Ottawa, Ontario, Canada

Eastern Cereal and Oilseed Research Centre

Expression of different Rp genes to common rust of corn in Ottawa, Ontario

Zhu, X., L. M. Reid, T. Woldmariam, C.Voloaca, and J. Wu

Twenty-eight H95 background inbred lines with different *Rp* resistance genes for common rust of corn (*Puccinia sorghi*), and resistant inbred B108 and B113; four inbred lines (H95, A679, CO348, and CO4428) with different level of resistances were evaluated for the expression of resistance in our common rust nursery at Agriculture and Agri-Food Canada (AAFC), Eastern Cereal and Oilseed Research Centre (ECORC), Ottawa, Ontario in 2002 and 2010. Each genotype was planted in a one-row plot with single row plots of 15 plants. To achieve good infection levels plants are inoculated twice, once at the 6-8 leaf stage and again at the 10-12 leaf stage. Two ml of urediniospores suspension ( $2.5 \times 10^5$  spores/ml) is injected into the whorl (plant tissues are not damaged) of each plant each time by using a graduated, 10 ml, self-refilling, automatic vaccinator attached to a 2.5 L backpack container (Nasco Co., Fort Akinson, WI). If there was no rain after inoculation, the plots were irrigated for 10-15 minutes every day to provide suitable environment for disease to develop.

*P. sorghi* is an obligatory parasite, so the pathogen must be kept alive and cannot be stored in a dormant state as with the other pathogens outlined in this guide. Sweet corn, which is quite susceptible to common rust, is a useful overwintering host when planted in the greenhouse. Urediniospores can survive up to six months in the refrigerator (4-6  $C^{\circ}$ );

so from January to mid-June, urediniospores are collected from greenhouse grown sweet corn plants. Prior to inoculation, all the collected spores are mixed together and divided into two batches for inoculation at two stages of plant development. Before inoculation, the spores are suspended in sterilized water with 1 ml of 0.5% Tween 20 added to every 500 ml of water to aid followed by agitation (stirring) for 15 minutes. The suspension is then filtered through two layers of cheesecloth to discard any big particles or plant tissues and diluted with sterilized water to  $2.5 \times 10^5$  spores/ml.

At the soft dough stage about 3 weeks after silk emergence, plants are rated for both specific (based on pustule type) and general resistance (based on disease severity). Six pustule types (Fig. 8) are classified as: I = immune, no infection symptoms; HR = hypersensitive response, yellowish or brownish pin-point fleck symptoms; R = resistant, vellowish or brown necrotic borders, no typical pustules and urediniospores; MR = moderately resistant, small pustules covered by the host cuticle, no broken-out urediniospores; MS = moderately susceptible, some pustules and urediniospores broken out of the leaf cuticle; and S = susceptible, large pustules, mostly link together and broken out with lots of urediniospores. Disease severity is evaluated by using a rating scale based on the percentage of leaf area with visible symptoms where 1 = no symptoms, 2 = < 1%, 3 = 1 - 5%, 4 = 6 - 20%, 5 = 21 - 50%, 6 = > 50% of leaf area with symptoms, and, 7= plant is dead. It can be difficult to rate the percentage of leaf area infected; a more simple and practical scale to use is: 1= no symptoms; 2= a few lesions; 3= many lesions, but they are not linked together; 4= a lot of lesions, some linked together to form a necrotic (dead) area; 5= necrotic areas linked together and a few leaf tips are dead; 6= 50% of the leaf tips are dead; and, 7 = most of the leaves are dead.

2002 H95 serial of Rp lines were donated from the Kansas State University to AAFC. Most lines were uniform in agronomy traits, such as plant height, ear height, tassel shape and silk color. However, they are somewhat different with H95 lines, might be not enough back-cross generations. Their resistance to common rust also showed some separation (Table 1). Best resistant plants from each Rp lines were selfed for future breeding use. Their resistance had some changes in 2010 (Table 1).

H95(*Rp3-A*), H95(*Rp-G15c*), H95(*Rp-G5*), H95(*Rp-G5JCa*), H95(*Rp-G6J1*), and H95(*Rp-GFJ*) were immune lines with very few and small hypersensitive response. H95(*Rp1-B*), H95(*Rp1-Dj4*), H95(*Rp1-Jc13a*), H95(*Rp1-JFC1*), H95(*Rp1-K*), H95(*Rp1-Kr3*), H95(*Rp1-Kr4*), H95(*Rp5*), and B113 showed excellent HR or R pustule type and disease severity rating 2; H95(*Rp1-A*), H95(*Rp1-C*), H95(*Rp1-D*), H95(*Rp1-M*), and B108 showed R or MR pustule types and disease severity rating 3; H95(*Rp1-J*), H95(*Rp1-Kr1-J92*), H95(*Rp4-A*), H95(*Rp4-B*), H95(*Rp5-D*), H95(*Rp7*), H95(*Rp8-A*), and CO428 showed MS or S pustule types, however, they still have some resistance because of less percentage of leaf area infected.

*Rp-G5, Rp-G5JCa, Rp-G15c, Rp-G6J1, Rp1-Jc13a,* and *Rp1-K* were used in AAFC corn program, back-crossed to CO325, CM105, and CO388. Early maturity resistance lines to common rust will be released in 2011 and 2012.

Table 1. Common rust of corn screening results of inbred lines with different resistance

Genotypes	2002		2010	
	Rating	Pustule types*	Rating	Pustule types*
H95	5	S		
H95( <i>Rp1-A</i> )	3	MR/R	3	R
H95( <i>Rp1-B</i> )	2	R	2	R
H95( <i>Rp1-C</i> )	3	HR/R	3	R
H95( <i>Rp1-D</i> )	3	R/MR	3	R/MR
H95( <i>Rp1-Dj4</i> )	2	HR/R	2	HR
H95( <i>Rp1-FJ6q</i> )	2	R	2	MR/MS
H95( <i>Rp1-J</i> )	4	MR	3	MS
H95( <i>Rp1-Jc13a</i> )	2	I/HR	2	I/HR
H95( <i>Rp1-JFC1</i> )	2	HR	2	R
H95( <i>Rp1-K</i> )	2	HR	2	HR
H95( <i>Rp1-Kr1-J6</i> )	3	HR		
H95( <i>Rp1-Kr1-J92</i> )	3	MR/MS	3	MR/MS
H95( <i>Rp1-Kr3</i> )	2	R	2	R
H95( <i>Rp1-Kr4</i> )	2	R	2	R
H95( <i>Rp1-M</i> )	3	MR/MS	3	MR
H95( <i>Rp3-A</i> )	2	I/HR	1	Ι
H95( <i>Rp4-A</i> )	4	MS	4	MR/MS
H95( <i>Rp4-B</i> )	3	MR	3	MR/MS
H95( <i>Rp5</i> )	4	MR	2	R
H95( <i>Rp5-D</i> )	4	MS/S	3	MS/S
H95( <i>Rp6</i> )	2	HR		
H95( <i>Rp7</i> )	5	MS	4	MS
H95( <i>Rp8-A</i> )	4	MS	3	MR/MS
H95( <i>Rp-G15c</i> )	2	I/HR	1	Ι
H95( <i>Rp-G5</i> )	2	I/HR	1	Ι
H95( <i>Rp-G5JCa</i> )	2	I/HR	1	Ι
H95( <i>Rp-G6J1</i> )	2	I/HR	1	Ι
H95( <i>Rp-GFJ</i> )	2	I/HR	1	Ι
B108			3	R
B113	3	R	2	R
A679	5	S	4	S
CO348	5	S	5	S
CO428	4	S	3	S

in 2002 and 2010 in Ottawa, Ontario, Canada

\* I = immune, no infection symptoms; HR = hypersensitive response, yellowish or brownish pin-point fleck symptoms; R = resistant, yellowish or brown necrotic borders, no typical pustules and urediniospores; MR = moderately resistant, small pustules covered by the host cuticle, no broken-out urediniospores; MS = moderately susceptible, some pustules and urediniospores broken out of the leaf cuticle; and S = susceptible, large pustules, mostly link together and broken out with lots of urediniospores.