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# Center for Smart Agriculture

## Mass Production of AM inoculants



**Module 1:**  
**Session 3:**



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# Preparation of trap culture inoculum of arbuscular mycorrhizal (AM) fungal spores

- Trap of healthy arbuscular mycorrhizal fungi (AMF) spores
- Establish inoculum from monospecific cultures
- Spore trap cultures can also aid in AMF identification
- The spores may appear differently, due to weathering and intrinsic soil environment effect, either physical, chemical or biological. Trap cultures are important in the following situations



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## Procedures

- Collect Rhizosphere soil with shoots of trap plant cut at the crown
- Chop finely the roots and mix with the soils using a sharp chopper.
- Mix the chopped roots and soil at 1:1 ratio (v/v) with autoclaved coarse sand in a mechanical mixer, or massaged well in a durable plastic bag
- Transfer the soil mix to a 15 cm plastic pot.



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## Procedures

- Plant seeds of suitable trap plant such as tropical signal grass, *Brachiaria decumbens*, into the pot.
- Maintain the pot cultures in a greenhouse for at least 3 months, and check sporulation from time to time
- Sanitary tests may also be carried out to ensure no contamination from parasitic fungi occurs.



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## Procedures

- Keep fertilizer application to a minimum, to encourage AMF proliferation.
- Trap culture pots are later left to dry under shade for up to 2 weeks.
- Harvest the spores using the sieving and decanting techniques or the density-gradient centrifugation technique.
- The monospecific spores are ready for inoculation onto seedlings of the desired crops



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## Inoculation of AMF

- The seedlings are gently uprooted singly on in a small bunch, and have a gentle stream of water sprayed onto the roots so that they stick together.
- Spores collected from 3.3.1 are suspended in water and about 200  $\mu\text{l}$  of the spore suspension are pipetted onto the moist roots.
- The inoculated seedlings are immediately transplanted into containers of suitable size, containing sterilized soil.



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## Inoculation of AMF

- The soil is topped with a sterile growth medium, watered gently under shade, before transferring into the greenhouse.
- To encourage colonization of AMF onto seedling, fertilizers are not given during the early growth stage of the seedlings.



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## **Problems and potential for AMF inoculum production and utilization**

- Situations where effective indigenous AMF population is low.
- Inoculation is best for transplanted crops, where soil disturbances has reduces AMF inoculum potential.



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**Thank you...**