CROSS-INOCULATION RELATIONSHIPS OF PSOPHOCARPUS TETRAGONOLOBUS AND ITS RHIZOBIUM WITH OTHER LEGUMES AND RHIZOBIA

R. P. T. ELMES*

ABSTRACT

Representative strains of Rhizobium *from six cross-inoculation groups were inoculated onto winged bean* (P. tetragonolobus) *and an isolate from* P. tetragonolobus *was inoculated onto legumes from five cross-inoculation groups.* P. tetragonolobus *was effectively nodulated by two isolates from itself and a* Rhizobium *of the Cowpea Group. It was ineffectively or partially effectively nodulated by Rhizobia of the Lupin, Medic, Pea and Clover cross-inoculation groups.* Stylosanthes guianensis *was effectively nodulated and* Phaseolus vulgaris *ineffectively nodulated by an isolate from* P. tetragonolobus. *The results show that* P. tetragonolobus *belongs to the cowpea cross-inoculation miscellany and is promiscuous with regard to nodulation by* Rhizobium.

INTRODUCTION

It has been realized recently that the winged bean, *Psophocarpus tetragonolobus* (L.) DC., offers exceptional promise as a source of edible vegetable protein that can be grown in the humid tropics. Because the varieties now cultivated have a climbing growth habit and require staking to produce high yields of pods and seeds, and because not all pods mature at the same time, it is not likely to be suitable for large-scale commercial production in the near future. The fact that the pods do not all mature at the same time, however, makes the winged bean well suited to subsistence agriculture and market gardening (Anon. 1975).

P. tetragonolobus apparently nodulates well without inoculation of the seed in Papua New Guinea. The aim of this experiment was to determine the cross-inoculation relationships of *P. tetragonolobus* and its *Rhizobium* with other legumes and Rhizobia.

MATERIALS AND METHODS

Rhizobial Isolates

Two isolates from *P. tetragonolobus* were tested. Strain NGR 156 was isolated by Trinick in 1963 from a plant growing in the Waghi Valley near Mount Hagen, Western Highlands Province. This strain effectively nodulated *Macroptilium bractearum* (Trinick, unpublished). Strain NGR 258 was isolated by Elmes in 1974 from a plant growing at Laloki near Port Moresby.

The other strains of *Rhizobium* tested were strains commonly used to inoculate legume seed and represented six cross-inoculation groups: WU 425—*Lupinus* spp.; U 45—*Medicago* spp.; CC 511—*Phaseolus vulgaris*; SU 391—*Pisum sativum*; NGR 156 and NGR 258—*P. tetragonolobus;* CB 756—Cowpea Group; TA1—*Trifolium* spp. These are also shown in the *Table.*

All strains were maintained on yeast mannitol agar (YMA) (Vincent 1970).

Seed

It is necessary to scarify the seed coat of certain legumes before they will germinate. *Medicago sativa* and *Trifolium repens* were immersed in concentrated sulphuric acid for 10 minutes and *Stylosanthes guianensis* for 20 minutes. The seed of *P. tetragonolobus* was sandpapered. *Phaseolus vulgaris* and *Pisum sativum* were not treated. After treatment with acid the seeds were washed in six changes of tap water.

All seeds were sterilized by immersion for three minutes in 0.1 per cent acidified mercuric chloride followed by washing in six changes of sterile tap water.

*Plant Pathologist, Department of Primary Industry, Konedobu, Papua New Guinea.

It was intended to test *Lupinus* sp. also but no viable seed could be obtained.

Growing Assemblies

Different assemblies were used depending on seed size, as follows: bottle-jar assembly (*Ph. vulgaris*); 18 oz MacCartney bottle (*P. tetragonolobus*); 255 mm x 50 mm tube (*Pi. sativum*); 8 oz MacCartney bottle (*S. guianensis*); 150 mm x 25 mm tube (*M. sativa* and *T. repens*).

The method used for the bottle-jar assemblies was as described by Vincent (1970) under modified Leonard bottle-jar.

The method used for the 18 oz and 8 oz MacCartney bottles was described by Trinick (1968) for 8 oz flat dispensing bottles.

The method used with the 255 mm x 50 mm tubes followed the principle of the 8 oz bottle method in which the plant tops were unenclosed. The tubes were filled to within about 3 cm of the top with seedling nutrient agar. A sterile, pregerminated seed was placed on the agar surface in each tube and a sterile, inverted jar placed over the mouth of the tube. When the seedling had grown up into the jar, the jar was removed and a loose, sterile cotton-wool plug placed in the mouth of the tube.

The 150 mm x 25 mm tubes were used in the way described by Vincent (1970). Twelve ml of agar were placed in the bottom of the cotton-wool plugged tube and the plant grown entirely within the tube.

Seedling Media

The nutrient solution for the bottle-jar assemblies and the nutrient agar were modified from Trinick (1968). In the non-nitrate media 0.407 g potassium chloride was used instead of 0.141 g. In the nitrate control media 0.141 g potassium chloride was used as per Trinick. The bottle-jar nutrient solution was half the concentration of the nutrient agar.

Experimental Design

All strains of *Rhizobium* listed in the *Table* were inoculated onto *P. tetragonolobus*. The following legumes were inoculated separately with their own strain and strain NGR 258 from *P. tetragonolobus*: *M. sativa*, *Ph. vulgaris*, *Pi. sativum*, *S. guianensis* and *T. repens*.

There were uninoculated controls and nitrate controls for each legume and there were three replications of all treatments.

RESULTS

The *Table* gives the nodulation of *P. tetragonolobus* and the other legumes by the strains tested. The results are based on the abundance, size and internal colour of the nodules.

For *P. tetragonolobus* there were no differences in nodulation between plants inoculated with CB 756 and those inoculated with the *P. tetragonolobus* isolates NGR 156 and NGR 258. All three strains appeared fully effective on this legume. Fairly abundant nodules with white or sometimes light pink interiors were formed on *P. tetragonolobus* by strain WU 425. Strains U 45 and SU 391 formed small, white to light-pink nodules, each in one replication only. TA 1 formed ineffective nodules on *P. tetragonolobus*. Under the conditions of the experiment the nodules were approximately spherical. Effective nodules were 2 to 6 mm diameter and ineffective or partially effective nodules were 1 to 4 mm diameter.

M. sativa was effectively nodulated by U 45 but not nodulated by NGR 258.

Ph. vulgaris was effectively nodulated by CC 511 and ineffectively nodulated in two replications by NGR 258. The other replication inoculated with NGR 258 was not nodulated.

Pi. sativum was ineffectively nodulated by SU 391 in two replications only. It is likely that temperatures were too high for effective symbiosis with this legume.

S. guianensis was effectively nodulated by CB 756 and NGR 258. There did not appear to be any difference in effectiveness between these two strains.

T. repens was effectively nodulated by TA 1 but not nodulated by NGR 258.

DISCUSSION

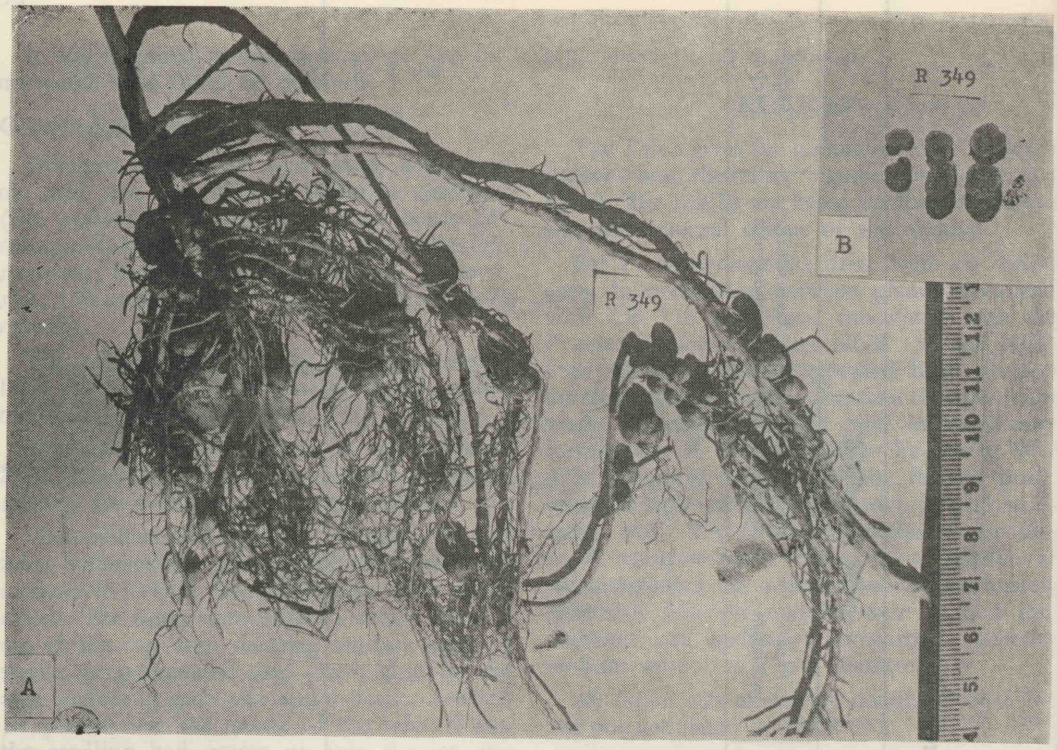
The results indicate that *P. tetragonolobus* belongs to the cowpea cross-inoculation miscellany of legumes. It was effectively nodulated by CB 756, a strain of *Rhizobium* that nodulates many cowpea miscellany legumes, as well as by its own isolates NGR 156 and NGR 258. Strain NGR 258 effectively nodulated *S. guianensis*, a representative of the cowpea miscellany.

The ineffective or partially effective nodulation of *P. tetragonolobus* by strain WU 425

Table.— Nodulation of P. tetragonolobus and other legumes

Rhizobium strain and recommended host(s)	Legume Inoculated					
	Medicago sativa	*Phaseolus vulgaris*	*Pisum sativum*	*Psophocarpus tetragonolobus*	*Stylosanthes guianensis*	*Trifolium repens*
WU 425 (*Lupinus* spp.)				I*, e†		
U 45 (*Medicago* spp.)	E			e†, —*		
CC 511 (*Ph. vulgaris*)		E		—		
SU 391 (*Pi. sativum*)			I*, —†	e†, —*		
NGR 156 (*P. tetragonolobus*)				E		
NGR 258 (*P. tetragonolobus*)		I*, —†	—	E	E	—
CB 756 (Cowpea Group)				E	E	
TA 1 (*Trifolium* spp.)				I		E
Uninoculated control	—	—	—	—	—	—
Nitrate control	—	—	—	—	—	—

E = effective nodulation
e = partially effective nodulation
I = ineffective nodulation
— = no nodules
* = 2 replications
† = 1 replication



Plate—A. Nodulated roots of winged bean (*P. tetragonolobus*) from a plant inoculated with *Rhizobium* strain NGR 258 and grown in soil. B. Halved nodules of winged bean (*P. tetragonolobus*) showing red-brown interiors which indicate effective nitrogen fixation

(*Lupinus* spp.), U 45 (*Medicago* spp.), SU 391 (*P. sativum*) and TA 1 (*Trifolium* spp.) is interesting as these belong to other cross-inoculation groups. Ineffective nodules were also formed on *Ph. vulgaris* by strain NGR 258. The results of the experiment thus contradict to a certain extent the usual concept of cross-inoculation groups. Wilson (1939), however, reported a number of cases of nodulation between legumes and bacteria in different cross-inoculation groups. For instance an isolate from *T. pratense* nodulated *Desmodium polycarpum, Centrosema virginianum* and *Pueraria hirsuta*, three cowpea miscellany legumes. In Papua New Guinea Trinick (1968) obtained nodulation of *M. sativa* by an isolate from *Leucaena leucocephala* which was also capable of nodulating legumes in the cowpea miscellany. The results obtained here suggest that *P. tetragonolobus* is a promiscuous legume, and that NGR 258, an isolate from it, is also promiscuous in that it nodulated (ineffectively) *Ph. vulgaris*.

CONCLUSION

The results support the field observation in Papua New Guinea that *P. tetragonolobus* usually nodulates well without inoculation. Presumably it would be capable of being nodulated by strains of *Rhizobium* from many tropical legumes of the cowpea cross-inoculation miscellany. Two isolates from *P. tetragonolobus* were no more effective on this host than CB 756. It appears reasonable, therefore, to recommend CB 756 for inoculation of *P. tetragonolobus*.

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