Preharvest-Applied Fungicides & Postharvest ClO$_2$ Gas for Reducing Diplodia Stem-end Rot on Fresh Grapefruit

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Control Options

• Preharvest - No reliable replacement yet for Benlate or Topsin
  – However, copper, Aliette, and phosphorous acid products to reduce Brown rot

• Postharvest control measures
  – Good sanitation practices
  – Careful handling
  – Use of fungicides
    • Must be effective against latent organisms such as Diplodia and Anthracnose
Fruit Decay - on Tree!

- Diplodia (*Lasiodiplodia theobromae*) causes fruit stem-end rot and was:
  - Consistently detected in the abscission zone and juice of HLB-infected fruit
  - Greater abundance of Diplodia was positively correlated with lower fruit detachment force
  - Fruit ethylene production is positively correlated with Diplodia infection levels
  - Quadris Top = multiple applications gave intermittent control under HLB conditions

Zhao et al., 2015, 2016
Materials and Methods (2019-2020 season)

➢ Materials were hand-sprayed on individual grapefruit within the tree canopy at 2 locations (groves)

➢ Fruit were harvested 2 and 14 days after fungicide treatments and subjected to 6-day degreening conditions (5 ppm ethylene, 85°F, and 90% RH)

➢ The fruit were then incubated at 75°F with 90-95% RH for up to 15 days and evaluated for Diplodia SER and other decays every 3 days
**Treatments:**
Control = Water
Thy = Thymeguard (Thyme oil)
Men = Mentor EC (Propiconazole)
Qua = Quadris Top (now Amistar Top) (Azoxystrobin + Difenconazole)
Hea = Headline (Pyraclostrobin)
Mer = Mertect 340F (Thiabendazole)*
Gra = Graduate A+ (Azoxystrobin + Fludioxonil)*
Top = Topsin (thiophanate-methyl)*
*Not labeled preharvest for Florida grapefruit

Dr. Jiaqi Yan
Dr. Jiuxu (John) Zhang
Cuifeng Hu
2020-21 Preharvest Trials (4 red grapefruit fields)

- CONTROL - WATER
- Topsin 4.5 FL*
  - thiophanate-methyl (45%)
- Graduate A+ *
  - fludioxonil (20.6%) + azoxystrobin (20.6%)
- Switch 62.5 WG
  - fludioxonil (25%) + cyprodinil (37.5%)
- Miravis Prime*
  - fludioxonil (21.4%) + pydiflumetofen (12.8%)
- Miravis Top
  - difenconazole (11.5%) + pydiflumetofen (6.9%)
- Headline
  - pyraclostrobin (23.6%)
- Thyme Guard (Thyme)
  - thyme (23%)
- Citrus Fix (2,4-d) *
  - 2, 4-D (45%)
- Quadris Top (now Amistar Top)
  - azoxystrobin (18.2%) + difenoconazole (11.4%)

*not labeled preharvest for FL grapefruit
2021-22 Preharvest Trials (3 red grapefruit fields)

- CONTROL - WATER
- Topsin 4.5 FL*
  - thiophanate-methyl (45%)
- Cannonball *
  - fludioxonil (50%)
- Miravis Prime*
  - fludioxonil (21.4%) + pydiflumetofen (12.8%)
- Abound
  - Azoxystrobin (22.9%)
- Headline
  - pyraclostrobin (23.6%)

- Cannonball* + Abound
- Cannonball* + Headline
- Luna Sensation
  - Fluopyram (21.4%) + trifloxystrobin (21.4%)
- Veltyma
  - Pyraclostrobin (17.56%) + mefentrifluconazole (17.56%)

*not labeled preharvest for FL grapefruit
Summary

• **Topsin** 4.5FL demonstrated the best Diplodia SER control
  – but it is NOT registered
• **Graduate A+** controlled Diplodia SER
  – But it is NOT registered, and component fungicide concentrations are much higher than what would be allowed preharvest
• Strobilurin-based fungicides (such as **Abound** and **Headline**) moderately reduced Diplodia SER when decay pressure
• **Miravis Prime** (not yet registered for grapefruit) consistently showed significant and moderate Diplodia SER control and appears to be a good candidate for grapefruit registration for Diplodia SER control
ClO$_2$ Gas for Reducing Postharvest Diplodia Stem-end Rot on Grapefruit
Chlorine Dioxide (ClO$_2$) Gas

- Strong oxidizing agent, with about 2.5 times higher activity than chlorine
- Broad biocidal effectiveness
- Previously tested in water solutions as a sanitizer
- Previous work in our lab (Zhong et. al. 2021) showed that gaseous ClO$_2$ treatments (ICA TriNova, LLC.) reduced Diplodia SER on mandarins
- The objective of the current studies were to evaluate the potential of ClO$_2$ gas to control postharvest Diplodia SER on grapefruit
Materials and Methods

• **In vitro** antifungal activity tests:
  – Place fungal PDA plugs and “fast” ClO$_2$-releasing materials at various rates in sealed plastic containers
  – Incubate for 24 h
  – Measure mycelial growth by incubating fungal plugs for an additional 24 h after treatment
Without fruit:

Control

0.25 g

0.5 g

0.75 g

1.0 g

1.25 g

1.5 g

With fruit:

Control

0.5 g

1.0 g

1.5 g

2.0 g

2.5 g

3.0 g

Complete inhibition
The graph shows the concentration of ClO$_2$ (in ppm) over the incubation time (in hours) for different fruit weights.

- **10g + 10kg fruit**
- **15g + 10kg fruit**
- **20g + 10kg fruit**
- **10g, no fruit**
- **15g, no fruit**
- **20g, no fruit**
Materials and Methods

• **Inoculated grapefruit** (4 h prior):
  – Treated with fast ClO₂ gas release materials at three concentrations for 8 h in sealed plastic containers
  – ClO₂ concentrations were monitored using a PortaSens II ClO₂ gas detector
  – Decay was evaluated 3 and 6 days after fruit were incubated at 75°F with 90-95% RH
Materials and Methods

• Naturally infected grapefruit:
  – Placed in a sealed plastic container
  – “Fast” ClO₂ gas release materials applied:
    • at three concentrations at 75°F for 8 h, or
    • At one concentration (1 g/kg fruit) for 2, 4, or 8 h
  – After degreening (5 ppm eth) for 3 days, the fruit were incubated at 75°F with 90-95% RH for up to 3 weeks and decay evaluated weekly
A. Diplodia SER (%)

<table>
<thead>
<tr>
<th>ClO₂ release materials (g/kg fruit)</th>
<th>Control</th>
<th>1.0g/kg</th>
<th>1.5g/kg</th>
<th>2.0g/kg</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diplodia SER (%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>80</td>
<td>a</td>
<td>b</td>
<td>b</td>
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</tr>
</tbody>
</table>

B. Diplodia decay index

<table>
<thead>
<tr>
<th>ClO₂ release materials (g/kg fruit)</th>
<th>Control</th>
<th>1.0g/kg</th>
<th>1.5g/kg</th>
<th>2.0g/kg</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diplodia decay index</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>4.0</td>
<td>a</td>
<td>b</td>
<td>b</td>
<td>b</td>
</tr>
</tbody>
</table>
A

![Graph A](image)

B

![Graph B](image)
Summary

• Grapefruit treated with 1.0g/kg fruit of a ClO₂ releasing product for 8 h significantly reduced Diplodia SER of inoculated and naturally infected fruit without causing fruit peel damage and or altering other quality attributes (e.g., weight loss, firmness, acids, soluble solids)

• This ClO₂ gas treatment shows potential to be included as a component of Florida’s fresh citrus decay control strategy

• However, this system requires further testing under more commercial conditions
Maximum Residue Limits (MRLs)

- Industry vigilance is required when MRLs for export markets are lower than U.S. MRLs
  - Countries change MRLs periodically
  - Limited knowledge of how fast residues of various compounds decline under different production/postharvest conditions
http://irrec.ifas.ufl.edu/postharvest/

Or simply search for

“UF Postharvest”
Pesticide Residues & Maximum Limits

UF/IFAS Publications

- Maximum Residue Limits (MRLs) for FL Citrus
  For U.S. & Selected Export Markets
  Condensed list of recent changes in Korean MRLs

Pesticide MRL Web Resources

- BCGlobal (Global MRL Database) - Bryant Christie, supported by the USDA Foreign Agricultural Service (FAS).
- U.S.A. Code of federal regulations - Go to Title 40, Part 180 found here for the official list of U.S. MRLs.
  - See Subpart D—Exemptions From Tolerances
  - Substances Generally Regarded as Safe (GRAS)
  - GRAS Overview Website
- European Union - EU-MRL Pesticide Database--European Commission Directorate General for Health and Consumers. Quick jump to: Grapefruit, Orange
  - USDA EU Early Alert Notifications
<table>
<thead>
<tr>
<th>Chemical Name</th>
<th>Brand or Trade Names (Examples only, not exhaustive)</th>
<th>U.S. Citrus</th>
<th>CODEX Citrus</th>
<th>Canada Citrus</th>
<th>EU (G &amp; O only)</th>
<th>Great Britain (G &amp; O only)</th>
<th>Japan (G &amp; O only)</th>
<th>Korea (G &amp; O only)</th>
<th>Taiwan (G &amp; O only)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2,4-D (2,4-Dichlorophenoxyacetic acid)</td>
<td>Citrus Fix, Hivol</td>
<td>3</td>
<td>1</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>2 (3 proposed)</td>
<td>0.15</td>
<td>0.01</td>
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<tr>
<td>Abamectin</td>
<td>Agri-Mek, Clinch, Zephyr, ABBA, Epi-mek, Reaper: Mineo Pro</td>
<td>0.02</td>
<td>0.02</td>
<td>0.02</td>
<td>0.04</td>
<td>0.04</td>
<td>0.1</td>
<td>0.01 (G); 0.05 (O)</td>
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<td>Acequinocyl</td>
<td>Kanemite</td>
<td>0.35</td>
<td>0.35</td>
<td>0.2 (G); 0.4 (O) (0.6 proposed)</td>
<td>0.2 (G); 0.4 (O)</td>
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<td>0.01 (G); 0.7 (O)</td>
<td>0.2</td>
<td>0.3</td>
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<tr>
<td>Beta-cyfluthrin</td>
<td>Baythroid XL, Brigade, Capture, Telstar, Fanfare</td>
<td>0.2</td>
<td>0.3</td>
<td>0.1</td>
<td>0.02 (E)</td>
<td>0.02</td>
<td>2 (1 proposed)</td>
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<td>0.3</td>
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<tr>
<td>Bifenthrin</td>
<td>Brigade, Capture, Telstar, Fanfare</td>
<td>0.05</td>
<td>0.05</td>
<td>0.1</td>
<td>0.05 (E)</td>
<td>2</td>
<td>0.01 (G); 0.5 (O)</td>
<td>0.5</td>
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<tr>
<td>Bromacil</td>
<td>Bromo, Hyvar</td>
<td>0.1</td>
<td>0.1</td>
<td>0.01</td>
<td>0.01</td>
<td>0.1</td>
<td>0.01</td>
<td>0.01</td>
<td>0.5</td>
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<tr>
<td>Buprofezin</td>
<td>Applaud, Centaur</td>
<td>4</td>
<td>1</td>
<td>0.1 (G, L, P), 0.1 (O, T)</td>
<td>0.01</td>
<td>0.01</td>
<td>3 (G), 2 (O)</td>
<td>0.01 (G), 2.5 (O)</td>
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<tr>
<td>Carbaryl</td>
<td>Sevin</td>
<td>10</td>
<td>15</td>
<td>10</td>
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<td>0.01</td>
<td>5</td>
<td>0.01 (G), 7 (O)</td>
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<tr>
<td>Carpinphos-ethyl</td>
<td>Aim</td>
<td>0.1</td>
<td>0.1</td>
<td>0.02</td>
<td>0.01</td>
<td>0.1</td>
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<td>0.1</td>
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<td>Clothianidin</td>
<td>Belay</td>
<td>0.07</td>
<td>0.07</td>
<td>0.1</td>
<td>0.06 (E)</td>
<td>0.06</td>
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<td>0.01 (G); 0.3 (O)</td>
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<tr>
<td>Cyfluthrin</td>
<td>Baythroid</td>
<td>0.2</td>
<td>0.3</td>
<td>0.1</td>
<td>0.02</td>
<td>0.02</td>
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<td>0.01 (G); 0.3 (O)</td>
<td>0.3</td>
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<tr>
<td>Diflubenzuron</td>
<td>Micromite</td>
<td>3</td>
<td>0.5</td>
<td>0.1</td>
<td>0.01</td>
<td>0.01</td>
<td>3</td>
<td>0.01 (G); 2 (O)</td>
<td>1</td>
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<tr>
<td>Dimethoate</td>
<td>Dimethoate, Cygon</td>
<td>2</td>
<td>5</td>
<td>1.5</td>
<td>0.01</td>
<td>0.01</td>
<td>2</td>
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<tr>
<td>Diuron</td>
<td>Diuron, Direx, Karmex</td>
<td>0.05, 0.5 (L)</td>
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<td>0.01</td>
<td>0.8 (G), 0.05 (O)</td>
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<td>0.05 (G), 0.2 (O)</td>
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<tr>
<td>Fenbutatin Oxide</td>
<td>Vendex</td>
<td>20</td>
<td>5</td>
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<td>5</td>
<td>0.01</td>
<td>2</td>
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<tr>
<td>Ferbam</td>
<td>Ferbam</td>
<td>4</td>
<td>10 (T), 2 (O)</td>
<td>0.1</td>
<td>0.01</td>
<td>0.01</td>
<td>2</td>
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<td>Fuseloy-aluminum</td>
<td>Alliete</td>
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<td>50 (T), 20 (O)</td>
<td>9</td>
<td>75</td>
<td>75</td>
<td>150</td>
<td>0.01 (G); 4 (O)</td>
<td>10</td>
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<tr>
<td>Glyphosate</td>
<td>Roundup, Durango, Touchdown, and others</td>
<td>0.5</td>
<td>0.1</td>
<td>0.1 (G) 0.5 (O)</td>
<td>0.1 (G) 0.5 (O)</td>
<td>0.5</td>
<td>0.05</td>
<td>0.01 (G), 2 (O)</td>
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<tr>
<td>Imazalil</td>
<td>DECCOZIL EC-289, Freshgard 700, Fungafior 500EC</td>
<td>10</td>
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<td>8 (O), 15 (L)</td>
<td>5</td>
<td>4</td>
<td>4</td>
<td>15</td>
<td>5</td>
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<td>Malathion</td>
<td>Malathion, Atrapa, Fyfanon</td>
<td>8</td>
<td>7</td>
<td>0.1</td>
<td>2 (E)</td>
<td>2</td>
<td>2</td>
<td>7</td>
<td>0.01 (G); 4 (O)</td>
</tr>
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<td>Taiwan (G &amp; O only)</td>
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<tr>
<td>Metalaxyl-M, Mefenoxam</td>
<td>Ridomil Gold, Subdue, UltraFlourish</td>
<td>1</td>
<td>5</td>
<td>5</td>
<td>0.7 (E)</td>
<td>0.7</td>
<td>0.7</td>
<td>0.01</td>
<td>0.5</td>
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<tr>
<td>Metaldehyde</td>
<td>OR-Cal Slug &amp; Snail Bait</td>
<td>0.26</td>
<td>0.1</td>
<td>0.1</td>
<td>0.05</td>
<td>0.05</td>
<td>0.7</td>
<td>0.01</td>
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<td>Norflurazon</td>
<td>Slicam</td>
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<td>0.1</td>
<td>0.1</td>
<td>0.01</td>
<td>0.01</td>
<td>0.2</td>
<td>0.01</td>
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<tr>
<td>Oryzalin</td>
<td>Oryzalin, Surflan</td>
<td>0.05</td>
<td>0.1</td>
<td>0.1</td>
<td>0.01</td>
<td>0.01</td>
<td>0.08</td>
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<td>Oxamyl</td>
<td>Vydate</td>
<td>3</td>
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<td>0.1</td>
<td>0.01</td>
<td>5</td>
<td>0.01</td>
<td>0.5 (G), 1 (O)</td>
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<tr>
<td>Paraquat Dichloride</td>
<td>Paraquat, Gramoxone, Boa</td>
<td>0.05</td>
<td>0.02</td>
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<td>0.02</td>
<td>0.02</td>
<td>0.05</td>
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<tr>
<td>Piperonyl Butoxide</td>
<td>Evergreen EC</td>
<td>8 (O)</td>
<td>5 (O)</td>
<td>8 (O)</td>
<td>0.01 (G), 4 (O) (E)</td>
<td>0.01 (G), 4 (O)</td>
<td>3 (5 proposed)</td>
<td>0.01 (O)</td>
<td>5 (O)</td>
</tr>
<tr>
<td>Propargite</td>
<td>Comite, Omite</td>
<td>5 (G, L), 10 (O)</td>
<td>3</td>
<td>5</td>
<td>0.01 (G), 4 (O)</td>
<td>0.01 (G), 4 (O)</td>
<td>3 (5 proposed)</td>
<td>0.01</td>
<td>5</td>
</tr>
<tr>
<td>Pyrethrins</td>
<td>Pyrellin (+ Rothenone), Evergreen (+ Piperonyl Butoxide)</td>
<td>1 (O)</td>
<td>0.05 (O)</td>
<td>1 (O)</td>
<td>1 (O)</td>
<td>1 (O)</td>
<td>1 (O)</td>
<td>0.01 (O)</td>
<td>0.05 (O)</td>
</tr>
<tr>
<td>Pyridaben</td>
<td>Nexter</td>
<td>0.9</td>
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<td>0.3</td>
<td>0.3</td>
<td>1</td>
<td>0.01</td>
<td>2</td>
</tr>
<tr>
<td>Pyriproxyfen</td>
<td>Distance, Esteem, Knack</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
<td>0.6</td>
<td>0.6</td>
<td>2</td>
<td>0.01</td>
<td>0.3 (G), 0.5 (O)</td>
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<tr>
<td>Sethoxydim</td>
<td>Past Plus</td>
<td>0.5</td>
<td>0.1</td>
<td>0.1</td>
<td>0.1 (0.01 proposed)</td>
<td>0.1</td>
<td>0.5</td>
<td>0.01</td>
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</tr>
<tr>
<td>Simazine</td>
<td>Simazine, Princep, Sim-Trol</td>
<td>0.25 (G, O)</td>
<td>0.1</td>
<td>0.1</td>
<td>0.01</td>
<td>0.01</td>
<td>0.2</td>
<td>0.01</td>
<td></td>
</tr>
<tr>
<td>S OPP (2 Phenylphenol, O-phenylphenol, OPP)</td>
<td>DECCOSOL 125, FreshGard 5</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>0.01</td>
<td>10</td>
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<tr>
<td>Spinetoram</td>
<td>Delegate</td>
<td>0.3</td>
<td>0.07 (O), 0.15 (T)</td>
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<td>0.2 (0.02 G; 0.07 O proposed)</td>
<td>0.2</td>
<td>0.7</td>
<td>0.01 (G), 0.05 (O)</td>
<td>0.2</td>
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<tr>
<td>Trifluralin</td>
<td>Trifluralin, Treflan, Trilin</td>
<td>0.05</td>
<td>0.1</td>
<td>0.1</td>
<td>0.01</td>
<td>0.01</td>
<td>0.05</td>
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<td>zeta-cypermethrin</td>
<td>Mustang</td>
<td>0.35</td>
<td>0.5 (G, P), 0.3 (O, T, L)</td>
<td>1</td>
<td>2 (E)</td>
<td>2</td>
<td>2</td>
<td>0.3</td>
<td>2</td>
</tr>
</tbody>
</table>
Thank You!

• For more information, visit the UF Postharvest Website

http://irrec.ifas.ufl.edu/postharvest/