

Disease severity was recorded 2 weeks after planting when seedlings were still at the primary leaf stage. In all of the rows planted with inoculated seeds, some of the plants were symptomless and others had leaf spots and puckering symptoms characteristic of bacterial brown spot. No difference in type or severity of symptoms was noted in any of the treatments. None of the treatments resulted in severe disease at this early stage, but all caused either slight or moderate levels of disease. None of the uninoculated plants had any brown spot symptoms. Symptoms were not recorded later because of concern that they could have been induced by inoculum that had spread from plants inoculated by other means.

The peat carrier with the high level of bacterial suspension was used in a preliminary study of the comparative ability of four strains of the pathogen to cause disease on the susceptible and resistant snap bean cultivars Bush Blue Lake 47 and Bush Blue Lake 94, respectively. In this trial, the inoculum was applied to seeds of a susceptible cultivar that were planted in "spreader rows" between rows of the susceptible and resistant cultivars. The pathogen spread from inoculated plants to uninoculated plants in adjacent rows and induced disease. The results of this trial were inconclusive. However, the inoculation procedure worked extremely well. Therefore, we recommend using laboratory-produced inoculum instead of dried-leaf inoculum in breeding trials because it overcomes the disadvantages described earlier for leaf inoculum.

References:

- 1) Daub, M.E. and D.J. Hagedorn 1981. Epiphytic populations of *Pseudomonas syringae* on susceptible and resistant bean lines *Phytopathology* 71:547-550.

A Modified Procedure for Assaying Bean Seeds for the Pathogen Causing Bacterial Brown Spot and Results of Assays of Commercial Bean Seeds

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Bacterial brown spot caused by *Pseudomonas syringae* pv. *syringae* has been a problem in snap bean fields in New York state for the past several years. As part of our research program on this disease we have investigated possible sources of inoculum, including the possibility that contaminated bean seeds are being brought into the state. To determine if we could detect the pathogen on seeds to be planted in New York in 1989 and 1990, we assayed untreated snap bean seeds provided by three seed companies.

The first year we used a procedure described by Mohan and Schaad in 1987 (1). In this paper they described a selective medium, referred to as KBC, that we have found to be excellent to isolate the pathogen from seed and plant samples. After isolating and identifying colonies as putative *P.s.* pv. *syringae*, we used a pod assay (2) to determine if the strains were capable of causing bacterial brown spot because most strains of *P.s.* pv. *syringae* are not pathogenic on beans.

In 1989, 32 seedlots were assayed and none were found to contain the pathogen. Several methods were then tried to increase the sensitivity of the assay. These included soaking the seeds at temperatures from 5 to 30 C at 5 C increments (5 C was standard in Mohan and Schaad's assay); incubating the seeds in liquid for various lengths of time (standard was 20 hours); and

agitating or aerating the liquid while soaking the seeds. We also used an enrichment technique in combination with these methods whereby KBC broth was substituted for saline, which was used as the liquid to soak the seeds in the standard procedure. Several antibiotics were also evaluated for their ability to suppress the growth of contaminants.

For these trials, a known quantity of *P.s. pv. syringae* in the form of dried-leaf inoculum was added to the seedlots to compare the efficiency of recovery of the experimental procedures to the standard assay. The best method was the standard procedure except that the seeds were soaked at 20 C instead of 5 C and the saline was aerated by means of an aquarium pump and airstone. The improved recovery of the pathogen by this method is shown in Table 1.

Increasing the temperature at which the seeds were soaked to 20 C and aerating the saline greatly increased the sensitivity of the assay. Therefore, in early 1990 the modified procedure was used to assay 55 lots of commercial snap bean seeds that were planted in New York state that summer. The pathogen was not recovered from any of these samples, indicating either that the pathogen was not present or it was below the level of detection.

Table 1.

Comparison of the Sensitivity of a Standard and Modified Procedure to Detect Strains of *P.s. pv. syringae* Capable of Causing Bacterial Brown Spot in the Presence of Naturally Occurring Contaminants on Snap Bean Seeds

Seedlot Number	Initial Population of Pathogen Added ^a	Population Recovered Using Following Procedure:	
		Standard ^b	Modified ^c
560	5,500	0	405,000
14950	5,500	1,500	111,000
6345	8,000	900	180,000
39050	8,000	1,800	1,800,000
6136	20,000	1,350	720,000
11950	20,000	900	570,000

^a Dried bean leaves with brown spot symptoms were ground to a fine powder and added to the saline used to soak the seeds; populations were determined immediately thereafter using KBC agar medium and standard microbiological methods. Values are total colony forming units in 5 L of saline.

^b 1 kg of seeds were soaked in 5 L of saline at 5 C for 20 hours before the population was enumerated.

^c Standard procedure except that seeds were soaked at 20 C and the saline was aerated.

References:

1. Mohan, S.K., and N.W. Schaad 1987. An improved agar plating assay for detecting *Pseudomonas syringae* pv. *syringae* and *P.s. pv. phaseolicola* in contaminated bean seed. *Phytopathology* 77:1390-1395.
2. Cheng, G.Y., D.E. Legard, J.E. Hunter, and T.J. Burr 1989. Use of a modified bean pod assay to detect strains of *Pseudomonas syringae* pv. *syringae* that cause bacterial brown spot of snap bean. *Plant Dis.* 73:419-423.