Bacillus subtilis, Strain QST 713
Use in Integrated Pest Management

D. W. Edgecomb
D. Mankar
AgraQuest, Inc. Davis, CA
Bacillus subtilis, Strain QST 713

- Isolated from Soil – CA Organic Peach Orchard
- Rod-shaped, Gram Positive, Aerobic, Motile Bacterium
- Naturally Occurring, No Genetic Modifications
- Unique – Patented Bacillus Strain
- Distinguished by Previously Unknown Metabolites
- First “Effective” Broad Spectrum Bio-fungicide
- Presidential Green Chemistry Award – 2003
- World Technology Award for Environment – 2004
- Now Registered Globally – Trade Name Serenade®
Global Registration Status

Bacillus subtilis (Strain QST 713)

<table>
<thead>
<tr>
<th>Year</th>
<th>Global Registration Status</th>
</tr>
</thead>
<tbody>
<tr>
<td>1999</td>
<td>Chile</td>
</tr>
<tr>
<td>2000</td>
<td>U.S.</td>
</tr>
<tr>
<td>2001</td>
<td>Mexico</td>
</tr>
<tr>
<td>2002</td>
<td>N.Z.</td>
</tr>
<tr>
<td>2003</td>
<td>P.Rico</td>
</tr>
<tr>
<td>2004</td>
<td>Japan</td>
</tr>
<tr>
<td>2005</td>
<td>Brazil</td>
</tr>
<tr>
<td>2006</td>
<td>Ecuador</td>
</tr>
</tbody>
</table>

Bold = Currently registered

Countries: Germany, Spain, Greece, Australia, Canada, Peru, Thailand, China, UK.
Bacillus subtilis, Strain QST 713

Registration Milestone

EU Annex 1 Inclusion

Bacillus subtilis strain QST 713, the active ingredient of biofungicide product Serenade® was voted for inclusion into Annex 1 of Directive 91/414/EEC at the EU Standing Committee on the Food Chain and Animal Health meeting on July 14, 2006.
Bacillus subtilis, Strain QST 713
Major Global Crops / Diseases

- Grapes
  - Gray Mold (Botrytis cinerea)
  - Powdery mildew (Uncinula necator)
- Bananas
  - Black Sigatoka (Mycosphaerella fijiensis)
  - Yellow Sigatoka (Mycosphaerella musicola)
- Tomato / Pepper
  - Bacterial Leaf Spot (Xanthomonas spp.)
  - Early Blight (Alternaria solani)
  - P. Mildew (Leveillula taurica)
- Cucurbitas
  - P. Mildew (Erysiphe / Sphaerotheca spp.)
- Lettuce
  - Leaf Drop (Sclerotinia spp.)
- Pome Fruit
  - Fire Blight (Erwinia amylovora)
  - Scab (Venturia inaequalis)
- Mango
  - Anthracnose (Colletotrichum gloeosporioides)
- Beans
  - White Mold (Sclerotinia sclerotiorum)
Bacillus subtilis, Strain QST 713

- Formulations: (AS) Aqueous Suspension
  (WP) Wettable Powder
- Active Ingredients Spec. = Minimum $1 \times 10^9$ cfu / gram
- Organic Formulations (OMRI, IMO, BCS, JAS)
- > Two Year Storage Stability
- Excellent Suspensibility
- Physical / Biological Tank Mix Compatibility with standard fungicides
Bacillus subtilis, Strain QST 713

**Applied Just Like Other Products**

**Novel Modes of Action**

- *B. subtilis* spores cover leaf surface & prevent fungal spores from penetrating and infecting the plant.
- Lipopeptide metabolites break down pathogen cell membranes, causing pathogen to collapse & die.
- Lipopeptides prevent pathogens from multiplying.

**SAR - Role in MOA**

Academic or Significant Contribution?

**No Special Equipment / Handling or Storage**
Profile of *B. subtilis*, QST 713 Metabolites

**LC-MS Profile of QST 713**

- **Iturins**
- **Surfactins**
- **Agrastatins/Plipastatins**

**Profile of B. subtilis, QST 713 Metabolites**
Botrytis 0% inhibition
Iturins 10ppm or
Agrastatins 50ppm

Botrytis 90% inhibition
Iturins 10ppm + Agrastatins 2.5ppm
Synergistic Action of Metabolites
**Bacillus subtilis, Strain QST 713**

**Novel / Multiple Modes of Action**

- *B.s.* spores – Colonize and Out-compete Pathogen
- Fungicidal Metabolites – Lipopeptides
  Present in Formulation / Produced by Colonizing *B.s.*
  Destroy Pathogen Spore & Mycelium Membranes
  Provide Barrier to Infection by Pathogen
- Optimal Use = Preventative Control Program
- New / Unique Resistance Management Tool
Bacillus subtilis, Strain QST 713

Use in Integrated Programs with Conventional Fungicides
**Bacillus subtilis, Strain QST 713**

Use in Integrated Disease Control Program - Japan

**IPM Program for Cucurbit Powdery Mildew (Sphaerotheca fuliginea)**

50% Reduction in Chemical Fungicide

<table>
<thead>
<tr>
<th></th>
<th>0 Day</th>
<th>7 Day</th>
<th>13 Day</th>
<th>20 Day</th>
<th>% Control</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Untreated</strong></td>
<td>Water</td>
<td>Water</td>
<td>Water</td>
<td>Water</td>
<td>---</td>
</tr>
<tr>
<td><strong>Chemical Repeating</strong></td>
<td>Std Fungicide</td>
<td>Std Fungicide</td>
<td>Std Fungicide</td>
<td>Std Fungicide</td>
<td>98.8</td>
</tr>
<tr>
<td><strong>Biological Repeating</strong></td>
<td>QST 713</td>
<td>QST 713</td>
<td>QST 713</td>
<td>QST 713</td>
<td>86.9</td>
</tr>
<tr>
<td><strong>Chemical + Biological</strong></td>
<td>Std Fungicide</td>
<td>QST 713</td>
<td>Std Fungicide</td>
<td>QST 713</td>
<td>98.6</td>
</tr>
<tr>
<td><strong>Chemical + Biological</strong></td>
<td>QST 713</td>
<td>Std Fungicide</td>
<td>QST 713</td>
<td>Std Fungicide</td>
<td>98.4</td>
</tr>
</tbody>
</table>

Chemical standard fungicide = Quinomethionat

---

50% Reduction in Chemical Fungicide
### Bacillus subtilis, Strain QST 713

#### Cucurbit Powdery Mildew (Sphaerotheca fuliginea) - USA

<table>
<thead>
<tr>
<th>Treatment (Amt/100 gal/acre)*</th>
<th>% Powdery Mildew</th>
<th>% Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chlorthalonil (2.25 lbs ai) alt with Azoxystrobin (0.25 lb ai)</td>
<td>2</td>
<td>4</td>
</tr>
<tr>
<td>QST 713 at 4 lbs (5 applns); followed by Azoxystrobin at 0.15 lb ai (3 applns)</td>
<td>1</td>
<td>5</td>
</tr>
<tr>
<td>QST 713 at 4 lbs</td>
<td>5</td>
<td>10</td>
</tr>
<tr>
<td>Untreated control</td>
<td>17</td>
<td>38</td>
</tr>
</tbody>
</table>

*Applied weekly on 5/3, 5/8, 5/15, 5/22, 5/30, 6/6, 6/13, and 6/19

---
Bacillus subtilis Strain QST 713 - Bananas

Black Sigatoka (*Mycosphaerella fijiensis*)

Requirement = 6 Functional Leaves at Harvest
### Bacillus subtilis, Strain QST 713 – “Sustainable” Black Sigatoka (Mycosphaerella fijiensis) Control Program in Bananas

Mindanao – The Philippines (March to August 2003)  
Small Plot / RCB Trial

<table>
<thead>
<tr>
<th>Treatment (rate / hectare)</th>
<th>Visible Streaks On Leaves</th>
<th>Functional Leaves At Harvest</th>
</tr>
</thead>
<tbody>
<tr>
<td>B. subtilis 2 liters</td>
<td>3.9 a</td>
<td>7.8 a</td>
</tr>
<tr>
<td>B. subtilis + mancozeb 2 + 0.9 liters</td>
<td>3.9 a</td>
<td>7.4 a</td>
</tr>
<tr>
<td>Standard Program Mancozeb – 1.8 liters</td>
<td>4.1 a</td>
<td>8.5 a</td>
</tr>
<tr>
<td>Untreated</td>
<td>3.4 b</td>
<td>4.9 b</td>
</tr>
</tbody>
</table>
**Product Positioning – Features**

- Effective “Non-Chemical” Option for Disease Control Programs
- Broad Spectrum Disease Control – Fungal and Bacterial Pathogens
- Tank mix and Rotation with Protectants / Systemics in Programs
- MRL Management / Sustainable / Organic Production Tool
- Resistance Management
- Single Site Compounds – e.g., Strobilurin and Triazole Fungicides
- Exempt From Tolerance / 0-Day PHI / No CODEX
  - Late Season, Pre-Harvest Option / No Residues
- Integrated Pest Management
- Not toxic to Non-target Organisms - *Honeybees, Lady Beetles, Lacewings, Parasitic Wasps, and Earthworms*
- Safe to Workers / Environment