

VA MYCORRHIZAL INOCULATION OF FIELD CROPS

C. LI. POWELL and D.J. BAGYARAJ

Ruakura Soil & Plant Research Station, MAF, Hamilton

ABSTRACT

The agricultural relevance of previously reported VAM field crop trials is assessed and recent VAM field crop trials in New Zealand and in India are described. We concluded that most VAM field crop trials have suffered from many design faults and few now have any relevance to practical agriculture. The main faults have been use of small unreplicated plot trial designs, inappropriate use of transplanted seedlings, use of field soils with unnaturally low populations of indigenous mycorrhizal fungi, use of very high (20-30 tonnes/ha) inoculum rates hand placed below the seed, lack of prior selection of best fungal inoculants, lack of calibration of VAM growth responses against responses possible with P fertiliser and lack of testing for longevity of VAM response. In one field trial with onions at Pukekohe in 1981-82, machine drilled VAM inoculum at 1.9 tonnes/ha increased onion bulb yield by up to 8.4 tonnes/ha. In field trials in southern India, VAM inoculation in the nursery bed increased the yield of the transplanted crops, finger millet and chilli, by more than 50%, equivalent to, or greater than, the response to 19 and 75 kg P/ha respectively. The relevance of these results and the directions for future VAM field research are discussed.

Additional Key Words: maize, onions, finger millet, chilli, agricultural machinery, seedling transplant

INTRODUCTION

General

It is well known that vesicular-arbuscular mycorrhizas (VAM) are present in almost all agricultural soils and that mycorrhizal inoculation of plants grown in sterilised soil in pots greatly stimulates plant growth (Mosse, 1973; Gerdemann, 1975). Yost and Fox (1979) also showed that the yield of field crops is highly dependent on these naturally occurring VAM. They grew a range of crops in plots on a P deficient oxisol after half the plots had been sterilised with methyl bromide which killed the indigenous VAM. At a soil solution P level of 0.012 $\mu\text{g/ml}$, soil sterilisation reduced the yield of cassava, soybean, cowpea, onions, stylosanthes and leucaena by 35-95%. This effect was almost entirely due to the lack of VAM. Likewise, Ross (1971) had recorded mycorrhizal growth responses of 122-12% in soybeans grown in sterilised field soil at P fertiliser rates ranging from 0 to 176 kg P/ha. Similar mycorrhizal responses have been found in sterilised soils or planting media with citrus, apples, avocados and a variety of ornamental and tree crops (Maronek *et al.*, 1981).

Despite the proven effects of VAM inoculation on plant yield in sterilised soils, relatively few field trials have been carried out internationally and fewer still are able to meet all of our current objectives for VAM field trials:

- (a) To demonstrate increased plant yield in the field with the same amount or less fertiliser,
- (b) To enable farmers and growers to inoculate their crops using existing machinery and labour,
- (c) To show that VAM inoculation is worthwhile economically.

Past field trials

Most field trials have suffered from a range of poor design features including:

- (a) Small plot sizes,
- (b) Neither treatment replication nor statistical analysis,
- (c) Inappropriate use of preinoculated seedlings transplanted to field plots (e.g. for maize and wheat),
- (d) Use of test soils with unusually low soil P levels to accentuate the mycorrhizal growth response,
- (e) Prior fallowing of test soil for several months or years to reduce (artificially) the inoculum level of the indigenous VAM,
- (f) The use of very high rates of soil inoculum (up to 20-30 tonnes/ha) usually placed by hand in the seed furrow,
- (g) The failure to carry out prior fungal selection trials to find the optimum fungal inoculant for the test crop and soil,
- (h) Lack of knowledge about persistence of mycorrhizal growth responses from year to year in annual or perennial crops,
- (i) Failure to equate mycorrhizal growth responses with those obtainable from P fertiliser alone.

Most of these field trials were carried out in the past when the vital role of mycorrhizal fungi in the uptake and cycling of nutrients (especially P) in soil was neither accepted nor understood by agronomists. Now that the potential benefits from VAM inoculation have been shown quite conclusively (despite some lack of agricultural reality), all future field trials must concentrate on showing that mycorrhizal inoculation can be a useful management tool in current agriculture practice (objectives (b) and (c)).

OUR CURRENT WORK

Mechanical placement of inoculum in the field

(a) **Onions.** *Allium cepa* L. In August 1981, with Graham Wilson's help, we set out two onion trials on Patumahoe clay loam (Olsen P 80 μ g/ml) at Pukekohe Horticultural Research Station. Granulated or loose unprocessed inoculum soil at 57 g per row metre (equivalent to 1.9 tonnes/ha) was drilled into the seed bed of each of the 7 x 1.5 m plots, at 30 or 60 mm depth, using a self propelled Oyjord cone seeder at the standard row spacing for onions of 200 mm between rows and five rows per 1.0 m wide raised bed. There were five replicates per treatment. Control plots received no inoculum. The mycorrhizal inoculum was poured down the cone and both forms (especially the granulated inoculum) flowed easily through the seed pipes and through the coulters into the soil. Onion seeds (cv. Pukekohe Long Keeper) were sown at 30 mm depth in the seed bed and above or with the VAM inoculum, by tractor-drawn Stanhay drill. The onion crops received the standard herbicide and pesticide treatments and bulbs were wrenched by hand in January 1981 and weighed without topping in February 1981. In the first trial, onions were also fertilised with phosphorus at 50 and 100 kg P/ha but this trial was abandoned because of garden symphilitid damage to the plants soon after seed sowing and responses to P or mycorrhiza could not be properly assessed.

In the second trial, undamaged by symphilitids, mycorrhizal infection levels in the roots were significantly higher in inoculated than uninoculated (control) plants during crop growth and at harvest (Table 1) although even the control plants were well infected (35% of mycorrhizal roots) by the indigenous VAM. The most effective inoculum treatment was loose inoculum at 30 mm (same depth as the seed) resulting in an 18% increase in bulb yield over controls, equivalent to an additional 8.4 tonnes/ha and significant at $p < 0.05$. Granulated inoculum sown at 30 mm gave a 13% increase in bulb yield, significant only at $p < 0.10$. Inoculum placed at 60 mm in the seed bed, 30 mm below the seed, had no apparent or significant effect on bulb yield. In this and most field experiments where seed bed inoculation has stimulated crop yield, a second inoculation later in crop life would probably have little effect on mycorrhizal infection levels or growth responses.

We stress that these encouraging results are from one trial only and that further field trials are now in progress at Pukekohe to test the effect of a range of inoculum rates (0, 500, 1000 and 2000 kg inoculum soil/ha) and P fertiliser rates (0, 70, and 140 kg P/ha) on responses to VAM inoculation in a second growing season. Nevertheless this is a milestone experiment for VAM field research as it showed that mycorrhizal inoculum could be accurately drilled into the seed bed at reasonably low rates using currently available agricultural machinery. It should be possible to achieve successful onion inoculation at the much lower inoculum rates of 500 or 1000 kg inoculum/ha as Powell (1981) found that 170 mg of soil inoculum per seed was sufficient to stimulate shoot and bulb yield in unsterilised

soil in pots whereas the 57 g per row metre inoculum rate used in this trial was equivalent to approximately 2 g inoculum per seed.

TABLE 1: Effect of formulation and depth of placement of soil inoculum on responses of onions to mycorrhizal fungi.

Inoculum treatments	Onion bulb weight (tonnes/ha)		% mycorrhizal infection at harvest
	Grade 1	Grade 2	
Control	48.1	3.2	51.3
Granules, 30mm	54.0	2.9	56.9
Loose, 30mm	56.5	2.9	59.4
Granules, 60mm	50.1	2.7	52.8
Loose, 60mm	48.9	2.9	51.8
LSD 10%	5.4	0.6	4.9
5%	6.5	0.8	6.0

All inoculum treatments except loose, 60 mm significantly increased mycorrhizal infection levels (**, $p < 0.01$) on angular transformed data.

Grade 1 onions > 40 mm diameter

Grade 2 onions < 40 mm diameter

(b) **Maize.** *Zea mays* L. We have adapted a tractor drawn maize seeder (model Nodet) to drill VAM inoculum into the maize seedbed along with seed, fertiliser and insecticide. In this case, we geared up the drive shaft to two of the fertiliser bins (by an eight fold speed increase) so that granulated VAM inoculum placed in these two bins could be accurately banded in the seed bed above, below or beside the seed at rates ranging from 0 to 2000 kg/ha. In a trial at Morrinsville, mycorrhizal inoculum drilled at 400 kg/ha resulted in a statistically significant increase in grain yield of 15%. Further trials will be carried out to examine minimum rates of inoculum required and optimum inoculum placement in the seed bed.

(c) **Nonmechanised countries.** Here, the appropriate technology for potential VAM introduction to field soils is vastly different and the animal drawn plough is often the sole means of primary tillage and seed (and fertiliser where used) are placed directly into the plough furrow (Harwood, 1979). A major advance for farmers in the semiarid tropics has been the research development of the animal drawn wheeled tool carrier (Anon., 1980), which can be fitted with implements such as plough, grader blade, seed unit, fertiliser bin, etc. It is quite possible that mycorrhizal inoculum from another bin could be drilled at the appropriate rate and position in the seed bed.

Hand placement of inoculum in nursery beds

(a) **Finger millet** (*Eleusine coracana* (L) Gaertn.) and **chilli** (*Capsicum annum* L.). The first practical application of VAM technology in third world countries will undoubtedly be for transplanted field crops, such as finger millet, chilli and tobacco which are routinely raised from seed in small nursery beds in the farmer's field and transplanted to the field when the plants are 4-8 weeks old. Mycorrhizal inoculum in small quantities can be used to inoculate large numbers of plants in the nursery beds and there are no additional labour or material costs in inoculation.

In a pot trial in India, Govinda Rao *et al.* (1983a) selected three strains of VAM fungi efficient at stimulating the growth of finger millet from a total of 15 field isolates collected. They then set up a field trial in which finger millet seedlings were grown in field beds (1.0 x 0.8 m), as done by farmers, after inoculation with the selected VAM strains (Govinda Rao *et al.*, 1983b). The seedlings were transplanted to the field plots (of 2.0 x 1.0m, with 70 plants/plot) given P fertiliser at 0, 19 or 38 kg P/ha. Available soil P levels before fertiliser application were 2.4 ppm P. At harvest there was an 18 % increase in grain yield over all P fertiliser levels to the inoculant strain M6 (Table 2) but most importantly, the M6 inoculated plants reached the optimum grain yield at 19 kg P/ha while the uninoculated plants (which became infected with the indigenous VAM in the field soil) needed 38 kg P/ha to reach the same yield. Bagyaraj and Sreeramulu (1983) found similar results with chilli also grown in field plots of 3.0 x 1.35 m (available soil P level 6.0 ppm) at three P fertiliser rates after seedlings were raised in nursery beds. At harvest the most efficient VAM inoculant (I4) increased shoot weight and fruit yield of unsterilised plants by 43%.

TABLE 2: Effect of mycorrhizal inoculants and P fertiliser rate on (a) shoot DM (g/plant) and (b) grain yield (kg/plot) of field-grown finger millet plants (from Govinda Rao *et al.* 1983b).

Mycorrhizal inoculant	P fertiliser rates kg P/ha		
	0	19	38
(a) Shoot DM			
Control	26.9	30.5	38.9
M ₆	33.8	39.4	40.3
M ₁₄	28.8	36.1	36.2
Glomus	28.7	38.3	39.3
LSD = 2.37			
(b) Grain yield			
Control	1.09	1.39	1.74
M ₆	1.38	1.73	1.88
M ₁₄	1.35	1.65	1.76
Glomus	1.25	1.53	1.73
LSD = 0.11			

LSD values at $p < 0.05$ are for significant inoculant x fertiliser interactions.

This increase due to inoculation was equal to or greater than the response to a P fertiliser application of 75 kg P/ha (Table 3). Mycorrhizal responses were even larger at the 37.5 kg P/ha fertiliser rate. These trials are good examples of appropriate technology — the use of tested VAM strains, in low quantities in typical nursery plots with VAM growth response compared to that available from P fertiliser alone (Tables 2 and 3). These results from the University of Agricultural Sciences farm at Bangalore, India are being investigated in similar trials with large field plots on the five major soil types throughout the region before the selected fungal inoculants can be recommended for use by farmers.

TABLE 3: Effect of mycorrhizal inoculants and P fertiliser rate on (a) shoot DM (g/plant) and (b) fruit yield (kg/plot) of field-grown chilli plants (from Bagyaraj and Sreeramulu, 1983).

Mycorrhizal inoculant	P fertiliser rates kg P/ha		
	0	37.5	75
(a) Shoot DM			
Control	29.4	31.1	37.0
Glomus	31.8	37.4	
I ₁₄	34.4	38.6	
I ₆	31.4	37.1	
I ₄	42.0	51.2	
LSD (inoc.) = 5.0, LSD (fert.) = 3.8			
(b) Green fruit yield			
Control	1.23	1.30	1.81
Glomus	1.33	1.56	
I ₁₄	1.44	1.61	
I ₆	1.31	1.55	
I ₄	1.76	2.14	
LSD (inoc.) = 0.22, LSD (fert.) = 0.14			

All LSD values significant at $p < 0.01$. Control yields at 75 kg P/ha not included in statistical analyses. No significant inoculant x fertiliser rate interactions.

(b) Asparagus. A very recent asparagus seedling trial in sterilised soil has shown that, in the field, asparagus needs VAM for good growth even at fertiliser levels as high as 200 kg P/ha. Where crowns have been raised in nursery beds in unsterilised soil there should be no problem as their roots will be infected by the indigenous VAM in the soil. Mycorrhizal inoculation is needed however for seedlings raised in nonmycorrhizal peat and sand mix in the glasshouse and transplanted as 3 month old plants to the deep-ploughed trenched soil in the field. Here, the non-mycorrhizal seedlings are often placed by chance in B horizon soil (very low mycorrhizal infectivity) and may remain non mycorrhizal (and stunted) for several months. We have glasshouse trials underway and field trials planned to investigate rates of inoculum required but even at a 1% amendment to the peat/sand mix only 30 kg of mycorrhizal soil will be required to inoculate enough seedlings to cover 1 hectare. If successful, this inoculation procedure can be immediately adopted by growers.

CONCLUSIONS

We have shown in recent field trials in India and New Zealand that VAM inoculation of field crops can successfully increase plant crop yield and that inoculation methods can be made suitable for local farming methods. We emphasise that, in future, all VAM field crop trials be carried out under agriculturally relevant conditions and that hand placing of heavy VAM inoculum rates in a seed furrow is totally impractical and has no relevance in

agriculturally mechanised countries. Future field trials must concentrate on finding out the minimum rates of inoculum required and optimum placement of VAM inoculum for seed-sown field crops.

ACKNOWLEDGEMENTS

We acknowledge the involvement of Graham Wilson (Pukekohe Horticultural Research Station) in this preliminary report of our joint onion trial at Pukekohe and the field assistance of Glenn Clark, Dave Thomsen and the late Bob Kidd (Ruakura Soil and Plant Research Station).

REFERENCES

- Anonymous 1980. Proceedings of the International Symposium on development and transfer of technology for rain fed agriculture and the SAT farmer. ICRISAT, Patancheru, India. p. 324.
- Bagyaraj, D.J., Sreeramulu, K.R. 1983. Inoculation with VA mycorrhiza improves growth and yield of chilli grown in field and saves phosphatic fertiliser. *Plant and Soil*: in press.
- Gerdemann, J.W. 1975. Vesicular-arbuscular mycorrhizae. In "The Development and Function of Roots" Eds. J.G. Torrey and D.T. Clarkson, Academic Press, London. pp. 576-591.
- Govinda Rao, Y.S., Bagyaraj, D.J., Rai, P.V. 1983a. Selection of an efficient VA mycorrhizal fungus for finger millet I. Glasshouse screening. *Zentralblatt fur Mikrobiologie*: in press.
- Govinda Rao, Y.S., Bagyaraj, D.J., Rai, P.V. 1983b. Selection of an efficient VA mycorrhizal fungus for finger millet II. Screening under field conditions. *Zentralblatt fur Mikrobiologie*: in press.
- Harwood, R.R. 1979. Small farms development — understanding and improving farming systems in the humid tropics. Westview Press, Boulder, Colorado. p. 160.
- Maronek, D.M., Hendrix, J.W., Kiernan, J. 1981. Mycorrhizal fungi and their importance in horticultural crop production. *Horticultural Reviews* 3: 172-213.
- Mosse, B. 1973. Advances in the study of vesicular-arbuscular mycorrhiza. *Annual Review of Phytopathology* 11: 171-196.
- Powell, C.Ll. 1981. Inoculation of barley with efficient mycorrhizal fungi stimulates seed yield. *Plant and Soil* 62: 231-239.
- Ross, J.P. 1971. Effect of phosphate fertilisation on yield of mycorrhizal and nonmycorrhizal soybeans. *Phytopathology* 61: 1400-1403.
- Yost, R.S., Fox, R.L. 1979. Contribution of mycorrhizae to P nutrition of crops growing on an oxisol. *Agronomy Journal* 71: 903-908.