

FINAL REPORT

WTFRC Project # AH-01-76

Organization Project #4248

Project title: Tree vigor & date of fruit maturity as affected by virus.

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Objectives:

1. Determine if differences in desirable horticultural attributes (early ripening) associated with 'Yataka' clones are due to graft transmissible agents or due to other causes.
2. Compare horticultural effects of standard latent virus isolates, individually and in combination, with those of the virus isolates from 'Yataka' and from apple green crinkle affected trees.
3. Segregate constituent viruses from 'Yataka' and from apple green crinkle diseased trees. Compare them to standard isolates of the common latent viruses.

Significant findings:

- No obvious symptoms or horticultural differences developed after inoculating 'Fuji' with graft-transmissible agents from early ripening 'Yataka' clones or from green crinkle diseased sources.
- In disagreement with last year's results, sugar and starch tests indicated that fruit maturity on 'Fuji' inoculated with early-ripening 'Yataka' was slightly delayed over that of non-inoculated 'Fuji'.
- Similar to last year, virus inoculations reduced vigor compared to non-inoculated controls. The green crinkle inoculated trees were among the least vigorous.
- The block of trees established to determine the cause of apple green crinkle disease grew well this year and should start fruiting in 2003.
- Apple chlorotic leaf spot (ACLSV) strains from apple green crinkle diseased trees were separated to help in future studies to identify the cause of this disease.

Methods:

1. Evaluate tree vigor and fruit characteristics of 'Fuji' trees that were graft-inoculated with tissue from virus-infected early-ripening 'Yataka' and from apple green crinkle affected trees. These attributes are then compared with those from non-inoculated virus-certified trees. This replicated field trial will resolve whether the beneficial horticultural characteristics associated with the early-ripening 'Yataka' and apple green crinkle disease (vigor control) are graft transmissible. Tree vigor will be monitored by measurements of trunk cross sectional areas. Fruit maturity and quality data will be collected from all inoculated trees.
2. Tree vigor and fruiting characteristics of 'Fuji' trees that have been graft-inoculated with tissue from virus-infected early-ripening 'Yataka' and from apple green crinkle affected trees will be compared to 'Fuji' inoculated with standard isolates of the common latent viruses. This comparison will help determine whether the attributes associated with the early ripening 'Yataka' and vigor control of apple green crinkle disease are induced by common strains of the latent viruses or by another agent. Other agents might include a different strain of these viruses or an entirely different pathogen.

Results and discussion:

1. *Is early ripening associated with 'Yataka' clones due to graft transmissible agents?* Fruit maturity differences between Fuji trees inoculated with tissue from early-ripening 'Yataka' and those left non-inoculated varied with the season. In 2000 maturity on inoculated trees was advanced. In 2001 it was delayed. A couple more years are needed to determine what, if any, significant effects these inoculations have on harvest date.

Table 1. Maturity as estimated by starch and brix tests of apples from 'Fuji' trees graft-inoculated with tissue from early ripening and virus-free 'Yataka' trees in October 2000 and 2001.

<u>Virus</u>	<u>Sugar</u>		<u>Starch</u>	
	00	01	00	01
Treatment				
'Yataka'- early-ripening (virus infected)	14.1	13.8	2.4	3.2
'Yataka' – virus free	13.8	14.2	2.9	3.6
Healthy Control	13.7	15.1	2.8	3.0

2. *Comparison of horticultural effects of 'latent' apple viruses with viruses isolated from early ripening 'Yataka' and from apple green crinkle affected trees.* Early ripening 'Yataka' clones were found to be infected with three latent apple viruses, namely *Apple chlorotic leafspot virus* (ACLSV), *Apple stem pitting virus* (ASPV) and *Apple stem grooving virus* (ASGV). Pairs of virus-free 'Fuji' trees were inoculated in 1997 with standard isolates of these three viruses. These viruses are also found in trees with apple green crinkle disease. The viruses were introduced individually and in combination; each treatment was replicated three times in a randomized manner. No obvious leaf or fruit symptoms resulted from any of these inoculations. Trunk cross-sectional area measurements are made at the end of each growing season to evaluate vigor. Virus-inoculated trees in general had less vigor than the non-inoculated virus-free control trees. The green crinkle-inoculated trees and the trees inoculated with all three latent viruses were the least vigorous (Table 2). We anticipate that these differences will continue to become more pronounced with each subsequent growing season.

Table 2. Vigor of virus-inoculated Fuji trees as estimated by growth in trunk circumference (mm) from the fall of 1998 to the fall of 2001.

<u>Treatment</u>	<u>Initial Size</u>	<u>Trunk Circumference Increase 1998-2001</u>
Healthy Control – set 1	210	96
Healthy Control – set 2	223	90
ACSLV, ASGV & ASPV	221	85
Green crinkle diseased	223	73

Fruit characteristics from trees inoculated with the various viruses and virus combinations were fairly consistent across all treatments. Brix readings averaged between 13.4 to 15.6% soluble solids and starch between 2.8 to 4.0 (Fuji starch chart 0-6). Based on the width-to-height ratio of fruit, apples from treatments containing both ASGV and ACLSV were the most flat (Table 3), but the differences were not great.

Table 3. Average fruit quality parameters on apples from virus-inoculated Fuji trees.

Virus	Weight	Sugar	Starch	W/H ratio
Healthy Control – set 1	0.57 lb	15.1	3.0	1.15
Healthy Control – set 2	0.62	14.6	3.2	1.16
ACLSV	0.61	14.3	2.9	1.16
ASGV	0.56	13.9	2.9	1.10
ASPV	0.56	14.8	2.8	1.15
ACLSV & ASGV	0.51	13.9	4.0	1.17
ACLSV & ASPV	0.58	14.7	3.5	1.15
ASGV & ASPV	0.57	14.6	3.4	1.15
ACSLV, ASGV & ASPV	0.54	13.4	3.7	1.17
Green Crinkle	0.53	15.6	3.4	1.15

3. *Segregation and comparison of viruses from early ripening 'Yataka' and from apple green crinkle-diseased trees.* During the past two years, the virus complexes within 'Yataka' and green crinkle affected trees were separated to help determine their individual effects. They were primarily separated using heat therapy. From that process 44 'Golden Delicious' trees with individual viruses and combinations of viruses were established and planted in a field plot in 2000. The trees grew well during the past growing season and will be observed over the next few years for symptom development. These observations will help identify the causal agent of apple green crinkle disease and to assist in determining if the fruit deforming agent of apple green crinkle disease can be separated from the agent that provides vigor control.

As described previously, there is evidence that a particular strain of ACLSV may be the causal agent of apple green crinkle disease. It appears that we've successfully transferred this and the common ACLSV isolate to herbaceous plants, characterized them, and returned them to apple by using herbaceous-woody plant grafting. Tissue from these newly infected apple seedlings will be utilized to inoculate Golden Delicious apple, which will then be observed over the next few growing seasons for green crinkle symptom development. If our hypothesis is correct, one of the ACLSV isolates will induce the disease while the other will induce no obvious symptoms. Success in this effort would allow us to develop rapid laboratory tests to detect the apple green crinkle agent in a few days, a process that now takes 3 to 4 years on biological indicator plants. Such a test would be a great asset in helping to keep this disease from adversely affecting orchard operations.

Budget:

Project duration: 1999-2001

Current year: FINAL REPORT

Original budget request:

Year	Year 1 (1997)	Year 2 (1998)	Year 3 (1999)	Year 4 (2000)	Year 5(2001)
Total	12,000	12,000	12,000	12,000	2,500

TOTAL: \$50,500