Population Changes of Pseudomonas glycinea on Germinating Soybean Seeds

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ABSTRACT

Significantly larger populations of two races of *Pseudomonas glycinea* developed on germinating seeds of a susceptible cultivar, but not on a resistant soybean cultivar, thus demonstrating pre-emergence host-pathogen specificity. Host emergence was reduced in some race-cultivar combinations in the greenhouse, but not in the field.

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Additional key words: seedborne bacteria, resistance, bacterial blight of soybean.

Bacterial blight of soybean [*Glycine max* (L.) Merr.] caused by *Pseudomonas glycinea* Coerper is a widespread seedborne disease of soybean in the upper midwest (3). Although we were unable to locate published information on growth and survival of bacterial pathogens on pre-emerged seedlings of their hosts, germinating seeds have been shown to produce substances which inhibit growth of microorganisms in general (1, 4). On the other hand, most seeds exude materials which are suitable for the growth of bacteria (2). At least one report indicates severe stand reductions of soybeans due to *P. glycinea* (5) and since the fate of pathogenic bacteria residing within or upon seeds has implications to the epidemiology of soybean blight as well as stand acceptibility and seedling quality, we monitored population changes of *P. glycinea* on susceptible and resistant soybean cultivars during seed germination. This effort was made to also ascertain the earlier period at which race-cultivar specificity might be manifested.

MATERIALS AND METHODS.—A supply of sterile seeds was obtained by harvesting pods 2-3 wk prior to maturity, drying at room temp for 3-4 wk, surface disinfesting in 2.5% sodium hypochlorite for 5 min and storing in covered sterile beakers. Seeds of cultivars Acme and Chippewa 64 were removed from pods and dipped in bacterial suspension (10^8 cells/ml) of race 1 or race 2 of *P. glycinea,* blotted, and placed on moist sterile filter paper in petri dishes. From this lot, washings (1 hr agitation in sterile water) of each of four seeds were assayed via dilution plates for numbers of bacteria per seed. After 48 h, an additional four seeds were chosen from the incubated group and assayed in the same manner. The experiment was done at two time periods: the first 48 hours of germination (0-48 h) and the second 48 h (48-96 h) and at two temp (18 C and 27 C). The entire experiment was repeated twice, and the data were reexpressed logarithmically and combined for statistical analysis.

RESULTS.—*Pseudomonas glycinea* was not detected on the control (noninoculated) seeds. Both races were able to increase on germinating seeds of each cultivar. Race 2 was able to achieve a significantly higher population change in a 48-h test period than was race 1, regardless of seed substrate. Also, population changes were almost 100 times greater on Acme (susceptible to race 1) than on Chippewa 64 (resistant to race 1) by the end of 48 h, regardless of the race involved. The first 48 h of germination were also more conducive to a higher population change than was the second 48-h period. Temperature had the largest effect on population change

and was involved in every significant interaction (Table 1).

Based on these results, the data were further analyzed by division into temp treatment groups. Variance within experiments at 18 C was too great to assign significant differences; however the trends appeared similar to those found at 27 C.

Inoculated seeds were also planted in vermiculite in a greenhouse (about 22 C). The experiment was repeated twice, resulting in eight replications of 20 seeds per treatment. Inoculation with race 1 resulted in a 20% reduction in emergence of Acme (susceptible) and no emergence reduction of Chippewa 64 (resistant). Race 2 inoculation resulted in a nonsignificant emergence reduction of Acme (susceptible) and a 12% emergence reduction of Chippewa 64 (susceptible). These reductions were significant at the 0.05 level based on Tukey's test for comparison of means. No significant differences were observed in field emergence of inoculated seeds. Field tests also included the cultivar Merit (which has the same disease reaction as Chippewa 64), and race 5, to which Acme is resistant and Chippewa 64 and Merit are intermediate.

TABLE 1. Population change of *Pseudomonas glycinea* on germinating soybean seeds as affected by main effects and significant interactions

Treatment	Mean log population change	
	27 C	18 C
Cultivar		
Acme	7.399 A[a]	4.560 A
Chippewa 64	7.036 B	0.342 A
Race		
Race 1	7.118 B	0.838 A
Race 2	7.317 A	4.407 A
Time period		
0-48 hr	7.047 B	4.779 A
48-96 hr	7.332 A	1.172 A
Cultivar × race		
Acme-Race 1	7.420 A	3.263 A
Acme-Race 2	7.377 A	5.862 A
Chippewa 64-Race 1	6.815 B	−1.584 A
Chippewa 64-Race 2	7.257 A	2.956 A

[a]Means followed by different letters are significantly different, $P = 0.05$ based on Tukey's test for comparison of means. Comparisons are made within treatments and temp groups. Means are based on four replications in each of two experiments.

DISCUSSION.—Although we were able to show cases of significantly reduced stands in the greenhouse, we were unable to demonstrate such a reduction under field conditions, based on combinations of three races and three cultivars. Our results establish that *P. glycinea* can multiply on soybean seeds during germination regardless of the inherent resistance or susceptibility of adult plants, thus increasing the initial reservoir of inoculum before emergence. This pre-emergence increase may influence efficiency of transmission of the pathogen from seed to seedling. The presence of a statistically significant cultivar × race interaction at 27 C appears to indicate an expression of race specificity during the first 96 h of germination.

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