COWPEA-GROUP *RHIZOBIUM* IN SOILS OF THE SEMIARID TROPICS

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Summary

Population of "cowpea-group" *Rhizobium* in fields at ICRISAT were estimated by the most probable number(MPN) method using siratro (*Macropulium atropurpureum)* as host. There was usually a large variability in *Rhizobium* numbers between sampling sites in the same Held. The populations were more consistent in Alfisols (range from 10^4 to 3 x 10^5/g soil) than in Vertisols (0 to 10^6/g soil) and decreased with depth. In paddy fields, the numbers were very low. Pigeonpea cultivars ICP-7332 (small seeded) and ICP-1 (medium sized), grown in test tubes, could also be used for the MPN method. In four of five soils tested, counts with pigeonpea as host were less than when siratro was used.

INTRODUCTION

Grain legumes such as groundnut, pigeonpea, and green gram are important agricultural crops in the semiarid tropics. Though nodulated by "cowpea-group" rhizobia, they have sometimes been shown to be poorly nodulated in farmers' fields (Nair, Ramaswamy & Porumal, 1971; Rewari, Kumar & Subba Rao, 1980; J.V.D.K. Kumar Rao & P.J. Dart, unpublished data), perhaps due to low numbers of the appropriate *Rhizobium* in soil. Because there is little data available on number of *Rhizobium* in arable soils of the semiarid tropics, we have examined the number and distribution of cowpea-group rhizobia in some soils typical of the region, using siratro (*Macroptilium atropurpureum)* and pigeonpea *(Cajanus cajan)* as trap hosts in most probable number (MPN) counts.

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MATERIALS AND METHODS

The sampling unit for 0-30 cm depth samples was generally an area of approximately 0.1 ha, from which a number of random samples, each nearly 100 g, was collected, bulked, and subsampled to provide a final sample of 100 g. Of this amount, about 10 g was suspended in water and the remainder used for moisture determination. When soils were relatively loose, a split soil sampling tube or 4 cm mechanical corer were used. When soil was hard, pits were dug.

Rhizohium populations in soil samples were estimated by a serial dilution, plant infection method using siratro and/or pigeonpea as the test plant. Siratro seedlings were grown aseptically from sterilized seed sown directly into 18 mm x 150 mm test tubes containing Jensen's nitrogen (N) free agar medium (Vincent, 1970). Pigeonpea seeds were pre-germinated in plates before transfer to 25 mm x 200 mm tubes with the same medium. When the plants were about one week old, each tube was inoculated with a 1 ml aliquot of a 10-fold serial dilution of soil and incubated in a light chamber with a 16 h light and 8 h dark period at a temperature of 28 ± 2°C for 30 days. The MPN counts of rhizobia present in samples were calculated from the proportion of plants that nodulated, using MPN tables (Fisher & Yates, 1963; Brockwell *et al.*, 1975).

Siratro is normally used as a trap host for cowpea rhizobia. Initial tests of pigeonpea as a trap host favored the use of a small-seeded cultivar ICP-7332 (100 seed weight, 5.3 g), but subsequently cv. ICP-1, a commonly grown cultivar (100 seed weight, 10 g), was also found to grow and nodulate satisfactorily in 25 x 200 mm tubes. When siratro and pigeonpea were used to count the rhizobia in artificial soil/*Rhizobium* mixtures, MPN counts correlated well with plate counts (see Table 1). It is evident from Table 1 that nodules form when only a few rhizobia are present in the aliquot. This method

TABLE 1: Counts of *Rhizobium* added as pure cultures to ICRISAT soil using the plate count and MPN count methods.

Method of counting	Strain used		
	MP-147	IHP-195	IHP-224
Plate count	5.0×10^8	1.0×10^7	5.0×10^6
MPN using pigeonpea	1.0×10^9	4.0×10^6	1.0×10^7
MPN using siratro	1.7×10^9	2.0×10^6	4.2×10^6

was then used to count the number of *Rhizobium* in fields at ICRISAT, to determine how distribution varied with depth, and to evaluate the cowpea-group and pigeonpea *Rhizobium* in a number of soils in India.

RESULTS AND DISCUSSION

Rhizobium populations in the fields at ICRISAT

Samples from 15 Vertisols and 8 Alfisols were collected, and MPN values were calculated using siratro as host. MPN counts for these 23 non-paddy soils are shown in Table 2. The MPN counts in Alfisols proved fairly uniform, but in the Vertisols, counts ranged from 0 to more than 10^6 rhizobia/g soil. There was no obvious relationship between population and present crop, nor any apparent reason why some fields should have such low numbers. The exception was field BA-10, where salinity was a problem. Samples were also taken from paddy soils at ICRISAT. *Rhizobium* populations in paddy soils

TABLE 2: Populations of cowpea group rhizobia (log_{10} MPN/g dry soil) in some Vertisols and Alfisols of ICRISAT, Hyderabad.

Vertisols				Alfisols			
Sample No.	Field	Log_{10} MPN		Sample No.	Field	Log_{10} MPN	
1	BW-2	6.1	(25)[1]	1	RW-2D	5.4	(3)[1]
2	BA-25	5.1	(34)	2	RA-17	5	(11)
3	ST-1	5.1	(32)	3	RA-25	5	(16)
4	BW-4	4.3	(25)	4	RW-2	5.0	(5)
5	BW-7	4.3	(23)	5	R-10	5.0	(3)
6	B-5	4.3	(22)	6	RA-26	4.7	(8)
7	M-14	4.2	(5)		(Healthy		
8	BW-6	3.7	(21)		Pigeonpea)		
9	BW-3	3.3	(25)	7	RA-26	4.2	(10)
10	B-2	3.1	(29)		(Sterility		
11	B-4	2.3	(29)		mosaic)		
12	BW-5	2.3	(26)				
13	BW-8	2.0	(15)	8	R-l	4.2	(11)
14	BA-10	1.7	(18)				
15	BW-1	0	(21)				
Mean over all Vertisols		3.4		Mean over all Alfisols		4.8	

[1] Moisture percentage of soil sample.

were low; generally less than 100/g of soil. In many rice growing areas of India it is common practice to grow a legume after the main crop of paddy, if water is limiting. If pigeonpea or other members of the cowpea inoculation group of legumes are planted after a paddy crop, it may be necessary to re-inoculate in order to ensure adequate nodulation.

Variation in *Rhizobium* population with depth

Pigeonpea is a deep-rooted crop, and roots grow to a depth of 200 cm. We examined the distribution of cowpea-group *Rhizobium* at soil depths ranging from 0-160 cm in small areas of different fields (see Table 3). In one Alfisol field (A), the *Rhizobium* population remained high (10^4/g dry soil) throughout most of the profile, whereas in field B, the population declined rapidly with depth, especially below 100 cm. Similar differences were observed in Vertisol fields. It is not known whether pigeonpea rhizobia travel along with the root system in the rhizosphere as the root grows through the soil. Further studies on the relationship of soil populations to nodulation and the response to inoculation are being initiated.

TABLE 3: Population of cowpea group rhizobia ($\log_{10}$ MPN/g dry soil) at different depths of two Alfisol and two Vertisol fields.

Soil depth (cm)	Alfisol		Vertisol	
	Field A[1]	Field B[2]	Field C[3]	Field[4]
0-5	3.2 (10)[5]	4.5[6] (9)	3.2 (2)	5.4[6] (21)
5-10	4.3(10)		3.2 (25)	
20-30	5.0(7)	4.0 (9)	3.8(24)	4.9 (21)
50-60	4.7 (11)	2.5 (12)	2.8(28)	4.6 (14)
100-110	4.2(13)	1.7 (12)	1.6(34)	3.0[7] (13)
150-160	3.3(13)	0 (17)	1.6(30)	2.8 (19)

[1]Average of 2 replications on a 4 x 16 m grid covering 0.1 ha in RW2B field, 1CRISAT site, Patancheru.
[2]Average of 10 replications in Nursery field, ICRISAT, Patancheru.
[3]Average of 4 replications on a 4 x 16 m grid covering 0.1 ha in BW 4 field, ICRISAT site, Patancheru,
[4]Average of 3 replications in M-l1 field, ICRISAT site, Patancheru.
[5]Value in brackets is moisture percent of sample on dry wt basis.
[6]Sample collected from 0-10 cm soil depth.
[7]Sample collected from 90-100 cm soil depth.

Specificity of pigeonpea

In the above studies, siratro was used as the trap legume for cowpea-type rhizobia. Subsequently both siratro and pigeonpea were used in MPN counts of cowpea-group rhizobia in different soils. With the marked exception of the soil from ICRISAT, where MPN estimates with the two legumes were similar, the estimates obtained using siratro as host were always higher than when pigeonpea was used (see Table 4). This suggests some degree of specificity between pigeonpea and cowpea-group rhizobia.

In the present study, rhizobial numbers varied within the same field, with soil type and depth: there was little relationship between present crop and *Rhizobium* population. Given this variation, seed inoculation could be worthwhile insurance, even in fields where the population of *Rhizobium* appears to be relatively high.

TABLE 4: Soil populations of cowpea *Rhizobium* when tested on siratro and pigeonpea.

Source of soil (India)	Numbers of rhizobia/g soil nodulating	
	Siratro	Pigeonpea
Kashmir	190,000	3270
Hissar (1)	3440	64
Hissar(2)	4300	0
Maharashtra	43000	92
ICRISAT	19300	19300

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