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Comparison of the Pour, Spread, and Drop Plate Methods for Enumeration of *Rhizobium* spp. in Inoculants Made from Presterilized Peat†

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The pour and spread plate methods are extensively used for viable counts with pure cultures of *Rhizobium* spp. The drop plate method (2, 3), however, is less frequently used, and no information exists on this method with regard to its relative recovery efficiency in comparison to the pour and spread plate methods.

To compare these plating methods, we recovered rhizobia from 10 different peat inoculants. Each strain of *Rhizobium* was grown individually in 100 ml of yeast mannitol broth (3) in 250-ml Erlenmeyer flasks. Flasks were shaken on a rotary shaker (100 rpm); the incubation temperature was 28°C.

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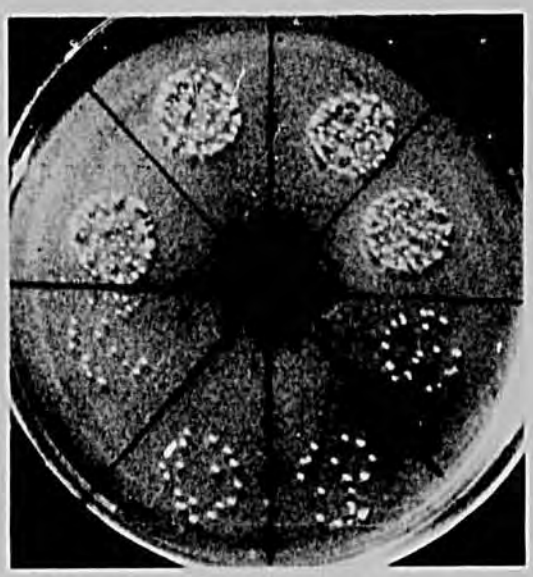

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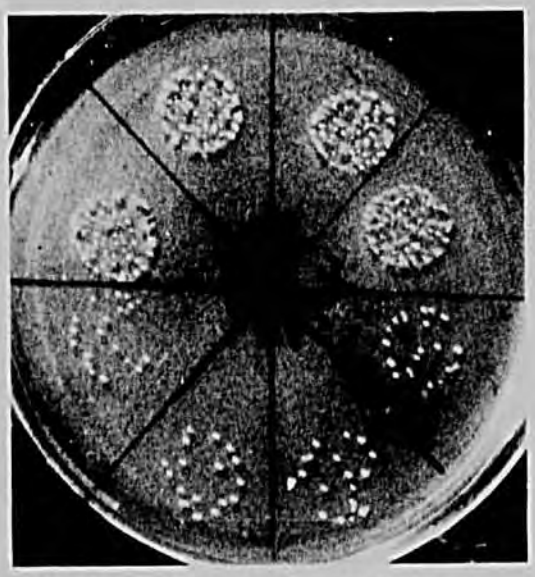

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Spread	9.82	8.78	9.93	9.24	10.06	8.81	9.60	9.56	9.88	**9.97**
Pour	**9.68**	**8.65**	**9.80**	9.21	**9.95**	8.80	9.51	9.50	9.77	9.88

[a] Mean of five replications. Means in boldface type within each column are significantly different ($P \leq 0.05$) according to the Duncan multiple range test. Laboratory sources were as follows: TAL, NifTAL Project, University of Hawaii, Honolulu; ICRISAT, International Crop Research Institute for the Semi-Arid Tropics, Hyderabad, India; NIT, Nitragin Co., Milwaukee, Wis.; CB, Commonwealth Scientific and Industrial Research Organization, Brisbane, Australia; and USDA, U.S. Department of Agriculture, Beltsville, MD. The rhizobia were isolated from nodules of the following legumes: TAL22, *Phaseolus lunatus*; TAL82, *Leucaena leucocephala*; TAL437, *Vigna radiata*; ICRISAT3889, *Cicer arietinum*; TAL1000, *Arachis hypogaea*; TAL182, *Phaseolus vulgaris*; NIT176A22, *Vigna unguiculata*; CB756, *Macrotyloma africanum*; USDA136b, *Glycine max*; and TAL999, *Vigna unguiculata*.

Of the three methods, the drop plate procedure was the most preferable because more counts could be made on one plate (Fig. 1). This was because the division of one plate into eight sectors was equivalent to producing eight spread or eight pour plates. Furthermore, with duplicate plating of each dilution, four dilutions could be plated on each plate. There was considerable economy in the use of available incubator space, and the drop method was also less laborious to carry out than the other two methods. These advantages were significant enough to cause us to adopt the drop plate method for quality control of inoculants prepared from presterilized peat. In situations in which inoculants have been exposed to high temperatures, the plant infection method may be preferred over plating methods for a reliable estimate of the numbers of infective rhizobia (4).

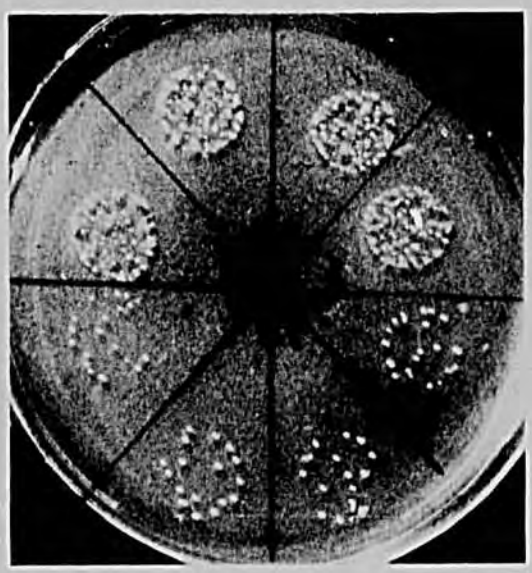

FIG. 1. Growth of colonies of *Rhizobium* sp. (TAL437) from drops plated by the drop method. Two dilutions (10^{-6} and 10^{-7}) of the peat inoculant were plated in quadruplicate and incubated at 28°C for 7 days. Note the reproducibility in all four drops of the 10^{-6} dilution.

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