

BOTRYODIPLODIA THEOBROMAE - A NEW GRAPEVINE PATHOGEN

George Leavitt, Dr. Donald Munnecke,
Dr. Robert Endo, Dr. John Menge

ABSTRACT

Arm death continues in the Koch's Postulates trial at the Kearney Agricultural Center. 6-7 are now dead and 30-40 exhibited very poor growth last year.

The natural infection trial at Coachella continues to become more infected each year. Several more years of observation will be needed before results are conclusive.

The fungicide pruning wound paints appear to work as infection by the inoculated strain was held to a minimum by all fungicides applied. Latex paint did not work. A time of pruning in relation to wound healing and subsequent infection was established. Results are not yet available.

Sampling from various vineyards statewide continues to strengthen the hypothesis that B. theobromae is a grapevine pathogen and is more widespread than Eutypa dieback.

OBJECTIVES

1. Continue observation trial and confirm Koch's postulates on Thompson seedless.
2. Prevention of infection at pruning wounds using different fungicides and formulations.
3. Continue monitoring of natural infection plot in Coachella on Perlette.
4. Establish a time of pruning and susceptibility of wounds to infection.
5. Continue spore trapping to determine time and condition of spore release in relation to cultural practices.

PROCEDURES

1. Continue monitoring Koch's Postulates begun at the Kearney Agricultural Center in 1983 to observe the development of the disease and its subsequent effect on vine growth. Pruning weights will be recorded from inoculated arms in late winter 1987.
2. A pruning wound fungicide trial was established at the Kearney Agricultural Center on Thompson seedless in February 1986. The treatments were as follows:

Treatment Number	Material	Rate of Formulated Material per gal	Number of spores Applied Post Treatment
1.	Benlate	3.2 oz	plus 500
2.	Rovrol	3.2 oz	plus 500
3.	Rovrol + white latex paint	3.2 oz	plus 500
4.	White latex paint	-	plus 500
5.	Captan	3.2 oz	plus 500
6.	Topaz 1E	195.5 ml	plus 500
7.	Control 1 (Benlate)	3.2 oz	plus 0
8.	Control 2 (No Chemical)	-	plus 500
9.	Control 3 (Natural infection)-	-	plus 0

The vines were pruned leaving a 3-4 inch spur on the arms which were to be inoculated. The spurs were removed just prior to treatment to create a fresh pruning wound. Each treatment was applied to thoroughly flood the wound surface and allowed to dry. After drying, the appropriate number of spores for each treatment (0 or 500) was also applied distributing the spores over the entire wound surface. The spores were collected from an isolate which demonstrated rapid growth at 36 C and produced red pigmentation (see 1984-1985 reports). This provided a rapid means of identifying the strain used for inoculation from the natural contaminants. Eight single vine replicates for each treatment were used with at least 3 arms per vine. The plot was established in February 1986 and harvested one year later. Each arm was removed from the vine and given a number. Each arm was prepared for sampling by removing all the loose bark and immersing for 10 minutes in a 9:1 solution of water and household bleach (sodium hypochlorite 5.25%).

After air drying a flame sterilized knife was used to expose the internal vascular tissue. Small pieces of wood were sampled from each arm at the inoculation site and at one inch intervals below the point of inoculation. Each sample was kept separate. Whenever a pruning wound site or another branch occurred on the sample below the point of inoculation, sampling was discontinued at that point due to possible contamination. Four pieces of wood from each sample were put onto sterile potato dextrose agar (PDA) plates and cultured using 12 hour light and dark intervals.

After 10 - 14 days each plate was checked for the presence of B. theobromae. Each positive culture was transferred to fresh plates to obtain pure cultures. Eventually each positive sample was grown at 36 C to separate the inoculated isolate from the natural contaminants.

3. One hundred twenty Perlette vines in the Coachella Valley were closely examined at every spur position for 3 years. No pruning wound fungicides are being applied. These vines will be checked each year to determine the "natural infection rate".
4. A time of pruning and its effect on wound susceptibility to infection was also established. Thirty-six vines were pruned on each of three dates: December 20, 1985, January 17 and February 14, 1986. Six single vine replicates were established for the following treatments on each date.

<u>Number</u>	<u>Treatment</u>
1	Check - No treatment
2	Prune - inoculate with 500 spores
3	Prune Wait 1 week - inoculate with 500 spores
4	" " 2 " " " " "
5	" " 3 " " " " "
6	" " 4 " " " " "

Those wounds to be inoculated were marked with bright paint at the time of pruning. The same fungal isolate used in the fungicide trial was also used here to differentiate natural contamination from the treatments.

- Two spore traps began operation again in November 1986. Spore distribution can possibly be monitored. Last years date (1984-1985) is still being analyzed to determine environmental conditions which will cause spore distribution in vineyards. One trap was in a vineyard to monitor the "natural" conditions. The second trap was placed in an open area but surrounded by a ring of old stumps and vines removed from vineyards. This "baited" trap is to determine if old wood is a source of spores.

RESULTS

- The Kochs postulates trial is progressing well. 6-7 arms have died (as of winter 85-86) and many more are very weak. Pruning weights have not yet been recorded for this year as it has not yet been pruned. One-third of the trial will be harvested next month for reisolation and analysis.
- Preliminary data indicates all of the fungicides used were effective in reducing the incidence of wound infections. Statistical data is not yet available. It also appears a high rate of natural infection can still occur. Latex paint used alone or in combination with a fungicide was not effective in reducing infection.
- The natural infection study in Coachella has been monitored for 3 years. The vines are now in full production and more arms are infected. Not all infections become immediately evident. In fact, most do not become obvious for several years. Those which I have found (6-3) will be observed yearly for symptoms and to watch disease progression.
- No results are yet available as this experiment is currently being harvested.
- Spore traps will continue until the end of May. No analysis is yet available. As this fungus is not readily moved by wind the numbers on the trap tapes could be very low. Other methods of spore dispersal (insects) will be briefly investigated.