We’re pleased to learn of your interest in the benefits of utilizing MycoApply® mycorrhizal inoculants to improve your professional growing operation and establish the essential microbes necessary for plant uniformity, health, and sustainability. This Trials Protocol provides a general overview of mycorrhizal fungi, outlines the basics of trialing these products, and provides compatibility and cultural protocols, in order to optimize your ability to best utilize these products, observe and document the benefits, and ultimately experience the return on investment (ROI) that these symbiotic organisms provides professional growers.

**General Information**

- Mycorrhizal fungi have evolved with plants for over 450 million years to develop symbiotic relationships benefitting both the fungus and the plant. Nearly 95% of all major crop species on Earth have mycorrhizal relations that provide carbon sugars to the fungi, and in return provide nutrients, like nitrogen and phosphorus, to the plant, as well as other benefits.

- Endomycorrhizae are the most common, and have a mutualistic association with approximately 85% of plant species. They colonize plant roots intracellularly (inside the root cell) and are responsible for enhanced uptake of all major and minor elements, thereby improving plant growth and productivity.

- Ectomycorrhizae form with about 10% of the plant species on earth. They colonize plant roots extracellularly (outside the root cell). Conifers, and many North American hardwood trees, have ectomycorrhizal relationships.

- Two common horticultural families, Ericaceae and Orchidaceae, form mycorrhizal relationships with other types of mycorrhizae, which are not commercially available. Additionally, approximately 5% of plant species are non-mycorrhizal.

- Prior to commencing your trial, review the “Types of Mycorrhizal Plants” list and determine whether your crops are endomycorrhizal (ENDO) or ectomycorrhizal (ECTO) to identify which MycoApply product would be best for your crops. If you are growing both endo and ectomycorrhizal plants, then request an ENDO/ECTO blend which will cover both. (Unlike endomycorrhizal fungi, which are generalists, ectomycorrhizae are often specific to certain tree species. If you are running your trial on ectomycorrhizal plants, please contact a Mycorrhizal Applications representative to determine which product is best for the plants you are growing.)

- The next task is to decide what form of product that you plan to use, a dry granular/powder formulation or a suspendable powder. Use the dry granular/powder to incorporate into potting soils or when mixing with landscape soils. The suspendable products can be added to water to form a mycorrhizal suspension, and then used as a dip, drench, etc. The suspension will require agitation, and the particles will pass a 50-mesh screen (though we do not recommend using a horticulture injector dosing system with the MycoApply suspendable powder products).
Mycorrhizal Trialing Basics

• We use the term “propagules” instead of spores to determine how much product to apply per pot or per plant. However, different products have different counts of propagules and it is the quantity of propagules that is important to the success of inoculation for the plant. For drenching, the desired propagule count per container varies depending on the size of the container, the maturity of the plant, and the crop time. Refer to the “Drench Recommendation Chart” for more information. If you plan to incorporate the mycorrhizae into your soil, refer to the “Soil Incorporation Chart” for more detailed information. Mycorrhizal propagules may survive for many years in the soil before germinating and colonizing the host plant. Thus, it is important to place the propagules in the root zone, near actively growing roots. These root tips produce exudates that signal the fungi for colonization.

• If the crop is going to be sub-irrigated or drip-irrigated at low volumes, then it is better to either incorporate the propagules into the container substrate or apply a heavy drench prior to placing the potted plants in these irrigation systems for the best distribution of the propagules in the growing substrate. The recommended drench rate, for optimal distribution of the propagules, represents about 8-10% of the container volume for peat or bark-based substrates, assuming that the mix is initially moist. Dry soils will require a greater quantity of water, but not more propagules. Saturated soils should be avoided because they will experience very little downward movement of propagules into the root zone.

• Mycorrhizae begin to colonize newly emerging plant roots within a couple of weeks after inoculation. The benefits of inoculation start to become more prominent approximately 8 weeks after inoculation. For mycorrhizal evaluation trials, it is best to test trial crops with a total crop time longer than 8 weeks. This delayed response presents a challenge for a grower that wants to see benefits before the product ships or is sold. To ensure the grower gets the maximum benefits, it is important to start the timeline early in the production process. We suggest applying mycorrhizae during propagation phase or after you receive plugs or liners, so you can start the process sooner and see positive results earlier. A grower of young plants (plugs & liners) will need about twice as many propagules per tray due to the many cells and their small volume to ensure adequate colonization. Applying the mycorrhizae earlier also reduces costs, since you are applying the mycorrhizae to a smaller soil volume. Once treated during propagation, the mycorrhizae stays with the plant throughout its entire life, and moves with the plant if it gets bumped up into a larger container. The benefits of mycorrhizae will become more apparent when the plant is moved from the plug/propagation tray into the finished container.

• The simplest trial is to grow plants with and without the mycorrhizae treatment, and care should be taken to avoid too many variables. However, some of the recognized benefits of mycorrhizae are in production systems using reduced inputs of fertilizer and water. Under optimal growing conditions found in most commercial operations today, a benefit may not be obvious. Reduced input costs and reduced shrinkage are major considerations in determining a “return on investment” (ROI) for any evaluation. To avoid biological and environmental variables, use large numbers of plants (100 or more if possible) for each treatment. Initially, it may be too complicated to reduce inputs but it should be a goal for future trials.

• Using fewer propagules than recommended will slow colonization and delay desired responses. Applying more propagules than recommended will have no adverse effect on plant performance but it will increase costs and may adversely alter your ROI.
### Cultural Protocols

#### Fertilizers

- Most greenhouse and nursery crops are over-fertilized and this results in soft, vegetative growth with high nitrogen levels, reduced carbohydrates, reduced root development and greater disease susceptibility of roots, stems, leaves and flowers. High levels of water-soluble nitrogen and phosphorus suppresses most mycorrhizal activity because it reduces the mutual needs of the host and the fungus. Plants that experience stress during production often develop a stronger relationship with their associated mycorrhizae.

- Controlled release and organic forms of fertilizers release their nutrients very slowly and do not increase the water-soluble nutrients in the substrate excessively. If you are accustomed to using a liquid fertilizer, we recommend using a fertilizer that is low in phosphorus and has a high percentage of nitrate-nitrogen. For example, instead of using a 20-20-20 fertilizer, consider using a 20-10-20 fertilizer, or even better a 15-5-15 or 15-2-15 fertilizer. If you can evaluate other variables, consider reducing the 200-300 ppm N to 100-150 ppm N for a comparison. If you must apply a high phosphorus analysis fertilizer, then apply it three or more weeks after inoculation to avoid inhibiting the mycorrhizal colonization.

#### Fungicides

- Mycorrhizal fungi are soil-borne fungi that may be susceptible to certain fungicide products. The "Horticulture and Turf Fungicide Effects on Mycorrhizal Fungi" table is a list of our present knowledge of the compatibility of mycorrhizal fungi and fungicides. Many fungicides, including all biological fungicides, are compatible with mycorrhizal inoculation and development. The inoculation stage is the most susceptible period. The longer the wait to apply any fungicide after mycorrhizal fungi inoculation, the better for the mycorrhizal development. We suggest waiting at least two weeks to avoid slowing the mycorrhizal inoculation.

- Most foliar applied fungicides (except systemics, such as Bayleton or Strike) have little effect on the mycorrhizal fungi.

- Seeds treated with fungicides typically do not impair mycorrhizal development since the germinating roots carry the mycorrhizae away from the treated seed.

- Terms and interpretations used in the "Horticulture and Turf Fungicide Effects on Mycorrhizal Fungi" table are as follows:
  - "OK" – These fungicides can be safely used and should maximize the impact of the mycorrhizal inoculation.
  - "Avoid Use" – If these fungicides are used as a part of the planned production:
    1. Apply the fungicide and wait a month.
    2. Apply the MycoApply mycorrhizal inoculant.
    3. Allow the mycorrhizal fungi time to colonize, approximately 6 weeks, before applying an “avoid use” fungicide.
  - “Insufficient Data” – This product has not been tested to determine its impact on mycorrhizae. We suggest you avoid use, until compatibility is confirmed.

#### Other Chemical Treatments

- Mycorrhizal fungi are generally not harmed by herbicides, nematicides or insecticides.

#### Mixed Tank

- Although mycorrhizal fungi can be mixed with fertilizers and most other amendments, for trial purposes, it is best not to complicate the application process by adding another variable to the trial. Do not combine with “avoid use” fungicides at any time.