Research Paper :

Evaluation of AM spores and mycorrhizal root fragment as fungal inoculum for establishing colonization on the Ri – T DNA transformed hairy roots

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SUMMARY

Arbuscular mycorrhizal fungi are ubiquitous and form symbiosis with roots of a majority of higher plants for establishing *in vitro* cultures. Hairy roots can serve as potential host under root organ culturing. Lab experiments were conducted to evaluate the potential of AM spores as well as AM roots to establish colonization in hairy roots. *Glomus mosseae, Glomus intraradices* and *Glomus caledonium* in spores as well as colonized roots pieces were used to develop colonization in Ri-TDNA transformed hairy roots of cowpea and tomato using Modified White's Medium (MWM) and Minimal medium (MM). Inoculated plants were kept under dark at 25^{0}C for about 3-4 weeks. When surface sterilized spores were used as inoculum for the establishment of AM colonization in Ri T-DNA transformed hairy roots, *G. intraradices* produced in M medium the maximum colonization in cowpea and tomato hairy roots, respectively. When mycorrhizal root fragments were used as inoculum, *Glomus mosseae* produced more colonization in M medium (30.3 %) than others in hairy roots of both cowpea and tomato. While comparing the efficiency of two sources two to three fold increase in root colonization was observed in cowpea and tomato hairy roots with the inoculation of AM spores rather than AM root pieces.

Key words : AM fungi, Mycorrhizal root fragment, AM spores, *Glomus,* Hairy roots

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Arbuscular Mycorrhizal (AM) fungi are a unique group of ubiquitous soil microorganisms known to form symbiotic association with roots of economically important crop plants. The symbiosis between the two biotropic organisms is mainly characterized by bi-directional transfer of nutrients which gives access for the plant to low mobile elements like phosphorus (Smith and Gianinazzi – Pearson, 1988). Compatibility with AM fungi enable plants to explore and conquer a novel ecosystem and continues to provide a selective advantage because of the nutritional benefit it provides to plants (Karandashov and Bucher, 2005).

AM fungi produce structures such as, vesicles and arbuscles in cortical roots (Bowen, 1987). AM propagules as isolated spores, vesicles and sheared mycorrhizal roots are virtually able to initiate AM symbiosis and establish the pre-symbiotic phase with the transformed root. Chlamydospores of *Glomus* sp. (Mosse and Hepper, 1975; Mugnier and Mosse, 1987) and non-sporocarpic azygospore of *Gigaspora margarita* (Becard and Fortin, 1988; Becard and Piche, 1989; Diop *et al.*, 1992) are also preferred as starter inoculum.

Spores, used as propagules to initiate monoxenic culture, harbor many saprophytic microorganisms that can influence both spore germination and AM formation and thus, require sterilization (Fracchia *et al.,* 1998). For all AM propagules, proper selection and efficiency of sterilization process are keys for the success of axenic or monoxenic AM fungal cultures.

Mycorrhizal roots used to initiate monoxenic cultures come from plants grown in pot cultures, with field collected soil or AM propagules. Leek (*Allium porrum* L.) plants are widely used because of their susceptibility to colonization. Two sources of AM inoculum *viz.*, spores and AM roots were tried for establishing the colonization in hairy roots of cowpea and tomato using three different species of AM fungi.

MATERIALS AND METHODS

AM spores as fungal inoculum for establishing colonization on the Ri – T DNA transformed hairy roots :

Hairy root sources :

Tomato and cowpea explants (tomato hairy roots produced with *A. rhizogenes* strain 2364 and cowpea hairy roots with *A. rhizogenes* strain 532 were used in this experiment). The hairy roots produced in