Outline

• Mapping and confirmation of the location of an RB resistance gene in the Japanese cultivar ‘Hyuuga’

• Identifying genetic variation in MG 000 to X germplasm accessions
Types of Lesion

**Tan**
Susceptible lesion
Dillon

**RB**
Resistance lesion
Hyuuga

Antirust Consortium, Embrapa-Soja, Brazil
Attapulgus, GA
Dillon x Hyuuga
Attapulgus, GA

• Planted 3 Sept 2005
• 117 RILs + 6 checks
• 2 reps in a RCBD
• Natural and artificial infection
• Rated 1 Dec 2005
• Evaluate 10 leaflets / row
• 10X magnification

Dillon (S)         Hyuuga(R)
Dillon x Hyuuga
Severity Scale

Score = 1
No yellowing

Score = 3

Score = 4

Score = 5
Yellowing at the top
Dillon x Hyuuga
Greenhouse Confirmation

• Feb-April 2006

• Evaluate 117 RILs

• 3 replications in RCBD

• Manual inoculation

• Humidity chamber
Dillon x Hyuuga
Greenhouse Confirmation

- Observations
  - using 2-3 leaflets
  - lesion type
  - number of lesions
  - sporulation
RB Lesion Type Maps to LG C2

Consensus Map

<table>
<thead>
<tr>
<th>cM</th>
<th>Satt289</th>
<th>Satt134</th>
<th>Satt100</th>
<th>Satt708</th>
<th>Satt460</th>
<th>Satt307</th>
<th>Satt202</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.4</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1.2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1.5</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2.3</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3.5</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>4.9</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Satt289
Satt134
Satt100
Satt708
Satt460
Satt307
Satt202

Satt100
Satt134
Satt460
Rpp?(Hyuuga)
Satt307
Satt202
Verification
Benning x Hyuuga

• Screen 92 $F_{5:6}$ lines with markers in 4 cM region on LG-C2

• Select 16 lines homozygous for each parental allele

• Planted 3 reps in a RCBD

<table>
<thead>
<tr>
<th>Homozygous</th>
<th>Expect T:Mx:RB</th>
<th>Observed T:Mx:RB</th>
</tr>
</thead>
<tbody>
<tr>
<td>Benning</td>
<td>16:0:0</td>
<td>15:1:0</td>
</tr>
<tr>
<td>Hyuuga</td>
<td>0:0:16</td>
<td>0:0:16</td>
</tr>
</tbody>
</table>

T = tan; M = mix; RB = red-brown
# Benning x Hyuuga Lesion Type and Sporulation

<table>
<thead>
<tr>
<th>Lesion No.</th>
<th>Sporulation</th>
<th>Benning</th>
<th>Hyuuga</th>
<th>Tan RILs</th>
<th>RB RILs</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean(^{§}) (cm(^{-2}))</td>
<td>Range (cm(^{-2}))</td>
<td>Mean(^{§}) (cm(^{-2}))</td>
<td>Range (cm(^{-2}))</td>
<td>Mean(^{§}) (cm(^{-2}))</td>
<td>Range (cm(^{-2}))</td>
</tr>
<tr>
<td>Benning</td>
<td>6.9(^{x})</td>
<td>5.4 – 8.5</td>
<td>4</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Hyuuga</td>
<td>2.5(^{y})</td>
<td>1.5 – 4.5</td>
<td>0</td>
<td>4</td>
<td></td>
</tr>
<tr>
<td>Tan RILs</td>
<td>6.1(^{a})</td>
<td>5.4 – 6.9</td>
<td>16</td>
<td>0(^{a})</td>
<td></td>
</tr>
<tr>
<td>RB RILs</td>
<td>3.3(^{b})</td>
<td>1.9 – 5.6</td>
<td>2</td>
<td>14(^{b})</td>
<td></td>
</tr>
</tbody>
</table>

\(^{§}\) Means followed by a different letter are different at p = 0.05.
Markers Close to \( Rpp?Hyuuga \) on LG-C2

Consensus Map

\[
\begin{array}{c|c|c|c|c|c|c|c|c}
\text{cM} & \text{Satt289} & 0.5 & \text{Satt134} \\
0.4 & \text{Satt134} & 0.5 & \text{Satt100} \\
1.2 & \text{Satt100} & 3.9 & \text{Satt460} \\
1.5 & \text{Satt708} & 0.1 & \text{Satt079} \\
2.3 & \text{Satt460} & 1.9 & \text{Staga001} \\
1.0 & \text{Satt079} & 0.5 & \text{BARC-010457-00640} \\
1.1 & \text{Staga001} & 1.4 & \text{Satt307} \\
1.4 & \text{Satt307} & & \\
\end{array}
\]
## Graphical Genotypes of Benning x Hyuuga

### cM Positions

<table>
<thead>
<tr>
<th>Gene</th>
<th>Position</th>
</tr>
</thead>
<tbody>
<tr>
<td>Satt134</td>
<td>0.6</td>
</tr>
<tr>
<td>Satt319</td>
<td>0.6</td>
</tr>
<tr>
<td>Satt100</td>
<td>3.5</td>
</tr>
<tr>
<td>Sat_238</td>
<td>0.3</td>
</tr>
<tr>
<td>Satt460</td>
<td>0.1</td>
</tr>
<tr>
<td>Satt079</td>
<td>1.9</td>
</tr>
<tr>
<td>Staga001</td>
<td>0.05</td>
</tr>
<tr>
<td>SNP10457</td>
<td>1.4</td>
</tr>
<tr>
<td>Satt307</td>
<td>24.2</td>
</tr>
<tr>
<td>Satt371</td>
<td>6.4</td>
</tr>
</tbody>
</table>

### Phenotypes

<table>
<thead>
<tr>
<th>LT</th>
<th>Spor.</th>
<th>Spor.</th>
<th>Mix</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tan</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Tan</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Tan</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Tan</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>RB</td>
<td>0</td>
<td>1</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>RB</td>
<td>0</td>
<td>1</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Mix</td>
<td>0</td>
<td>0</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>RB</td>
<td>0</td>
<td>1</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>RB</td>
<td>0</td>
<td>1</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>RB</td>
<td>0</td>
<td>1</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>RB</td>
<td>0</td>
<td>1</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>RB</td>
<td>0</td>
<td>1</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>RB</td>
<td>0</td>
<td>1</td>
<td>0</td>
<td>1</td>
</tr>
</tbody>
</table>

### DNA Types

- **Hyuuga homozygous DNA**
- **Benning homozygous DNA**
- **Heterogeneous**

### Reference

- Graphical representation showing the genotypes at various cM positions with Benning and Hyuuga DNA types and phenotypic traits.
2006 Attapulgus Soybean Asian Rust Screening

- Planted 17Aug 06
- (24-hour photoperiod from planting to 15 Sept 06)
- Treated with agricultural grade streptomycin 10 times
- Natural and artificial infection (5 Oct, 12 Oct, 19 Oct, 24 Oct, 31 Oct)
- Rated 16-17 Nov 06
Rust Incidence Ratings

• 2005 rating scheme
  5 = rust lesions in upper canopy
  4 = rust lesions in middle canopy
  3 = rust lesions in lower canopy
  2 = only a few lesions on lower canopy
  1 = no apparent rust lesions

• 2006 rating scheme
  5 = 4 or more leaves with many lesions
  4 = 1 to 3 leaves with many lesions
  3 = 4 or more leaves with a few lesions
  2 = 1 to 3 leaves with a few lesions
  1 = no apparent rust lesions

<table>
<thead>
<tr>
<th>Cultivar</th>
<th>Mat Gp</th>
<th>ASR incidence</th>
<th>ASR lesion type</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dunbar</td>
<td>III</td>
<td>--- (4.3)</td>
<td>T</td>
</tr>
<tr>
<td>5601T</td>
<td>V</td>
<td>4.0 (4.7)</td>
<td>T</td>
</tr>
<tr>
<td>Dillon</td>
<td>VI</td>
<td>4.5 (3.0)</td>
<td>T</td>
</tr>
<tr>
<td>Musen</td>
<td>VI</td>
<td>4.5 (4.7)</td>
<td>T</td>
</tr>
<tr>
<td>Haskell</td>
<td>VII</td>
<td>5.0 (5.0)</td>
<td>T</td>
</tr>
<tr>
<td>Benning</td>
<td>VII</td>
<td>5.0 (5.0)</td>
<td>T</td>
</tr>
</tbody>
</table>
2006 Rust Incidence < 2.3
(25 of 45 accessions)

- MG I (3)
- MG II (1)
- MG III (1)
- MG V (1)
- MG VII (2)
- MG VIII (6)
- MG IX (11)

- No lesions (2)
- RB lesions (10)
  - RB w/o spores (4)
- Tan lesions (9)
- Mixed lesions (4)

- Russian Fed. (1)
- Japan (5)
- China (4)
- Vietnam (3)
- Indonesia (9)
- South Korea (1)
- Brazil (1)
- Moldova (1)
Acknowledgements

• Jose T. Yorinori
• Jose F. Toledo
• Randy Nelson
• Brian Diers
• Dan Phillips
• David Walker
• Layla Sconyers
• Attapulgus research staff (Billy Mills)

Tinker Foundation
United Soybean Board
American Seed Trade Association
GA Agricultural Experiment Stations