

## I. AETIOLOGY, EPIDEMIOLOGY AND CONTROL OF MEASLES

Annual Report; March, 2000

### AMERICAN VINEYARD FOUNDATION / VITICULTURE CONSORTIUM

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#### III. Summary:

Pathogenicity trials have been done with Thompson Seedless, Flame Seedless, and Cabernet varieties inoculated with spore suspensions of two species of *Phaeoacremonium* and *Phaeomoniella chlamydospora* (*Phaeoacremonium chlamydosporum*). Additional experiments are also underway to stimulate early fruit production on certain varieties of grape to study the measles symptoms on the berries. Spore traps have been placed in ten production locations select vineyards where measles have occurred. Lab procedures have been developed to facilitate the recovery of *Phaeoacremonium* and *Phaeomoniella* spores from these traps. In vitro methods are being used to screen materials that may be effective against these pathogens.

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#### IV. Objectives and Experiments conducted:

1. Determining the cause of Measles. Grapevine measles is characterized by yellowing and crinkling of the foliage, chlorosis and necrosis of foliage, shoot dieback, and distinct spotting of the berries. Internally, vascular tissue is discolored and often occluded. Severe infections can result in death of the plant (apoplasty).

Past work determined that a new genus, *Phaeocremonium*, was isolated from plants exhibiting measles symptoms. This genus includes numerous species, at least three having been recovered from grapevines in California; *P. chlamydosporum*, *P. aleophilum*, and *P. inflatipes*. One of these species, *P. chlamydosporum*, has recently been reclassified into a new genus, *Phaeomoniella*, with a slightly different species name, *chlamydospora*. It is suspected that the disease involves a predisposition to symptom development by some type of stress (water deficit, nutrient stress, or cultural practice). Two of the species (*P. aleophilum* and *P. inflatipes*) may be endophytic, living in the plant without necessarily causing a problem until specific conditions arise.

Pathogenicity tests have been conducted with the three species mentioned above to determine if these fungi can attack the plant through pruning wounds. Other studies were done to evaluate the effect of infection by these pathogens, as well as the site of entry.

2. Study the biology of the pathogens and epidemiology of the disease. Since the cause of measles has not been completely elucidated, all species of *Phaeocremonium* and *Phaeomoniella* as well as other suspect fungi are being studied for their relationship to this disease.

Spore traps were placed in select vineyards where measles have been known to occur or suspected (El Dorado, Napa, Sonoma, Mendocino, Yolo, Contra Costa, Alameda, Tulare, Madera, Kern, and Solano counties). The traps are designed to catch spores of *Phaeocremonium* (and *Phaeomoniella*) when the fungus sporulates. These spore traps consist of vaseline-coated (both sides) glass microscope slides mounted in a ball of clay and affixed to a cordon of a plant that had or is suspected of having measles. There are three slides per "trap", oriented in three planes, so that the slides are exposed to all six "directions"; north, south, east, west, up, and down. After a week or so, depending on the weather, the slides were collected, taken or sent to our lab, and rinsed with sterile distilled water. The rinsate is then passed through filters to remove most of the other fungal spores, and plated onto media conducive to growth of *Phaeocremonium* species. We have already recovered spores of *Phaeocremonium inflatipes* from one site indicating the airborne nature

of the pathogen and implicating pruning wounds as a natural mode of entry into the vine. This work is continuing and will conclude at the end of rainfall for the season.

3. Develop strategies to control fungi suspected of causing measles. Since the use of sodium arsenite has been banned on grapes, other materials need to be found that can control this disease.

We have had some tentative success with controlling the growth of *Phaeoacremonium* and *Phaeomoniella* on agar plates, but we need to repeat and confirm our initial results before we can go to in vivo studies.

- V. Major research accomplishments and results: Pathogenicity tests have shown that both species of *Phaeoacremonium*, (*P. aleophilum* and *P. inflatipes*), and *Phaeomoniella chlamydospora* were reisolated from cuttings exposed to inoculum in infested sand. These results showed that all three species were able to infect dormant grapes through cut wounds exposed to infested sand. These tests also showed that there were significant reductions in plant height, number of internodes, number of roots, and root length in plants inoculated with these pathogens, compared to the control (non-inoculated) plants. Additionally, these tests reproduced the black discoloration of vascular tissue upon inoculation with all three of these pathogens. These fungi were all re-isolated from the test plants, indicating that Koch's postulates were fulfilled thus identifying them as pathogens of grapevine.

We have also found that in California vineyards, vascular streaking below pruning wounds on two year-old wood resulted from infections primarily from *Phaeomoniella chlamydospora*, with minor infections from *P. aleophilum* and *P. inflatipes*. Testing has shown that *P. chlamydospora* is primarily a wound pathogen whereas the other two species attack mainly through roots. However, *Phaeoacremonium spp.* can also invade through pruning wounds and *Phaeomoniella chlamydospora* can invade through roots but is a better pruning wound pathogen.

We have reproduced foliar symptoms in the greenhouse on both table and wine grape by inoculating plants with *Phaeoacremonium inflatipes* and *Phaeomoniella chlamydospora*.

We have recovered *Phaeoacremonium inflatipes* spores with our spore traps from one site. This indicates that the fungus resides on the grapevine or some other host in the vineyard. We suspect that it is on the vines themselves and current work is being done to try to identify what the structure is that is producing these spores.

VI. Outside presentation of research:

Vine Decline in California; epidemic or ruse. Gubler et.al. First International workshop on grapevine trunk diseases. October, 1999. Submitted manuscript to *Phytopathologia Mediterranea* and presented paper at the meeting in Siena, Italy.

Grape growers report losses to black-foot and grapevine decline. Scheck, et.al. *California Agriculture*, July-August, 1998.

Pathogenicity of *Phaeoacremonium spp.* On grapevine in California. Khan, et.al. in press; *Phytopathologia Mediterranea*.

Fungal species associated with Spanish Measles of grapevine in California. Gubler, et. al. In preparation.

VII. Research success statements:

Measles is still a significant problem for both table and wine grape growers in California (and the rest of the world). Reduction of vine vigor and fruit production (quality and quantity) continues to keep our interest in determining the cause of this disease and developing control measures. Since this disease occurs sporadically to some extent, and seems predicated on certain weather situations (wet springs and hot summers), we suspect that other factors than just the presence of the fungus are involved in the development of the disease and the expression of measles symptoms. By determining when spores are released by the fungus we may be able to target control measures to certain times, thereby reducing the number of spray applications necessary. Grapevine measles, (Esca, Apoplexy), is a complex disease involving several pathogens; although we have made progress in solving this puzzle, additional work is needed to further our understanding of pathogen biology and disease epidemiology.

VIII. Fund status:

Funds depleted.

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