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SUMMARY

Isolation of endomycorrhiza (vescicular arbuscular mycorrhiza) spores from five semi arid sites of Kenya revealed that the soils differed in their spore populations. No relationship was found between the spore populations and the pH nitrogen and phosphorus content of the soils. Inoculation of M. volkensii seedlings with vescicular arbuscular mycorrhiza (VAM) derived from Mwingi and Kiambere soils significantly increased height growth. Mycorrhiza infection was also high on inoculated seedlings in sterile soil. Slight infection was found on seedlings in unsterile soils that were not inoculated. The results indicate that inoculation of seedlings may be useful especially under adverse soil conditions.

INTRODUCTION

Mycorrhizae are symbiotic associations between certain fungi and roots of plants. There are two major types of mycorrhiza associations, ectotrophic and endotrophic or vescicular arbuscular mycorrhiza (VAM). Mycorrhizae have been found to improve the growth of plants by enhancing the uptake of nutrients especially phosphorus and water. About 95% of trees in tropical forests form endomycorrhiza (Le Tacon et al, 1986).

VAM associations have been found on trees growing in the dry areas especially on Acacias. Inoculation of Acacia tortilis with VAM has been found to enhance its survival and growth (Wilson et al, 1991). However studies on mycorrhiza associations in dry areas are few especially on some of the important indigenous tree species such as *M. volkensii.* Observations of root samples of this species from the field revealed that it had VAM association (Mwangi and Munga, 1990). This study was therefore undertaken to determine the effects on inoculation of *M. volkensii* seedlings with VAM derived from soils from some semi-arid areas of Kenya.

MATERIALS AND METHODS

Preparation of Inoculurn

Soil samples were collected from five semi-arid sites; Mwingi, Kiambere, Kibwezi, Marsabit and Kwavonza. Maize was grown in these soils as bait plants for mycorrhiza. Since VAM do not produce spores in fruit bodies like ectomycorrhiza, infected roots or soilborne spores are usually used as inoculum. After three months in the glasshouse, maize roots were obtained and thoroughly washed in water. The roots were cut into small pieces of one centimetre lengths, mixed and used as inoculum.

Isolation of Spores and Soil Analysis

Fifty grammes of soil samples were taken from soils from each site for isolation of spores following the method described by Gerdemann and Nicolson (1963). The isolated spores were then transferred into grided petri dishes containing water and counted. Soil samples were also analysed for N and P as described by Olsen and Cole (1954). The soil pH in water was also determined.

Inoculation and assessments

Forest soil was sterilized by autoclaving at 121 °c at 103.35Kpa (15psi) for one hour. Part of the sterile soil was transferred into polythene tubes and filled to two thirds. Two grammes of inoculum was spread evenly in each tube after which more sterile soil was added. However only inoculum from Kiambere and Mwingi were used which were selected to represent soils differing in spore populations and P content. The control treatment consisted of sterile uninoculated soil. An additional treatment was set using unsterile soil without inoculum. Seeds of M. ***volkensii,*** pre-germinated on Kimpak in growth chambers, were placed into the tubes and covered with a thin layer of soil. The treatments were arranged in a complete randomized block design consisting of ten replicates in a glasshouse.

The seedlings were assessed monthly for height growth. Mycorrhiza infection was assessed after four months. For mycorrhiza assessments, roots were washed using tap water, cleared and stained following the method of Phillips and Hayman (1970). They were then transferred in water in grided petri dishes to determine the infection using the grid line intersect method (Giovannetti and Mosse, 1980).

Results

The results of spore isolation and soil analysis are shown in Table 1.

Table 1. Mean Number of Spores, Soil pH, P and N Content of the Soils.

Soil source	Soil pH(H_2O) 1:2.5	N (%)	P (PPM)	Mean No. of spores (50g soil)
Mwingi	8.1	0.167	21.78	110
Kiambere	7.2	0.104	2.94	23
Kibwezi	5.0	0.097	1.32	138
Marsabit	7.8	0.391	106.60	26
Kwavonza	7.1	0.055	1.32	31

The result in Table 1 show that there were significant differences among some soils in spore population, N and P content and pH. Soils from Mwingi and Kibwezi had significantly highest spore populations:' Most of the spores were yellow to dark brown in colour and probably belong to the genus Glomus. However species identification was not done.

The growth of *M. volkensii* seedlings was significantly increased (P >0.01) after inoculation with VAM in sterile soil (Table 2). This improved growth was also reflected by the high percent mycorrhiza infection observed on inoculated seedlings. A large number of vesicles were also found on inoculated but not in the uninoculated seedlings. There was no infection on uninoculated seedlings.

Table 2. Mean height and mycorrhiza infection of *M. volkensii* seedlings.

Treatment	Mean height (cm)		% mycorrhiza infection	
	Mwingi	Kiambere	Mwingi	Kiambere
Sterile soil (inoculated)	20.4	25.9	79.0	70.4
Sterile (uninoculated)	12.2	18.0	None	None
Unsterile soil(uninoculated	11.9	16.4	3.0	4.5

DISCUSSION

The difference in spore populations found among the soils from the five sites do not seem to relate to soil pH, N and P content for all the soils. This shows that spore populations in soils may depend on other factors not analysed in this study. Several other factors e.g. soil moisture, weeds, organic matter and host species influence sporulation of VAM (Dodd et al, 1990). However N and P levels in the soil are also known to influence mycorrhiza infection and hence could also have an effect on spore populations in the soil.

Height growth of seedlings that were inoculated with VAM increased significantly. Similar effects have been reported by several researchers in both trees and crops. The study seems to indicate that there is no relationship between the quantity of spores in soil and mycorrhiza infection. Hayman (1970) found that VAM infection was most abundant in plots containing more Endogone spores. However, Read et al, (1976) found no correlation between spores population and level of mycorrhiza infection. These conflicting results may be due to problems in determining the amount of viable spores in the soil as some may be dead and hence ineffective. The study however indicate that there may be a critical amount of VAM needed to improve the growth of seedlings which may not be met by indigenous mycorrhiza. This was shown by the similar growth of seedlings in both sterile and unsterile uninoculated soils. The unsterile soil would be expected to contain indigenous VAM spores which was shown by

the infection observed on the roots of seedlings but was absent on those from the sterile uninoculated soil.

CONCLUSION

Soils from five semi-arid sites were found to contain varying amount of VAM spores. However,the spore populations could not be directly related to pH, N and P level in the soil and other factors may be involved. The study has established that there is potential for improving tree growth in dry areas through inoculation with VAM.

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REFERENCES

Dodd, J.C., I. Arias, F. Koomen and D.S. Hayman, 1990. The management of populations of vesicular arbuscular mycorrhizal fungi in acid infertile soils of a Savannah ecosystem. Plant and soil, 122:241-247.

Gerdemann, J.W. and T.H. Nicolson, 1963. Spores of mycorrhizal Endogone species extracted from soil by wet sieving and ecanting method. Trans. Brit. Mycol. soc. 46:397-418.

Giovannetti, M and B. Mosse, 1980. An evaluation of techniques for measuring vesicular arbuscular mycorrihizal infection in roots. New Phytologist 84:489-500.

Hayman, D.S. 1970. Endogone spore numbers in soil and vesicular arbuscular mycorrhiza in Wheat as influenced by season and soil treatment. Trans. Brit. Mycol. Soc. 54:53-63.

Le Tacon, F.J. Garbaye and G. Carr. 1986. The use of mycorrhizas in temperate and tropical forests. 18th. IUFRO Congress. Ljubljana P.513-524.

Mwangi L.M. and F.M. Munga, 1990. Recent advances in mycorrhiza research and its potential for forestry development in Kenya. Proc. of 1st KEFRI Annual Conference. (in preparation).

Olsen S.R. and C.V. Cole, 1954. Estimation of available phosphorus in soils by extraction with Sodium bicarbonate. USDA Circ. 939.

Phillips J.M. and D.S. Hayman, 1970. Improved procedures for clearing roots and staining parasitic and vesicular arbuscular mycorrhiza fungi for assessment of infection. Trans. Brit. Mycol. Soc. 55:158-161.

Read D.J., H.K. Koncheki and J. Hodgson, 1976. Vesicucular arbuscular mycorrhiza in natural vegetation systems. New Phytologist 77:641-665.

Wilson J., K. Ingleby, P.A. Mason, J. Jefwa, P.N. Muthoka, J. Mcp Dick and R.R.B. Leaky, 1989. Tree establishment in semi-arid lands of Kenya. Role of mycorrhizal inoculation and water retaining polymer. Forest Ecology and Management. 45:153-163.