and the coefficient of variation per determination is 2 per cent.

As a check on accuracy the calcium content of various samples has been determined gravimetrically as the oxalate, by conventional flame photometry using selfstandardization, and by the method outlined here. results are given in Table 1.

An investigation into the efficiency of the plasma-jet in overcoming other matrix effects might be worth while.

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## Effect of Inoculation of Legumes with Rhizobium Strain labelled with Phosphorus-32

EVEN to-day some authors recommend inoculation with Rhizobium bacteria only for soils where legumes have never been grown or at least not for many years past.

Although results reported so far support the view that inoculation may lead to positive results even if Rhizobia of the legume being dealt with are present, we desired to trace the Rhizobium bacteria introduced in the soil by means of radioactive isotopes.

A Rhizobium strain of high productivity, isolated from red clover and tested previously for efficiency, was used which had been incubated for 48 h at 27° C on a bean agar surface of 1.5 mc. activity per 15 ml. medium. Growth of the bacteria on this medium was satisfactory. After having inactivated the surface of the bacteria by washing, the activity amounted to 40,000 imp./min. For the control plants a solution of similar activity has been produced that, however, contained no Rhizobium bacteria. Both from this solution and from the bacterium suspension 0.2 ml. was applied to each grain. The test was made in plastic pots of alluvial soil which had been proved to contain originally a high amount of Rhizobium bacteria. In the course of the eighth week of growth every 8-10 days 2 plants were taken out with roots and subjected to autoradiographic examinations by the method recom-mended by Bernert<sup>1</sup> and Manigault<sup>2</sup>. "Technofortix-S" X-ray films were used to produce the radiographs.

|                                     |                                  |      | Table 1        |       |       |       |                                  |
|-------------------------------------|----------------------------------|------|----------------|-------|-------|-------|----------------------------------|
| Seeding date                        | Production of the autoradiograph |      |                |       |       |       |                                  |
| Aug 27, 1960                        | 1                                | II   | $\mathbf{III}$ | IV    | v     | VI    | $\mathbf{v}\mathbf{I}\mathbf{I}$ |
| Plants taken<br>out<br>Films put on | 12.9                             | 19.9 | 26.9           | 5.10  | 14.10 | 14.10 | 31.10                            |
| the plant<br>Films                  | 13.9                             | 20.9 | 28.9           | 7.10  | 19.10 | 27.10 | 3.11                             |
| developed                           | 19.9                             | 4.10 | 12.10          | 20.10 | 12.11 | 16.11 | 25.11                            |

Table 1 presents the date when the plants were uprooted, when the film was put on the dry plants and when the films were developed.

In the radiographs of the first sampling only the noninoculated active control plants showed very high activity. The inoculated plants proved to be inactive, so no radiograph could be produced from them. In the 20-day old plants uprooted at the second sampling, however, the plants inoculated with active Rhizobium also showed certain activity, but while in the non-inoculated active

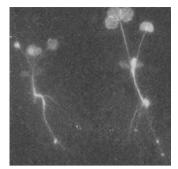


Fig. 1. Non-inoculated plants after 28 days

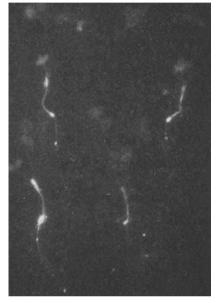


Fig. 2. Inoculated plants showing higher activity at certain centres

control plants activity was equally very high in roots, stems and leaves, in those inoculated with active Rhizobia activity was observed only at some centres and concentrated around a few nodules. In the stems and leaves of inoculated plants the activity was considerably lower than in the non-inoculated controls. The effect of inoculation was very evident also in the difference of growth since the inoculated plants were more vigorous even after three weeks' growth. Also the radiograph of the 28-day old noninoculated plants showed stronger activity (Fig. 1) whereas in the inoculated plants activity appeared concentrated in some spots and to a lesser extent (Fig. 2). In further samplings the difference in activity between inoculated and non-inoculated plants gradually diminished and completely disappeared after 8 weeks.

Thus the results obtained suggest that Rhizobium culture injected into the soil is not destroyed but enters into competition with the local Rhizobium bacteria naturally present in the soil, forming nodules on the roots. of the plants and taking part in the fixation of nitrogen.

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<sup>&</sup>lt;sup>1</sup> Bernert, T., Die künstliche Radioaktivität in Biologie und Medizin (Deuticke, Wien, 1949).

<sup>&</sup>lt;sup>2</sup> Manigault, P., Proc. Second U.N. Intern. Conf. Peaceful Uses of Atomic Energy, Geneva, 27 (1958).