Evaluation of Quantitative Resistance to Soybean Rust (*Phakopsora pachyrhizi*) in Vietnam

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ABSTRACT

Soybean rust (*Phakopsora pachyrhizi* Sydow) has been reported throughout the southern states and a few Midwestern states in the USA. Many efforts have been made to screen soybean germplasm for resistance to the disease, and race-specific and non race-specific resistance has been identified. The objective of this study was to compare quantitative resistance using different disease severity assessments. Twelve soybean accessions, including pre-selected plant introductions (PIs) and local checks, were studied in repeated experiments conducted in 2005 and 2006 at NPRI. Three severity assessments were used to evaluate the resistance. Data was used to calculate area under disease progress curve (AUDPC) values for each assessment method. In addition, AUDPC of each canopy levels and correlations with overall AUDPC were also computed for the evaluation of simplicity and reliability. The results demonstrated that all methods were useful in identifying resistant accessions. One of the methods differentiated disease levels among soybean genotypes better than the other two methods. Disease severity assessment of the mid and upper canopy provided better separation of accessions in terms of quantifying resistance than did assessments of the lower canopy.

MATERIALS AND METHODS

***Plant materials*:**

Ten plant introductions (PIs) and cultivars with various levels of resistance and two local checks were planted in the fall 2005 and repeated in spring 2006 at NPPI. The experimental design was randomized complete block (RCB) with four replicates. Each entry was grown in 1-m plots with spacing between and within rows of 60 and 10 cm, respectively.

Seedling stage in the field

Field plot at NPPI, 2005

Inoculation method:

Plants were inoculated twice at growth stages V6 and R1 with a spore suspension ($5x10^4$ spores/ml) of a local unpurified isolate of the fungus. Prior to inoculation, the plots were thoroughly irrigated and covered with plastic sheets to maintain humidity for 12–16 h. The following morning, humidity inside was checked prior to removing the plastic sheets. Five plants each entry were used for disease rating.

Leaf showing severe infection

Incubation with plastic sheets

Disease assessments:

Method 1: Using a 3-digit rating system adopted by the International Workshop Group on Soybean Rust (IWGSR) in 1976. Disease severity was assessed three times, 1 week prior to flowering, 15 days post flowering, and 10 days post pod formation. Infected leaf at lower, middle, and upper canopy (Fig. 1) on each plant was scored for density of lesions using a 1-4 scale (Table 1). Sporulation of pustules was also scored.

Method 2: Disease severity was assessed at the reproductive growth stages R3, R4, R5, and R6 on five plants at three leaf positions (Fig. 1), using a 1-4 scale (Table 1).

Method 3: Disease severity was also assessed at the same reproductive growth stages, R3, R4, R5, and R6, at each leaf position (Fig. 1), using percentage of infected leaf areas (Miles et al., 2006).

In each disease assessment, overall AUDPC (Shaner and Finney, 1977) of each accession was computed for statistical analysis using the GLM procedure of SAS 9.1 (SAS Institute, Cary NY). In addition, AUDPC of each leaf position was also computed and utilized for correlation analysis between overall AUDPC and AUDPC of leaf position.

Figure 1. Canopy levels

Table 1. Disease rating of a 1-4 scale.

Scale	Disease severity
1	no symptoms
2	light number of lesions (1-100)
3	moderate number of lesions (101-500)
4	heavy number of lesions (>500)

RESULTS AND DISCUSSION

Table 2. Area under disease progress curve (AUDPC) values of 12 soybean accessions using in three disease assessment methods.

Method 1			Method 2			Method 3		
Accession	AUDPC		Accession	AUDPC		Accession	AUDPC	
PI189402	249	a	PI189402	401	a	PI189402	4596	a
PI398998	205	b	PI459025F	355	b	PI459025F	4487	a
Pana	200	bc	Williams 82	317	c	Williams 82	3650	b
PI594172A	199	bc	PI200492	274	d	PI200492	2981	c
PI200492	199	bc	Pana	268	d	PI462312	2748	cd
PI459025F	196	bcd	PI462312	265	d	PI594172A	2722	cd
Williams 82	196	bcd	PI398998	254	de	Pana	2641	cd
PI462312	195	bcd	PI561287A	254	de	PI561287A	2423	cde
Cao Bang	194	bcd	DT2000	243	e	PI398998	2300	de
PI230970	187	cd	PI594172A	242	e	Cao Bang	1858	ef
PI561287A	183	d	Cao Bang	215	f	DT2000	1615	f
DT2000	144	e	PI230970	206	f	PI230970	1459	f
LSD .05	*14.0*		*LSD .05*	*21.3*		*LSD .05*	*605*	

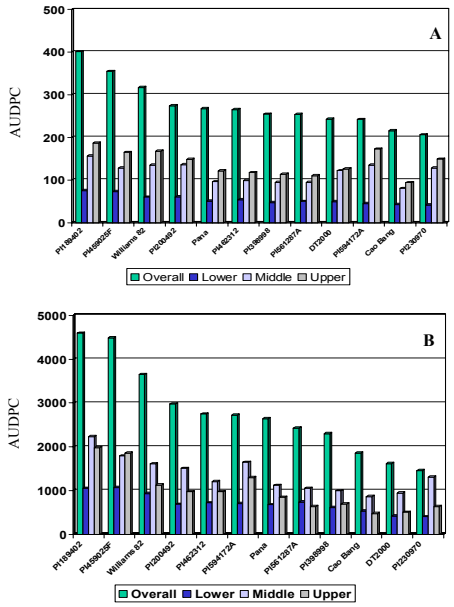

Figure 2. Overall area under disease progress curve (AUDPC) and AUDPC of each canopy position of 12 soybean genotypes evaluated Method 2 (Panel A) and Method 3 (Panel B).

Table 3. Correlation coefficients between overall AUDPC and disease severity at different canopy positions in Method 2 and 3.

Method 2				
	Overall	Lower canopy	Middle canopy	Upper canopy
Overall	1	0.378**	0.706***	0.820***
Lower canopy		1	0.207 ns	0.104 ns
Middle canopy			1	0.905***
Upper canopy				1
Method 3				
	Overall	Lower canopy	Middle canopy	Upper canopy
Overall	1	0.394**	0.898***	0.936***
Lower canopy		1	0.229*	0.206 ns
Middle canopy			1	0.894***
Upper canopy				1

Single-gene resistance in soybean was well documented, but has not been found durable when these resistant sources were deployed in the field (Kochman, 1977; Bromfield, 1984). It was believed that the fungus *P. pachyrhizi* with considerable variation in virulence among isolates was able to effectively overcome single-gene resistance in soybean (Hartman et al., 2005). Partial resistance to soybean rust was reported in soybean (Wang and Hartman, 1992). However, identification and utilization of the partial resistance may be time-consuming and difficult to incorporate into breeding programs because of its evaluation methods, especially in the field conditions.

Most of greenhouse assays, which were developed for the identification of race-specific resistance with purified isolates of the pathogen, used soybean seedlings at 14-18 days old; meanwhile partial resistance needs to be quantitatively measured for adult soybean in the field, where rust disease severity normally increases during the reproductive growth stages.

Area under disease progress curve (AUDPC) (Shaner and Finney, 1977) has been commonly utilized for quantitative assessments of disease resistance in crop plants. In the present study, we compared three disease severity assessment methods based upon AUDPC values to evaluate partial resistance to rust in the field condition. Ten selected soybean accessions, along with local checks, were grown in a disease nursery of NPPI, Vietnam.

The results (Table 2) showed that disease progress were significantly differentiated among accessions studied. However, when comparing the differentiation by each method, the greatest difference in AUDPC values between cv. Williams 82 (the susceptible) and DT2000 (the resistant) was observed with Method 3, while less differences in AUDPC were observed with Method 2 and 1. It indicates that Method 3, in which AUDPC were calculated based on the percentage of infected leaf area at the reproductive growth stages, enabled us to effectively evaluate disease reactions to soybean rust in the field.

In order to simplify the rating process, correlations of disease severity rating at each canopy position and overall severity were calculated (Table 3). The results suggest that severity assessments at lower canopy position did not have considerable contributions to overall AUDPC values of accessions. In contrast, severity assessments at middle and upper positions contributed to greater differentiations in terms of quantifying disease resistance to soybean rust (Fig. 2).

In summary, the use of percentage of infected leaf areas for AUDPC estimation (Method 3) was useful to effectively evaluate quantitative resistance to soybean rust in the field conditions. Severity assessments at either middle or upper canopy position were significant contributions to AUDPC values. It helped rating process in the field with less time consumption without a sacrifice of reliability of resistance evaluation.

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