

IDENTIFICATION, BIOASSAY AND EVALUATION OF MYCORRHIZA FOR UTILIZATION IN CASSAVA AND SWEETPOTATO PRODUCTION

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ABSTRACT

The mycorrhizal fungi observed in various places in Leyte were found in cassava, sweetpotato, legume, vegetable and plantation crops. *Gigaspora* and *Glomus* were the most commonly isolated local mycorrhizal genera. Higher infection was noted in cassava (Golden Yellow) than in sweetpotato (BNAS-51). Isolates obtained from cassava cultivar (Colombia) were infective on sweetpotato, with *Gigaspora* sp. causing the highest infection. The most suitable age of sweetpotato and cassava for inoculation with vesicular-arbuscular mycorrhiza (VAM) was found to be 2 and 3 wks after planting, respectively. In sweetpotato, root-mycorrhizal association was observed 6 wks after inoculation while in cassava, it was 4 wks after inoculation. The use of rhizosphere soil mixed with mycorrhizal spores as inoculum resulted in higher root infection than with the use of spores (in paper) and infected roots.

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INTRODUCTION

Many land areas in the Philippines are phosphorus-deficient but their phosphorus requirement may be reduced with the proper application of mycorrhiza. Both ecto- and endomycorrhizae generally increase the phosphorus uptake by plants, especially in phosphorus-deficient soils. In addition, mycorrhizal fungi also serve as biological control agents against soilborne pathogens in feeder roots of plants. Utilization of effective mycorrhizal strains on agricultural plants can, therefore, represent

tremendous savings in fertilizer and pesticide input of farmers. Since root crops need a substantial amount of phosphorus for root and tuber formation, mycorrhizal application can provide this requirement.

Research on mycorrhiza in the Philippines has been conducted on forest trees and agronomic crops (Halos *et al.*, 1982; Ilag, 1987). Except for an initial study on mycorrhiza in cassava (Almendras, 1982), no other report on mycorrhizal association with root crops in the Philippines is available.

This paper presents the identity of mycorrhiza associated with agricultural crops and the various techniques used in inoculating mycorrhiza.

MATERIALS AND METHODS

Collection, identification and bioassay of mycorrhizal fungi

Root samples and rhizosphere soils from areas planted to cassava and sweetpotato as well as to leguminous, solanaceous, pasture and plantation crops were collected from different places. The soil samples were passed through several sieves to collect mycorrhizal spores while the root samples were washed to remove the debris and later fixed for microscopy.

Microscopic examinations were conducted to identify mycorrhizal spores with the use of references like synoptic keys of Trappe (1982). Samples were also sent to CIAT, Cali, Colombia for identification purposes. About 20-50 spores were used in the identification of each species. Mycorrhizae from different sources were tested for infectivity on sweetpotato and cassava cultivars through bioassay experiments.

Evaluation of mycorrhizal inoculation techniques

The funnel technique (Menge and Timmer, 1982) was used for the production of inoculum in corn. The fungus collected from this method was further mass produced in pots and used for the evaluation of mycorrhizal inoculation techniques. Different inoculation techniques and methods (Menge and Timmer, 1982) were tried and/or developed on sweetpotato (BNAS-51) and cassava (Golden Yellow). The suitable age of plants for inoculation with VAM was also determined by measuring the elongation of sweetpotato and cassava roots in 36-cm diameter pots. A test was likewise done to identify the earliest time and amount of mycorrhizal rhizosphere soil needed for infection to occur in sweetpotato and cassava after inoculation.

Four inoculation techniques for mycorrhiza were tried using the following procedure: 1) *Rhizosphere soil under stakes/cuttings*. Five hundred grams of rhizosphere soil were placed at the center of sterilized soil in a pot where cassava stake or sweetpotato cutting was to be held. 2) *Spores in paper laid under stakes/cuttings*. At least 100 spores were spread on tissue paper which was then wrapped around the base of stakes/cuttings. 3) *Chopped mycorrhiza-infected roots*. Two grams of mycorrhiza-infected roots were placed at the center of the pot with sterilized soil where stake/cutting was planted. 4) *Rhizosphere and sterilized soil mixture*. Rhizosphere soil (150 g) was mixed with 550 g of sterilized soil in pots.

The following forms of inocula were tested on sweetpotato and cassava: rhizosphere soil, infected roots and spore inoculum. In all these different methods, root samples were gathered, stained and examined under the microscope for mycorrhizal infection following the rating scale below:

Low	=	Small colonization sites widely scattered along the roots
Moderate	=	Large colonization sites with more uniform distribution throughout the colonized roots, but rarely coalescing
High	=	Solid colonization with few easily identified isolated patches of colonization.

Ten randomly selected lateral root tip segments per plant taken from the primary root were examined under the microscope.

RESULTS AND DISCUSSION

Collection and identification of mycorrhizal fungi

Results of the survey showed that mycorrhizal fungi are present in many areas in Leyte. The following legume, vegetable and plantation crops were found to have mycorrhizal fungi: cacao, ipil-ipil (*Leucaena leucocephala* (Lam.) de Wit), kudzu (*Calopogonium muconoides* Desv.), eggplant, horse radish, kadios (*Cajanus cajan* (L.) Merr.), calamansi (*Citrus mitis* Blanco.), rambutan (*Nephelium lappaceum* L.), okra, mungo and sweetpotato/cassava accessions or varieties. One factor that may contribute to the prevalence of mycorrhiza in the surveyed areas in Leyte is the clayey nature of the soils which are mostly acidic and low in nutrient content (Ferraren, personal communication). With the use of the Synoptic Keys to the Genera and

Table 1. Vesicular-arbuscular mycorrhiza (VAM) collected locally, its associated plant and place of collection.

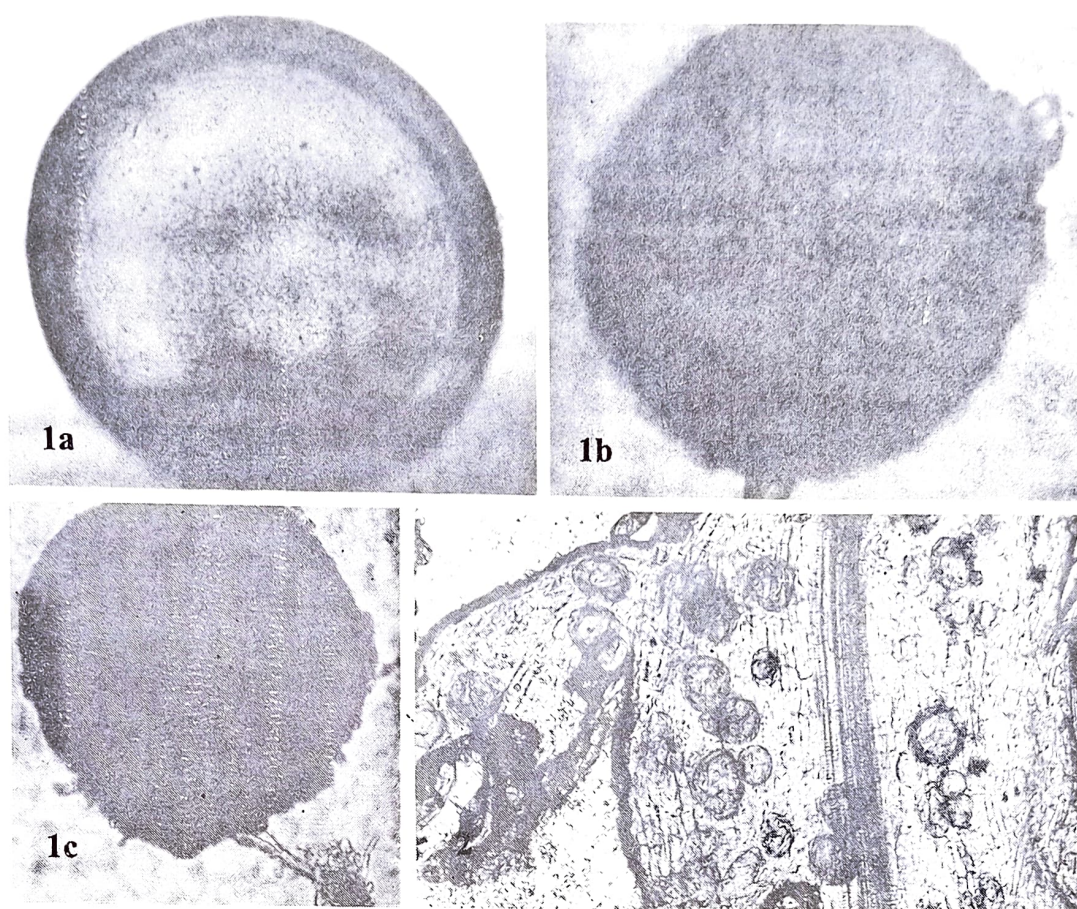
VAM Species	Associated Plant	Place Collected
<i>Gigaspora margarita</i> Becker & Hall	Kudzu (<i>Caloponium muconoides</i> Desv.)	Gaas, Baybay
<i>Gigaspora margarita</i> Becker & Hall	Sweetpotato (<i>Ipomoea batatas</i>) Cassava (<i>Manihot esculenta</i>)	Bunga, Baybay
<i>Glomus geosporum</i> (Nicol & Gerd) Walker	PRS 153 Sweetpotato (<i>Ipomoea batatas</i>)	PRCRTC Sweetpotato Germplasm, ViSCA
<i>Glomus constrictum</i> Trappe	Pineapple (<i>Anonas comosus</i>)	RCRC Pineapple Area, ViSCA
<i>Glomus constrictum</i> Trappe	Kakawati (<i>Gliricidia sepium</i>)	Forestry, ViSCA

Species of Zygomycetous Mycorrhizal Fungi (Trappe, 1982), a local coding of the VAM species was derived. It can be noted that two species were *Gigaspora* while three were of *Glomus* (Table 1). The spores of the identified local species are shown in Figures 1 and 2. Their presence in different kinds of plants shows that the vesicular-arbuscular (VAM) or fungal symbionts are not host-specific. The differences in sources or place collected also indicate that VAM is ubiquitous in nature.

Bioassay of mycorrhizal fungi

Out of 40 types of mycorrhizal fungi taken from the different field and plantation crops in the locality and inoculated to BNAS-51, only 10 were able to infect the roots. The small number of VAM species infecting the test plants indicate some degree of fungal specificity.

Microscopic examination also showed that out of 32 mycorrhizal fungi inoculated to cassava, 28 were able to produce infection. Cassava roots infected with mycorrhiza taken from sweetpotato Acc. 4 had the highest degree of infection. Infection was characterized by the presence of vesicles and arbuscules penetrating the host tissue. The roots of BNAS-51 inoculated with the five collections received from CIAT, Cali, Colombia showed infection with VAM (Table 2).



Figures 1-2. Vesicular-arbuscular mycorrhizae and their associated crops: *Gigaspora margarita* in kudzu (1a), *Glomus constrictum* in pineapple (1b), *Glomus geosporum* in sweetpotato (1c). A vesicular-arbuscular infection by *Gigaspora margarita* in BNAS-51 sweetpotato (2).

Table 2. Bioassay of five CIAT vesicular-arbuscular (VAM) isolates on BNAS-51 sweetpotato variety.

VAM Species	Infection on BNAS-51
<i>Acaulospora foveata</i> (C-314)	Few vesicles observed
<i>Gigaspora versiforme</i> (C-141)	Arbuscules/vesicles plenty
<i>Glomus manihotis</i> (C-172)	Profuse mycelia observed on root surface but no vesicles
<i>Gigaspora margarita</i> (C-21)	Arbuscules/vesicles plenty
<i>Glomus occultum</i> (C-122)	Mycelia observed on root surface but no vesicles

Table 3. Infection of cassava and sweetpotato using various techniques of mycorrhizal inoculation.

Technique	Infection ¹
Rhizosphere soil placed under stakes/cuttings	Moderate
Spores in paper laid under stakes/cuttings	Moderate
Chopped mycorrhizal roots poured into hole	Moderate
Rhizosphere soil mixed in sterilized soil	Moderate

¹ See text for details

Mycorrhizal inoculation techniques

The most suitable age of sweetpotato and cassava for VAM inoculation was on the second and third week after planting, respectively. Moreover, the funnel technique for production of inoculum on corn resulted in moderate infection and sporulation of *Gigaspora margarita*.

In sweetpotato, root-mycorrhizal association was observed six weeks after inoculation using 100, 300, 400 and 500 g of rhizosphere soil per 36 cm diameter pot of soil with mycorrhiza; in cassava, it was observed four weeks after inoculation using 300, 400 and 500 g of rhizosphere soil per 36 cm diameter pot of soil. The result indicates that cassava is better suited for VAM infection than sweetpotato. Although the level of VAM inoculum has no effect on VAM infection (Mosse, 1972), the effectiveness of a certain amount is affected by the cultivars used. All the inoculation techniques tested resulted in moderate infection in cassava and sweetpotato (Table 3).

Table 4. Effect of using three forms of mycorrhizal inocula on the infection of cassava and sweetpotato.

Inoculum	Infection ¹	
	Cassava	Sweetpotato
Rhizosphere soil (with spores)	High	High
Spores (in filter paper)	Moderate	Moderate
Infected roots	Moderate	Moderate

¹ See text for details.

In the test on the use of three forms of inocula, rhizosphere soil mixed with mycorrhizal spores gave higher root infection than with the use of spores (in paper) and infected roots (Table 4). The result corroborates the report of Menge and Timmer (1982) wherein rhizosphere soil with mycorrhizal spores was shown to be the most natural method for inoculating plants.

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