THE CULTURE AND INOCULATION OF ARBUSCULAR MYCORRHIZAL FUNGI (AMF) FOR NURSERY AGRICULTURE IN THE TROPICS

BY

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Abstract

This study examines the potential exploitation of perhaps the most widespread plant-microbial interaction found in nature, the Arbuscular mycorrhizal fungal (AMF) symbiosis. This symbiosis may be manipulated and utilised to increase crop production in the Tropics, where fertiliser availability and/or uptake is often limited. Possible approaches for the exploitation of AMF are the inoculation of selected AMF and/or the establishment of growth environments more conducive to a positive AMF effect. If AMF inoculation is to become economically practical, however, a culture material must be identified which not only supports the proliferation of infective propagules, but also enables the storage and transfer of inoculum thereafter. It is equally necessary that the substrate be cheap and readily available to enable the large quantities of inoculum needed for field application to be produced. This thesis, therefore, represents the progress of two lines of research: a) the identification of an AMF inoculum culture system for the commercial production of AMF inoculum and b) an investigation of the factors contributing to AMF effectiveness.

This work identified coir fibre pith (CFP) as a suitable culture substrate for AMF, thereby replacing conventional materials such as sand and soil. The relative stability of CFP to decomposition, and its known unique physicochemical properties, allowed it to be compressed to seven times its original volume, and then expanded at the time of inoculation by the addition of water. As this compression did not adversely effect propagule viability, CFP can be considered a cost-effective storage and transport medium for AMF inoculum. However, the choice of CFP needs careful evaluation, as young material, containing

relatively high levels of available P (*ca.* 40 ppm P), produced lower colonisation levels compared to older CFP, which contained lower P levels (*ca.* 5 ppm P). Although spore number was unaffected by the lower colonisation level in the young CFP, any reduction in the length of colonised root would mean a reduction in the quantity of root propagules available for subsequent inoculation.

Our work did not show whether CFP was a superior carrier to conventional media as infected roots pieces of *Glomus clarum* and *Glomus constrictum* did not maintain infectivity after one month of storage at any storage temperature tested (room temperature, 4 °C and -20 °C). Further work is required on these two AMF species in order to validate these findings, as the loss of root propagule viability after storage may have been affected by the presence of contaminanting fungi, and may not be due to physiological characteristics of the AMF themselves.

This study produced promising results concerning spore activation. We found that constitutionally dormant spores may be induced to germinate by subjecting whole spore-inoculum to repeated freeze-thaw cycles. Further work is needed to determine the percentage of the total spore population that was 'activated' but initial indications suggest that this procedure can be used immediately prior to inoculation in order to increase the inoculum potential of the inoculum.

Regulation of the available P level of the growth medium was necessary to promte an AMF effect as AMF effectiveness was found to occur only within a certain, maximum level of

available P. This maximum level varies with the particular AMF/host combination tested. The water-holding capacity of coir fibre pith was used to target available P concentrations. Targeted levels of available P were maintained in a coir fibre pith growth medium even after 10 months of plant growth, provided P levels were periodically replaced. The use of coir fibre pith adjusted to defined available P levels increases the ease with which AMF/host/available P screening trials are carried-out, eliminating the prior use of adsorption isotherms as CFP had no P adsorption characteristics.

Available P mediated the effect of *G. constrictum* with the tea clone TRI 2023, from mutualism at lower available P levels, to antagonism at higher P levels (> 6 ppm P). Due to the accumulation of P in plant tissue, without corresponding increases in plant dry matter production, or pronounced changes in C allocation between the roots and shoots, it was suggested that tissue P levels may be inferior to available P levels as boundary indicators of AMF effect, although it may still be suitable for broad classification of % colonisation. *Glomus clarum*, however, showed no positive effect at any P level. This suggested that some AMF species are intrinsically incapable of functioning positively with certain hosts. As colonisation was maintained with no benefit to the host, *G. clarum* seems to be capable of aggressively acquiring host carbon. These factors will also affect the relationship between AMF and other microbes. This study found no positive effect of a mixed species AMF inoculum on the *Bradyrhizobium* strain TAL 45. As this AMF inoculum was not effective in P uptake, it may not be capable of acting synergistically with a *Rhizobium* species.

Although available P may be considered the single most important soil characteristic for the determination of AMF effect, and one which can mediate the AMF effect from positive to negative, ultimately, however, AMF effectiveness with a given host may depend on the AMF species itself, and whether it is intrinsically specific, or compatible, with its host.