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Resistance of *Phaseolus vulgaris* L. Cultivars to Hypocotyl Inoculation with *Rhizoctonia solani* Kuehn[1,2]

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Abstract. **Seven-day-old hypocotyls of 149 bean cultivars were inoculated with *Rhizoctonia solani* and incubated in a growth chamber for 72 hours at 30°C. Based on the mean size of the largest lesion, and 10 most resistant cultivars were: 'Dwarf Horticultural Long Pod', 'Yellow Eye', 'White Kidney', 'Romano Pole', 'French Horticultural', 'Red Kidney', 'Manitou', 'Sutter Pink', 'Wren's Egg', and 'Red Mexican UI 37'. The mean largest lesion for 'Dwarf Horticultural Long Pod' (the most resistant) was 3.0 mm² while that for the Bush Blue Lake 274 (susceptible) was 64.4 mm². These 10 cultivars were further tested using 5 different isolates of *R. solani*. 'Yellow Eye' was the most resistant to all 5 isolates with a mean largest lesion size of 10.0 mm² while 'Bush Blue Lake' had a mean lesion size of 90.9 mm². The root rot ratings were highly correlated with the areas of the largest lesion, and the number of lesions. 'White Kidney' was the only white-seeded resistant cultivar.**

Rhizoctonia root and hypocotyl rot caused by *Rhizoctonia solani* is a serious disease of *Phaseolus vulgaris* in many areas of the United States and the world. Plants are attacked in the seedling stage and become progressively more resistant with age. Chemical control of rhizoctonia root rot is possible but erratic due to environmental variability and is usually uneconomical (3). Genetic resistance of beans to *R. solani* is the ideal long term solution and resistance has been reported in some field and greenhouse tests (4, 5, 6, 8). This study was initiated to determine the resistance of bean cultivars to hypocotyl inoculation with *R. solani*.

Materials and Methods

Fungal isolates. Initial examinations were made using *R. solani* 75-1 isolated from beans grown in naturally infested soil from the Virginia State University Randolph Farm. Isolate 75-1 was the most pathogenic of those isolated in 1975. It was maintained on PDA slants at room temperature and transferred periodically. This isolate was stored in sterile soil to maintain pathogenicity and reisolated from beans planted in the infested soil when necessary. Isolates 175842, 175859, 226865, and 278692 were isolated in 1978 from field grown Plant Introductions (PI) with the same numerical designation.

Preparation of inoculum. Inoculum was grown on Weinholds A medium (7) consisting of 1.75 g KH_2PO, 0.75 g $MgSO_4 \cdot 7H_2O$, 37 mg $CaCl_2 \cdot 2H_2O$, 0.8 mg $CuSO_4 \cdot 5H_2O$, 1.0 mg $FeCl_3 \cdot 6H_2O$, 0.5 mg $NaMoO \cdot 2H_2O$, 0.9 mg $ZnSO_4 \cdot 7H_2O$, 0.3 mg $MnSO_4 \cdot H_2O$, 2.0 g Asparagine and 20 g glucose in 1 liter of distilled water. The fungus was grown in 9 cm diameter Petri dishes for 5 days at 28°C before use. Mycelial discs 3 mm in diameter were cut from the mycelia mat.

Preparation of plants. Plants to be inoculated were germinated in 7.6 cm plastic pots containing vermiculite, in a growth chamber at 30°C and 14 hr daylength. The minimum time required for cotyledon development was 7 days. Seedlings were gently uprooted at this time and washed.

Inoculation and incubation. Horticultural potting soil, 7 mm deep, was sprinkled on glass plates 25 × 18 cm. The soil moistened and 10 plants were placed on the plate. Each plant was then inoculated with a 3 mm mycelial disc of *R. solani* placed on the transition area of the plant. The roots and transition areas were then sprinkled with enough soil to cover the roots and hypocotyl. The plants and soil were then moistened and the inoculated plates were wrapped in aluminum foil. The plates were then placed upright in plastic pans and incubated in a growth chamber at 30°C and 14 hr daylength. After incubation for 72 hr, the length and width of the largest lesion per plant was measured and the size of the lesion (mm²) calculated. In addition, the number of lesions per hypocotyl and root rot ratings were determined. Root rot ratings were made on a scale of 1 = healthy, 2 = slightly diseased, 3 = moderately diseased, 4 = severely diseased and 5 = dead. 'Bush Blue Lake 274' was used as a known susceptible entry.

Results and Discussion

The 10 most resistant of 149 cultivars tested and 'Bush Blue Lake 274', based on mean largest lesion, are listed in Table 1. 'Dwarf Horticultural Long Pod' had an average largest lesion

Table 1. Means of the largest lesions, root rot ratings, and number of lesions of the 10 most resistant of 149 cultivars of *Phaseolus vulgaris* and 'Bush Blue Lake 274', 72 hr after hypocotyl inoculation.

Cultivar	Mean largest[x] lesion (mm²)	Root rot rating	Mean no. of lesions
Dwarf Horticultural Long Pod	3.0 d[y]	1.7 c	1.8 d
Yellow Eye	7.7 cd	2.2 b	3.1 bcd
White Kidney	8.6 bcd	2.2 b	3.5 bcd
Romano Pole	8.7 bcd	2.2 b	3.1 bcd
French Horticultural	9.2 bcd	2.0 bc	1.9 cd
Red Kidney	9.3 bcd	2.3 b	3.6 bcd
Manitou	10.3 bcd	2.2 b	4.1 bc
Sutter Pink	10.9 bcd	2.1 b	3.7 bcd
Wren's Egg	11.2 bcd	2.2 b	2.5 bcd
Red Mexican UI 37	11.3 bcd	2.3 b	3.2 bcd
Bush Blue Lake 274	64.4 a	3.1 a	5.3 a
All cultivars	44.6	2.8	4.8

[y]Mean separation in columns by Duncan's multiple range test, 5% level.
[x]Means of 'Bush Blue Lake 274' (control) from 870 plants; all other figures based on 60 plants.

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Table 2. Five most resistant of 149 *Phaseolus vulgaris* cultivars plus 'Bush Blue Lake 274' infected with 5 isolates of *Rhizoctonia solani*.

Cultivar	75-1	175842	175859	226865	278692	Mean for cultivars[x,y]
			Mean size of largest lesion (mm^2)			
Dwarf Horticultural Long Pod	2.2 d	14.4 d	9.7 d	18.9 d	14.7 d	12.0 b
Yellow Eye	6.2 d	4.2 d	13.4 d	20.0 d	6.5 d	10.1 b
French Horticultural	8.2 d	22.9 d	11.9 d	9.9 d	11.6 d	12.9 b
White Kidney	8.5 d	10.3 d	18.8 d	16.9 d	9.9 d	12.9 b
Romano Pole	7.7 d	4.7 d	13.0 d	16.2 d	13.3 d	11.0 b
Bush Blue Lake 274	94.0 ab	102.6 ab	76.5 bc	54.7 c	122.9 a	90.9 a
Means for isolates	21.2 a	26.5 a	23.9 a	22.8 a	29.8 a	--
			Root rot ratings			
Dwarf Horticultural Long Pod	1.7 hi	2.3 c-i	2.1 d-i	2.6 cd	2.3 c-h	2.2 b
Yellow Eye	2.1 d-i	1.6 i	2.2 c-i	2.3 c-i	2.0 d-i	2.0 c
French Horticultural	1.7 f-i	2.5 cde	2.0 d-i	2.0 d-i	1.9 e-i	2.0 c
White Kidney	2.2 c-i	2.3 c-h	2.6 cd	2.4 cde	2.3 e-g	2.4 b
Romano Pole	2.2 c-i	1.7 ghi	2.1 c-i	2.4 c-f	2.2 c-i	2.1 bc
Bush Blue Lake 274	3.2 ab	3.2 ab	2.8 bc	2.6 cd	3.4 a	3.0 a
Means for isolates	2.2 a	2.3 a	2.3 a	2.3 a	2.3 a	--

[x]Mean separation by Duncan's multiple range test, 5% level.

[y]Each mean for variety x isolate is based on 30 plants.

size of only 3.0 mm^2; the lowest root rot rating, 1.7; and the fewest number of lesions, 1.8 per plant. This is in contrast with the average for all 149 cultivars tested in which the mean size of the largest lesion was 44.6 mm^2, the mean root rot rating was 2.8, and the mean number of lesions was 4.8. It is also in contrast with the mean of the susceptible 'Bush Blue Lake 274' control in which the mean size of the largest lesion was 64.4 mm^2, the mean root rot rating was 3.1 and the mean number of lesions was 5.3. Lesion sizes of all 10 cultivars were not significantly different from one another but were significantly different from the 'Bush Blue Lake 274' control (Table 1). When root rot ratings were compared. 'Dwarf Horticultural Long Pod' and 'French Horticultural' had significantly lower ratings than the remaining 8 cultivars (Table 1). Significant differences in the mean number of lesions per plant among the 10 cultivars evaluated occurred only between 'Dwarf Horticultural Long Pod', 1.8 lesions, and 'Manitou', 4.1 lesions per plant.

The 5 most resistant of the 149 cultivars tested were further examined using 5 isolates of *R. solani* (Table 2). Based on their mean largest lesions, none of the 5 resistant cultivars were significantly different from each other. However, their mean largest lesions were significantly smaller than 'Bush Blue Lake 274'. No significant differences in mean largest lesion size among the 5 isolates were observed but isolates differed significantly on 'Bush Blue Lake 274'. Lesions produced by isolate 226865 were significantly smaller than those produced by isolates 278692, 175842, and 75-1.

The mean root rot ratings of the 5 resistant cultivars infected with different isolates of *R. solani* are also presented in Table 2. The 5 resistant cultivars means for root rot ratings were all significantly less than that of 'Bush Blue Lake 274'. 'French Horticultural' and 'Yellow Eye' had significantly lower root rot ratings than 'White Kidney' and 'Dwarf Horticultural Long Pod'. No significant differences in root rot ratings among the 5 isolates used were observed. There were, however, significant interactions between cultivars and isolates. The lowest root rot rating (1.6) was observed for 'Yellow Eye' inoculated with isolate 175842, whereas the highest rating (3.4) was observed when 'Bush Blue Lake 274' was inoculated with isolate 278692.

The correlation coefficients among root rot rating, number of lesions, and area of lesions of bean cultivars infected with *R. solani* are presented in Table 3. Correlation coefficients were highly significant between root rot ratings and number of lesions, and area of lesions. The correlation coefficients were significant whether based on 149 cultivars and 1 isolate or 10 cultivars and 5 isolates.

Many of the cultivars listed in Tables 1 and 2 have relatively large robust plants with large diameter and fiborous hypocotyls. This confirms the idea expressed by several authors (4, 9) that cultivars with woody stems are more resistant to *R. solani* than cultivars with succulent hypocotyls. With the exception of the 'White Kidney', all had colored seeds. Pigmented seed coats have previously been associated with resistant to rhizoctonia root rot (1, 4, 5, 6) but Dickson and Boettger (2), found that resistance was not always tightly linked to seed color.

Out of the cultivars tested, none of the bush snap beans were found to be resistant and only 1 pole snap bean, 'Romano Pole', was resistant. The most resistant cultivars were either dry or shell beans. The rhizoctonia root rot problem is more serious on snap bean than on dry beans but there is no reason to anticipate that the resistance identified cannot be incorporated into snap beans.

Table 3. Correlation coefficients among number of lesions, area of lesions, and root rot rating of *Phaseolus vulgaris* L. cultivars infected with *Rhizoctonia solani*.

Variable	Correlation coefficient: Area of lesions	Correlation coefficient: Root rot rating
Number of lesions		
149 cultivars, 1 isolate	.19	.38**
10 cultivars, 5 isolate	.09	.51**
Area of lesions		
149 cultivars, 1 isolate	--	.83**
10 cultivars, 5 isolate	--	.78**

Literature Cited

1. Deakin, J. R. 1974. Association of seed color with emergence and seed yield of snap bean. *J. Amer. Soc. Hort. Sci.* 99:110-144.
2. Dickson, M. H. and M. A. Boettiger. 1977. Breeding for multiple root rot resistance in snap beans. *J. Amer. Soc. Hort. Sci.* 102:373-377.
3. Hoch, H. C. and D. J. Hagedorn. 1974. Studies on chemical control of bean root and hypocotyl rot in Wisconsin. *Plant Dis. Rptr.* 58: 941-944.
4. McLean, D. M., J. C. Hoffman, and G. B. Brown. 1968. Greenhouse studies on resistance of snap beans to *Rhizoctonia solani. Plant Dis. Rpt.* 52:486-488.
5. Prasad, K. and J. L. Weigle. 1969. Resistance to *Rhizoctonia solani* in *Phaseolus vulgaris* (snap bean). *Plant Dis. Rptr.* 53:350-352.
6. _____ and _____ 1970. Screening for resistance to *Rhizoctonia solani* in *Phaseolus vulgaris. Plant Dis. Rptr.* 54:40-44.
7. Weinhold, A. R., T. Bowman, and R. L. Dodman. 1969. Virulence of *Rhizoctonia solani* as affected by nutrition of the pathogen. *Phytopathology* 59:1601-1605.
8. Zaumeyer, W. J. and J. P. Meiners. 1975. Disease resistance in beans *Annu. Rev. Phytopath.* 13:313-334.
9. _____ and H. R. Thomas. 1957. A monographic study of bean diseases and methods for their control. *U.S. Dept. Agr. Tech. Bul.* 868.

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Diabroticite Beetle Responses to Cucurbitacin Kairomones in *Cucurbita* Hybrids[1]

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Abstract. **Two hybrid cucurbits were produced that combined the genetic production of cucurbitacins as found in the wild bitter gourds, *Cucurbita andreana* Naud and *C. texana* Gray, with the high fruit yields characteristic of the domesticated cultivars of *C. maxima* Duchesne and *C. pepo* L. Both the *C. andreana* x *C. maxima* and *C. texana* x *C. pepo* hybrids produced relatively high yields of fruit with high cucurbitacin content. Both hybrids showed promise as attractants for population estimation of corn rootworm beetles (*Diabrotica undecimpunctata howardi* Barber and *D. virgifera* LeConte) or for use in poisoned baits using methomyl or trichlorfon with the bitter cut fruits or fruit homogenates.**

The fruits of wild species of *Cucurbita* contain substantial amounts of a series of oxygenated tetracyclic triterpenes, the cucurbitacins (10, 14). The cucurbitacins in such wild species as *C. andreana, C. ecuadorensis* Cutl. & Whit., *C. foetidissima* HBK, *C. martinezii* Bailey, *C. okeechobeensis* Bailey, *C. palmata* Wats. *C. palmeri* Bailey, *C. pedatifolia* Bailey, and *C. texana* Gray, are responsible for not only their extremely bitter taste but also for their high toxicity to higher anaimals (7, 17). In contrast, fruits of the edible, domesticated *C. ficifolia* Bouche, *C. maxima* Duchesne, *C. mixta* Pangalo, *C. moschata* Duchesne ex Poir, and *C. pepo* L. either lack or have extremely low concentrations of cucurbitacins.

Most herbivores, including man will not feed on plants containing cucurbitacins which taste bitter in dilutions as low as 1 ppb. It seems likely that these substances were selected through evolution to protect the plants against attack by both invertebrate and vertebrate herbivores. The edible species of *Cucurbita* must have been domesticated through centuries of selection of the least bitter forms by early man. On the other hand, a large group of Diabroticite beetles (Coleoptera, Chrysomelidae, Gallerucinae) have coevolved with *Cucurbita* so that the cucurbitacins have become kairomones, acting as arrestants, and feeding stimulants (4, 6, 9, 12). These beetles including the spotted cucumber beetles *Diabrotica undecimpuncta howardi* Barber, and *D. u. undecimpunctata* Mannerheim, the banded cucumber beetle *D. balteata* LeConte, the western corn rootworm *D. virgifera* LeConte, and the striped cucumber beetle *Acalymma vittata* (Fabricius) and its western relative *A. trivittata* (Mannerheim) are important pests of squash, cucumbers, and muskmelons in North America and *D. undecimpunctata howardi* and *D. virgifera* are severe pests of corn as well. These beetles are compulsive feeders on bitter *Cucurbita* spp. destroying blossoms, leaves, and fruits. They also feed on pure cucurbitacins B, D, E, I, and F-glycoside and can detect amounts of cucurbitacin B as small as 1 ng (12).

Cucurbitacin kairomones are important tools in studying Diabroticite coevolution and behavior (11). They also have possible utility in integrated pest management programs for the cucumber beetles and corn rootworms: (a) as sampling agents for monitoring beetle populations, (b) as ingredients in poison baits, and (c) as trap crops. Large scale field investigations and possible commercial utilization will depend upon the availability of sufficient quantities of bitter *Cucurbita* fruits and of the cucurbitacins. Wild *Cucurbita* spp. do not provide dependable sources of cucurbitacins as they are more difficult to grow and yield less than domesticated spp. and in some cases fruiting is dependent upon photoperiod. Therefore, we have investigated the transfer of cucurbitacin controlling genes from wild species to domesticated species. Suitable germplasm should produce maximum amounts of cucurbitacins and high yields of fruit, together with other factors conferring maximum activity to the *Diabroticites.*

Materials and Methods

Plant materials. The initial crosses consisted of *C. texana* x *C. pepo* cv. Zucchini, and *C. andreana* x *C. maxima* cv. Macre

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