MYCORRHIZAL INOCULATION DURING PLANT PROPAGATION

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Mycorrhizae are the symbiotic associations of certain soil fungi and plant roots. Hyphae of mycorrhizal fungi are in close association with the cells in the outer region of the root and extend outward into the soil, effectively increasing the surface area of the root system. Mycorrhizae have been documented for nearly all plants in their natural habitats.

Since this is a symbiotic relationship, both the plant and the fungi benefit. The plant benefits primarily from increased nutrient and water uptake, while the fungi benefits from the use of the plant as a source of carbohydrates and plant exudates.

Research with ornamental plants has focused on both ectomycorrhizae and endomycorrhizae. Ectomycorrhizae form a visible hyphal sheath around root tips and are responsible for the characteristic branching of ectomycorrhizal roots. Within the root, the hyphae surround, but do not penetrate individual cells. Unlike ectomycorrhizae, endomycorrhizae are not visible without magnification. In endomycorrhizal relationships, the hyphae penetrating the roots actually enter individual cells. Endomycorrhizae are characterized by arbuscles and vesicles structures. Arbuscles are concentrations of fine hyphae which form and dissolve, and may be a means of exchange between the two symbionts. Vesicles are lipid droplets which when thin-walled serve a function of storage, and when thick-walled can act as resting spores.

There is increasing evidence that mycorrhizal inoculation may effectively increase plant growth under cultural regimes typical of commercial production (2, 4, 7, 8, 9). This paper will focus on the importance of mycorrhizal inoculation during plant propagation.

CUTTING PROPAGATION

When endomycorrhizal inoculum is included in a propagation medium, substantial gains in the establishment of roots can result (7). Cuttings of *Viburnum dentatum* were rooted under mist in a medium of perlite:vermiculite (1:1, v/v), or in the same medium amended with inoculum at a ratio of 7 medium: 1 inoculum (5 spore/cm^3).

Inoculum consisted of isolated spores of *Glomus fasciculatum*. Cuttings were 20 cm in length, had 6 leaves and were treated with 0.1% IBA. Number and weights of roots were measured after 4, 5, 6, 7, and 8 weeks of rooting.

Inoculum significantly increased the number of root initials penetrating the stems of cuttings after 5 weeks of rooting, with smaller increases on subsequent weeks (Table 1). This effect on root initiation occurred only after roots had begun to form and could be infected. This increase in number of roots is probably mediated through an effect of the fungus on plant metabolism, rather than a fungal exudate into the rooting medium.

Table 1. Effect of the mycorrhizal fungus, *Glomus fasciculatum*, on number of root initials formed by cuttings of *Viburnum dentatum*.

	Number of root initials	
Week	Inoculated	Control
4	5.25 c	2.25 c
5	33.75 ab	8.25 c
6	33.50 ab	25.00 b
7	49.75 a	34.25 ab
8	42.75 ab	27.50 ab

Numbers followed by the same letter are not significantly different at the 5% level.

Glomus fasciculatum also increased the fresh weight of roots of these cuttings after 7 weeks of rooting (Table 2). Soon after roots could be infected by the mycorrhizae, growth enhancements began.

There is no evidence from this work that fungal exudates are present in large enough concentrations to influence rooting prior to fungal infection. This supports the theory that roots first must be present before infection can occur and there can be any beneficial effect of the inoculum (Fig. 1).

Table 2. Effect of the mycorrhizal fungus, *Glomus fasciculatum*, on fresh weight of roots formed by cuttings of *Viburnum dentatum*.

	Root fresh weight (g)	
Week	Inoculated	Control
4	0.0000 d	0.0000 d
5	0.0083 d	0.0033 d
6	0.0000 d	0.0000 d
7	0.5091 b	0.2793 c
8	0.7736 a	0.2605 c

Numbers followed by the same letter are not significantly different at the 5% level.

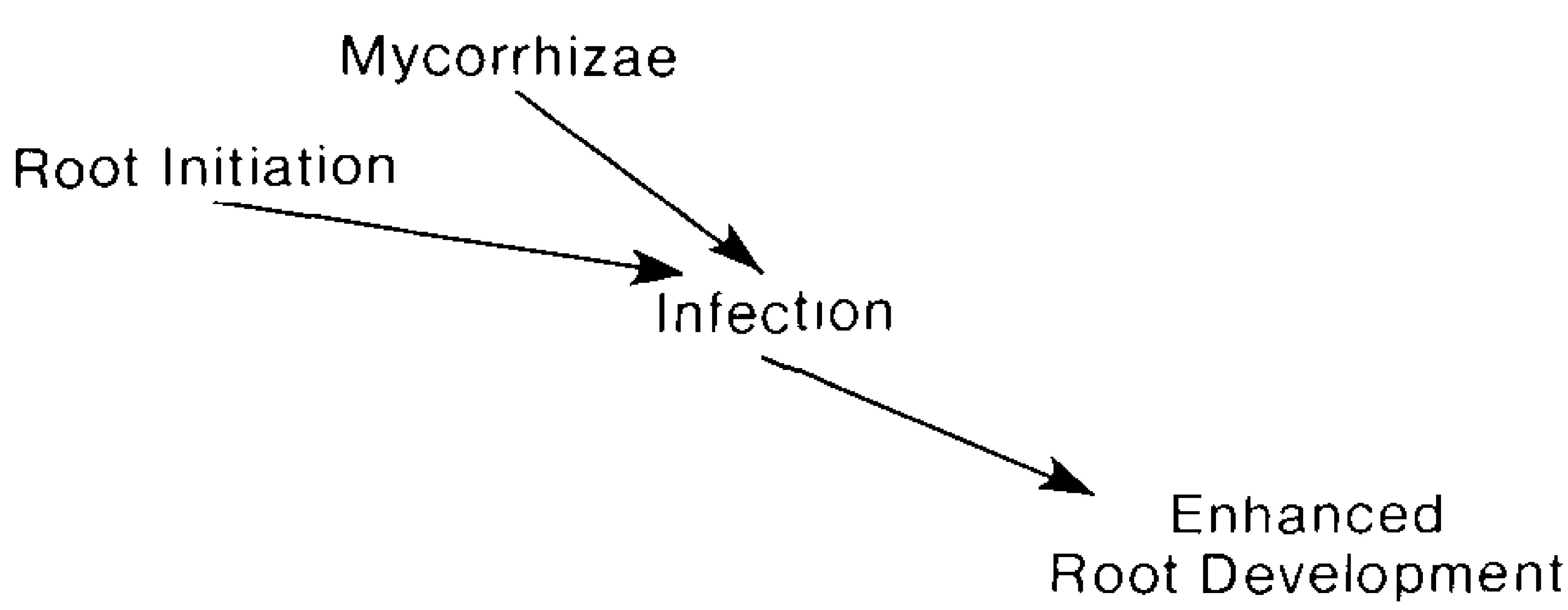

Figure 1. Sequence of events which would require infection occur prior to a beneficial effect of mycorrhizae on root growth.

Work by Linderman and Call (3) with ectomycorrhizae indicated an effect of inoculum on rooting before, or in the absence of fungal infection. This enhancement of rooting may be due to the exudation of auxin by the fungi (Fig. 2). This theory is supported by the fact that the appearance of an ectomycorrhizal root system can be induced by auxin treatment (6). Although these theories about the effect of mycorrhizae on rooting have not been tested, they are not mutually exclusive because ectomycorrhizae may have higher auxin exudates than endomycorrhizae.

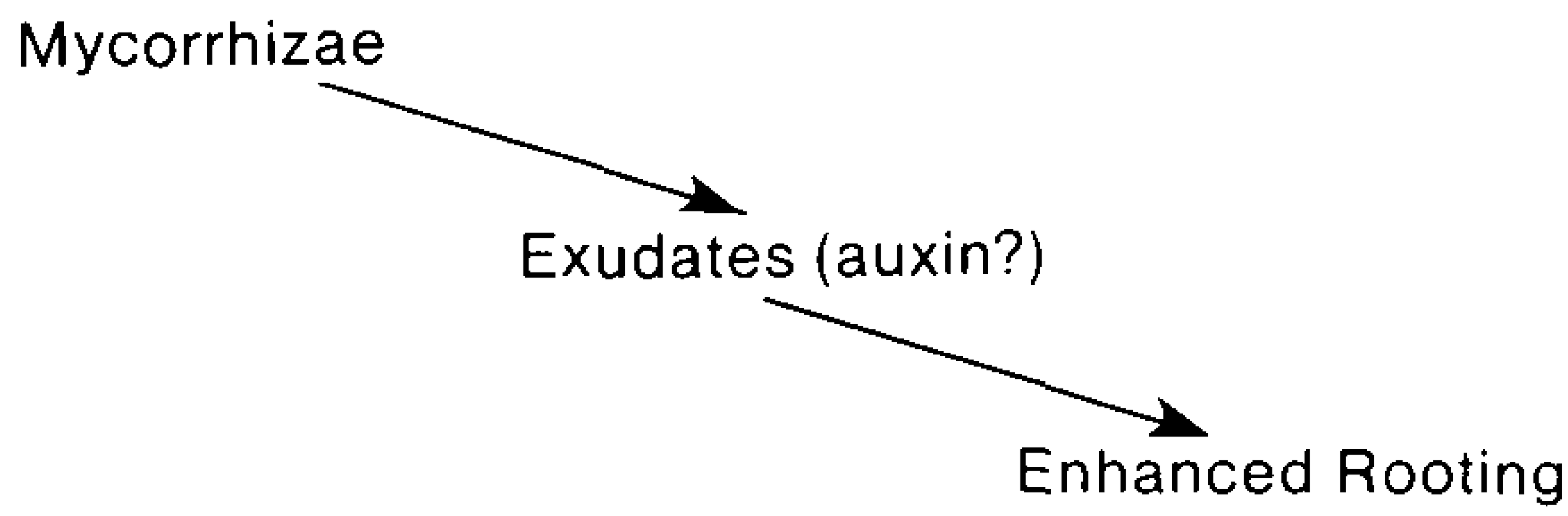

Figure 2. Sequence of events in which mycorrhizae could have a beneficial effect on rooting prior to, or in the absence of, rooting.

SEEDLING PROPAGATION

Seedlings can also benefit from the incorporation of mycorrhizal inoculum into the propagating medium. The objective of this experiment was to examine the extended effects of mycorrhizal inoculation during propagation on growth of seedlings following transplanting. Seedlings of *Cornus sericea* were germinated in media inoculated with *Glomus fasciculatum* or *Glomus macrocarpum*. Seeds were sown 0.25 cm deep in medium of perlite:vermiculite (1:1, v/v) or the same medium inoculated with isolated spore of *Glomus fasciculatum* or *Glomus macrocarpum* at a rate of 4 spores/cm^3. After 16 weeks the seedlings were transplanted into 1-quart (116 cm^3) pots and grown for 16 additional weeks. Data included plant height and dry weight of shoots.

Seedlings inoculated with mycorrhizal fungi attained a greater height and dry weight of shoots than non-inoculated plants (Figs. 3 and 4). Plants inoculated with *Glomus fasciculatum* had the greater height and dry weight of shoots than those inoculated with *Glomus macrocarpum*. Mycorrhizal infection of seedlings during propagation promoted seedling growth and had beneficial effects which extended beyond the propagation phase of production.

MICROPROPAGATION

It is possible to introduce fungal spores directly into tissue culture (1); however, this can present some potentially very serious problems with contamination. It is currently more feasible to inoculate stage IV plants while root systems are expanding. These plants

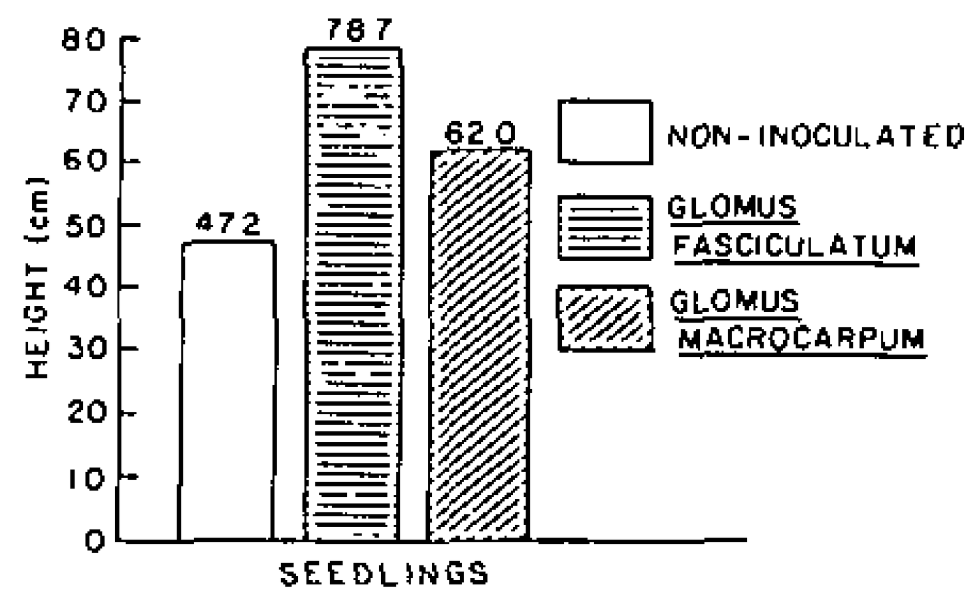

Figure 3. Effect of *Glomus fasciculatum* and *Glomus macrocarpum* on height of *Cornus sericea* seedlings.

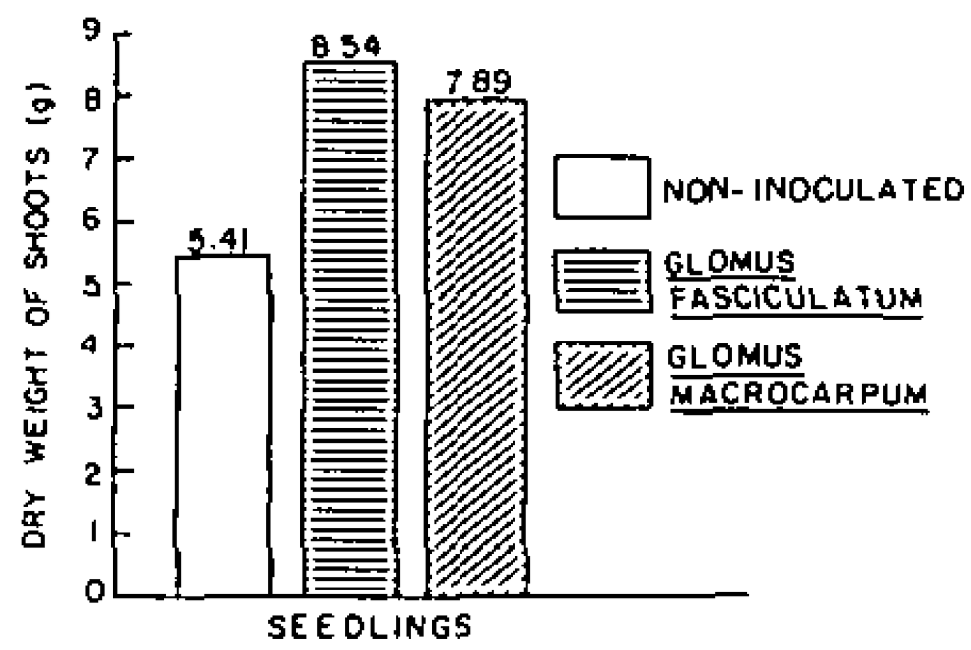

Figure 4. Effect of *Glomus fasciculatum* and *Glomus macrocarpum* on shoot dry weight of *Cornus sericea* seedlings.

could be inoculated in much the same manner as cuttings and seedlings.

CRITICAL STAGE

An important point to consider is that propagation is the critical stage of production for mycorrhizal inoculation. Propagation is the most logical stage of production for mycorrhizal inoculation. Inoculation during propagation enables the earliest possible infection and resulting benefits. It also facilitates efficient inoculation because a large number of plants can be inoculated in a small area. Cuttings, seedlings and tissue culture plants also can all be inoculated in propagation and handled the same way, using the propagation media as a carrier for inoculum. For these reasons, I am confident that if mycorrhizal inoculation is to become routine for production of any plants, propagators will be at the heart of this activity.

INOCULATION PROCEDURE

Figure 5 illustrates a possible scheme for inoculation with endomycorrhizae. Endomycorrhizae are considered obligate symbionts, and therefore must be grown with plants if they are to be grown for any period of time. In this scheme, inoculum sources are built up by "pot cultures" which enable the number of spores and hyphae to increase (5). The soil containing the roots, hyphae and spores can be used as inoculum, or spores can be isolated and used as inoculum. The use of isolated spores reduces the risk of a contaminant being present in the inoculum.

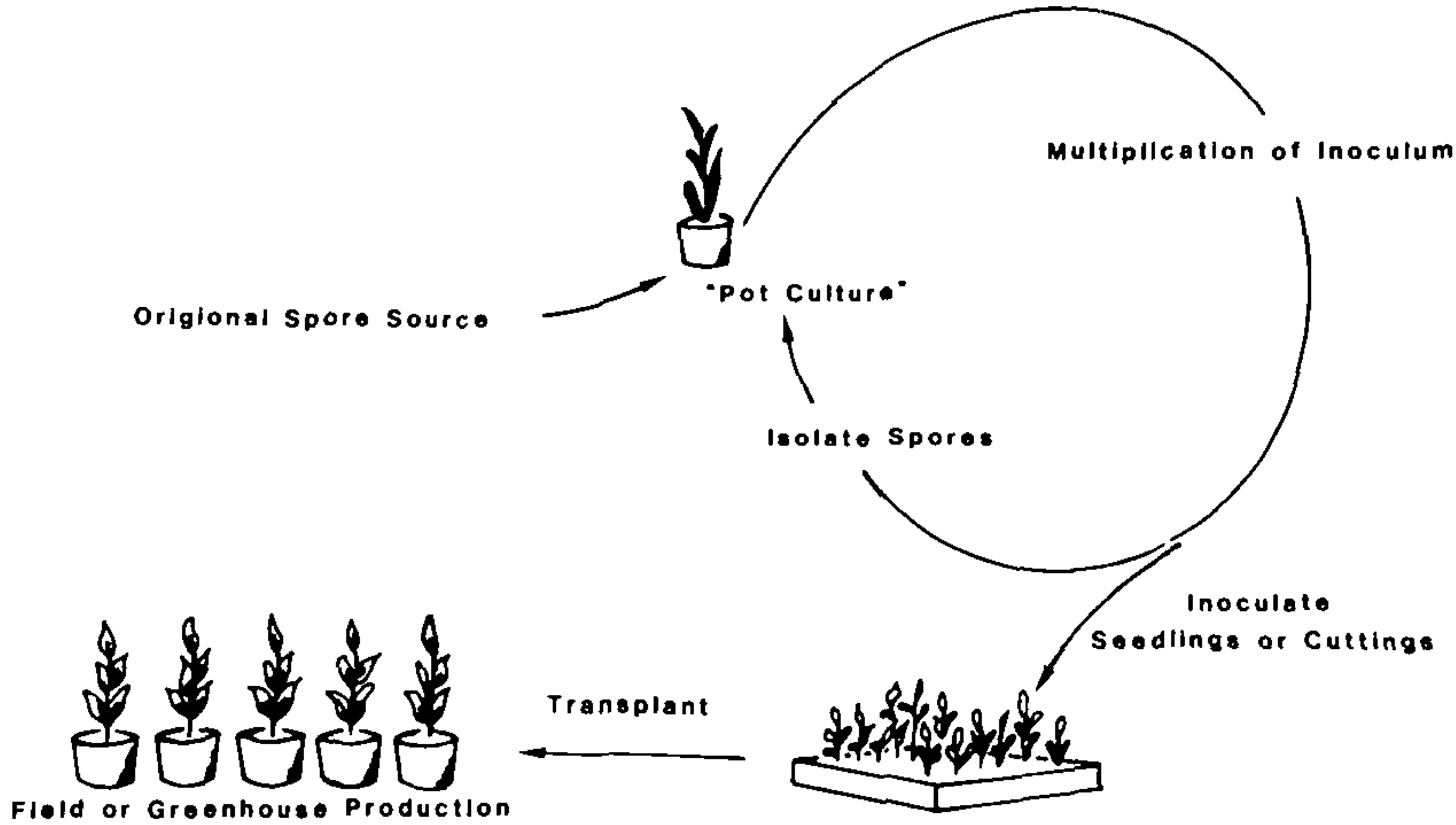

Figure 5. Diagram depicting a scheme for endomycorrhizal inoculum production and inoculation during production.

Inoculation is most efficient when the plant density is high, as in propagation. A determined volume of inoculum should be mixed into the propagation medium to facilitate infection as the roots develop. As propagation is achieved, the plants can become mycorrhizal and will benefit from inoculation during propagation, during field production, and when growing in the landscape.

A small amount of inoculum must be retained to perpetuate the "pot cultures" which serve as the inoculum source for future crops. Prior to initiating new pot cultures the spores must be isolated to reduce the risk of perpetuating any contaminants which may have become established.

CONSIDERATIONS FOR USE

Nurseries that experiment with mycorrhizal inoculation in a production system should be sure to:

1) Use the fungal species which will maximize the growth of the plant you are growing.
2) Use selective pesticides to prevent an unwanted effect on the mycorrhizal fungi.
3) Determine the cost of maintaining an inoculum source and the cost of the added step of inoculating.
4) Determine the value of the growth increases obtained

through inoculation. This value must be greater than the cost of inoculation for the inoculation to be practical.

Will mycorrhizal inoculation become a routine practice in nursery production? The answer to this question will be best answered by nurserymen. Researchers and nurserymen will need to cooperate to conduct research trials on actual nursery sites using commercially important plant species to identify and solve any problems which might occur due to the incorporation of mycorrhizal inoculation into production. Only actual attempts to use this new technology will truly answer the question of the potential for mycorrhizal inoculation in nursery production.

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