Inheritance of Downy Mildew Resistance in Two Plant Introductions of Lactuca sativa

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ABSTRACT

Resistance to lettuce downy mildew (*Bremia lactucae*) was detected in the field in two plant introductions (P.I. 164937 and P.I. 164939) of *Lactuca sativa*. Resistance was confirmed in seedlings inoculated in a dew chamber and grown for 6 days in the growth room. Segregation for disease reaction in seedling progenies from susceptible × resistant crosses indicated that resistance is controlled by a single dominant gene in both cases. P.I. 164937 is resistant to all the races of *Bremia lactucae* prevalent in Wisconsin; P.I. 164939, however, is susceptible to a new race of the fungus that appeared in 1968. Phytopathology 61:578-579.

Additional key words: lettuce cultivars.

Head lettuce (*Lactuca sativa* L.) is one of the most important vegetable crops in the United States; its cash value is surpassed only by potatoes and tomatoes. All commercial head lettuce cultivars grown in Wisconsin (Great Lakes 659, Fulton, Minetto) are highly susceptible to downy mildew caused by *Bremia lactucae* Regel. When weather conditions are ideal for development of the fungus, particularly in late summer, downy mildew seriously affects the quality of lettuce produced in Wisconsin. Control is attempted by means of expensive and ineffective weekly fungicide applications. Also, fungicide applications cannot be made close to harvest, at a time when the appearance of lettuce can be affected most seriously by the disease. A more practical means for control would be the development of resistant cultivars for the north central states.

Resistance to several races of *B. lactucae* was reported in a collection of *Lactuca serriola* L. from Russia (P.I. 104854) and in a French cultivar (Grosse blonde d'hiver Bourguignonne); in both, resistance appears to be conferred by a single dominant gene (2, 9), but it was not determined whether common loci were responsible for resistance. Cultivars derived from crosses of P.I. 104854 × Imperial D (9) have been used in California and Texas, but they do not head properly during the summer in the Northern United States. Moreover, these cultivars are susceptible to race 6 of the downy mildew fungus in the Lower Rio Grande Valley of Texas (6). Additional sources of resistance from Holland have been described (8), but their performance in the United States has not been determined. There is need, therefore, for additional sources of resistance to the races of *B. lactucae* that predominate in the United States.

During routine testing of more than 200 accessions of *L. sativa* (received from the Regional Plant Introduction Station, USDA, Ames, Iowa) for resistance to corky root rot, two lines, P.I. 164937 and P.I. 164939, remained free from downy mildew in the field in 1963 and 1964. Procedures were developed for testing seedling reaction to *B. lactucae* under growth room conditions. Initial inoculum was obtained from infected leaves of field-grown Great Lakes 659 which were placed in a Percival dew chamber at 18 C. The conidiophores that developed on the leaves overnight were removed with a spatula and shaken in double distilled water. This spore suspension was sprayed to runoff on 6- to 8-day lettuce seedlings of the susceptible cultivar Oakleaf which had been grown in vermiculite at 24 C and 2,000 ft-c on a 12-hr photoperiod provided by General Electric cool-white fluorescent lights. Inoculated seedlings were placed in a dew chamber at 18 C for 24 hr in the dark, transferred to the growth chamber, and grown for 6 days. By placing the seedlings back in the dew chamber for 6 hr in the dark, conidiophores and conidia were formed profusely on the adaxial surfaces of the cotyledons and on petioles and blades of primary leaves (3, 5).

Spore suspensions prepared from Oakleaf seedlings were used to inoculate large populations of seedlings of the resistant lines P.I. 164937 and P.I. 164939. For comparison, seedlings of the cultivar Valverde, known to be resistant to at least five physiologic races of the downy mildew fungus (6), were inoculated also. All three cultivars were uniformly resistant to the race of the fungus that predominated in 1963 in Wisconsin (Table 1). Seedling progenies of resistant × susceptible crosses were inoculated in a similar manner, except that susceptible plants were counted and removed, and those apparently resistant were reinoculated twice at weekly intervals. Six days after each inoculation, the plants were transferred to the dew chamber for 24 hr, and any additional plants on which sporulation could be detected were removed and counted.

Crosses of P.I. 164938 (susceptible) and the two resistant P.I. lines were made by standard hybridization techniques (4); the susceptible line was used as the female parent. In the cross Valverde × Fulton, the resistant line (Valverde) was the female parent. Pollen from recently opened flowers was removed from the stigmas by means of a gentle water stream and replaced with pollen from the resistant parent. With this method, or with alternative hand pollination methods, it was not possible to prevent selfing entirely and seeds from a flower which had been cross-fertilized invariably yielded a certain number (up to 50%) of selfed plants along with the F_1 plants. Hybrids could be distinguished from selfed plants because of their resistance to downy mildew (if the female parent was susceptible), hybrid vigor, and morphological features. In those instances in which the male parent produced black seed (as in P.I. 164939 and Fulton) and the female parent was white-seeded (as in P.I. 164938), production of black seed by resistant F_1 plants confirmed the fact that they were hybrids (7). Because of the large number of selfed

TABLE 1. Incidence of downy mildew (*Bremia lactucae*) in susceptible-resistant parental lines of *Lactuca sativa* and in progenies from crosses between them

Generation	Pedigree	No. diseased	No. healthy	X^2 3:1	X^2 P
P_1	164938-S[a]	>400	0		
P_1	164939-R[a]	0	>500		
F_2	(164938-S × 164939-R) × self	273	889	1.41	0.25
P_1	164938-S	>400	0		
P_1	164937-R	0	>400		
F_2	(164938-S × 164937-R) × self	71	227	0.23	0.60
BC_2[a]	[(164938-S × 164937-R) × Fulton-S] × self	168	460	1.03	0.30
P_1	Valverde-R	0	>400		
P_1	Fulton-S	>400	0		
F_2	(Valverde-R × Fulton-S) × self	44	117	0.46	0.50

[a] S = Susceptible; R = resistant. BC = Backcross.

plants obtained along with the hybrid progeny, however, it was not possible to determine if the total F_1 was resistant to downy mildew, as would be inferred for a dominant character if the parents were homozygous for either resistance or susceptibility. The fact that all resistant plants also produced black seed, however, would imply that all F_1 plants were resistant. Resistant F_1 plants were allowed to self-pollinate, and ratios of resistant to susceptible plants in the F_2 progeny were determined. The observed ratios approximated those expected for a single dominant gene in both P.I. lines, as well as in the cultivar Valverde (Table 1).

Because segregation for disease reaction could not be determined accurately in F_1 progeny of a simple backcross, the single gene hypothesis could not be tested in this manner. Instead, a resistant F_1 plant from the cross P.I. 164938 × P.I. 164937, presumably heterozygous for resistance (genotype *Rr*), was used as the female parent in a cross with the susceptible cultivar Fulton (genotype *rr*). Resistant plants in the F_1 progeny (genotype *Rr*) were separated from susceptible self progeny after inoculation with the pathogen and then allowed to self-pollinate. Segregation for disease reaction in the F_2 approximated the expected 3:1 ratio for a single, dominant gene (Table 1).

The results indicate that resistance to the race of *B. lactucae* prevalent in 1963 in Wisconsin is controlled by a single gene in both P.I. lines, as well as in the cultivar Valverde. No genetic analyses have been performed to determine if the same gene confers resistance in all three instances, but the results of field and controlled inoculation experiments suggest that different genes are involved. In 1968, a second race of the downy mildew fungus, capable of attacking P.I. 164939, but not Valverde or PI 164937, appeared in Wisconsin. Tests carried out under field conditions at the Texas Agricultural Experiment Station, Weslaco, Texas, indicate that P.I. 164937 is resistant to the six races of the downy mildew fungus (6) that predominate in that area, but both Valverde and P.I. 164939 are susceptible to race 6. Work is now in progress to determine if the three genes involved in resistance to the different races of the fungus are different alleles or genes at different loci.

Race denomination in the downy mildew fungus (1, 8, 9) is confusing because standard differential cultivars have not been used at all locations. With the use of the system suggested by Tjallingii & Rodenburg (8), we have determined that the race prevalent in Wisconsin since 1968 is race 1. The other race present in Wisconsin has not been identified as yet. Since all commercial lettuce cultivars grown in Wisconsin are highly susceptible to these races of *B. lactucae,* the dominant gene resistance found in P.I. 164937 is of importance in the development of resistant cultivars. Several lines with high resistance to downy mildew and good horticultural qualities, derived from a breeding program involving crosses of Fulton × P.I. 164937, followed by extensive backcrossing to Fulton, are now being tested and appear promising for future release.

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