

## Protection of Cucumber against *Colletotrichum lagenarium* and *Cladosporium cucumerinum*

R. Hammerschmidt, S. Acres, and J. Kuć

First author, Department of Botany and Plant Pathology, Purdue University, W. Lafayette, Indiana 47907. Second author, 2529 North B, Elwood, Indiana 46036. Third author, Department of Plant Pathology, University of Kentucky, Lexington 40506.

Research supported in part by the Herman Frasch Foundation and Cooperative States Research Service Grant No. 316-15-51, under P. L. 89-106 of the U. S. Department of Agriculture.

Journal Paper No. 5954 of the Purdue Agricultural Experiment Station, W. Lafayette, Indiana 47907; and Journal Paper No. 75-11-106 of the Kentucky Agricultural Experiment Station, Lexington 40506.

Accepted for publication 16 December 1975.

### ABSTRACT

HAMMERSCHMIDT, R., S. ACRES, and J. KUC. 1976. Protection of cucumber against *Colletotrichum lagenarium* and *Cladosporium cucumerinum*. *Phytopathology* 66: 790-793

Cucumber cultivars resistant to *Cladosporium cucumerinum*, when inoculated with *C. cucumerinum*, were protected against disease from subsequent inoculation with *Colletotrichum lagenarium*. Cucumber plants susceptible to *C. cucumerinum*, when inoculated with *Colletotrichum lindemuthianum*, were protected against disease from

subsequent inoculation with *C. cucumerinum*. *Cladosporium cucumerinum* and *C. lagenarium* are pathogens of cucumber, whereas *C. lindemuthianum* is a pathogen of bean. Effective mechanisms for disease resistance to a pathogen may exist in apparently susceptible cultivars.

*Additional key words:* induced resistance, cucumber, anthracnose.

The systemic and local protection of bean against pathogenic races of *Colletotrichum lindemuthianum* (Sacc. et Magn.) Scribner by nonpathogenic races of the fungus and other *Colletotrichum* spp. nonpathogenic on bean has been demonstrated (2, 3, 4, 7, 8). In addition, *Helminthosporium carbonum* Ullstrup, an *Alternaria* sp. and heat-attenuated pathogenic races of *C. lindemuthianum* also protect (5, 6, 7). The first two fungi are nonpathogens of bean.

This investigation was undertaken to determine whether: (i) *Cladosporium cucumerinum* Ell. & Arth., a pathogen of cucumber, would elicit protection in a resistant cultivar to *Colletotrichum lagenarium* (Pass.) Ell. & Halst.; and (ii) *Colletotrichum lindemuthianum*, a pathogen of bean, would protect cucumber against *C. cucumerinum*. An abstract of this work has been published (1).

### MATERIALS AND METHODS

Cultures of *C. cucumerinum* were maintained on potato-dextrose agar at 20 C, and cultures of the  $\beta$  race of *C. lindemuthianum* and race 1 of *C. lagenarium* were maintained at 24 C on green bean juice agar. Conidial suspensions were prepared from 7- to 10-day-old cultures grown in petri plates. Suspensions were filtered through cheesecloth and the spore concentration determined with a hemocytometer.

The following cucumber (*Cucumis sativus* L.) cultivars were used: Marketmore, SMR 12, SMR 18, SMR 58, and Victory (scab-resistant); and Marketer and Straight 8 (scab-susceptible). Seeds were surface-sterilized by soaking in 1% sodium hypochlorite for 5 minutes followed by several washes with sterile water.

Plants in one series of experiments were grown in plastic pots containing sand in a greenhouse (23-28 C) with a 16-hour photoperiod. They were watered daily with a nutrient solution consisting of 50 ml 1.26%  $\text{Ca}(\text{NO}_3)_2 \cdot 4 \text{H}_2\text{O}$ , 50 ml 3.69%  $\text{MgSO}_4 \cdot 7 \text{H}_2\text{O}$ , 50 ml 2.45%  $\text{KH}_2\text{PO}_4$ , and 850 ml of  $\text{H}_2\text{O}$ . When the first true leaves were opened, the plants were sprayed with water or a conidial suspension of *C. lindemuthianum*  $1 \times 10^5$  spores/ml and placed in a humidity chamber for 24 hours at 19 C. At the end of the incubation period the plants were allowed to dry for 2-3 hours, sprayed with water or *C. cucumerinum* ( $5 \times 10^4$  spores/ml), and returned to humidity chambers for 2-3 days at 19 C. After the second incubation period, the atmosphere in the chamber was gradually equilibrated with that in the laboratory and the plants were maintained at 19 C with a 16-hour photoperiod until symptoms developed.

In a second series of experiments, plants were grown in sand as described above and sprayed with water or a conidial suspension of *C. cucumerinum* ( $2 \times 10^5$  spores/ml). They then were held in a humidity chamber at 19 C for 2 days, dried, sprayed with water or *C. lagenarium* race 1 ( $6 \times 10^4$  spores/ml), and returned to a humidity chamber at 22 C for 24 hours. The chamber then was opened partially, the atmosphere in the chamber gradually equilibrated for 24 hours with that in the laboratory, and the plants were removed and maintained in greenhouse with a 16-hour photoperiod.

In a third series of experiments, seeds were planted in autoclaved "rag dolls" made of two layers of moistened rolled germination paper covered with aluminum foil. After approximately 8 days at 23 to 25 C the seedlings were well above the top of the dolls; the dolls were unrolled, seedcoats removed, and the plants pulled down

below the top edge of the doll. The plants were sprayed with water or a conidial suspension of *C. cucumerinum* ( $1 \times 10^5$  spores/ml) and the dolls were rolled. After 24 hours at 19 C the hypocotyls were dried, sprayed with water or *C. lagenarium* ( $5 \times 10^4$  spores/ml), and kept at 23-24 C.

In a fourth series of experiments, plants were grown in a soil substitute, "Redi-Earth" (Grace Products, Cambridge, Massachusetts 02140). When the first leaf was opened, the plants were sprayed with water or *C. cucumerinum* ( $1 \times 10^5$  spores/ml) and placed in a humidity chamber for 24 hours at 19 C. The plants then were dried, sprayed with water or *C. lagenarium* ( $1 \times 10^4$

spores/ml), and returned to a humidity chamber maintained at 22-24 C for 2 days. After the incubation period, the plants were kept in a greenhouse with a 16-hour photoperiod.

## RESULTS

Scab-susceptible cucumber cultivars inoculated with the  $\beta$  race of *C. lindemuthianum* were protected from damage by subsequent inoculation with *C. cucumerinum* (Table 1). Cucumber cultivars resistant to *C. cucumerinum*, when inoculated with *C. cucumerinum*, were protected against damage from subsequent

TABLE 1. Protection of cucumber plants against *Cladosporium cucumerinum* by *Colletotrichum lindemuthianum*<sup>a</sup>

Cultivar <sup>b</sup> and treatment <sup>c</sup>	Percent of total plants in each damage category		
	Leaf and cotyledon area covered by lesions <sup>d</sup>		
	0-10 (%)	> 10-60 (%)	> 60 <sup>e</sup> (%)
Victory -	W/W	100	0
	Cl/W	100	0
	W/Cc	100	0
	Cl/Cc	100	0
Marketer -	W/W	85	0
	Cl/W	87	0
	W/Cc	5	0
	Cl/Cc	52	0
Straight 8 -	W/W	91	0
	Cl/W	100	0
	W/Cc	4	0
	Cl/Cc	50	25

<sup>a</sup>Plants grown in sand. Results are based on three experiments containing 10 to 25 plants per treatment in each experiment.

<sup>b</sup>Cultivar victory is resistant and cultivars Marketer and Straight 8 are susceptible to *C. cucumerinum*.

<sup>c</sup>Plants sprayed with water (W) or *C. lindemuthianum* (Cl) spore suspension ( $1 \times 10^5$  spores/ml), and, after 24 hours in humidity chambers, the plants were resprayed with water or *C. cucumerinum* (Cc) spore suspension ( $5 \times 10^4$  spores/ml).

<sup>d</sup>Determined 4-5 days after the second treatment.

<sup>e</sup>Plants with > 60% damage are usually dead 6 to 7 days after the second treatment. Treatments W/W and Cl/W caused injury to some plants if the plants were removed from moist chambers without adequate time for equilibration of atmospheres in the laboratory and chamber.

TABLE 2. Protection of cucumber plants against *Colletotrichum lagenarium* by *Cladosporium cucumerinum*<sup>a</sup>

Cultivar <sup>b</sup> and treatment <sup>c</sup>	Percent of total plants in each damage category		
	Leaf and cotyledon area covered by lesions <sup>d</sup>		
	0-10 (%)	> 10-60 (%)	> 60 <sup>e</sup> (%)
Marketmore	W/W	100	0
	Cc/W	100	0
	W/C. lag	0	21
	Cc/C. lag	47	33
Victory	W/W	100	0
	Cc/W	100	0
	W/C. lag	0	40
	Cc/C. lag	44	50

<sup>a</sup>Plants grown in sand. Results are based on two experiments using 10-25 plants per treatment in each experiment.

<sup>b</sup>Cultivars Victory and Marketmore are resistant to *C. cucumerinum* and susceptible to *C. lagenarium*.

<sup>c</sup>Plants sprayed with water (W) or *C. cucumerinum* (Cc) spore suspension ( $2 \times 10^5$  spores/ml), and, after 2 days in humidity chambers, the plants were resprayed with water, or *C. lagenarium* (C. lag) spore suspension ( $6 \times 10^4$  spores/ml).

<sup>d</sup>Determined 4-5 days after the second treatment.

<sup>e</sup>Plants with > 60% damage are usually dead 6-7 days after the second treatment.

inoculation with race 1 of *C. lagenarium*. This was true for plants grown in sand (Table 2) and "Redi-Earth" (Table 3) in the greenhouse as well as for plants grown in rag dolls (Table 4). Protection was evident in both a reduction in lesion number and size of lesions. Plants grown in sand and "Redi-Earth" remained protected for at least 3 weeks after challenge at which time the experiments were terminated. It is imperative to reduce humidity in humidity chambers gradually after incubation. In early experiments this was not done and some damage was apparent in uninoculated plants and plants inoculated with nonpathogens.

### DISCUSSION

Effective mechanisms for disease resistance may be expressed in plants apparently susceptible to a pathogen. Thus, cucumber cultivars susceptible to *C. cucumerinum* were protected by the bean pathogen, *C. lindemuthianum*, and cucumber cultivars resistant to *C. cucumerinum* were protected against *C. lagenarium* by *C. cucumerinum*. In the host-parasite interactions studied, it appears that resistance is a condition elicited after interaction of the host and infectious agents. Once elicited, the condition can be effective against successful pathogens of the host. Though the resistance mechanism elicited by a nonpathogen may differ from that elicited by a nonpathogenic race of a pathogen, it nevertheless is a highly effective mechanism. This work further supports

the contention that some types of resistance may not be determined by the presence or absence of a genetic potential for resistance, but rather the ability of the potential to be quickly expressed with sufficient magnitude. The expression of this potential by chemical agents may permit the plant breeder to minimize the use of resistant, but otherwise agronomically inferior, plant selections in breeding programs.

TABLE 3. Protection of cucumber plants against *Colletotrichum lagenarium* by *Cladosporium cucumerinum*<sup>a</sup>

Cultivar <sup>b</sup>	No. plants alive/Total plants <sup>c</sup>	
	W/C. lag <sup>d</sup>	Cc/C. lag
Marketmore	5/50	21/51
Victory	2/47	32/44
SMR 12	4/44	41/48
SMR 18	3/46	28/33
SMR 58	18/53	50/50

<sup>a</sup>Plants grown in "Redi-Earth" in plastic trays. Results are summaries of four experiments with at least five plants per treatment in each experiment.

<sup>b</sup>Cultivars all resistant to *C. cucumerinum* and susceptible to *C. lagenarium*.

<sup>c</sup>Damage recorded 4-5 days after second treatment.

<sup>d</sup>Plants sprayed with water (W) or *C. cucumerinum* (Cc) spore suspension ( $1 \times 10^5$  spores/ml) followed in 24 hours by *C. lagenarium* (C. lag) spore suspension ( $1 \times 10^5$  spores/ml).

TABLE 4. Protection of cucumber plants against *Colletotrichum lagenarium* by *Cladosporium cucumerinum*<sup>a</sup>

Cultivar <sup>b</sup> and treatment <sup>c</sup>	Percent of total plants in each damage category				
	Hypocotyl area covered by lesions <sup>d</sup>				
	0-10 (%)	> 10-30 (%)	> 30-60 <sup>e</sup> (%)	> 60 <sup>e</sup> (%)	
Marketmore Cc/-	88	12	0	0	
	Cc/C. lag	90	10	0	
	W/W	100	0	0	
	W/C. lag	0	0	17	83
	C. lag/-	0	0	3	97
Victory Cc/-	71	17	10	2	
	Cc/C. lag	3	75	18	4
	W/W	100	0	0	0
	W/C. lag	3	0	25	72
	C. lag/-	0	0	28	72

<sup>a</sup>Plants grown in rolled germination paper (rag dolls). Results are based on four experiments with at least 10 plants per treatment in each experiment.

<sup>b</sup>Marketmore and Victory are resistant to *C. cucumerinum* and susceptible to *C. lagenarium*.

<sup>c</sup>Plants sprayed with water (W), *C. cucumerinum* (Cc) spore suspension ( $1 \times 10^5$  spores/ml) or *C. lagenarium* (C. lag) spore suspension ( $5 \times 10^4$  spores/ml), and, after 24 hours sprayed with water, *C. lagenarium*, or unsprayed (-).

<sup>d</sup>Determined 4-5 days after the second treatment.

<sup>e</sup>Hypocotyls with > 60% damage usually dead 6-7 days after the second treatment.

### LITERATURE CITED

- ACRES, G. S., R. E. HAMMERSCHMIDT, and J. KUC. 1975. Induction of resistance in cucumbers and recognition of the presence of a compound fungitoxic to *Cladosporium cucumerinum*. Proc. Indiana Acad. Sci. 85:(In press).
- BERARD, D. F., J. KUC, and E. B. WILLIAMS. 1973. Relationship of genes for resistance to protection by diffusates from incompatible interactions of *Phaseolus vulgaris* with *Colletotrichum lindemuthianum*. Physiol. Plant Pathol. 3:51-56.
- ELLISTON, J. E. 1975. A histological and biochemical study of local and systemic protection of *Phaseolus vulgaris* against *Colletotrichum lindemuthianum* as elicited by fungi. Ph.D. Thesis, Purdue University, Lafayette, Indiana. 271 p.
- ELLISTON, J. E., J. KUC, and E. B. WILLIAMS. 1971.

- Induced resistance to bean anthracnose at a distance from the site of the inducing interaction. *Phytopathology* 61:1110-1112.
5. RAHE, J. E. 1973. Phytoalexin nature of heat-induced protection against bean anthracnose. *Phytopathology* 63:572-577.
  6. RAHE, J. E., and J. KUC. 1970. Metabolic nature of the infection-limiting effect of heat on bean anthracnose. *Phytopathology* 60:1005-1009.
  7. RAHE, J. E., J. KUC, C. CHUANG, and E. B. WILLIAMS. 1969. Induced resistance in *Phaseolus vulgaris* to bean anthracnose. *Phytopathology* 59:1641-1645.
  8. RAHE, J. E., J. KUC, C. CHUANG, and E. B. WILLIAMS. 1969. Correlation of phenolic metabolism with histological changes in *Phaseolus vulgaris* inoculated with fungi. *Neth. J. Plant Pathol.* 75:58-71.