IDENTIFICATION OF RACES OF PSEUDOMONAS PHASEOLICOLA FROM QUEBEC BEAN FIELDS[1]

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Abstract

Races 1 and 2 of Pseudomonas phaseolicola were isolated from green beans (Phaseolus vulgaris) grown near Ste. Martine, Quebec in 1966. The races were identified on the basis of their pathogenicity on 'Red Mexican UI No. 3'. Twenty-six of the pathogenic isolates tested were of race 2, and four were of race 1.

Until recently bean seed produced in the semi-arid western United States, particularly in Idaho, has been free from the halo blight organism Pseudomonas phaseolicola (Burkh.) Dows. Similarly, halo blight has occurred rarely in California, where the annual rainfall is low and splash dispersal is at a minimum. However, in 1961, 1962, and 1963, the quality of seed produced in Idaho was impaired because of infection by P. phaseolicola. The incidence of the disease also increased in seed crops grown in 1965 and 1966 in California, where overhead sprinkler irrigation caused considerable splash dispersal. Furrow-irrigated fields planted with seed from the same source showed a very low incidence of disease (2). With the advent of an epiphytotic of halo blight in areas formerly regarded as excellent for the production of disease-free seed, studies relating to the identity of the organism in the seed were initiated. The presence of two physiologic races was determined (9), and serological reactions have been studied (3, 4).

Most of the bean seed used to produce canning, freezing, and field bean crops in Canada is grown in Idaho and California. Field surveys have revealed that the bacterial blight pathogens (Xanthomonas phaseoli and Xanthomonas phaseoli var. fuscans are frequently present in the field bean crop in Ontario (7, 10). Halo blight, however, has been found only occasionally, (8). In 1966, an epiphytotic of halo blight was present in the Ste. Martine area of Quebec where extensive acreages of beans are grown for canning. The seed used to produce this crop was grown in Idaho. Because two races of halo blight have been described in the United States (9) and Europe (1), experiments were set up to identify the races occurring in Quebec.

Materials and methods

Isolations were made from blighted leaves collected from green bean (Phaseolus vulgaris L.) fields in the Ste. Martine area of Quebec in 1966. The leaves were cut into 5 mm sections, which were surface sterilized for 2 minutes in 2% sodium hypochlorite solution, plated on nutrient agar in 90 mm disposable petri dishes, and incubated at 27C. Forty-seven Pseudomonas-like cultures were obtained in this manner and were grown on slants of yeast-dextrose-carbonate agar. Cultures of races 1 and 2 were obtained from Dr. J. Natti, New York Agricultural Experimental Station, Geneva, and were used for comparative purposes in all tests.

For the identification of races 1 and 2, several methods were examined and the following two were selected:

'Red Mexican UI No. 3' and 'Kinghorn Wax' plants were grown from seed in pots of soilina growth chamber maintained at 20C and 94% relative humidity, and providing 1250 ft-c of fluorescent and incandescent light for 16 hr/day. Two-week-old plants were inoculated by atomizing an aqueous bacterial suspension onto the underside of the unifoliate leaves. After 2–3 weeks, the plants were examined for symptoms of halo blight.

In the second method, pods at the edible stage were surface sterilized for 2 min in a solution of sodium hypochlorite. Three pods of each variety were inoculated with each isolate by touching the tip of a sterilized dissecting needle onto a 24-hr-old agar slant culture of the organism, and then pricking the pod at a number of locations (Fig. 1). The inoculated pods were placed on moist filter paper in uncovered 150 ml petri dishes and maintained in a growth chamber under environmental conditions

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similar to those used in the first method. Pods were examined for symptoms of halo blight after two weeks.

Results and discussion

In preliminary tests with the spray inoculation method, considerable difficulty was encountered in distinguishing races 1 and 2 on thr basis of the resistance and susceptibility of the variety 'Red Mexican UI No. 3'. However, after familiarization with the technique and repeated inoculations, the various isolates could be designated as belonging to races 1 or 2.

The pod inoculation tests were easier to interprrt. Differences in the size of lesions were attributed to differences in the virulence of isolates belonging to the same race. Susceptible and resistant reactions were easily distinguishable on 'Red Mexican UI No. 3', because of the large diffuse-type of lesion produced by racr 2 (Fig. 1c).

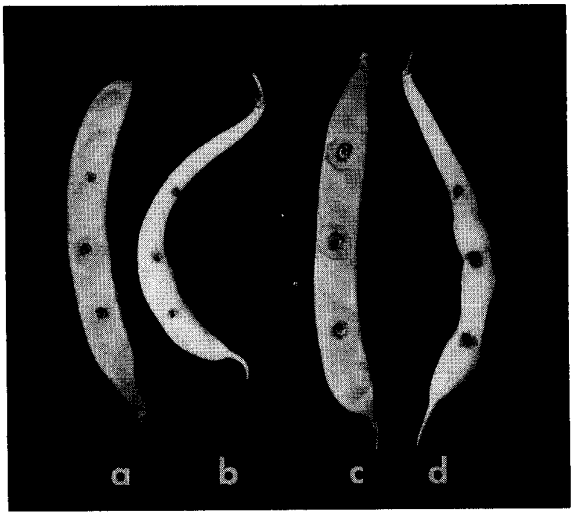

Figure 1. Differentiation of races 1 and 2 of *Pseudomonas* phoseolicolo by pod inoculation. Race 1 reaction: (a) 'Red Mexican UI No. 3', resistant; (b) 'Kinghorn Wax', susceptible. Race 2 reaction: (c) Red Mexican UI No, 3', susceptible; (d) 'Kinghorn Wax', susceptible.

Of the 47 isolates tested, 26 were of race 2, four of race 1, and 19 isolates failed to produce symptoms of either 'Kinghorn Wax' or 'Red Mexican UI No. 3'. The prevalence of race 2 is in agreement with the findings of Guthrie and Fenwick (5). who showed that of 20 isolates of Idaho origin 17 belonged to race 2. Epton and Deverall (1) showed that of 7 isolates of P. phaseolicola from Europe and Africa, 4 were classified as race 1, and 3 as race 2. Patel and Walker (6) demonstrated that most isolates of foreign originwere of race 1, whereas all isolates from Wisconsin were of race 2.

Because the seed planted at Ste. Martine was grown in Idaho, where race 2 is believed to have originated (5), it is not surprising that the majority of the isolates from Quebec were of race 2. This study illustrates the ease by which races of pathogenic organisms can be distributed from country to country by seed and establishes the presence of both races 1 and 2 of Pseudomonas phaseolicola in Canada.

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