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## Abstract

Charcoal rot of soybean [*Glycine max* (L.) Merr.], caused by *Macrophomina phaseolina* (Tassi) Goid causes yield loss in soybean production worldwide. Development of resistant varieties has been limited, in part, due to a lack of consistent and reliable *M. phaseolina* assessment methodology. Growth stage at assessment is an important consideration. Four soybean lines (DT97-4290, DS97-84-1, Magellan, and A5979) were evaluated for their disease response at four stages of development (R6, R7, R8, and R8 + 1 wk) in order to determine the optimal developmental stage for disease assessment. The soybean lines were sown on 6 May 2005 at Stoneville, MS and artificially inoculated with *M. phaseolina* at planting. The experimental design was a 4 x 4 factorial in a completely randomized design, where each line-growth stage combination had a minimum of 16 replications. Single-plant assessments were made by estimating disease severity present in root and stem tissues using a scale of 1 (resistant) to 5 (susceptible). Over all lines, ratings taken at R8 and R8 + 1 wk had higher disease severities than those taken at R6 and R7. Ratings at R7 had higher severity than those taken at R6. Over all growth stages, DS97-84-1 was most susceptible and DT97-4290 was most resistant. There was no difference among lines at R6 but significant differences among lines occurred at the other three stages. Analysis indicated that stages R7 and R8 were best for distinguishing genotypic differences. Selecting which of the two stages to use may depend on the specific objectives of the research. For example, differentiating among levels of resistance might be most effective at R8, when DT97-4290 was more resistant than A5979, Magellan, and DS97-84-1. And differentiating among levels of susceptibility might be most effective at R7, when DS97-84-1 was more susceptible than Magellan, A5979, and DT97-4290.

Table 1. Mean growth stage scores for root and stem severity (RSS) ratings of charcoal rot over all soybean lines (DS97-84-1, Magellan, A5979, and DT97-4290) at Stoneville, MS in 2005.

Growth Stage	Mean RSS Scores
R8 + 1 week	3.0 a
R8	2.7 a
R7	1.6 b
R6	1.1 c

Mean RSS scores followed by the same letter are not different at  $P \leq 0.05$ .



Figure 1. Soybean plant inoculated with *M. phaseolina* at planting and dying prematurely prior to R8

## Introduction

Charcoal rot of soybean [*Glycine max* (L.) Merr.], caused by *Macrophomina phaseolina* (Tassi) Goid causes yield loss by early plant death (Figure 1) in soybean production worldwide. Development of resistant varieties has been limited, in part, due to a lack of consistent and reliable *M. phaseolina* assessment methodology. Growth stage at assessment is an important consideration when conducting disease severity assays. The purpose of this research was to determine the effect of soybean growth stage on disease assessment for charcoal rot and then to compare multiple soybean lines and determine at which growth stage maximum differentiation of disease severity is possible.

Table 2. Soybean line mean root and stem severity (RSS) scores for charcoal rot over all growth stages (R6, R7, R8, R8+1 week) at Stoneville, MS in 2005.

Soybean Line	Mean RSS Scores
DS97-84-1	3.0 a
Magellan	2.1 b
A5979	2.0 b
DT97-4290	1.4 c

Mean RSS scores followed by the same letter are not different at  $P \leq 0.05$ .

## Materials and Methods

Four soybean lines (DT97-4290, MG IV; DS97-84-1, MG IV; Magellan, MG IV; and A5979, MG V) were evaluated for their disease response at four stages of development [R6, R7, R8 (Fehr and Caviness, 1977), and R8 + 1 wk] in order to determine the optimal developmental stage for disease assessment. The soybean lines were sown on 6 May 2005 at Stoneville, MS in the field in a fine-loamy Bosket soil and artificially inoculated with *M. phaseolina* at planting. Five seed per plot were sown with seed jabber and thinned to one plant per plot at the V2 growth stage (Fehr and Caviness, 1977). Single-plant plots were 0.31 m apart in rows 0.61 m wide. The experimental design was a 4 x 4 factorial in a completely randomized design, where each line-growth stage combination had a minimum of 16 replications. Single-plant assessments were made by estimating disease severity present in root and stem tissues (root and stem severity; RSS; Mengistu et al., 2007) using a scale of 1 (resistant) to 5 (susceptible). Assessments were made after plants were uprooted from the soil at the appropriate growth stages and longitudinally split along the stem and tap root. Each stem was visually rated for intensity of discoloration and microsclerotial load covering the vascular and cortical tissues (Figure 2). The 1-5 RSS scale was divided into four classifications (Paris et al., 2006) for use at R7; resistant = 1, moderately resistant > 1 and ≤ 2, moderately susceptible > 2 and < 3, and susceptible ≥ 3. Analysis of variance was performed on the data and mean RSS scores were differentiated using Tukey's multiple pairwise comparison procedure. Although the 1-5 rating scale might be considered non-parametric, the "central limit theorem" states that means tend to be normally distributed even when the data that comprise them are not. Means based on a sufficient number of replications closely approximate normality so as to allow the use of appropriate parametric methods. All data were analyzed using Statistix 8 (Analytical Software, Tallahassee, FL).

Table 3. Mean root and stem severity (RSS) scores for charcoal rot according to soybean line and growth stage at Stoneville, MS in 2005.

Soybean Line	R6	R7	R8	R8 + one week
DT97-4290	1.1 f	1.0 f	1.5 ef	1.9 def
A5979	1.0 f	1.5 ef	3.0 bcd	2.4 cde
Magellan	1.0 f	1.0 f	2.9 bcd	3.3 abc
DS97-84-1	1.2 f	2.8 bcd	3.5 ab	4.4 a

Mean RSS scores followed by the same letter are not different at  $P \leq 0.05$ .



Figure 2. Lower stems and roots of soybean plants inoculated at planting with *M. phaseolina* and split longitudinally after harvest at R7. RSS ratings from left to right are 1, 3, and 5.

## Results and Discussion

Over all lines, ratings taken at R8 and R8 + 1 wk had higher disease severities than those taken at R6 and R7 and ratings at R7 had higher severity than those taken at R6 (Table 1). Over all growth stages, DS97-84-1 was most susceptible and DT97-4290 was most resistant, whereas Magellan and A5979 were intermediate (Table 2). There was no difference among lines at R6, but significant differences among lines occurred at the other three stages (Table 3). Analysis indicated that stages R7 and R8 were best for distinguishing genotypic differences. Selecting which of the two stages to use may depend on the specific objectives of the research. For example, differentiating among levels of resistance might be most effective at R8, when DT97-4290 was more resistant than A5979, Magellan, and DS97-84-1. And differentiating among levels of susceptibility might be most effective at R7, when DS97-84-1 was more susceptible than Magellan, A5979, and DT97-4290. In any case, great care needs to be exercised so that the same growth stage is used when lines are compared for disease severity.

## References

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