Possibilities for Manipulating Mycorrhizal Associations in Crops

K.K. Lee[1] and S.P. Wani[1]

Abstract

Vesicular-arbuscular mycorrhizae (VAM) occur in the roots of most crops and are known to play an important role in crop growth. Legumes are quite responsive to VAM, especially in soils with low available phosphorus (P) levels. Possible approaches to manipulating VAM associations may be (1) inoculation with selected VAM fungi, (2) selection of plant genotypes that are conducive to colonization by efficient VAM fungi, and (3) establishment of a soil environment that favors increased VAM number and activity. This chapter evaluates the feasibility of each of these possible approaches, using appropriate examples from the literature and from our experience.

Manipulating VAM associations through inoculation is not feasible on a field scale unless pure VAM fungi can be grown in large quantities by standard microbiological techniques. There is also increasing evidence that the magnitude of VAM activity differs among plant genotypes, and numerous studies have indicated that VAM fungal status can be altered by soil management. Our experience in exploiting the VAM fungus along with the use of rock phosphate in West Africa has revealed the possibility of improving crop production in low-P soils, using rock phosphate as a P fertilizer.

Introduction

Vesicular-arbuscular mycorrhizae (VAM) occur in the roots of most crop species, and VAM fungi are known to be ubiquitous in agricultural soils. The VAM fungus is an important component of the rhizosphere and creates a mutually beneficial root-fungus association, which has been extensively documented in recent years (e.g., Hayman 1974; Mosse 1980). It is well established that VAM infection can markedly improve plant growth in soils where nutrient concentrations are suboptimal. Of all the nutrients, P is the one most studied in relation to VAM, because improved growth by VAM infection is most often correlated with P uptake by the plant. Thus the extent of VAM benefit is often assessed in terms of improved P uptake rather than of increased biomass production.

It would be most beneficial if VAM associations could be manipulated and utilized to increase crop production in areas where fertilizer availability is limited. Possible approaches to manipulating VAM fungi associated with annual crops may be (1) inoculation with selected VAM fungi, (2) selection of plant genotypes that preferentially favor colonization by efficient VAM fungi, and (3) establishment of soil environments more conducive to proliferation of VAM fungi in the rhizosphere and their enhanced symbiotic function.

We assess each of these approaches here by reviewing the literature and by drawing on our experience in using VAM in West Africa. The first approach is discussed at length, as inoculation with VAM fungi in the field is often considered a feasible way of manipulating the VAM-crop association. This review is

1. Resource Management Program, International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Patancheru, A.P. 502 324, India.

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Inoculum Density and Rate and Extent of VAM Infection

Rapid and extensive infection is an important factor in determining both the effectiveness of a VAM fungus in increasing plant growth, and its ability to compete with other fungi for infection (Wilson 1984a). Formation of primary infection points is therefore important, and their number is known to be controlled by inoculum level, more specifically, by inoculum density, that is, the number of propagules g^{-1} of soil. Many inoculation trials have been conducted with unknown quantities of inoculum or inoculum concentrations based on spore number. However, the spore number alone does not determine the number of infection sites or the extent of infection. The number of propagules available for infection is more relevant to the measurement of infectivity or root colonization. Haas and Krikun (1985) investigated the relationship of root colonization with inoculum density. Root colonization by VAM fungus was correlated with inoculum density, which in turn was correlated with the height and mass parameters measured in bell pepper (*Capsicum annuum* L.) seedlings.

As inoculum density is an important parameter in assessing the quality of inoculum, it is useful to compare fungi at similar inoculum densities for selecting fungal species with high infectivity and high rates of infection development. Wilson (1984a) demonstrated that the infectivity and rate of infection development of *G. fasciculatum* were greater than those of *G. decipiens*.

Efficiency of VAM Fungus

An introduced VAM fungus that can compete with indigenous VAM fungi and initiate infection is not necessarily successful; it should also be efficient in enhancing plant growth. This efficiency is difficult to measure, because enhanced plant growth results from the combined effects of the fungus and many processes in infected plant and in the soil. In general, the efficiency of the VAM is measured by comparing the growth of plants inoculated with various VAM species and strains.

The responsiveness to VAM inoculation differed with plant species, VAM species, and even with isolates of the same species (e.g., Clarke and Mosse 1981; Daft 1983; Van Nuffelen and Schenck 1984; Jensen 1984). In the strict sense, comparison of efficiency should be made with known inoculum density. As discussed earlier, Haas and Krikun (1985) tested isolates of *G. macrocarpum*, at known numbers of propagules, for their efficiency in enhancing plant growth of bell pepper. Efficiency varied considerably, both among *G. macrocarpum* isolates collected from different soils and among isolates collected from the same soil. From these studies, it seems evident that efficient VAM for particular host-plant species are available and that it is possible to select the most appropriate fungi for an inoculation program.

Selection of Plant Genotypes More Responsive to Indigenous VAM

Although the extent of root colonization and responsiveness to VAM fungi are known to differ from one plant species to another, much less is known about the differences among genotypes within a plant species. Heckman and Angle (1987) examined 15 soybean cultivars for colonization by a heterogeneous field population of VAM fungi at two P levels and found that colonization varied significantly with cultivar. Addition of P to the soil reduced colonization, but there was no cultivar × P interaction. Krishna et al. (1985) found that root colonization by indigenous VAM fungi differed among 30 pearl millet genotypes. In another experiment, with two male-sterile lines, restorer lines, and their derived crosses, Krishna et al. (1985) also found that root colonization differed significantly among pearl millet genotypes, suggesting that the trait for VAM colonization is heritable.

Because genotypes within a plant species show different levels of root colonization by indigenous VAM fungi, an important consideration is whether the level of colonization can be used as a selection criterion for responsiveness of plant growth to VAM fungi. However, VAM infection is not always beneficial to the host plant; it can be parasitic under poor light and temperature conditions (Hayman 1983). Azcon and Ocampo (1981) found that variation among wheat cultivars in root colonization by *G. mosseae* was not correlated with growth response. On the contrary, Wilson (1984a) proposed that the extent of infection is an important factor in determining the effectiveness of VAM fungi in increasing plant growth. In pearl millet, total mycorrhizal root length and percentage colonization were correlated with total root length, shoot and total dry matter, and total P uptake (Krishna et al. 1985).

At present, no conclusive and clear relationship is apparent between the levels of mycorrhizal infection and plant responsiveness to VAM fungi. As Hayman

infection, perhaps because higher P levels were applied in the field than those used in pot experiments. However, increased root infection with increasing P application has also been observed. For example, cassava roots showed increased infection by *Glomus manitrotis* with increasing P application up to 200 kg P ha^{-1}, although infection by other VAM fungi decreased with increasing P application (Sieverding and Howeler 1985). Clark and Mosse (1981) compared indigenous and introduced VAM fungi in their response to P application (82.6 kg P ha^{-1}) and found that in barley roots, added P depressed infection by indigenous VAM more than it depressed infection by the introduced VAM fungus.

The level of P fertilizer at which VAM development and activity are best enhanced depends on: the levels of P available to both plant and VAM in the soil, the host plant, the VAM species, and the soil environment. It should be noted that P levels adequate for growth of the host plant are not usually those suited for VAM development and activity. However, considering that P is essential for both the host plant and the VAM fungus, it is obvious that a certain level of available soil P should be ensured for the crop to derive maximum benefit from VAM associations.

A further question is whether sparingly soluble phosphates, such as rock phosphate, can enhance VAM growth. There are numerous reports that mycorrhizal plants can take up P from sparingly soluble phosphate more readily than nonmycorrhizal plants can (Mosse 1980). This is now attributed not to the ability of VAM fungus to dissolve relatively insoluble P but to its capacity to better scavenge the low levels of soluble P in and around rock phosphate particles. Rock phosphate might improve the residual value of applied P in the soil; in addition, it does not reduce the level of VAM infection as soluble P fertilizer does (Barea et al. 1980).

Organic manures have long been known to increase the population and the activity of microorganisms involved in soil fertility. However, information on the effect of organic manures on VAM growth is scarce. The effect of straw on VAM sporulation has been examined in relation to soil tillage (Kruckelmann 1975). Chopped straw applied before tillage over 10 years greatly increased VAM spores in shallow-plowed soil, but did not do so in soil tilled with a rotary hoe or under zero tillage. Harinikumar and Bagyaraj (1989) found that the application of farmyard manure (FYM) at 7.5 t ha^{-1} significantly increased root colonization of VAM and VAM spore numbers in the second season. This effect was carried over even to the third season without further application of FYM. The application of inorganic fertilizer alone at the recommended level (for example, 160 kg N, 33 kg P, and 31 kg K ha^{-1} for maize) reduced VAM infection, but fertilizer application at 50% of the recommended level together with FYM did not reduce VAM root colonization and spore numbers in comparison with the FYM alone treatment. Kruckelmann (1975) has reported that the effect of FYM on mycorrhizal status was not consistent and was influenced by soil type. In a silty clay loam soil at Rothamsted, the most VAM spores were found in the nonmanured plot, whereas in a loamy sand, fewer spores were found in the nonmanured plot. It is likely that organic manure affects VAM growth indirectly, by improving soil conditions, such as water-holding capacity, aggregate formation, and nutrient composition. However, direct effects may also be involved; for instance, *G. mossae* and *G. caledonium* were shown to survive and grow saprophytically as hyphae in organic matter (Warner 1984).

Tillage

Soil disturbance changes physical, chemical, and biological properties of the soil, and thus affects root growth and distribution. Consequently, tillage practices may affect the root-VAM association. Excessive secondary tillage and traffic reduced the colonization of common bean (*Phaseolus vulgaris*) root by VAM (Mulligan et al. 1985). An inverse relationship was found between VAM colonization in common bean and bulk density of the clay. The suggested mechanisms are that compacted soils limit root growth and root length, and therefore reduce the frequency with which roots encounter VAM spores. The decrease in VAM colonization in compacted soils may also result from reduced soil aeration. Although lowered oxygen (O_2) concentration may not be the sole cause of decreased VAM infection, it greatly influences the activity of VAM fungi. Saif (1983) found that the effect of soil O_2 was more pronounced in mycorrhizal plants than in nonmycorrhizal plants. The VAM fungi differed in their response to different O_2 levels, but increasing levels of O_2 increased the growth of all mycorrhizal plants in the same way, with an optimum O_2 concentration of 16%. Saif (1983) concluded that, to derive the greatest benefits from mycorrhizal associations, it is essential to maintain soil aeration at a high level.

The effects of tillage practices on VAM infection have been also studied in relation to P absorption by plants. Evans and Miller (1988) demonstrated that dis-

Each field was cropped with groundnut in 1988 under four treatments: (1) control, (2) rock phosphate (17 kg P ha^{-1}), (3) inoculation with *G. clarum*, and (4) rock phosphate plus *G. clarum*. Treatment 4 increased dry shoot mass and nodule number in the infertile soil, but had no effect in the virgin soil (Table 2). Overall spore numbers were generally higher in infertile soil. Spore numbers were the highest with rock phosphate plus *G. clarum* in virgin soil, but with *G. clarum* alone in infertile soil.

As discussed earlier, a potential for manipulation of VAM association through inoculation exists if host-VAM fungus features such as specificity, competitiveness, and efficiency are exploited. There are indeed reports on successful inoculation of nonsterilized soils in pots (e.g., Barea et al. 1980, Manjunath and Bagyaraj 1986) and in the field (e.g. Clarke and [illegible]se 1981; Hall 1984). It may be expected that inoculation responses can be obtained in carefully controlled pot and field experiments conducted by experienced scientists. However, farmers may face practical problems, particularly in selecting appropriate fungi and in using the correct inoculation techniques. It is almost impossible to select the best VAM fungus species for different soil environments by studying the literature. Until widely effective strains of VAM fungus are available, the controlled experiments, such as pot trials, are required to determine specificity, competitiveness, effectiveness, and efficiency of VAM fungus strains against indigenous VAM fungi.

Another immediate problem to be solved is the requirement for large quantities of inoculum. Since the VAM fungi are thought to be obligate symbionts on plant roots, because an artificial medium for independent growth of VAM fungus has not been identified, crops must be inoculated with VAM-infested roots or soil, or with VAM spores sieved from VAM-infested soil. Generally, annual field crops require inoculum rates of several tons per hectare, at least as VAM-infested soil. In our experiment in Niger, sieved spores mixed with local soil were used as inoculum for groundnut. However, the amount of original infested soil used in these trials was about 3 t ha^{-1}, as calculated by spore density in our stock culture. These inoculum rates seem impractical for wide utilization of VAM fungi on a field scale. The feasibility of VAM inoculation on a field scale thus depends

Table 2. Effect of rock phosphate[1] and vesicular-arbuscular mycorrhizal (VAM) inoculation on growth and VAM infection of groundnut, ICRISAT Sahelian Center, Sadoré, Niger, 1988.

	Control	Rock phosphate	VAM	Rock phosphate + VAM	SE
Shoot dry mass (t ha^{-1})					
Virgin soil	1.81	2.13	1.70	2.34	±0.103
Infertile soil	0.66	0.92	0.92	1.03	±0.054
[illegible]ule no. $plant^{-1}$					
[illegible]rgin soil	190	278	199	295	±9.2
Infertile soil	87	87	104	156	±9.5
Grain yield (t ha^{-1})					
Virgin soil	1.13	1.26	1.19	1.32	±0.350
Infertile soil	0.32	0.50	0.46	0.52	±0.372
VAM infection (%)					
Virgin soil	49	54	52	61	±1.2
Infertile soil	56	53	63	67	±2.9
VAM spores (no. g^{-1} wet soil)					
Virgin soil	12.6	9.0	11.5	18.5	±0.77
Infertile soil	9.3	7.1	10.2	10.2	±0.51

1. Rock phosphate applied at 17 kg P ha^{-1}.

Source: M.J. Daft et al., 1989, University of Dundee, personal communication.

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