

## Distribution of VA Mycorrhizal Spores in Sandy Beach Soils under Cashew

AZIZAH CHULAN (HASHIM), OTHMAN YAACOB, A.J.M. KAMAL and S. PARAMANANTHAN  
Soil Science Department,  
University Pertanian Malaysia, Serdang, Selangor, Malaysia.

**Key words:** VAM spores; distribution; sandy beach soils; cashew; Malaysia.

### RINGKASAN

Tanah berpasir dari horizon permukaan (0-15 cm) di bawah tanaman gajus telah disampel dari kawasan-kawasan pantai di Besut dan Kuantan (Malaysia) untuk spora-spora mikoriza. Pengasingan spora-spora mikoriza telah dibuat daripada kedua-dua siri Rudua dan Jambu di Besut dan juga dari siri Rudua dan Baging di Kuantan. Sampel-sampel tanah dari Kuantan mengandungi lebih banyak spora mikoriza dari tanah-tanah di Besut. Keputusan kajian ini telah menunjukkan kaitan positif di antara bilangan spora mikoriza yang diasingkan dengan peratus kandungan bahan organan yang sedia ada di dalam sampel tanah itu. Pengasingan jumlah spora yang tinggi dari kawasan Kuantan (berbanding dengan bilangan spora dari Besut) berkemungkinan mempunyai kesan ke atas pengambilan baja fosforus oleh pokok-pokok gajus. Ini selanjutnya mungkin dapat menerangkan mengapa berlakunya perbezaan hasil yang diperolehi dari kedua-dua kawasan pantai ini.

### SUMMARY

Surface horizons (0–15 cm) of sandy beach soils from Besut and Kuantan, (Malaysia) under cashew (*Anacardium occidentale* L) were sampled for Vesicular-Arbuscular Mycorrhizal (VAM) spores. Isolation of VAM spores were made from both the Rudua and Jambu series in Besut as well as the Rudua and Baging series in Kuantan. There were more spores isolated from the Kuantan than the Besut areas. The results also indicate a positive relationship between spore number to percent organic matter present in the topsoil. The abundant spores isolated from the Kuantan areas (in contrast to Besut) could have some significance in the P nutrition of cashew and probably explain the yield differences existing between these two areas.

### INTRODUCTION

Cashew (*Anacardium occidentale* L) which was at one time a neglected crop grown by small farmers on infertile, sandy soils is now being grown commercially by the Cashewnut Industry of Malaysia (CIMA).

At present, a total of more than 3,600 ha is under cashew: 2,400 ha at Besut and the other 1,200 ha at Kuantan (Zambri *et al.*, 1982). Since 1976, the overall yield from the Besut plantation has been lower than that from Kuantan (25.4 t.y<sup>-1</sup> compared to 83.9 t.y<sup>-1</sup> respectively). The depth of the spodic horizon and the organic matter content in the topsoil has been shown to affect the nutrition uptake of cashew (Borhan *et al.*, 1982; Yaacob and Kamal, 1983; Zambri *et al.*, 1982). In addition to these, some biological aspects, probably those associated with organic matter, may also

be factors influencing the yield and nutrition; and thus the difference between yields in the two locations.

The aim of the present study is to examine the distribution of spores of VA mycorrhiza in sandy beach soils (Rudua, Jambu and Baging Series) under cashew in the two areas in relation to the organic matter content, the spodic layer and the P content of the soil as well as the P content in the cashew leaves.

This preliminary study investigates the possibility of exploiting the beneficial effects of VAM fungi in cashew production.

#### *Background of soils*

The areas under which the cashew is grown by CIMA form part of the beach ridge complex running all along the East Coast of Peninsular

---

Key to authors' names: Azizah Chulan, O. Yaacob, A.J.M. Kamal and S. Paramanathan.

Malaysia. This ridge and depression complex has often been referred to as the "Bris Soils" and in the reconnaissance soil surveys it has been mapped as the Rudua-Rusila Association (Paramanathan, *pers. comm.*). All the well drained soils of the ridges were mapped as the Rudua Series; and all the soils in the depressions, which were often under water throughout the year, as the Rusila Series.

Detailed surveys of these beach ridges, however, have indicated that these ridges consist of a number of soil types (such as the Rudua and Jambu series) with varying properties. In the cashew plantations of Besut and Kuantan detailed surveys have indicated that only the major soils exist on the ridges, i.e. the Baging, Rudua and the Jambu series (Paramanathan, *pers. comm.*). A hypothetical cross-section of the ridge complex showing the relationship between these soils is shown in Figure 1. A summary of the soil properties is shown in Table 1.

**MATERIALS AND METHODS**

*Sampling area*

The soil samples were obtained from the Besut plantation by the technique as reported by Zambri *et al.*, (1982). Five subsamples of 200g of topsoil (0-15 cm) were taken from the

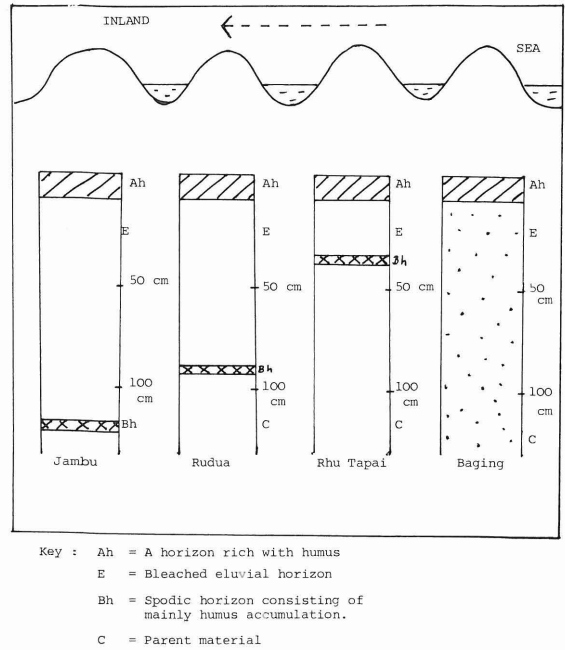


Fig. 1. Hypothetical Cross-Section Across The Beach Ridge Complex (Paramanathan, *pers. comm.*).

TABLE 1  
A summary of the soil properties

Soil Series	Surface Horizons	Subsurface Horizons	Other Properties
Baging	Grayish brown sand	Deep uniform light yellowish brown sand	Somewhat excessively drained soils found on ridges near the coast
Rudua	Dark grayish brown sand	Upper part white or light gray structureless sand extending to over 50 cm depths. Spodic horizon consisting of very dark brown sand. Cementation varies from weak to strong.	Well drained soils which may be flooded after heavy rains if spodic layer is cemented. These soils occur on second or third ridge from the coast
Jambu	Light brownish gray sand	Deep white to light gray structureless sand extending to over 100 cm depth. Very dark brown cemented spodic horizon only occurs below 100 cm depth	Excessively drained soils found on last and often furthest ridge from the coast

rhizosphere of each individual cashew tree. These subsamples were bulked to constitute one sample. A total of five soil samples were taken from each sub-section of the Besut plantation, from both the Rudua and Jambu series respectively.

The same sampling procedures were repeated for the Jambu and Baging series found in Kuantan. The soil samples collected were placed in labelled plastic bags, sealed, and brought back to the laboratory for isolation of VAM spores. The soil samples were collected between the months of December 1982 and January 1983.

#### *Sieving and counting of spores*

Between five to ten replicates, each of 100 g soil samples, were used for each soil series. The method of wet-sieving and decanting as described by Gerdemann and Nicolson (1963) was adopted for the isolation of spores from the soil particles.

The material retained on the sieves was further processed using the centrifugation technique of Tommerup and Kidby (1979). The supernatant containing the spores was then poured onto Whatman No. 1 Filter papers. The filter papers were observed under a dissecting microscope and the viable spores transferred to lactophenol in 25 mm diameter plastic petri dishes. The total number of viable spores from each soil sample was recorded.

The soil samples were also analysed for the organic matter and P content. The cashew leaves collected (third and fourth leaves) from the lower shaded part of the canopy were dried at 60°C for 72 hours and finely ground in a plant mill (Model Kinematica GMBH/Switzerland). The ground tissues were digested using concentrated sulphuric acid and 50% hydrogen peroxide (Thomas *et al.*, 1967) and analysed for their P content. The student's t-test was used for test of significance.

## RESULTS

The number of mycorrhizal spores isolated from the Kuantan areas exceeded the number of spores isolated from the Besut areas by two and a half times (Table 2). Root remnants present in the soil were also found to contain VA mycorrhizal infections. Very fine intricate hyphae, sometimes with two or three very young white spores still attached, were frequently seen on these roots under the 40x stereomicroscope.

Ninety-nine percent of the VAM spores isolated had diameters below 106  $\mu\text{m}$ . Most of these spores were dark-brown dead spores, not readily

identifiable (Mosse and Bowen, 1968). Only 1% of the total spores isolated had diameters greater than 250  $\mu\text{m}$ . These spores, of pale white to yellow in colour, with pronounced bulbous bases belong to *Gigaspora* (Hall, *pers. comm.*).

In Besut, soil of the Rudua series resulted in slightly higher organic matter content ( $0.99 \pm 0.60\%$ ) compared to  $0.92 \pm 0.50\%$  as present in the Jambu series. This corresponds to slightly higher spore count ( $29.20 \pm 19.31$  spores/g/soil) in the Rudua compared to  $26.17 \pm 14.74$  spores/g/soil in the Jambu series.

In contrast to the above, the soils in the Kuantan area had a much higher organic matter content and a higher spore count. The Rudua in Kuantan, with organic matter content of 2.19% gave  $82.61 \pm 72.98$  spores/g/soil while the Baging with 1.53% organic matter supported  $82.14 \pm 50.90$  spores/g/soil.

The amount of available P in the soil, and hence P concentration in the leaves of cashews growing in the Kuantan areas were higher than those found in the Besut areas (Table 2).

## DISCUSSION

The number of spores recovered (1 to 300 spores/g soil (data not shown)) is much higher than the number isolated from agricultural soils (0.1 to 5 spores/g soil – Mosse, 1979). However, there was much variability in the number of spores isolated from the soils within one section. Saif *et al.*, (1975) found that the numbers of VAM spores vary with soil texture: loamy soil was rich in VA mycorrhizal spores (with slight variation in frequency) while sandy soil contained the least number of endogonaceous spores, the range being from zero to about 150 spores/100g soil.

The present survey indicated the strong interdependence of the VAM spores and the organic matter content of the soil. The higher the percentage of organic matter present in the topsoil, the higher was the number of spores isolated (Table 2). These results are consistent with findings made by other workers. Sheikh *et al.*, (1975) found that the organic matter content of the soil is positively related to the endogonaceous spore population. The same workers also found more spores in the surface and subsurface horizons of the soil than in the sub-soil which could be correlated to the rate of oxygen diffusion.

TABLE 2  
 Number of VAM spores recovered from the various sand beach soils in relation to the organic matter and P content of the soil and P concentration in the cashew leaves

Soil series	Soil organic matter (%)	Spores recovered (per g soil)	P in the soil (ppm)	P in leaves (%)
BESUT				
Jambu	0.92±0.50*a	26.17±14.74a	10.33±9.75a	0.26±0.05a
Rudua	0.99±0.60a,d	29.20±19.31a,d	5.50±1.00a,d	0.26±0.05a,d
KUANTAN				
Rudua	2.19±1.18b,d	82.61±72.98b,d	13.28±6.22b,e	0.48±0.03b,e
Baging	1.53±0.62b	82.14±50.90b	13.49±5.81b	0.40±0.10c

\*Mean ± Standard deviation

N.B. Values that differ significantly are followed by different letter of the alphabet.

(P < 0.01)

The varying depths of the spodic horizon and levels of soil organic matter are also closely correlated with one another. Zambri *et al.*, (1982) in their work on the sandy beach soils found a negative relationship ( $r = -0.363$ ,  $P < 0.05$ ) between the varying depth of spodic horizon and levels of soil organic matter. This means that the level of the soil organic matter would decrease with increase in depth of the spodic layer (*Fig. 1*). The results obtained from this survey further showed that the distribution of mycorrhizal spores is inversely proportional to the depth of the spodic horizon and directly proportional to the amount of organic matter present in the soil (*Fig. 1* and Table 2). As seen in this study, in the Kuantan areas the higher the level of organic matter present in the soil, the more the number of mycorrhizal spores isolated. Spore production by these mycorrhizal fungi in the root zone has been shown to be closely related to the root infection and is known to vary with the plant species. (Manjunath and Bagyaraj, 1982).

Of the two soil series surveyed from the Kuantan areas, the number of spores isolated from the Baging series is slightly lower than that isolated from the Rudua series. This difference in spore number could perhaps be due to the fact that Baging being closer to the sea (*Fig. 1*) could have higher salinity because of the sea-sprays. The high salinity could have affected the activity of these endophytes. Koske (1975) in his investigation, found that VAM spores increased from about

26/10 g of sand in a marine foredune to 275/10 g sand in the second dune. However, the difference in spore numbers recovered from these respective soil series was not found to be significant.

Another important factor affecting the distribution of the endogonaceous spores is the soil moisture. Saif *et al.*, (1975) observed a change, in the VAM population with change in the water conditions of the soil. The maximum number of endogonaceous spores was found in soils with 40-60% water holding capacity. Similar findings were made by Khan (1974). Mosse and Bowen (1968) also found more microflora spores in the drier soil than in the permanently wet soil. The well-drained nature of the sandy beach soils surveyed in this study could probably account for the high spore count. However, the difference in rainfall intensity, and thus soil moisture content which prevailed in the Besut and Kuantan areas, might be an important factor in determining the number of spores isolated from these two areas. In Besut, the onset of the dry period occurs almost immediately after the monsoon season (in December) while in the Kuantan areas, moist conditions prevailed for another two to three months (i.e. until March the following year). The cumulative effect of the prolonged moist condition experienced in the Kuantan areas as well as the favourable aeration offered by the sandy soil could be a stimulatory factor for increased microbial and fungal growth.

In a field survey by Abu Bakar *et al.*, (1981), it was found that cashew seems to grow better on darker sandy Spodosols (indicating higher organic matter level) than those grown on lighter coloured sandy soils (with lower organic matter content). Zambri *et al.*, (1982) further showed that there is a positive correlation between the growth and nutrition of cashew trees and the level of organic matter present in the soil.

With reference to the P content in the leaves and in the soil in this study, it is interesting to note that the t-test showed that there is a highly significant difference ( $P < 0.01$ ) in the P in the leaves between the Rudua series in Besut and the Rudua series in the Kuantan area. This difference could perhaps be due to the significantly higher P content in the soil and also higher spore numbers (statistically not significant) in the Rudua series in the Kuantan area. Similarly, when the P content in the leaves was compared between the Rudua and Baging series in the Kuantan area, there was a highly significant difference ( $P < 0.01$ ). However, since there was no significant difference between the P content in the soils, the differences in P content in the leaves could perhaps be due to the higher spore numbers in the Rudua series even through the t-test did not show a significant difference in spore numbers in the two soil series perhaps because of the high variability of spore counts.

Studies in the last two decades have established that VA mycorrhizas improve plant growth by increasing the nutrient uptake by plants especially phosphorus (Mosse, 1973, 1977). Increased root and shoot weight and nutrient content, especially of phosphate, in plants inoculated with mycorrhiza compared to non-mycorrhizal plants has also been reported in a wide range of crop plants such as cotton, cowpea and finger millet (Bagyaraj and Manjunath, 1980); citrus (Menge *et al.*, 1978) and cocoa (Ragu and Azizah Chulan, 1983). The present study indicates that cashew might respond well to mycorrhizal inoculation. The response of cashew to specific mycorrhizal species is worthy of further investigation with a view to more efficient usage of phosphate fertilizers in cashew plantations.

#### ACKNOWLEDGEMENT

We would like to thank En. Kamaruddin Othman and En. Junaidi Jaafar for technical assistance and Miss Koh Bee Choo for typing this manuscript.

#### REFERENCES

- ABU BAKAR, O., ABU BAKAR, I. and GOPINATHAN, B. (1981): Semi-detailed soil survey of the Marang – Dungun coastal strip, Trengganu, *MARDI Report No. 3*.
- BAGYARAJ, D.T. & MANJUNATH, A. (1980): Response of crop plants to VA mycorrhizal inoculation in an unsterile Indian soil. *New Phytol.* **85**: 33-36.
- BORHAN, M.J., YAACOB, O. and A.J.M. KAMAL, (1982): Penentuan faktor-faktor tanah ke atas tumbesaran dan pemakanan gajus di kawasan tanah bris: Bahagian II. *Malays. appl. Biol.* **11**(2): 157-164.
- GERDEMANN, J.W. and NICOLSON, T.H. (1963): *Endogone* species extracted from soil by wet-sieving and decanting. *Trans. Br. mycol. Soc.* **46**: 235-244.
- KHAN, A.G. (1974): The occurrence of mycorrhizas in halophytes, hydrophytes and xerophytes, and of *Endogone* spores in adjacent soils. *J. of Gen. Microbiology*, **81**: 7-14.
- KOSKE, R.E. (1975): *Endogone* spores in Australian sand dunes. *Canadian J. Bot.* **53**: 668-672.
- MANJUNATH, A. and BAGYARAJ, D.J. (1982): Vesicular arbuscular mycorrhizas in three plantation crops and cultivars of field bean. Reprinted from *Current Science*, **51**: 707-708.
- MENGE, J.A., LABANAUSKAS, C.K. JOHNSON, E.L.V. and PLATT, R.G. (1978): Partial substitution of mycorrhizal fungi for phosphorus fertilization in the greenhouse culture of citrus. *Soil Sci. Soc. Am. J.* **42**: 926-930.
- MOSSE, B. (1973): Plant growth responses to vesicular-arbuscular mycorrhiza. IV. In soil given additional phosphate. *New Phytol.* **72**: 127-136.
- MOSSE, B. (1977): Plant growth responses to vesicular-arbuscular mycorrhiza. X. Responses of *Stylosanthes* and maize to inoculation in unsterile soils. *New Phytol.* **78**: 277-288.
- MOSSE, B. (1979): Vesicular – arbuscular mycorrhiza research for tropical agriculture. College of Tropical Agric., Univ. of Hawaii, *Misc. Publ.*
- MOSSE, B. and BOWEN, G.D. (1968): The distribution of *Endogone* spores in some Australian and New Zealand soils and in an experimental soil at Rothamsted. *Trans. Br. Mycol. Soc.* **51**: 485-492.
- RAGU, P. and AZIZAH CHULAN, (1983): Effect of vesicular-arbuscular mycorrhiza on *Theobroma cacao* L. seedlings. To be presented at the Conference on Impact of Microbiology in Tropical Agriculture. Universiti Pertanian Malaysia, 24-26 October 1983.
- SAIF, S.R., SHEIKH, N.A. and KHAN, A.G. (1975): Ecology of *Endogone*. I. Relationship of *Endogone* spore populations with physical soil factors. *Islamabad J. Sci.* **2**: 1-5.
- SHEIKH, N.A., SAIF, S.R. and KHAN, A.G. (1975): Ecology of *Endogone* II. Relationship of *Endogone* spore populations with chemical soil factors. *Islamabad J. Sci.* **2**: 6-9.

- THOMAS, R.L., SHEARD, R.N. and MEYER, J.R. (1967): Comparison of conventional and automated procedure for N, P and K analysis of plant. *Agronomy J.* 59: 240-243.
- TOMMERUP, I.C. and KIDBY, D.K. (1979): Preservation of spores of vesicular-arbuscular endophytes by L-drying. *Appl. Environ. Microbiol.* 37: 831-835.
- YAACOB, O., and A.J.M. KAMAL, (1982): Soil factors affecting growth and nutrition of cashew in Malaysia. Ninth International Colloquium on Plant Nutrition, England. In "Plant Nutrition 1982". C.A.B. 2, 728-733.
- YAACOB, O., and KAMAL, A.M.J. (1983): The nutrition of cashew on sandy soils of Malaysia. *Comm. Soil Sci. Plant Anal.* 14(8): (In press).
- ZAMBRI, M.P., YAACOB, O. KAMAL, A.J.M., and PARAMANATHAN, S. (1982): The determination of soil factors on growth of Cashew on Bris Soil: *Pertanika*, 5(2): 200-206.

(Received 1 August 1983)