PLANT SAMPLE COLLECTION GUIDE

Target Pathogens

Plant pathogens can be detected in different areas of the plant. Depending on the target pathogen, the site of sampling on the plant can change. Sampling from multiple locations on single plant will result in a more comprehensive assessment of infection. Follow the chart below to determine the number of samplings and locations required by to test for each pathogen

Target Pathogens	Samples
Hops Latent Viroid (HLVd) Triplex	2 leaves (top and middle) and 1 root sample (2+ cm)
Powdery Mildew	3 leaves (top, middle and bottom) per plant
Tobacco Mosaic Virus	3 leaves (top, middle and bottom) per plant
Fusarium	3 leaves (top, middle and bottom) per plant
Pythium spp.	3 leaf samples per plant

* *If testing for multiple targets (ex. Full Plant Pathogen package) then 3 leaves per plant is sufficient and 1 root sampling (5 cm)*

Sampling Method

There are a variety of sampling methods that can be used to select plants for testing. Growers should consider the size of their grow room, the environmental monitoring procedures in place that limit infection (i.e., sterilization of tools, use of fungicides etc.), recent introduction of new plants, visible signs of poor plant health and recent treatment for any of the pathogens. We suggest two types of sampling: **Selective/targeted** and **Haphazard sampling.**

Selective or Targeted Sampling involves selecting plants from the lot that are most likely to be infected. In this method, samples can come from plants that show signs of wilting, yellowing or splotchy leaves or have stunted growth. Alternatively, samples can be harvested from plants that have yet to enter the grow room. Thus, ensuring that these new plants are not introducing pathogens to their healthy neighbours.

Haphazard Sampling involves selecting samples randomly throughout the grow room without a specific randomizing process. This process is a good fit for grow rooms that have never been tested before or for verifying the efficacy of treatment meant to eliminate any of our target plant pathogens.

Testing 3-4 plants within the grow area is suggested depending on the size, in hopes of covering a larger area where conditions might differ.

* *The procedure outlined below can also be used for sampling for plant gender, however only one leaf per plant is required.*

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Plant Tissue Sampling

It is essential that plant samples are gathered in a way that reduces the chances of contamination. Follow the steps outlined below to avoid contamination.

Supplies

- Clean (preferably sterile) sample bags, small sample containers or vials
- Disposable gloves
- 10 % Bleach solution in a spray bottle and cup (Prepare 10% Bleach solution by combining 50 mL of bleach with 450 mL of water)
- Scissors or trimming tool
- Paper towel

General Protocol

- Before sampling label all sample bags and containers with the plant identification information (e.g., section number, plant id number etc.)
- Leaf samples from a single plant should be gathered and placed into 1 clean sample bag. Root samples even from the same plant should kept separately to avoid getting excess soil on the leaves
- Wash hands with antibacterial soap before sampling. Put on a pair of disposable gloves. Sterilize the gloves by spraying with 10% Bleach solution.
    - It is recommended to wear 2 pairs of gloves. This method allows for changing of the outer set between samples without running the risk of contact with bare hands.
- Soak the trimming tools in the bleach solution for 60 seconds. Spray any parts of the tool that cannot be submerged with the bleach solution spray.
- Completely sample leaves and roots from one plant before moving on to other plants.
- Sterilize hands and tools between each sample

Leaf Sampling

1. Sterilize gloves and trimmers with 10% bleach solution.
2. Use sterilized trimmers to cut leaves from the top, middle and bottom sections of plant.
3. Use the trimmers or scissors to place the leaves in the correctly labelled sample bag.
4. Close the sample bag and set aside. Begin root sampling for the same plant or discard gloves and repeat procedure for a new plant.

Root Sampling

1. Sterilize gloves and scissors with 10% bleach solution.
2. Lightly brush away soil to reveal surface root of plant.
3. Pull gently on the root to break it away from the root mass.
4. For soilless media, use sterilized scissors to cut small pieces of exposed root tissue.
5. Brush off any excess growing medium and place root tissue onto a clean paper towel and into the appropriately labelled container. Discard gloves and repeat procedure for additional samples.

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Sample Storage and Shipment

Samples should be shipped or delivered to the laboratory as soon after collection as possible. If it is necessary to store the sample before shipment, place within a refrigerator, preferably between 2-8°C.

Suggested shipping procedure: Place the original sample container within a large resealable bag then place the bag between a piece of folded cardboard to minimize damage to the leaves during shipment. Put the protected leaves into the shipping envelope or box. Mail via next day shipping to ensure samples are in good condition when they arrive.