The Analysis of Quantitative Resistance to Foliar Diseases of Maize

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Introduction

We are interested in the analysis of quantitative resistance - the dominant form of resistance utilized in cultivated maize. Very little is known about the molecular genetic or physiological basis of this type of resistance in maize or any other crop. Working with the foliar diseases gray leaf spot (GLS), southern leaf blight (SLB) and northern leaf blight (NLB), we are identifying and mapping new sources of resistance from diverse germplasm.

Use of a Disease Lesion Mimic Gene to Identify Components of the Defense Response

Rp1-D21 is an aberrant major disease resistance gene, which triggers the hypersensitive defense response (HR) in the absence of pathogen recognition, leading to constitutive necrotic spotting, called a disease lesion mimic phenotype. We have shown that the strength and quality of the phenotpye conferred by Rp1-D21 is variable, depending on the genetic background. **Table 1** below shows the **Diversity of the Rp1-D21 mediated HR in maize**. In this case the mutant tester - H95 heterozygous for Rp1-D21(called H95:Rp1-D21) - was crossed to each of the lines indicated. The height ratio is the average height of the mutant F1 segregants divided by the average height of the wild type F1 segregants

Table 1

Cross	Severity	Height ratio (M / WT)
B73 x Rp1-D21	4	177/245 = **0.71**
Mo17 x Rp1-D21	6	105/220 = **0.47**
B97 x Rp1-D21	2	190/235 = **0.81**
CML52 x Rp1-D21	5	145/240 = **0.6**
CM103 x Rp1-D21	5	140/230 = **0.61**
CML228 x Rp1-D21	5	148/235 = **0.63**
CML322 x Rp1-D21	7	100/225 = **0.44**
CML333 x Rp-D21	2	187/227 = **0.82**
HP301 x Rp1-D21	6	115/222 = **0.52**
IL14H x Rp1-D21	6	125/230 = **0.54**
Ki3 x Rp1-D21	5	150/250 = **0.6**
Ky21 x Rp1-D21	7	85/210 = **0.4**
M37W x Rp1-D21	10	dead/250 = **0**
M162W x Rp1-D21	8	65/245 = **0.26**
MS-71 x Rp1-D21	4	135/200 = **0.67**
NC350 x Rp1-D21	10	dead/230 = **0**
NC358 x Rp1-D21	6	124/236 = **0.52**
Oh7B x Rp1-D21	3	180/240 = **0.75**
Oh43 x Rp1-D21	2	155/200 = **0.79**
P39 x Rp1-D21	5	150/225 = **0.66**
TX303 x Rp1-D21	7	85/245 = **0.34**
Tzi8 x Rp1-D21	2	186/229 = **0.81**
Mo20W x Rp1-D21	10	dead/205 = **0**
A632 x Rp1-D21	1.5	190/205 = **0.92**

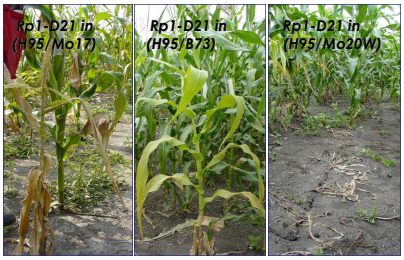


Fig 1. Effect of genetic background on the *Rp1-D21* phenotype

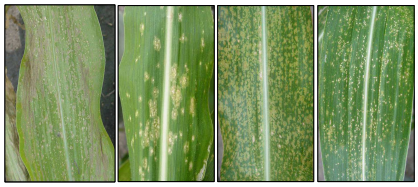


Fig 2. Different morphology, size and color of lesion of *Rp1-D21* lesions in different backgrounds.

We are now using this system to investigate the **genetic architecture of the defense response induced by *Rp1***. Since the Rp1-D21 phenotype is much stronger in an H95/Mo17 background than in an H95/B73 background, we were able to use the Mo17 x B73 –derived IBM population to map genomic regions that are responsible for this differential. Each of the IBM lines was crossed to H95:Rp1-D21. The resulting F1 seed segregated 1:1 for the presence of Rp1-D21 (since H95:Rp1-D21 is heterozygous for the gene). Each F1 population was scored in the field for several traits: for lesion severity on a 1-10 scale, average difference in height between wt and mt segregants and the average difference in flowering time between wt and mt segregants. The population was scored in replicated field trails in North Carolina and Indiana and in the greenhouse. The same major QTL, mapping on chromosome 10 and designated ***Hrml1***, for HR modulating locus-1 , was identified in each environment for each of these traits.

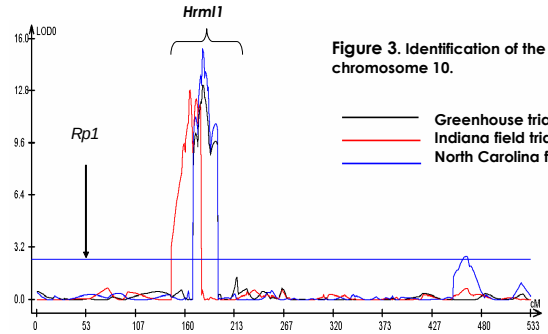


Figure 3. Identification of the *Hrml1* locus on chromosome 10.

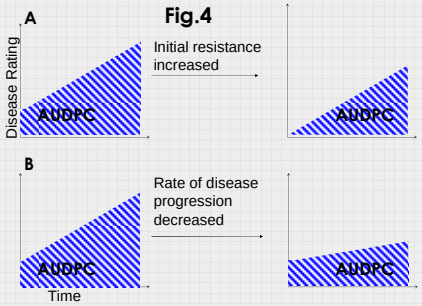
Greenhouse trial
Indiana field trial
North Carolina field trial

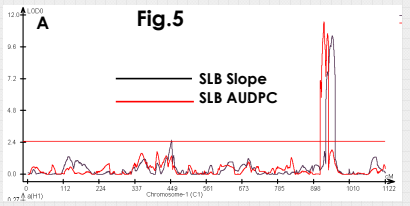
By a similar scheme, we are planning to perform more detailed analyses using the new nested association mapping (NAM) population to characterize the **genetic architecture of the defense response.**

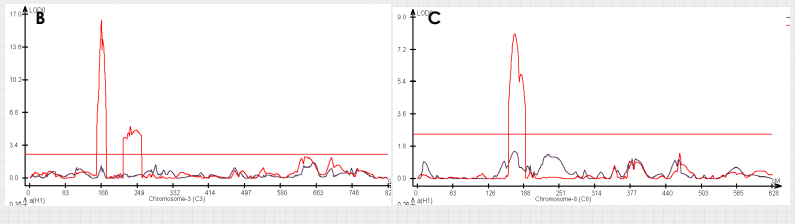
Dissection of Components of Quantitative Disease Resistance

We have mapped resistance QTL for SLB, GLS and NLB in several populations, including the IBM and NAM populations (see posters 215, 216, 220 and refs 1-5). In general we use a trait known as "Area under disease progress curve"- AUDPC as a measure of disease resistance. This is a measure of disease over time.

In general, there are three ways to decrease AUDPC (and increase resistance): **1**. Initial resistance could increase but rate of disease progression remains same (**Fig 4A**). 2. Initial resistance stays the same but rate of progression decreases (**Fig. 4B**). 3. Some mixture of the above. With this in mind we reanlysed our SLB data collected on the IBM population, **using slope (~disease progression) as a trait**. **Figure 5** shows the three major QTL for SLB AUDPC on chromosomes 1 (Fig. 5A), 3 (Fig. 5B) and 8 (Fig. 5C). The chm 1 SLB QTL colocalizes with the main QTL for SLB slope while there is no effect on slope at the other two SLB AUDPC QTL. From this it can be inferred that the **chm3 and chm8 SLB AUDPC QTL increase the initial level of resistance while the chm1 SLB AUDPC QTL may function by decreasing the disease progression rate**.







Using Image Analysis to Accurately Score Disease

One of the main challenges in the analysis of quantitative disease resistance is simply how to score the resistance trait accurately and efficiently. With a foliar blight disease, the basic trait scored is necrotic leaf area but it is challenging to accurately assess this quickly. We use a semi-qualitative scale (**Fig. 6**) which makes for relatively rapid scoring but is likely non-linear. We are investigatingusing image analysis to more acurately score disease phenotypes (**Fig. 7**). A segregating population derived from a B73 x De811 crosswas assessed using the normal scoring system and using image analysis. Comparion of QTL mapping analyses based on these two data sets indicate that different sets of QTL are identified (**Fig. 8**)

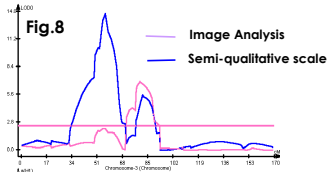
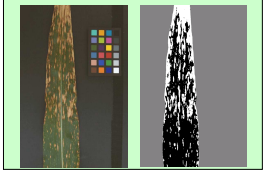


Fig.6 Guide for scoring SLB

9- No evidence of leaf blight
8- A few spots on the lower leaves
7-A few spots on the ear leaf
6-More spots on the ear leaf but the lesions don't coalesce
5-Lesions on the ear leaf have grown together , particularly at the tip of the leaf to give quite large necrotic areas
4-Lesions on the leaf above the ear leaf have grown together too
3-Leaf above the ear leaf almost completely dead
2-Almost all tissue on the plant dead
1-Everything brown

Fig.7 Image analysis used to determine total necrotic leaf area



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