Genetic Control of Soybean Innate Immunity

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Plants can interact with different microorganisms

(Soybean leaf infected by *Fusarium solani*)

(Pathogenesis)
How do plants defend against different pathogens?

Plant Immunity

**MAMP-Triggered Immunity (MTI)**

**Effector-Triggered Immunity (ETI)**

*Nat Rev. 2010. 11:539*  
*Current Op in Microbiol 2011. 14:54*
Soybean (*Glycine max*)

- 91 million metric tons of soybean was produced in the U.S. during 2010, representing ~$39 billion.

- Total yield loss from a variety of diseases totaled ~ 7% of the U.S. soybean production, which equates to approximately $5 billion.

What do we know about soybean innate immunity?

Is soybean innate immunity genotype dependent?

Can we harness soybean genetic diversity to better understand innate immunity and find soybean cultivars with broad disease resistance?
Identification of soybean genotypes

We were interested in genotypes with:

A divergent MTI response (strong or weak)
Availability of mapping populations

Sandra Thibivilliers performed this screening

<table>
<thead>
<tr>
<th>Parents of population</th>
<th>Trait(s) tested</th>
<th>No. of lines in population</th>
<th>ROS ratio between parental lines after PAMPs treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td>PI 88788 x Williams</td>
<td>Brown stem rot resistance</td>
<td>147</td>
<td>1.5</td>
</tr>
<tr>
<td>Ripley x Spencer</td>
<td>Sudden death syndrome</td>
<td>91</td>
<td>1.3</td>
</tr>
<tr>
<td>PI 567374 x Omaha</td>
<td>Sudden death syndrome</td>
<td>96</td>
<td>1</td>
</tr>
<tr>
<td>Ripley x EF59</td>
<td>Sudden death syndrome</td>
<td>81</td>
<td>2.8</td>
</tr>
<tr>
<td>PI 153282 x NKS19-90</td>
<td>White mold, bean pod mottle virus, alfalfa mosaic virus</td>
<td>174</td>
<td>1.8</td>
</tr>
<tr>
<td>LD0-2817 x LDX01-165</td>
<td>Soybean Cyst Nematode</td>
<td>128</td>
<td>3.4</td>
</tr>
<tr>
<td>PI 84674 x LD00-3309</td>
<td>Partial resistance to soybean rust</td>
<td>167</td>
<td>1.1</td>
</tr>
</tbody>
</table>
Soybean MTI analysis

Leaves from 3-weeks old soybean plants (LD02817, LDX01-165, Ripley and EF59)

MAMP treatment (flag22, chitin or ch+flg22)

Oxidative Burst Analysis

MAMP treatment (ch+flg22)

Gene Expression Analysis after 30 min of MAMP treatment
Variation in the oxidative burst across soybean genotypes
Identification of MAMP-responsive genes

In collaboration with Dr. Robert Goldberg at University of California

**Soybean IV-gene-chip array**
- 50% of the soybean genes are printed
- One probe set per gen
- Gene expression

**Soybean- WT- ST array**
- All soybean (c.a. 66,000) genes are printed
- One probe set per exon of each gene
- Gene expression and splicing variants
Variation in the transcriptional profile across genotypes
Soybean-WT- ST Array validation
Variation in the transcriptional profile across genotypes
Overview of the expression of receptor-like kinases in MAMP-treated leaves of different soybean genotypes
What can we conclude?

- MTI can vary across different soybean genotypes: Clear genotypic variation.

- Is there a correlation between the genotypic variation in the MTI and pathogen resistance?

- The genotypes LD0-2817P and LDX01-1-65 show a clear divergent MTI response.
Pathogen resistance triggered by MAMP treatment

letters to nature

Bacterial disease resistance in Arabidopsis through flagellin perception

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Figure 2 Treatment with different bacterial extracts limits subsequent growth of Pst DC3000 in Ler-0 and fls2-17 plants. Ler-0 (open bars) and fls2-17 (filled bars) plants were either left untreated, pretreated for 24 h with 1 μM flg22 or pretreated with one of the following bacterial extracts: Pseudomonas syringae pv. syringae (Pss) (3 mg ml−1), Pseudomonas syringae pv. tomato DC3000 (PstDC3000) (3 mg ml−1) or Agrobacterium tumefaciens (A. tum.) (10 mg ml−1). Subsequent leaf infection with 105 c.f.u. ml−1 Pst DC3000 was performed, and bacterial growth was assessed 2 days after infection. Results shown are means ± s.e.m. (n = 8).
Leaves from 3-weeks old soybean plants (LD0-2817P or LDX01-1-65)

Leaf disc treated with MAMPs for 24 hrs

*Pseudomonas syringae pv glycinea*

Infection level

Detached leaves treated with MAMP for 24 hr

White mold (*Sclerotinia sclerotiorum*)

Infection level

Pathogenesis assay

Dr. Glen Hartman
U of Illinois
MTI can induce pathogen resistance in soybean
What can we conclude?

- A clear interaction between genotype and MAMP treatment
- Genotypes with strong MTI can show resistance to *P.seudomonas* and *S. sclerotiorum*. 
- Resistance is genotype- and treatment-dependent.
Can we map Quantitative Trait Loci (QTL) associated to soybean MTI?
Identification of QTL associated to MTI

97 mapping lines from LD02817 X LDX01-1-65

ROS production data

Selection of 25 MAMP-responsive genes, whose fold-changes in LD0-2817P were 2-fold bigger than LDX01-1-65

ROS QTL

expression QTL
Identification of QTL associated to MTI

Four QTLs: Two major effect- and two minor effect QTL

Expression QTL (eQTL)

NADPH oxidase
MAPK kinase

LRR-receptors
Transcription Factors

Transporters
2° metabolism

LRR-receptors
Transcription Factors
What can we conclude?

The variation is quantitatively inherited and can be associated to QTL.
Is the variation in the pathogen resistance inherited by the progeny?
Broad pathogen-resistance triggered by MTI is inherited
Conclusion

Our results demonstrate that the MTI responses among the four soybean genotypes varied depending on MAMP treatment and pathogen. Results also indicated that MTI variation among soybean genotypes was quantitatively inherited.
Can we find additional regulators of the plant-pathogen interaction?

Gm_Argonaute5 expression across a soybean mapping population from LD x LX

Does Argonaute 5 regulate MTI or plant-pathogen interaction?
Argonaute 5 may be involved in the resistance to *Pseudomonas* infection

Over-expression level of Argonaute 5

**Soybean**

**Arabidopsis**

*Pseudomonas* infection level
Investigation of soybean cytosine methylation patterns

Dr. Joseph Ecker

Dr. Bob Schmitz

Talk: Patterns and heritability of epiallelic variation in *Glycine max* at the Soybean Genomics Workshop (Tuesday, 10:20 am)
Acknowledgements

Stacey Lab

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Dr. Jin Qiu (U of Missouri)

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