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Avocado Black Streak

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We are continuing Black Streak studies on several fronts. The seed transmission studies have been completed with interesting results. Tissue culture efforts continue and we are (as time permits) making more isolations from the lesion areas on affected trees to assure ourselves that we have not missed anything. A field study has been completed to determine if a correlation between Black Streak symptoms and dsRNA patterns exists in scion materials. Currently we are assaying collections from rootstock suckers from both healthy and Black Streak affected trees to determine if there are any differences between the two in dsRNA patterns.

We have picked up the mill that Mr. Shepherd located for us and by the time of the annual meeting we hope to have developed the techniques needed to grind the leaf tissue that we are extracting. Hopefully this will reduce the time needed to process the samples. We may have to develop new techniques because grinding is usually done in liquid nitrogen which may not be possible in the mill.

Seed Transmission Experiments

These experiments were designed to determine if the dsRNA patterns found in seedlings are the same as those patterns found in the parent tree and to determine the extent of transmission of those patterns to the seedlings.

Six different varieties of avocados were used with 100 seeds of each being planted. Of the resulting seedlings 43 to 76 were assayed depending on the variety and the time available. Seedlings were assayed for dsRNA patterns by polyacrylamide gel electrophoresis.

The results of this experiment turned out to be quite interesting. Duke 7 with a parent tree showing no dsRNA to be present also did not have any seedlings with any dsRNA patterns. This could mean that there is no seed transmission of the patterns, that there is no pollen transmission of the patterns, or that we were unable to detect dsRNA in this host for some reason. Since we have had no trouble detecting dsRNA in other Duke 7 selections the conclusion that neither type of transmission occurred seems to be the correct one. However, as time permits, it may be best to compare this Duke 7 selection with one that is known to have dsRNA patterns present.

RESULTS:

Seedling dsRNA Patterns	Parent dsRNA Pattern					
Teague	Duke 7	G-6	Zutano	Rincon	Ganter	
1-2-3	0	1	2	3	1-2-3	
1	0	36	1	1	0	0
2	0	0	17	0	24	0
3	0	0	0	22	3	29
1 & 2	0	2	1	1	1	2
1 & 3	0	36	0	2	0	3
2 & 3	0	0	28	15	14	20
1 & 2 & 3	0	2	4	10	1	3
Total Seedlings	50	76	51	51	43	57
Parent dsRNA Present	50	76	50	49	1	3
Percent Transmission of Parent dsRNA	100	100	98	96	2	5

The G-6 variety used in this experiment had pattern 1 in the parent tree. Results of the pattern analysis show that there was 100 percent transmission of pattern 1 either by itself or in combination with another pattern. Of interest is the fact that pattern 3 showed up in exactly half (38) of the seedlings and pattern 2 showed up in 2 seedlings. Neither pattern 2 or 3 showed up by itself but was always in combination with one or two other patterns. From these results we can see that pattern 1 was transmitted 100%. However, from this data it would be impossible to determine if the transmission was through the seed, by pollen, or a combination of the two. The most interesting aspect is the presence of two patterns that were not present in the seed parent. From this we can conclude that there was pollen transmission or that we were unable to detect those patterns in the seed parent. Of the two pollen transmission seems to be the most logical explanation.

Zutano variety with pattern 2 in the seed parent had 98% transmission of this pattern to the seedlings. It also had evidence of pollen transmission with a 64% transmission of pattern 3 and a 12% transmission of pattern 1.

Rincon variety had pattern 3 in the seed parent and had a 96% transmission factor of this pattern to the seedlings. It also had a 14% transmission of pattern 1 and a 51% of pattern 2 again supposedly by pollen transmission. It is interesting to note that the trees with patterns 1 and 3 did not have any transmission of pattern 2 alone but that it was always in combination with another pattern.

In the Ganter and Teague varieties used, the parent trees all had pattern 1, 2, and 3. It is interesting to note that in Ganter all three patterns transmitted together in only 2%

while in Teague they all transmitted only 5%. Pattern 2 transmitted by itself in Ganter at a 46% rate and not at all by itself in Teague. Pattern 3 transmitted by itself at 51% in Teague and only at 7% in Ganter. In both varieties pattern 1 failed to transmit by itself. In both varieties it would be impossible to determine if pollen transmission occurred from these data.

Another area that we have been active in is in collecting and assaying rootstock sucker material from trees known to have a past history of Black Streak. Ten samples were taken from suckers of trees that had been cut down but known to have had Black Streak. Ten samples of such suckers were taken from the rootstock suckers of trees that had been cut down but had no history of Black Streak. In this case an effort was made to choose trees that did not have adjacent trees with Black Streak. Ten samples were taken from scion material of known Black Streak trees and ten were taken from scion material of trees thought to be healthy and again not adjacent to Black Streak affected trees. These samples are currently being processed and we hope to have the results by the meeting.

Tissue culture continues as we try to work out the techniques needed to obtain virus free material. The fact that seed transmission is not 100 percent leads me to believe that we will be able to obtain virus free material. Further work needs to be done but there is no evidence to date that the dsRNA patterns can be pollen transmitted to another existing tree, only to the seedling. If this is true then we may well be able to maintain virus free trees in the field unless there are other vectors present for transmission. Before enlarging the tissue culture operations we are working with a few to work out the techniques needed for success. We will expand that project when we are confident that we have a good chance of success.

We have also decided to look over some of the past work to make sure we have not bypassed an area that should be investigated further. We are sampling trees from a diverse area and isolating for fungi and/or bacteria. This project has just started and we have no results to report yet.

As previously reported, we sampled scions of a grove of 343 trees of known Black Streak distribution and assayed them for their dsRNA patterns. We were unable to find any correlation between patterns in the scion and Black Streak distribution in the grove. Due to the lack of suckers we were unable to test for patterns in the rootstock. We must also be aware that the possibility exists that our current techniques may not be able to detect low levels of virus in avocado.