Fungicide Sensitivity and the Management of Tart Cherry Pathogens

Objectives
- Evaluate available chemistries for possible use on CLS (SI resistance)
- Evaluate the sensitivity of ABR to Indar

Methods
- Establish baseline, or current in vitro sensitivity levels to relevant chemistries
  - Track changes over time
  - Evaluate fungicide resistance management techniques
- Determine field efficacy to give perspective to sensitivities measured in the lab

Control of Cherry Leaf Spot
- As of 2004, statewide resistance to SI fungicides was reported (Proffer et al., 2006)
- Recent NWHRS trials demonstrated that cover sprays of SI chemistries have either failed or provided significantly reduced control of CLS

2005 Spray Trial Results

<table>
<thead>
<tr>
<th>Fungicide and rate per acre</th>
<th>Timing</th>
<th>CLS Infected leaves (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bravo 82.5WDG 3 lb</td>
<td>bloom, hook split</td>
<td></td>
</tr>
<tr>
<td>Flint 50WG 2.5 oz</td>
<td>first cover</td>
<td></td>
</tr>
<tr>
<td>Doule (Sy &amp; (6Y) 2 lb</td>
<td>second, third cover</td>
<td>55.1 a</td>
</tr>
<tr>
<td>Elite 45WG 6 oz</td>
<td>fourth cover</td>
<td></td>
</tr>
<tr>
<td>Bravo 82.5WDG 3 lb</td>
<td>bloom, hook split</td>
<td></td>
</tr>
<tr>
<td>Flint 50WG 2.5 oz</td>
<td>first cover</td>
<td></td>
</tr>
<tr>
<td>Copper Sulfate (Cuprine D. 40DF) 3.5 lb</td>
<td>second, third cover</td>
<td>33.3 b</td>
</tr>
<tr>
<td>Elite 45WG 6 oz</td>
<td>fourth cover</td>
<td></td>
</tr>
<tr>
<td>Bravo 82.5WDG 3 lb</td>
<td>bloom, hook split</td>
<td></td>
</tr>
<tr>
<td>Flint 50WG 2.5 oz</td>
<td>first cover</td>
<td></td>
</tr>
<tr>
<td>Copper Hydrosite (Keclidean 2000 35DF) 4 lb</td>
<td>second, third cover</td>
<td>53.6 a</td>
</tr>
<tr>
<td>Elite 45WG 6 oz</td>
<td>fourth cover</td>
<td></td>
</tr>
<tr>
<td>Untreated control</td>
<td></td>
<td>64.3 a</td>
</tr>
</tbody>
</table>

* Means followed by the same letter are not significantly different according to Fisher’s Protected LSD (α = 0.05).
**CLS Spray Trial, 2006**

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Cherry Leaf Spot (%)</th>
<th>Dicots (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>28-Jul</td>
<td>18-Sep</td>
</tr>
<tr>
<td>Control</td>
<td>61.50a</td>
<td>95.10a</td>
</tr>
<tr>
<td>Elite 45WG/0.045WG/0.060 + Captan 0.050W/0.030lb SI/Broad</td>
<td>5.1 b</td>
<td>95.3 a</td>
</tr>
<tr>
<td>Gem/006K 0.050SC/0.030fl 0.060oz Strobilurin</td>
<td>2.9 b</td>
<td>97.7 a</td>
</tr>
<tr>
<td>Sylitt 0.025fl 0.060oz Dodine</td>
<td>0.7 c</td>
<td>78.0 b</td>
</tr>
</tbody>
</table>

Based on the results, assessing dodine sensitivity in Michigan orchards became a relevant project.

**In vitro Dodine Assay Methods**

- 860 CLS isolates
- Isolates collected from managed and unmanaged sources
- Screened for minimum inhibitory concentration (MIC)

- 78.5% isolates MIC was 100 ppm or less
- 21.2% had an MIC of 200-400 ppm
- 0.2% MIC was greater than 400 ppm

**Conclusions**

- The vast majority of CLS isolates are highly sensitive to dodine in lab testing
- Practical field resistance hasn’t occurred but we are seeing a precursor for reduced sensitivity
- Dodine is still useful but needs to be managed for resistance and tank mixed with a broad-spectrum fungicide (captan)
**American Brown Rot**

- Infects cherries, mostly sweets
- Infects mainly mature fruit
- Reduces quality

**ABR Sensitivity Survey**

- Indar (fenbuconazole)
- 23 Michigan orchards
- 10+ isolates from each
- Single-spore isolated

**Establishing an Effective Dose**

**Effective dose (ED\(_{50}\)) values are the concentration of fungicide at which relative growth is reduced by 50%**

- Colony size is measured at increasing fungicide concentrations and a regression analysis is performed

**Regression Analysis**

**As Fungicide Concentration Increases**

Relative Growth Decreases

**Indar Assay Results**

- There is significant variability
- Up to a 10-fold difference
- Variability may be a precursor of reduced sensitivity
Is the Variability of ABR Indar-Sensitivity Significant?

Fruit Assay
- The most and least Indar-sensitive isolates were selected
- Cherries were briefly dipped in 3 concentrations of Indar (1/4, 1/2, and full rates)
- One 20µl droplet of fungal propagules, was placed on each sweet cherry
- In the case of tart cherries, inoculum was sprayed on using an atomizer
- Cherries were incubated for 2 weeks
- The number of cherries with ABR infections was then recorded

Tart Cherry Assay

Significant differences in infection rate based on isolate sensitivity

Sweet Cherry Assay

Significant differences in infection rate based on isolate sensitivity

Indar Fruit Assay Conclusions
- Uninoculated cherries did not develop ABR infection (no latent infections)
- The less Indar-sensitive isolates appear to be more virulent
- Based on ED50 values, there are significant differences between the infection ability of ABR isolates in the presence of Indar

Sequencing the CYP51 gene

- Michigan isolates are still sensitive to Indar in the field
- A significant mutation of the CYP51 gene is unlikely
- Characterizing the CYP51 genes of these sensitive isolates will be useful in future endeavors

Base pair differences between Michigan ABR isolates did not differentiate isolates based on sensitivity levels or confer changes to the amino acids

ABR Conclusions
- There is variability in the sensitivity of ABR to Indar
- This variability significantly affected the ability of the isolates to infect fruit in the presence of Indar
- Establishing the current sequence of the CYP51 gene in sensitive populations will aid in locating possible future mutations
Amended Cherry Fungicide Spray Calendar

- **Bloom: EBR**
  - Sterol inhibitor (captan)
- **Petal Fall: CLS**
  - Chlorothalonil
- **1st–4th Cover: CLS and PM**
  - Gem (Strobi) & boscalid
  - Pristine (Boscalid & strobi)
  - Copper (weather permitting)
  - Dodine & captan
- **+ 4th Cover ABR**
  - Sterol inhibitors (captan)
- **Post Harvest: CLS**
  - Chlorothalonil

Tank-mix & new options