Evaluation of Resistance of Cucumber Lines to Damping-Off Caused by *Rhizoctonia solani*

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The soil-borne pathogen *Rhizoctonia solani* can cause damping-off in cucumber seedlings. The rot originates at or near the surface of the ground and weakens the stem so that the plant topples over or damps off. Subsequently, the whole plant may decay, either from the primary cause or from secondary rot organisms (4). *R. solani* can be divided into 5 anastomosis groups (AG). Isolates of AG 4 exist at or near the soil surface and are generally responsible for damping-off as well as other plant symptoms such as fruit rots (2,3).

The purpose of this study was to develop a screening test for resistance to damping-off in cucumber seedlings and to determine whether the results were comparable to those of fruit rot tests of the plants at the fruiting stage.

Inoculum preparation. The isolates of *R. solani* AG 4 (R8C, R8D) used in this study were originally collected from cucumber fields in Arkansas. Inoculum was increased by transferring the fungus from stock cultures stored in paraffin oil in test tubes to potato dextrose agar. The final increase of inoculum took place on sterile oat grains as follows. Pieces of about 1 cm^2 of the colonized agar were transferred onto sterile oat grains in autoclavable bags, which were obtained by autoclaving 300 cm^3 of oat grains and 250 ml water 2 times for 1 hr. with at least 10 hr. between to ensure that all microorganisms were killed. After about 7 days at 20 to 25°C (shaking the bags every 2 days), when the oats were completely colonized by the fungus, they were dried and then stored at 4°C until needed.

Reaction to damping-off by *R. solani* was tested by planting seeds in flats of inoculated soil. The soil was inoculated by mixing it with a predetermined number of colonized oat grains per volume of soil. Soil was inoculated 7 days before planting the seeds to enable the fungus to colonize it completely.

Damping-off screening tests. The first test was conducted in a germination chamber held at 18°C as well as in a greenhouse (held at 25 to 30°C). Inoculum concentrations were 0, 40, 80, 160, 320, 640 and 1280 oat grains per 1000 cm^3 of one isolate (R-8C). Sixteen seeds of 2 cultivars (Marketmore 76 and Sumter) were planted and rated each day for 18 days. This experiment, replicated once, was conducted to determine the optimum temperature and inoculum concentration for future evaluation of damping-off resistance. A second test was run using 6 concentration (0, 10, 20, 40, 80, 160 oats per liter) of 2 isolates (R-8C, R-8D), and 7 cultivars (Addis, Clinton, Earlipik 14, Marketmore 76, Pacer, Sumter and Supergreen) which differed in resistance for fruit rot. The seeded flats were placed in a chamber at 20°C and 14 hr. daylength using fluorescent lights. Twenty seeds of each line per isolate per concentration were planted in 3 replications. The objective was to refine further the screening tests. The experimental design was a randomized complete block with 3 replications.

In the third test, seedlings of 35 lines were evaluated for their resistance to damping-off using one concentration (40 oat grains/1000 cm^3) of one isolate (R-8C). Treatments were arranged in a randomized complete block design with 4 replications. Twenty seeds of each line were planted for each treatment combination. Seedlings were grown in uninoculated flats (in the same experimental design, but with 3 replications) as a check on germination and emergence under non-stress conditions. All flats were kept at 25-30°C on greenhouse benches with a 12 hr photoperiod.

Data analysis. Days to emergence as well as the percentage of emergence of the seedlings was calculated by counting the number emerged each day until the seedlings were scored for final disease severity. After 18 days for the first test, and 20 for the second and third tests, the emerged seedlings were rated for damping-off. Counts were used to calculate percentage of infected seedlings. Also, a disease rating was given to the seedlings, using a scale ranging from 0 to 9, in which 0 = no disease, 1-3 = slight damage, 4-6 = moderate damage, 7-9 = plants dying. A corrected disease rating was calculated by correcting the disease rating for the percentage of emergence measured in the control treatments as follows:

corrected rating = [(rating) - (9(total - emerged))]/total
where: rating = disease rating for the seedlings at end of test,
total = total number of emerged seedlings in the control,
emerged = number of emerged seedlings in the inoculated flats.

The number of non-emerged seedlings (total - emerged) was multiplied by 9 because it was assumed that those seedlings were dead.

The results were compared to data for fruit rot resistance of these lines (data not shown), which were obtained from a field test at the Horticultural Crops Research Station, Clinton, N.C. and in a laboratory test run in a mist chamber at Raleigh, N.C. in July 1985. Correlation analysis was run for the damping-off scores and the percentage of the fruits covered with lesions in the fruit rot tests using means for each line (over replications and harvest dates).

Results. As a result of the first test, a concentration range from 0 to 160 oat grains/1000 cm^3 was chosen for further testing to find an optimum concentration. An incubation temperature of 20°C was chosen, because at a lower temperature, the percentage of germination of some lines was low and infections seem to be most severe at temperatures which were relatively less favorable for the pathogen than for the host (1). The second test indicated that an inoculum concentration of 40 oat grains/1000 cm^3 using isolate R-8C was optimum. A wide range among lines for percentage of diseased seedlings and for corrected disease rating was used as the criterion for the optimum inoculum concentration.

In the third test, the number of days before first emergence and the mean number of days for emergence were not effective variables for measurement of damping-off. That is shown by the nonsignificant F value for treated vs. control seedlings (Table 1). There was a large treatment effect and also a large line effect for percentage of emergence. In addition, those variables were not correlated with the disease ratings (Table 2). Correlation with the disease ratings was not high. Percentage of emergence was only moderately correlated with the corrected disease rating. There were significant treatment effects and line effects for mean disease rating and for corrected disease rating, and those were highly correlated with each other (Tables 1 and 2).

We believe the corrected disease rating to be the best variable to use because it includes a correction for the vigor of the seeds, which was measured in the control treatment. The mean of 3.6 for the corrected disease rating was not high, however, indicating some damping-off resistance in the lines tested. Also, the range among lines for corrected disease rating was not large, indicating a similarity in resistance to this disease.

Fruit rot correlations. Correlations between damping-off rating and percentage of the fruit damaged in the fruit rot tests was low and not statistically significant (Table 2). The damping-off test was faster and easier to run than the fruit rot test, but does not appear to be a useful substitute due to the low correlation. All tests were run using the same isolates and similar soil inoculation techniques, so the lack of correlation indicates different mechanisms of resistance are acting, or that the isolates of *R. solani* used differ in virulence as fruit rot or seedling pathogens. That result is not surprising since damping-off is mainly a root and hypocotyl response, whereas belly rot response relates to the fruit surface. Further studies are needed to evaluate the cucumber germplasm collections for resistance to damping-off, and to determine the effect of isolates from the different AG's on damping-off resistance.

Literature Cited

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Table 1. Percentage of emergence (PE), number of days to first emergence (DFE), mean days to emergence (DME), mean disease score[z] (MS) and corrected disease rating (CR) for seeds of 35 lines tested in uninoculated and inoculated soil (lines are ordered by CR).

	0 oat grains per/1000 cm³			40 oat grains per/1000 cm³				
Line	PE	DFE	DME	PE	DFE	DME	MS[y]	CR
PI 163216	-	-	-	38	7	7.5	0.9	-
National Pickling	97	7	7.8	96	7	8.1	1.3	1.5
Earlipik 14	55	9	11.7	44	8	11.4	1.1	2.7
DEXP 130	90	7	8.5	83	6	9.2	2.0	2.8
Raider	98	7	7.4	90	6	7.6	2.3	2.8
GY 14A	98	7	8.1	91	7	7.4	2.5	2.8
Sprint 440	70	7	8.6	75	7	8.5	2.8	3.0
Straight 8	93	7	8.1	93	7	8.0	2.7	3.0
Ashley	83	7	9.0	71	7	9.8	2.3	3.0
Sumter	98	7	7.7	85	7	8.2	2.4	3.1
Supergreen	72	7	8.3	60	7	8.4	2.7	3.2
Little Leaf	93	7	7.2	93	6	7.5	3.0	3.2
M 16	90	6	7.7	93	7	7.6	3.2	3.2
Carolina	80	7	9.1	88	7	9.0	3.1	3.3
Commander	48	8	9.4	49	8	10.5	2.3	3.3
Castlepik	82	7	7.7	93	7	7.6	3.0	3.3
Score	93	7	7.7	83	6	7.5	3.1	3.4
Pikmaster	100	6	6.7	96	7	7.1	3.3	3.7
Dasher II	90	6	7.3	83	6	7.5	2.9	3.5
M 23	95	7	7.1	89	7	8.6	3.0	3.5
Verino	92	7	10.1	76	7	8.9	2.5	3.6
Pioneer	92	7	6.7	81	6	7.5	2.8	3.6
M 15	87	7	8.5	84	7	8.1	3.4	3.6
Guardian	100	8	8.3	93	6	7.1	3.5	3.6
Pacer	98	7	8.7	93	6	7.5	3.5	3.7
Poinsett 76	100	7	9.3	80	7	8.5	2.8	3.7
Clinton	72	7	8.1	63	7	8.6	3.1	3.9
Calypso	85	7	10.6	74	7	8.8	3.2	3.9
SMR 18	88	7	7.9	83	7	7.7	3.7	4.0
Tamor	75	7	8.4	60	7	8.5	2.9	4.2
Castlemaster	88	7	9.3	68	9	10.3	3.1	4.3
M 21	75	7	9.3	56	8	9.9	3.2	4.4
Marketmore 76	73	7	8.5	50	8	9.9	2.8	5.4
GY 3	37	12	14.0	19	14	16.0	2.5	5.7
Addis	30	9	10.8	24	8	10.7	4.6	5.9
$\bar{x}$	83	7	8.6	75	7	8.8	2.9	3.6
LSD (5%)	22	2	2.0	17	2	1.5	1.4	1.8
F (0 vs 1)	-	-	-	18	1	0	777	728

[z]Data are means of 3 replications in the uninoculated treatments, and of 4 replications of each 20 seedlings tested with 40 infested oat grains/1000 cm³ of soil.
[y]The MS and CR for the control were 0 for all lines, so they were not listed in the table.

Table 2. Correlation among 5 variables for damping-off resistance to *Rhizoctonia solani* in cucumber[z].

Variable	Days to first emergence	Mean days to emergence	Mean rating	Corrected rating	Percent fruit damage field	Percent fruit damage Lab
Percentage of emegence	-0.58	-0.63	-0.16	-0.37	-0.09	-0.25
Days to first emergence		0.83	-0.11	0.11	0.16	-0.06
Mean days to emergence			-0.33	0.16	0.12	-0.10
Mean rating				0.89	0.00	-0.24
Corrected rating					-0.10	-0.19
% damage-Field						0.58**

[z]Correlations were calculated among 35 lines and 2 inoculum levels (0 and 40 oat grains/1000 cm^3) with 3 and 4 replications, respectively.
**,*Correlation significant at the 1 and 5% levels, respectively.