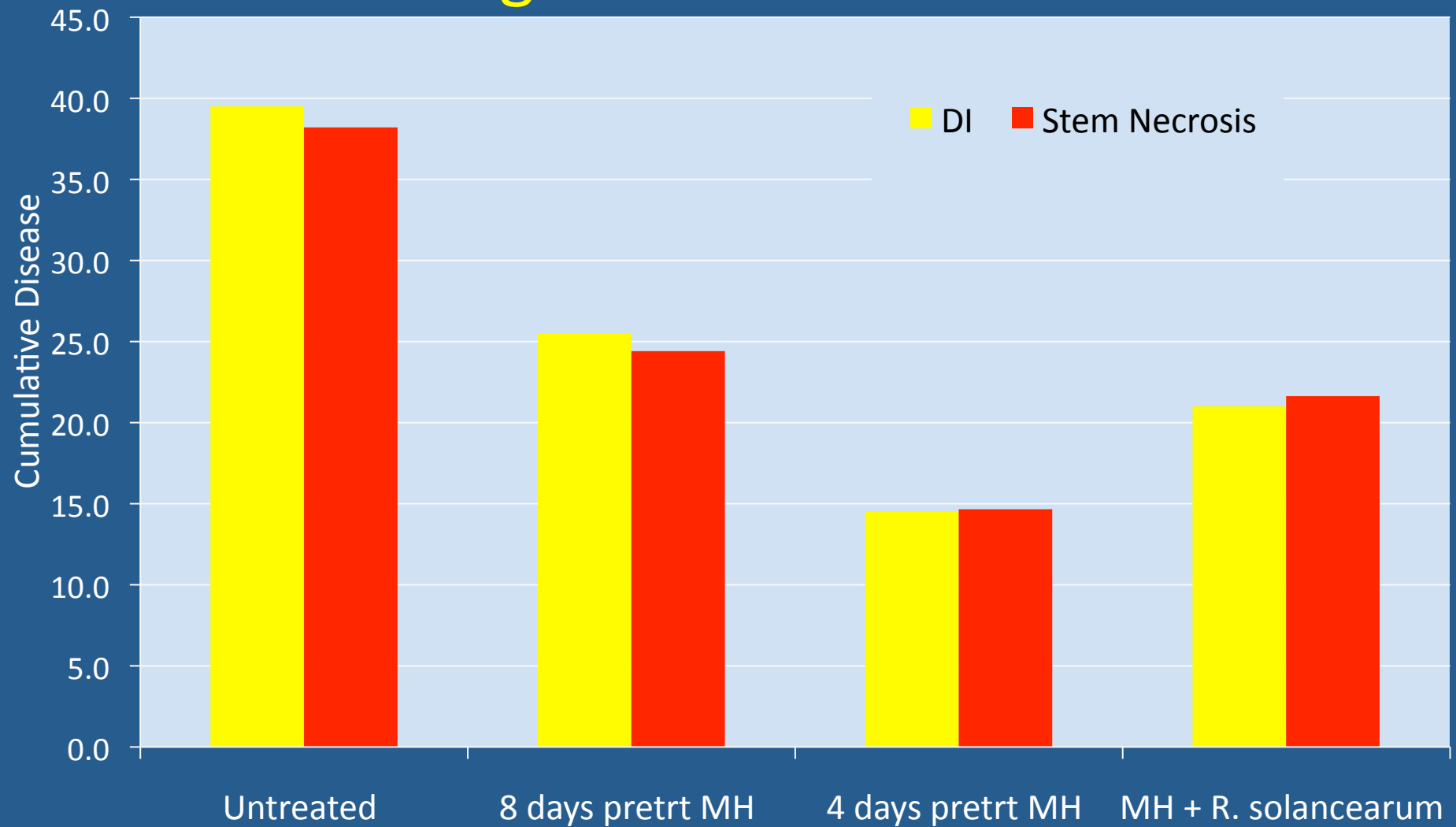
## Disease Assessment – Field Tests

- *R. solanacearum* was positively confirmed using immunological testing strips (Agdia Pathoscreen Kit).
- Plants were assessed weekly for disease severity starting 21 days following inoculation and rated on a 0 to 5 scale (0 = no visible symptoms, 5 = complete collapse of tissue).
- Stem necrosis was recorded on a 0 to 5 scale at final disease assessment date.
- Disease severity data were subjected to ANOVA using JMP software (SAS); AUDPC values were calculated for each treatment.

# Results - Field Topping Tests

Treatment	Disease Index	Separation Level	Stem Necrosis	Separation Level
Inoc. with <i>R. solanacearum</i> without MH	4.50	A	4.32	A
Very early MH 8 days pre – inoculation	3.50	AB	3.28	B
Simultaneous MH application	3.25	B	2.71	B
Early MH 4 days pre-inoculation	2.00	C	1.89	C

# Field Topping Tests - Area Under Disease Progress Curve Values



## Results - Field Harvest Tests

Treatment	Disease Index	Separation Level	Stem Necrosis	Separation Level
Inoc. with <i>R. solanacearum</i> without MH	3.50	A	4.62	A
Very early MH 14 days pre – inoculation	3.50	A	3.95	A
Late MH 4 days post-inoculation	3.75	A	3.92	A
Early MH 7 days pre-inoculation	2.75	AB	3.50	AB
Immediate post MH treatment	1.88	B	2.48	B



# Summary

- Replicated Field Studies indicate Maleic Hydrazide (MH) suppresses the severity of bacterial wilt.
- Disease suppression is strongly linked to MH application timing.