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THE INOCULATION OF DIFFERENT VARIETIES OF SOYBEANS BY VARIOUS STRAINS OF RHIZOBIUM JAPONICUM

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The symbiotic nitrogen fixing bacteria have usually been grouped on the basis of their ability to cause inoculation in certain species or groups of leguminous plants. In this system of classification the soybean bacteria (*Rhizobium japonicum*) are usually placed alone as no conclusive evidence of cross inoculation with any other legume has been secured. Within this species certain differences in physiological reactions may be noted. Variations in the efficiency of the soybean organisms both in their ability to infect the host plant and in their ability to fix nitrogen are quite generally recognized. The suggestion has also been made that certain varieties of soybeans are more difficult to inoculate than other varieties. Voorhees (1915) in some tests on New Jersey soils found that one variety of soybeans was not inoculated but that other varieties growing in the same field were well inoculated. This led him to believe that varieties differed in their ability to resist inoculation by symbiotic bacteria and that it might be necessary to use specific strains of organisms in order to secure inoculation of certain varieties of soybeans. Morse (1915) in discussing the work of Voorhees, states that he has often noted one variety uninoculated growing under the same conditions and in the same field with another variety which is well inoculated.

Leonard (1916) reported the successful inoculation of 19 varieties of soybeans using a culture of bacteria isolated from the nodules of a Medium Yellow soybean plant. Fred and Bryan (1922) found that, in both soil and sand cultures, the bacteria from one variety of soybean readily infected another and they considered that the variations noted by some investigators were probably due to some factors other than difference in bacteria. Perkins (1925) found a difference in the number of nodules produced on different varieties of soybeans but secured some nodules on all the varieties tested.

In some recent studies Erdman and Wilkins (1928) found that

different varieties of soybeans differed considerably in the extent of inoculation in the field. Certain varieties were readily inoculated while others receiving the same treatment showed very poor inoculation. The Peking variety was found to belong in the latter class. In one of the tests with this variety they reported that only 4 of the 33 cultures used gave as much as 50 percent inoculation. This, they believe, indicated that certain strains of bacteria are required to produce efficient nodulation on a given variety of soybeans.

In some preliminary work with soybean bacteria it was found advisable to test the stock cultures for nodule formation. Six varieties of soybeans were chosen and these were inoculated with the twelve strains of soybean bacteria kept as stock cultures in this laboratory. The varieties tested were: Manchu, Dunfield, Soysota, Peking, Pinpu and Habaro. The cultures used for inoculation had the following history:

401 — Commercial culture
402 — Commercial culture
403 — Commercial culture
404 — Isolated from Habaro variety
405 — Isolated from Mikado variety
406 — Isolated from Mikado variety
407 — Isolated from Dunfield variety
408 — Isolated from Dunfield variety
409 — Isolated from Pinpu variety
410 — Isolated from Pinpu variety
411 — Isolated from Pinpu variety
412 — Isolated from Soysota variety

These cultures have been kept in a cool place in the laboratory on yeast mannitol agar since their isolation. Transfers have been made to fresh slants several times since isolation.

The seeds of the six varieties of soybeans were sterilized by treating with 15 percent hydrogen peroxide solution for 30 minutes, according to the method proposed by Walker and Erdman (1925). This proved very satisfactory except in a few cases in which apparently there were molds under the seed coat in such a place that they were not exposed to the peroxide. It is doubtful if it would be possible to kill these molds by any of the ordinary methods of seed sterilization. After sterilization the seeds were washed twice with sterile water to remove the peroxide. In the first tests the seeds were germinated in sterile petri dishes before inoculation and planted in tumblers of sterile sand. This method was not satisfactory because of the molds which appeared in the petri dishes during germination. It was also difficult to keep moisture conditions at the optimum in the tumblers. Most of the tests

made by this method were repeated, the inoculation being made on the seed, by immersing it in a suspension of the bacteria. These seeds were then planted in 250 cc. Erlenmeyer flasks containing about 225 grams of sand. The sand had been moistened and sterilized in the flask. The flasks were kept plugged until the plants began to push up against the cotton. Crones nitrogen-free solution was added as needed to keep the sand moist.

The plants were grown for three or four weeks in the greenhouse, after which time the roots were carefully washed free from sand and examined for nodules. The results appear in table I.

Table I — Results of Inoculation Tests with Soybeans

Variety	Strain												Check	Ave.
	401	402	403	404	405	406	407	408	409	410	411	412		
Dunfield	2	1	1	0	1	3	2	2	3	1	1	2	0	1.58
Habaro	1	1	3	0	†	0	1	0	1	1	2	3	0	1.18
Manchu	†	1	†	0	1	1	†	1	3	1	0	1	0	1.00
Peking	1	2	3	0	1	3	2	1	1	1	2	3	0	1.67
Pinpu	2	1	2	0	1	3	2	1	3	†	2	2	0	1.73
Soysota	1	1	3	0	1	2	3	1	3	2	3	3	0	1.91
Av. for strain	1.4	1.17	2.4	0	1.0	2.0	2.0	1.0	2.33	1.2	1.67	2.33	0	

0 — No inoculation.
1 — Less than ½ plants inoculated.
2 — More than ½ plants inoculated.
3 — All plants well inoculated.
† — Plants not normal.

None of the plants in the check flasks showed any nodules so the sterilization of both seeds and sand was apparently adequate. The values given to show the relative efficiency of inoculation are only comparative. As there were only six or less plants per pot and some of the tests were made at a different time and using a slightly different proceedure, it is impossible to give exact values, but they represent, as nearly as can be judged, the comparative efficiency of inoculation. The averages listed in the right column and at the bottom of the table show the average value of inoculation for the variety and for the strain of bacteria respectively. In making these averages and in the discussion the tests marked † were not included because the plants growing in them were not normal.

EFFICIENCY OF STRAINS IN INOCULATION

If the average value for each of the strains on the six varieties is considered first, it will be noted that there are some which give weak inoculation and others which give good inoculation. The strains giving weak inoculation are 401, 402, 405, 408 and 410.

They did not rank high on any variety and have an average value of about 1.0. Strains 403, 406, 407, 409, 411 and 412 gave good inoculation on at least one variety each and have an average of about 2.0. With the weak strains there were four tests which failed to give inoculation while with the strong cultures there were only two. Culture 404, although isolated from Habaro soybeans failed to cause inoculation in any of the varieties tested. This culture has apparently lost its efficiency.

EFFECT OF VARIETY ON INOCULATION

If, as has been suggested, there is an adaptation to variety, then it would be expected that the strain would inoculate best the variety from which it was isolated. Strains 407 and 408 were isolated from Dunfield, but 407 shows better inoculation on Soysota and equal inoculation on Peking and Pinpu. Strain 408 does show better inoculation on Dunfield than on any of the other varieties tested. Strains 409, 410 and 411 were isolated from Pinpu variety soybeans. Strain 409 shows as good inoculation on Dunfield, Soysota and Manchu as on Pinpu, and strain 411 shows better inoculation on Soysota and equal inoculation on Peking and Habaro. The test on 410 on Pinpu is not included because the plants were not normal. Strain 412 was isolated from Soysota and shows equal inoculation on Peking, Habaro, and Soysota. These data do not indicate any marked adaptation of a strain to a variety.

A general consideration of the table and the average values for the varieties indicates that the Soysota variety is the easiest of the six varieties to inoculate, followed in order by Pinpu, Peking, Dunfield, Habaro and Manchu. The Habaro and Manchu seed were old and in all cases germination was poor. With better seed it is probable that much better inoculation would occur with these two varieties. The small differences between the other four varieties are not large enough to be of great significance. The failures in the few cases noted in the table are probably caused by unfavorable conditions for growth rather than varietal specificity.

SUMMARY AND CONCLUSIONS

1. Inoculation tests of twelve strains of Rhizobium japonicum on six varieties of soybeans are reported.
2. The strains are divided into groups on the basis of efficiency of inoculation — those giving weak inoculation and those giving good inoculation. One strain did not inoculate any of the varieties tested.

3. The differences found in the inoculation of the six varieties are probably due to factors other than adaptation to a particular variety.

LITERATURE CITED

ERDMAN, L. W. AND WILKINS, F. S. 1928. Soybean inoculation studies. Iowa Research Bulletin 114.

FRED, E. B. AND BRYAN, O. C. 1922. The formation of nodules by different varieties of soybeans. Soil Sci. V. 14, pp. 417-420.

LEONARD, L. T. 1916. Variations in nodule formation. Jour. Am. Soc. Agron. V. 8, pp. 116-118.

MORSE, W. J. 1915. Discussion. Jour. Am. Soc. Agron. V. 7, p. 140.

PERKINS, A. T. 1925. Regarding the possible adaptation of soybean radicicola to a specific host variety. Jour. Agr. Research V. 30, pp. 243-244.

VOORHEES, J. B. 1915. Variations in soybean inoculation. Jour. Am. Soc. Agron. V. 7, pp. 139-140.

WALKER, R. H. AND ERDMAN, L. W. 1925. Preliminary note on the sterilization of seeds of the Leguminosae with hydrogen peroxide. Proc. Iowa Acad. Sci. V. 33, pp. 91-95.

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