Plants/m^2	Unsprayed				Sprayed			
	4	8	33	67	4	8	33	67
Heliothis (larvae/m^2)	11	18	40	42	4	2	3	4
Pod damage (%)	18	19	20	24	3	1	0	0
Grain yield (g/m^2)	86	112	92	85	90	159	159	165

"economic injury level" studies and our host plant resistance screening methodology.

- *S. Sithanantham and W. Reed (ICRISAT).*

Microbiology

Counting Chickpea Rhizobia in Soils

Of the several causes of poor nodulation in farmers' fields one is that there are sometimes too few of the very specific *Rhizobium* strains that can nodulate chickpea. There is no reliable method for direct counting of *Rhizobium* bacteria in soil dilutions, as media specific for growth of *Rhizobium* alone are not available. We have adapted for chickpea rhizobia a method of estimating the most probable number (MPN) of *Rhizobium* in a sample from the pattern of nodulation of plants inoculated with a series of dilutions of the sample. The number is only an estimate because it is based on the probability that a single *Rhizobium* will form a nodule. The method requires that contaminating *Rhizobium* strains from other sources, such as in dust and in water, are kept away from the test plants. This is usually done by growing the plants under sterile conditions in a cotton-wool-stoppered test tube. Culture of large-seeded legumes, such as chickpea, is not easy in test tubes as they do not nodulate readily under these conditions. The logistics of keeping contaminants out and the requirement of space usually preclude the use of pot culture assemblies for such counts.

We obtain reliable nodulation of chickpea grown in test tubes by dwarfing the plants by cutting off most of their cotyledons. Surface-sterilized seeds are germinated on agar, and the cotyledons are excised when the radicle is 2 to 3 cm long. The plantlet is then grown in a 200 x 25 mm test tube with sand or sand plus vermiculite as the root medium. Plants are inoculated with 1 ml aliquots of a regular series of dilutions of the sample whose population is being counted.

It is necessary to prevent the temperature inside the test tubes from rising above 27°C, otherwise the nodulation becomes unreliable. This is usually very difficult to achieve in glasshouses, and we have designed a chamber for growing our plants in a room where temperature is controlled by an air conditioner and the test tubes are illuminated by fluorescent tubes (Fig. 1). Fans circulate the conditioned air over the fluorescent tubes to keep them cool. Plans of this facility are available from ICRISAT.

The concurrence between the MPN and a direct plate count for pure cultures of *Rhizobium* is excellent.

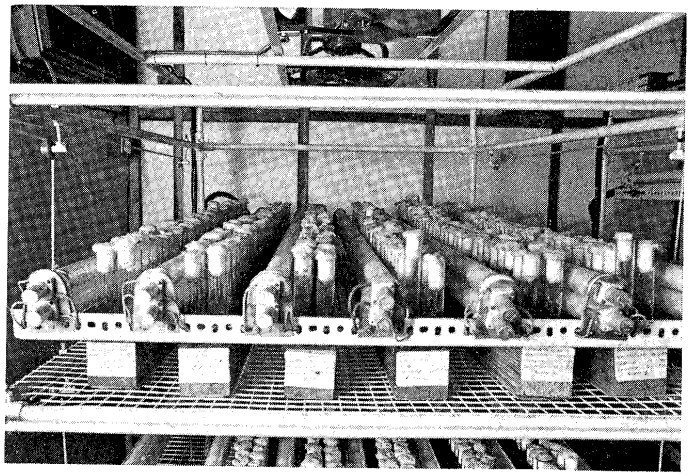

Fig. 1. Light chamber unit for growth of chickpea seedlings in test tubes. The lights can be racked away from the test tubes by a pulley system so that the wooden blocks holding the test tubes can be put in position or removed. The starters for the flourescent lights are on the frame holding the tubes, the ballasts are outside the room. The tubes are cooled by blowing conditioned air from the wall AC units over them, by two fans.

Using this method we counted 10^3 to 10^4 chickpea rhizobia/g soil in a vertisol field at ICRISAT Center, where chickpea normally nodulates well without inoculation. The number declined to fewer than 400/g below 60 cm. In a paddy field, rhizobia were fewer than 10/g soil. This method of counting will also be very useful in quality control during inoculant production, as it is the only reliable way to count the specific chickpea *Rhizobium* population in inoculants also containing other organisms. The potential usefulness of an inoculant depends primarily on the number of rhizobia it contains, and this should be at least 10^8/g of carrier.

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Research at ICARDA on Improving Nitrogen Fixation in Chickpea

The yield of chickpea in West Asia and North Africa has remained static around 750 Kg/ha for the last 10 years. This is much less as compared to the yields of *Vicia faba* (1164 Kg/ha) and soybean (1147 Kg/ha) grown in the region (FAO Production Year Book, 1977). Our surveys of farmers' fields in several countries of this region suggest that the low yields may be partly caused by the lack of effective nodulation. These surveys have helped us identify several problems associated with nitrogen fixation in chickpea, and we are now investigating these on the ICARDA farm at Tal Hadia in Syria. Some preliminary results are presented here.

Response to inoculation with *Rhizobium*

Chickpea nodulates poorly on the ICARDA farm at Tal Hadia. An experiment was conducted in the spring of 1978 to study the response of two chickpea cultivars - Syrian Local, a large seeded kabuli and NEC 2304, a small seeded desi type, to inoculation with eight strains of *Rhizobium* (CB-1189, CC-1192, Ca-7, DNRa-I, Pantnagar strain, IC-13, IC-20 and IC-26) obtained from ICRISAT but of diverse origin. The experiment was designed to identify suitable strains of *Rhizobium* for local conditions.

Three harvests at different stages of growth (early vegetative, early flowering and mid pod-fill) were made for nodulation studies before final grain yield harvest.

The uninoculated plants had, on an average, one nodule per plant. Two *Rhizobium* strains (DNRa-1 and IC-13) produced only 3 nodules per plant, but others produced 18 to 45 nodules. The two cultivars responded

Table 1. Effect of inoculation on nodulation of chickpea, Tal Hadia, Syria, 1978

Treatment	SYRIAN LOCAL			NEC 2304		
	Nodule No/plant[a]	Nodule dry wt. (mg/plant)[a]	Yield (Kg/ha)	Nodule No./ Plant[b]	Nodule dry wt. (mg/plant)[b]	Yield (Kg/ha)
Uninoculated	0.9	71	1255	0.8	83	1420
Inoculated	35.3	421	1286 to 1508[c]	28.7	-	1282 to 1798[c]
Uninoculated 120 Kg N/ha	0.4	29	-	0.2	13	-

[a] Harvested 66 days after germination
[b] Harvested 74 days after germination
[c] Range for six inoculum strains