Managing Fungal Trunk Diseases in Plant Nursery Stock

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Project Summary/Abstract

Briefly describe the long-term objectives for achieving the stated goals of the project.

The overarching goal of this project is to increase the quality of plant nursery stock. We propose to further profile pathogenic fungi in the rootstock and scion of nursery trees/vines and identify possible infection routes. This information will help establishing guidelines for improving the management of fungal trunk diseases in nurseries. In addition, it will facilitate the development of a cost-effective and sensitive PCR-based diagnostic method and its dissemination to stakeholders

Scope of Work

Describe the goals and specific objectives of the proposed project and summarize the expected outcomes. If applicable, describe the overall strategy, methodology, and analyses to be used. Include how the data will be collected, analyzed, and interpreted as well as any resource sharing plans as appropriate. Discuss potential problems, alternative strategies, and benchmarks for success anticipated to achieve the goals and objectives.

Certification programs give insurance that commercial plants sold by nurseries are free of pathogens. Viruses are commonly host-specific or have a narrow host range. For example, Grape Leafroll Disease (GLD) caused by viruses is specific to grapevine. A large collection of disease-tested wine, table, raisin and rootstock grape selections is part of the California Grapevine Registration & Certification (R&C) Program and has been successfully at controlling GLD. However, there is no certification program for fungal trunk pathogens, even if those have been clearly identified in nursery plant propagation pipelines. One reason is that the causal agents of trunk disease have a broad host range and the same species can infect many plant hosts. For example, *Diplodia seriata* infects several woody crops including *Vitis, Prunus, Malus* and *Juglans*. The goal of this proposal is **not** to establish the foundation for a certification program from fungi causing wood diseases because it is unrealistic. However, this project will benefit the nursery industry because it will improve the quality control of plant stock by developing standard operating procedures that eliminate or at least reduce the likelihood of fungal infections in the propagation pipeline.

Objective: Profile trunk disease pathogens in nursery stock.

Workplans and Methods

We hope to work with at least three nurseries that are members of the California Fruit Tree, Nut Tree, and Grapevine Improvement Advisory Board. The name of nurseries will not be disclosed in public domains.

We will initially test 100 commercial vines per nursery. Our previous results showed that there was high variability from batch to batch. So, each sample will represent 10 plants from one batch including both the scion and rootstock. This sampling is non-destructive so plants could still be sold commercially after sampling. We will focus our research efforts on popular wine grapes planted in California, including Chardonnay Clone 4 x 1103P rootstock (50 samples) and Cabernet Sauvignon FPS 8 x 1103P rootstock (50 samples). This way we will be able to compare data and limit variability due to scion and rootstock genotypes. Plant material will be tested using both culture-independent approach developed by the Cantu lab (Morales-Cruz et al. 2018) and standard culture-dependent diagnostic protocols routinely done in the Rolshausen lab (Doll et al., 2015; Rolshausen et al., 2013). For the culture-independent approach done in the Cantu lab, total DNA will be extracted from plant samples and fungal trunk pathogens will be profiled using the specific, sensitive and accurate metabarcoding ITS primers designed by Morales-Cruz et al. (2018). For the standard diagnostic culture dependent approach done in the Rolshausen lab, plant tissue samples will be plated on fungal culture medium and incubated for 10 days. Fungi identified as trunk pathogens based on morphology in culture will be further transferred to obtain a pure culture and DNA will be extracted. Fungal species will be identified based on Sanger DNA sequencing using universal ITS primers. If fungal infections are found above a certain threshold (see objective 2) in the 100 samples tested, we will increase the range of our sampling for each nursery in year 2 and 3 of the project (sampling design to be determined), and collect materials (plant, soil, water) starting with mother plants and moving downstream at every step of the plant propagation pipeline in order to pinpoint the infection routes. The information collected will be used to build a database of the fungal trunk pathogens occurring in California nursery**.**