

VA MYCORRHIZAE AND ASPARAGUS

C.L.I. Powell and D.J. Bagyaraj

Ruakura Soil and Plant Research Station, Ministry of Agriculture and Fisheries, Hamilton

ABSTRACT

Asparagus plants from 16 field plantations (mainly in Waikato and Manawatu) were sampled for the presence of VA mycorrhizal fungi. Root systems of 6-10 plants were sampled in each plantation and the mean mycorrhizal infection level over all sites was 68%. Infections were invariably dense and well established even in young plantations and were observed in all varieties examined (Mary Washington, UC72, Limbras, Lucullus and Rutgers Beacon).

In a seedling bed trial in (methyl-bromide) sterilised Horotiu sandy loam at Rukuhia Horticultural Research Area, non-inoculated Mary Washington plants weighed only 0.23 g/plant at 15 months after seeding in the field, while plants inoculated with VA mycorrhizal inoculum had a mean DM of 4.11 g/plant. Mycorrhizal inoculation also increased plant survival rate by 19% from 42 to 50 plants/m². There was no effect of P fertiliser at 0, 100, 200 or 400 kg P/ha on the growth or survival rate of mycorrhizal or non-mycorrhizal asparagus. It is suggested that asparagus has an extremely high mycorrhizal dependency in most soils.

Additional Keywords: nursery bed, field trial, P fertiliser, survey.

INTRODUCTION

VA mycorrhizal fungi are common soil fungi in New Zealand and throughout the world and rapidly colonise roots of agronomically important plants in virtually all agricultural soils (Mosse, 1973). Asparagus is an increasingly important export crop for New Zealand with 3000 ha currently planted nationally. We have recently carried out a detailed mycorrhiza research programme on the crop after a demonstration of large field responses to accidental inoculation occurred in 1982.

A block of land at Rukuhia Horticultural Research Area had been sterilised with methyl bromide so that the phytotoxic effects of some pre and post emergence herbicides on asparagus seedlings could be determined. The asparagus seed was drilled on January 1982 and by April it was obvious that the plants in the sterilised soil were severely stunted and phosphate-deficient. By contrast, asparagus plants in the guard plots around the outside of the sterilised block were large, healthy and vigorously growing. Mycorrhizal examinations showed that the stunted plants in the fumigated block were non-mycorrhizal while the large guard plants were highly mycorrhizal.

We therefore carried out a variety of nursery and field trials to see if we could use mycorrhizal inoculation to increase crop yields. This paper describes part of this programme in which a mycorrhizal inoculation x P fertiliser trial was carried out on the sterilised block to quantify the effects of mycorrhizal fungi on asparagus seedling growth. The experimental findings are directly applicable to nurserymen and all horticulturalists who sterilise soils before raising crops.

MATERIALS AND METHODS

Field Trial

(a) *Site.* On July 3 1981, the trial site on Horotiu sandy loam at Rukuhia Horticultural Research Area had a basal topdressing of hydrated lime, superphosphate and a 12:12:12 NPK fertiliser at 1.0, 1.1 and 0.9 tonnes/ha respectively. After a spring fallow, the soil was ploughed and rotary hoed. On December 20 1981, it was sterilised with methyl bromide at 100 g ai/m² under an inflated black polythene tent. The tent was removed after 4 days and the soil allowed to vent. The soil was rolled on January 15 1982 and asparagus seed (cv Mary Washington) machine drilled into 1.0 m wide beds with 5 rows at 200 mm spacings. Asparagus seed was also drilled into guard plots on non sterilised soil around the perimeter of the sterilised block.

(b) *Mycorrhizal inoculation.* The mycorrhizal inoculation x P fertiliser trial was superimposed on the existing asparagus seedlings in the sterilised bed on May 4 1982. At this stage, soil properties were pH, 6.0; Olsen P, 26 µg P/ml; P retention, 95%.

Phosphate fertiliser as lbex P was applied over each entire 2 x 1 m plot at rates equivalent to 0, 100, 200 or 400 kg P/ha. The fertiliser was mixed into the top 40 mm of soil by hand using a spintiller.

Mycorrhizal inoculum was sidedressed against the asparagus seedlings (now 80 mm high) at 0 or 140 g air-dried inoculum soil/metre of row. Only the inner 3 rows of the 5 row plots were inoculated (with the outer 2 rows as guards). In addition, inoculum was only applied to 1.4 m of the 2 m length of each plot leaving a 0.3 m guard strip at each end of the plot. Thus, 0.84 m² (1.4 x 0.6 m) of each 2

m² plot received inoculum, at the rate of 0 or 600 g/plot equal to 0 or 7.1 tonnes/ha. Inoculum was worked into the top soil next to the asparagus seedlings using the spintiller. Inoculum had been prepared by growing red clover for 3 months in methyl-bromide sterilised Horotiu sandy loam (Olsen P 15 µg P/ml), reinfested with a mixed starter culture of *Glomus* spp. Inoculum soil was dried, the plants removed and freed of large clover root masses before use as an inoculant for the asparagus trial.

TABLE 1: Mycorrhizal infection levels (% root segments infected) in roots from field-grown asparagus plants.

Grower No	District	Asparagus cultivar	Mycorrhizal infection ^b level (% roots infected)
1	Waikato	Mary Washington	83
2	Waikato	Mary Washington	66
3 ^a	Waikato	Mary Washington	90
4	Bay of Plenty	Mary Washington	89
5	Waikato	Mary Washington	82
6	Nelson	Mary Washington	42
7	Manawatu	Rutgers Beacon	57
8	Manawatu	Rutgers Beacon	46
9	Manawatu	Rutgers Beacon	61
10	Manawatu	Rutgers Beacon	68
11	Manawatu	Rutgers Beacon	78
12	Waikato	Rutgers Beacon	95
13	Waikato	Rutgers Beacon	82
3 ^a	Waikato	Limbras	68
14	Waikato	Limbras	46
15	Waikato	Limbras	80
16	Bay of Plenty	Lucullus	75
3 ^a	Waikato	UC72	39
Mean			69

a 3 cultivars sampled at same grower

b Values are means of 6-10 separate root samples from each grower

(c) Measurements

Mycorrhizal infection. Roots were cut from 2 plants at random in each plot on August 8 and December 3 1982 and at March 3 1983. Feeding roots were washed, cleared and stained (Phillips and Hayman, 1970) and root segments examined as before (Powell, 1982) for mycorrhizal infection.

Plant dry matter. At December 3 1982, 1 plant was randomly taken from each plot and oven dried to constant weight. At March 3 1983, the experiment was destructively harvested. All plants from the 0.84 m² treated area in each plot were dug up, the roots removed and the plant tops washed and dried. By the time of harvest, some of the non-inoculated plots had been contaminated with mycorrhizal fungi and all plants were graded as mycorrhizal or non-mycorrhizal before computing means and statistical analyses (see Table 2). Total plant density in the treated

area of each plot was measured. The shoot tissue from 5 randomly selected plants from each plot was analysed for percentage phosphorus concentration by a phosphomolybdic method after dry ashing and HCl extraction.

Results were analysed by 2 way ANOVA after log, angular and square root transformations of the dry matter, % mycorrhizal infection and plant density data respectively.

TABLE 2: Effect of mycorrhizal inoculation on dry matter production of asparagus seedlings at 7 and 10 months after inoculation.

Mycorrhizal inoculation	Shoot DM (mg/plant)		
	7 months	10 months ^a	10 months ^b
-	59	224	1324
+	69	4108	4108
Significance	ns	***	***
LSR ^c (5%)	1.36	1.29	1.77

a 20% of all plants in uninoculated plots were considered mycorrhiza-contaminated and these were excluded before statistical analysis and calculation of means.

b Mycorrhiza-contaminated plants in uninoculated plots included in plot totals for calculating means and statistical significance etc.

c Least Significance Ratio (LSR) is shown as data were log transformed before analysis.

Grower Survey

In conjunction with the field trial, asparagus root samples were taken in May-June 1982 from well established 2-5 year old crops on 16 growers' properties in the Waikato, Manawatu and Bay of Plenty regions. All growers except no. 3 only grew one cultivar (Table 1). For each grower, 6-10 root samples were processed and examined for mycorrhizal fungi and the infection level expressed in Table 1 as a mean figure over all samples.

RESULTS AND DISCUSSION

Grower Survey

Table 1 shows that mycorrhizal infection levels were generally high in all root samples, with an overall mean of 69%. There was no significant difference in infection levels of the different cultivars tested. Root infections were invariably very dense with many external hyphae and extensive fungal colonisation of the cortical cells. This confirmed that asparagus was similar to most crops in forming extensive mycorrhizal infections over a range of soils and soil fertility conditions (Powell, 1982). It also showed that the severe soil disturbance involved in establishing an asparagus crop (deep ploughing and trenching with crowns often planted into the B horizon) did not prevent buildup of extensive mycorrhizal infections.

Field Trial

Inoculation with mycorrhizal fungi greatly increased the growth of asparagus seedlings in the sterilised nursery bed (Table 2) but the growth response took several months to develop. The mean mycorrhizal infection levels in the inoculated plots in August and December 1982 and at March 1983 were 5%, 23% and 87% respectively. This seems to be a relatively slow initial spread of mycorrhizal inoculant into the existing roots with a rapid increase in infection over summer. This delay in infection spread probably occurred because the inoculum had to be applied (of necessity) above the active root zone and is unlikely to be due to residual effects of methyl bromide or indirect effects of sterilisation on microbial populations in the soil.

For mycorrhizal inoculation of a commercial asparagus nursery crop, inoculum soil should be directly layered into the furrow with the seed (Powell and Bagyaraj, 1982). In this case, inoculum rates could easily be reduced to 0.5-1 tonne/ha instead of the 7 tonnes/ha used in this trial.

Phosphate fertiliser rate had no effect on mycorrhizal infection level at any sampling date.

The development of the large growth response to inoculation was preceded by the rise in mycorrhizal infection level as found previously with other crops (Smith and Daft, 1977). In December 1982 (7 months after inoculation) when infection levels had risen to 23% in inoculated plots, there was still no effect apparent on plant growth (Table 2). By March 1983 (at harvest), when mycorrhizal levels had risen to 87%, inoculation resulted in a 17 fold increase in plant dry matter production.

By harvest time, an average of 20% of plants in non-inoculated plots had become mycorrhiza-contaminated. This was easily recognised in non-inoculated plots as rapidly enlarging islands of large green healthy plants amongst the mass of stunted plants (Klienschmidt and Gerdemann, 1972). When these plants were included in the individual plot yields, the apparent response to mycorrhizal inoculation was down to 210% (Table 2).

In commercial practice however, it is unlikely that widespread mycorrhizal contamination (from agricultural machinery, wind blown soil or unsterilised B horizon) could be relied on to inoculate the crop as it was not until a year after sterilisation that contamination became widespread: most asparagus seed crops are lifted (as crowns) 6-8 months after seeding.

Phosphate fertiliser at 100-400 kg P/ha had no effect on the growth or survival rate of non-inoculated or inoculated plants and was no substitute for mycorrhizal infection for asparagus. This is an amazing result as one would expect a heavy phosphate fertiliser application such as 400 kg P/ha to increase the growth of the severely stunted non-mycorrhizal plants. In fact, there was no effect at all of P fertiliser rate on percentage P concentration in asparagus shoot tissue at harvest (overall mean 0.204%). This suggests that non-mycorrhizal asparagus roots were completely unable to recover any of the phosphate applied to the soil as fertiliser. This in turn suggests that asparagus, with its poorly developed root system is almost wholly dependent on infection by VA mycorrhizal fungi for

effective P uptake and growth in P-retentive soils.

Mycorrhizal inoculation significantly increased plant survival rate by 19%, from 42 to 50 plants/m². This would almost certainly increase the number of saleable plants per square metre, although plants were not graded for this at harvest.

Although this trial was only carried out once at one site on a soil with high P retention, the response was so large that worthwhile responses to inoculation would be expected on most sterilised soil types. It should be noted that this demonstration of the need for mycorrhizal re-inoculation in sterilised nursery beds has been made with asparagus but similar responses would probably occur in other crops also raised in sterilised beds.

CONCLUSIONS

1. Methyl bromide sterilisation can kill VA mycorrhizal fungi in nursery soil beds.
2. Asparagus plants grown in sterilised nursery beds will need re-inoculation with mycorrhizal fungi in the seed furrow. Large additions of P fertiliser will probably be unable to compensate for the lack of mycorrhizal fungi.
3. Many horticultural crops currently raised or grown in sterilised soils will probably respond to mycorrhizal inoculation.

ACKNOWLEDGEMENTS

We wish to thank Mr P. Sanders for bringing our attention to the mycorrhiza problem in asparagus and for allowing us to use his trial site. We also acknowledge the technical assistance of Dr W. Bussell, Mrs K. Caldwell and Mr G. Clark in the field trial and grower survey.

REFERENCES

- Kleinschmidt, G.D., Gerdemann, J.W. 1972. Stunting of citrus seedlings in fumigated nursery soils related to the absence of endomycorrhiza. *Phytopathology* 62: 1447-1453.
- Mosse, B. 1973. Advances in the study of vesicular-arbuscular mycorrhiza. *Annual Review of Phytopathology* II: 171-196.
- Phillips, J.M., Hayman, D.S. 1970. Improved procedures for clearing roots and staining parasitic and vesicular-arbuscular mycorrhizal fungi for rapid assessment of infection. *Transactions of the British Mycological Society* 5: 158-161.
- Powell, C. Ll. 1982. Mycorrhizae. In 'Experimental Microbial Ecology' Eds Richard G. Burns and J. Howard Slater, Blackwell Scientific Publications, Oxford. pp 447-471.
- Powell, C. Ll., Bagyaraj, D.J. 1982. VA mycorrhizal inoculation of field crops. *Proceedings Agronomy Society of N.Z.* 12: 85-88.
- Smith, S.E., Daft, M.J. 1977. Interactions between growth phosphate content, and N₂ fixation in mycorrhizal and non-mycorrhizal *Medicago sativa*. *Australian Journal of Plant Physiology* 4: 403-413.