

2006 OSCC/WTFRC

Cherry Research Review

November 3-4, 2005

The Dalles, Oregon

Time	Page	PI	Project Title	Funding period
8:00		McFerson	Introduction & Technology Roadmap Update	
8:15	1	Iezzoni/Whiting	Breeding and genetics program for PNW sweet cherries	05-07
8:45	9	Olmstead/Iezzoni	Environmental and genetic influences on cherry fruit size	04-05
9:00	19	Iezzoni	Sweet cherry dwarfing rootstocks	05-07
9:10	25	Whiting	Clonal rootstock evaluation	04-06
9:15	34	Núñez-Elisea	Cultivars, rootstocks, training systems, and fruit quality	03-05
9:30	40	Azarenko	Horticulture management systems for fresh & brine cherries	90
9:45	47	Yin	Alternative nutrient, water and floor mgmt strategies	05-07
10:00			Break	
10:15	53	Núñez-Elisea	Tree water use, irrigation, & water management	03-05
10:30	58	Whiting	High density orchard systems	04-06
10:35	64	Whiting	Sweet cherry source-sink relations	04-06
10:40	70	Millard	Understanding N requirements for cherry production	05-07
10:45	76	Azarenko	Flowering, pollination & fruit set of sweet cherry	05-07
10:50	81	Elfvig	Branch induction in cherry trees in orchards & nurseries	05-07
10:55	85	Elfvig	Bioregulators to manage growth, flowering, and cropping	04-06
11:00		Bai	Edible coating to improve storage and marketing quality	04-06
11:15		Schrader	Improving cherry fruit quality & post harvest shelf life	05-06
11:20		Whiting	Trip report: ISHS Turkey Symposium	05
11:45			OSCC/WTFRC Committee Lunches	
1:30		Grove	Modeling and managing cherry PM	04-06
1:35		Calabro	Biology & control of fruit infection phase of PM (extension)	03-05
1:40		Eastwell	Virus control strategies to assist cherry production	04-06
1:45		Brunner	Biology and management of bark beetles	04-05
2:00		Hansen	Use of surfactants to remove surface pests	05
2:15		Yee	Insecticide effects on CFF biology	04-06
2:20		Yee	CFF feeding ecology and food-based lures and baits	04-06
2:25		Smith	CFF control options	04-05
2:40		Azarenko	Managing soil ecology to improve soil & tree function	05-07
2:55		Culbertson	California Update	

CONTINUING PROJECT
WTFRC Project #: CH-05-504

YEAR 1/3

Project Title: Breeding and genetics program for the PNW fresh market sweet Cherries
P.I.: Dr. Matt Whiting
Organization: Washington State University
Address: Prosser, WA
Cooperator: Jim Olmstead
Consultants: Dr. Amy Iezzoni & Dr. Fred Bliss

Commercial Goal: Develop a full-season series of sweet cherry cultivars that exceed current cultivars for a range of characteristics desired for current and future domestic and foreign market opportunities.

Scientific Goal: Initiate a broad-based research and development program on genetics and genomics of sweet cherry, apply the results to the plant breeding program, and integrate the information and materials into a multi-disciplinary tree fruit initiative.

Objectives to achieve goals:

1. Develop a comprehensive framework for a sweet cherry breeding and genetics program.
2. Produce through hybridization and selfing, genetically-variable sweet cherry selection populations that segregate for important target traits.
3. Select outstanding families and elite individual seedlings in multiple target environments for important production and fruit traits.
4. Establish elite selections in replicated multi-site locations for pre-commercial evaluation.
5. Install a data base for storage and analysis of critical information that will effectively support the breeding goals.
6. Acquire and develop molecular information and tools that facilitate rapid and efficient marker-assisted selection for production and fruit traits critical for commercial success.

Significant findings and accomplishments:

- A comprehensive oversight and communications framework was established and implemented. Oversight of the breeding program is provided by an Advisory Committee (AC) consisting of cherry growers from WA and OR.
- Germination of seeds from YR 2004 crosses in the seed beds at Willow Drive Nursery was extremely poor as the seeds and seedlings rotted with only 250 seedlings resulting from the 4,466 seeds (Appendices 1 and 2).
- In consultation with the AC we decided to bud the 250 YR 2004 seedlings on a precocious rootstock so that early flowering would be assured. Matt Whiting further determined that the budding be on MaxMa14 and not GI 6 as the bud take on GI 6 is reported to be extremely low.

- New strategies for seed germination were implemented with the seed from YR 2005 crosses. The vast majority of the seeds were handled using a standard stratification protocol (See Methods section). The germinated seedlings will be grown in the greenhouse.
- A germplasm collection trip by Matt Whiting was canceled. Instead germplasm was successfully imported by Amy Iezzoni as pollen (Appendix 1).
- Despite the consistently cold weather conditions, 84 crosses including reciprocal crosses were successfully accomplished at Prosser in spring 2005 resulting in 7,166 hybrid seed (Appendices 1, 3, and 4).
- Matt Whiting planted a new orchard to evaluate the potential for rootstock and nursery production system to hasten productivity. ‘Bing’ and ‘Tieton’ on Mazzard seedling, Gisela®6, and Maxma14 trees were planted at the Roza farm as sleeping eyes (i.e., fall-budded, spring dug and planted), and standard nursery trees (i.e., fall-budded, nursery-grown, spring dug and planted +1yr).
- The project to identify a molecular marker linked to the gene controlling powdery mildew resistance is two thirds completed. Because of a high level of genetic similarity, no candidate markers linked to *Pmr-1* have been identified to date.
- It was decided that the Oregon selection site (Obj. 3) will be located at the Hood River Station.
- Amy Iezzoni received a \$400,000 three year USDA-CSREES-NRI grant entitled “Genomic resources to improve fruit size and quality in sweet cherry”. This grant which started June 1, 2005, provides funds to obtain the baseline genetics/genomics research required for the breeding program. The grant team members are Drs. Wayne Loescher (fruit biochemistry/physiology), Dechun Wang (quantitative genetics) and Esther van der Knaap (molecular genetics).

Methods to be employed by objective:

1. Develop a comprehensive framework for a sweet cherry breeding and genetics program.

- 1.1 Complete a feasibility study to verify accelerated crossing strategy: Completed in 2004
- 1.2 Develop a breeding program strategy and implementation plan: Completed in 2004 and contained in Appendix 1 of the original proposal. Current modifications to the implementation plan are described under Objectives 2 and 3.
- 1.3 Present a project proposal to funding board for approval. This proposal was submitted on October 8, 2004.
- 1.4 Establish a defined working relationship with an industry Advisory Committee made up of members from the WA and OR industries: Completed in 2005.

2. Produce through hybridization and selfing, genetically-variable sweet cherry selection populations that segregate for important target traits.

Determine the final crosses taking into account seed quality, bloom time, self-fertility or incompatibility group, and virus status. Diverse germplasm will be obtained from Europe as pollen. The goal is to produce enough seed to generate 5,000 seedlings per year for each of the three years.

Methods modification: Seedling germination:

Rationale: The seeds from the YR 2004 crosses were planted in a seed bed at Willow Drive Nursery in October 2004. Germination was very poor and the vast majority of the seeds and seedlings rotted. Therefore, we implemented a standard seed germination procedure described below with the seeds from the YR 2005 crosses. In July 2005 we also attempted a second procedure whereby the seed coat was removed and the seeds were placed in sterile germination media. If successful, this technique will also be used with YR 2006 seed.

- Seed germination using standard stratification procedures: The seeds were cleaned and placed in plastic bags containing moist vermiculite mixed with a half-teaspoon of Captan. The seed bags were placed in 35F. Seeds will begin to germinate in the bags in December and January. Once the seed radical is over ½ cm long, the seeds will be removed and planted in the greenhouse in a standard potting mix.
- Seed germination following the removal of the seed coat: Seeds were cleaned using normal procedures, surface sterilized by soaking in a dilute Clorox solution. The seed coat was peeled off and the seeds were placed on sterile perlite in Petri plates. The plates were placed in the refrigerator. Germination is reported to occur in approximately 3 months. At that time the seedlings will be planted in the greenhouse.

3. Select outstanding families and elite individual seedlings in multiple target environments for important production and fruit traits.

3.1 Develop effective methods to accelerate seedling flowering and fruiting using grafting to Maxma 14 and two growth cycles in year 1. *NOTE:* These two strategies represent modifications to the original plant handling strategy.

YR 2004 seedlings:

Rationale: Because only 250 seedlings resulted from the YR 2004 crosses, these seedlings will be budded on a precocious rootstock to ensure that they will have abundant fruit as soon as possible. The budding is possible due to the cost savings associated with the low seedling number. Matt Whiting determined that it would be best to bud on MxM14 as the bud take on GI 6 can be extremely low.

- The 250 seedlings at Willow Drive Nursery will be labeled and dug in fall 2005. In winter 2006, budwood from each tree will be taken and bench grafted to MaxMa14. The resulting trees with a sleeping eye bud will be planted at Prosser in the spring along with the original seedlings grown at Willow Drive Nursery. Those MaxMa14 trees where the bud fails will be rebudded as necessary.

YR 2005 seedlings:

Rationale: Our goal is to bring trees into significant bearing as soon as possible to accelerate fruit evaluation. Obtaining the abundant fruiting needed for evaluation can require as long as 5 to 6 years on seedlings trees that have the genetic tendency towards a long juvenile period. Such is the case for the population NY 54 x Emperor Francis, where in year 4, only a few flowers were produced on ~ 20% of the seedling trees. Fortunately, sweet cherry breeding programs have inadvertently selected for the genetic tendency for a reduced juvenility period. For example, seedlings from crosses between Rainier, Bing, Van and PMR1, fruited profusely in Year 4 (Jim Olmstead, pers. comm.). When a seedling has this genetic tendency towards early blooming and fruits heavily in year 4, there is no time saving associated with budding on a precocious rootstock. This is because one year is lost when the seedling is chip budded onto the rootstock in the fall of the first year. Therefore, the **first new strategy** that we propose to reduce the flowering time involves budding the seedlings the first spring and obtaining grafted trees the first year. The **second strategy** that we propose to investigate does not involve grafting the seedlings. Instead the seedlings are cycled in and out of a cold room so that the seedlings experience two artificial growing seasons prior to field planting. If both these scenarios are successful, abundant fruiting should occur in year 3.

- **Shoot tip grafting onto GI6 (Fig. 1):** Plant 100 one-year-old GI 6 liners in the greenhouse in January 2006. Once the seedlings to be grafted and the rootstocks are growing vigorously, cut a succulent tip off of the seedling shoot and using a cleft graft, place the shoot tip on the rootstock. The succulent tip will start growing immediately and so the grafted plant is placed in a humid chamber for one or two weeks. The succulent tip is then adapted to normal atmospheric conditions as the vascular connections are initiated. This procedure will be done in collaboration with the NRSP-5 Heat Therapy technician who does this procedure routinely.
- **Two growth cycles in Year 1 (Fig. 2):** Grow the germinated seedlings to maximize growth using optimum growing practices and a pot size of at least 6 inches. We anticipate that we can grow the seedlings to two to three feet in height in January to the end of April. Move the seedlings to a cooler and over a 10 day period reduce the temperature to 5 C and the light to 8 hour days. Maintain the 5 C and the 8 hours days for four months (until the end of August). Water as needed. Hand defoliate the plants in the end of July. In September, move the seedlings into the greenhouse. Tip the terminal bud to encourage lateral growth. Grow the seedlings using optimum growing practices for four months. Our goal will be to obtain 2 to 3 side branches per seedling. Put the seedlings back in the cooler and follow the same acclimation and cultural procedures as above. Remove the seedlings from the cooler in April 2007 for field planting.

Strategy for future implementation:

We speculate that those seedlings which are genetically predisposed to a short juvenility period would respond well to the cycling strategy and would flower profusely in Year 3. However, those seedlings that are genetically predisposed to a longer juvenility period may need to be grafted to GI 6 to ensure early flowering. In order to determine what procedure to use for each seedling, we will explore the possibility of identifying molecular marker(s) linked to juvenility time. The onset of flowering and fruiting will be recorded in the Emperor Francis x New York 54 population and subjected to QTL analysis using the developing sweet cherry linkage map (part of Obj. 6). This initial QTL discovery objectively will be funded by the USDA-CSREES Cherry Genomics Project.

3.2 Establish which production and fruit traits are of the highest priorities as selection criteria for producing competitive new cultivars and then use these as a component of seedling and advanced selection evaluations.

- In consultation with the AC, finalize the descriptions of the target cultivars that are desired outcomes of the breeding program.

3.3 Implement a multi-site selection and testing strategy to emphasize adaptation to specific site and production requirements provide rapid, effective evaluation for commercial value and coordinate evaluation of scion/rootstock combinations.

- The first two selection sites have been determined: A. Prosser: WSU-Roza Experimental Farm, and B. Hood River Station. Identify the last two selection sites within the following locations: C. Wenatchee, mid-elevation 800-1500 ft, and D. Brewster, high elevation 1500-2500 ft (to be identified).
- Prepare the three non-Prosser test sites for planting ~ 500 seedlings each in Spring 2007.

4. Establish elite selections in replicated multi-site locations for pre-commercial evaluation.

Planting will begin in Spring 2007.

5. Install a data base for storage and analysis of critical information that will effectively support; 1) parental choices, 2) pedigree information, 3) selection criteria and progeny performances, 4) trait and tree performance at multiple sites, and 5) filing and prosecution for optimal Intellectual Property protection.

5.1 Acquire and activate a suitable data base for the breeding program.

Preliminary inquiries indicate that a suitable data base system will be publicly available. The system currently used by the WSU Apple Breeding Program may be able to be easily adapted for the Cherry Breeding Program.

6. Acquire and develop molecular information and tools that facilitate rapid and efficient marker-assisted selection for production and fruit traits critical for commercial success.

6.1 Develop and validate a DNA screening test for determining seedlings resistant and susceptible to powdery mildew:

Powdery mildew resistance: Progeny populations resulting from YR 2005 crosses will be screened in the greenhouse for PMR and selected populations will be genotyped using the marker(s) linked to PMR-1 to validate the markers across different genetic backgrounds. This is the final marker test prior to implementation.

6.2 Screen all selection populations segregating for mildew reaction using the DNA screen and complemented by pathology phenotyping: FY07.

6.3 Explore collaboration with 3rd party labs to assess feasibility and cost-effectiveness of applying known/emerging technology to sweet cherry marker development.

- Identify a lab that can do high-through put DNA extraction and marker screening.
- Test the efficiency of screening seedlings for self-fertility.

Summary List of YR 2006 Activities

- Acquire and activate a program database.
- Explore collaborations for DNA marker development and screening.
- Implement the multi-site testing strategy with the establishment of four test sites.
- In consultation with the AC, finalize the descriptions of the target cultivars that are desired outcomes of the breeding program.

YR 2004 seedlings

- Graft the 250 seedlings onto MaxMa14.
- Plant the own-rooted seedlings and the MaxMa14 sleeping eye trees at Prosser.

YR 2005 seedling

- Germinate and plant in the greenhouse and maintain the seedlings resulting from the 7,166 seeds.
- Plant 100 GI 6 liners in the greenhouse and use for shoot tip budding with a subset of seedlings.
- Implement the “two year cycle” regime with a subset of seedlings from YR 2005 crosses.
- Validate the candidate markers linked to PM resistance.
- Screen for powdery mildew resistance those seedlings that are from segregating populations.
- Test the efficiency of pre-screening for self-fertility using DNA markers.

YR 2006 crosses and seed handling

- Develop a crossing plan, import needed pollen and carry out the spring crosses.
- Harvest the fruit and clean and stratify the seed from the spring crosses.

Results and Discussion

A comprehensive oversight and communications framework was established and implemented. Oversight of the breeding program is provided by an Advisory Committee (AC) consisting of cherry growers from WA and OR. Quarterly executive summaries of progress will be and were provided to the AC on a quarterly basis. These quarterly summaries can be freely distributed. Matt Whiting established a secure web site to enhance communication with the AC. This web site also contains seedling and pedigree information that must remain confidential to protect IP. Communication with the entire grower community was accomplished through two articles in the Good Fruit Grower.

Due to logistic considerations the seed collection trip for 2005 was canceled. Instead pollen was imported from Europe (Appendix 1). To date, the project has seedlings resulting from 12 newly utilized foreign sweet cherry varieties (Appendix 1). These foreign selections were chosen specifically to add the following superior traits to the breeding program: heat tolerance, very late ripening time, fruit firmness, large fruit size and suitability for mechanical harvest. A list of the

pedigrees of the seedlings from the YR 2004 crosses and the seeds from the YR 2005 crosses are provided in Appendices 2 and 4, respectively.

The vast majority of the crosses intended for 2005 (Appendix 3) were completed. For certain crosses, the cross could not be done as the only pollen would have been available from trees in the NRSP-5 collection. Unfortunately the flower buds on many of these accessions were completely frozen in the spring. Additionally some crosses did not set presumably due to the cold windy weather. The pollen from Italy arrived after bloom and will be used in YR 2006 crosses. Those crosses that were unsuccessful will also be done in 2006.

The poor seed germination from the YR 2004 crosses resulted in a reduction in the projected number of superior seedlings selected from the material produced in the four years of crossing (Tables 1 and 2). However, if the percentage seed germination from the 7,166 seedlings is 70%, we will meet the seedling target of 5,000 seedlings from the YR 2005 crossing. For the sake of discussion, the projected number of superior seedlings from the four years of crosses was recalculated from that presented in the original proposal using a germination percentage of 50% for the YR 2005 seed. The adjusted number of superior seedlings is reduced to 1,370 from 1,730 (Tables 1 and 2).

Leaves were collected and DNA was extracted from the parents and 375 seedlings from crosses between PMR-1 with Van, Bing, and Rainier that had previously been screened for powdery mildew resistance. Also included were four other cultivars that are powdery mildew resistant, Venus, Moreau, Chelan and Hedelfingen. Resistant and susceptible bulk populations were designed and screened using AFLPs generated from 40 of the 64 possible E+AN and M+CNN selective primer pair combinations. Because of a high level of genetic similarity, no candidate markers linked to *Pmr-1* have been identified to date. However, twenty four primer combinations remain to be screened. The limited number of polymorphic fragments identified among the four sweet cherry cultivars (PMR-1, Bing, Van and Rainier) highlights the genetic uniformity present in sweet cherry cultivars.

The breeding project includes a budget line of \$10,000 for genotyping services, not the genetic research that would be required to elucidate the genetic control of complex fruit quality traits. The USDA-CSREES project was designed specifically to fill this void. The goal of the USDA grant is to develop the genomic resources required to implement marker-assisted selection in cherry (*Prunus* sp.) breeding programs. We plan to accomplish this goal using a QTL strategy focused on fruit size and quality traits, followed by QTL validation and allele mining using a newly-developed pedigree genotyping approach. The research consists of the following steps: (1) Construct a sweet cherry genetic linkage map for comparative mapping with the *Prunus* reference map and other *Prunus* linkage maps. (2) Identify QTL for fruit size and quality traits. (3) Fine map the major QTL identified and design markers for marker assisted selection. (4) Validate the QTL across genetic backgrounds and identify QTL alleles. This supplemental funding will allow us to greatly exceed our prior expectations for objectives 5 and 6, database capability and acquisition of molecular information, respectively. In addition, one of our team members, Dr. Wayne Loescher, is studying the biochemical basis of the differences in fruit quality using fruit from the varieties used as parents in the breeding program. This information will greatly enhance our selection and QTL discovery capabilities.

2006 BUDGET

Project Title: Breeding and genetics program for the PNW fresh market sweet Cherries

P.I.: Dr. Matt Whiting

Current year: 2006

Project total (3 years): \$ 235,255

Current year request: \$ 86,722

Year	Year 1 (2005)	Year 2 (2006)	Year 3 (2007)
Request	\$41,397	\$86,722	\$ 107,136

Budget

Item	Year 1 (2005)	Year 2 (2006)	Year 3 (2007)
1. Salaries			
a. P.I.	0	0	0
b. Post-doc	0	\$30,000	\$31,200
c. Benefits (%)	0	\$11,400	\$11,856
2. Hourly labor - WSU			
a. Regular employees	\$3,000	\$8,000	\$3,000
b. Commercial employees	\$500	\$500	\$500
3. Supplies	\$500	\$1,000	\$1,500
4. Equipment	\$500	\$500	\$1,500
5. Travel			
a. Within WA & OR	0	\$1,000	\$1,500
6. Tree/rootstock cost			
a. Seedling (@ \$1/tree)	\$200	0	
b. Grafted (@ \$10/tree)	\$3,000	0	
c. MxM14 sleeping eye		\$2,500	\$2,500
d. GI6 for shoot-tip grafting		\$200	\$400
7. Plot fees			
a. WSU – Prosser	0	0	0
b. Non-Prosser sites	0	1,200	7,080
8. Contract Services			
a. Breeding consultant	\$13,000	13,000	13,000
b. Genetics consultant (Bliss)	\$10,270	10,500	10,500
c. Virus indexing (NRSP-5)	\$1,000	\$1,000	\$1,000
d. DNA genotyping services	\$10,000	\$10,000	\$15,000
e. Shoot-tip grafting (NRSP-5)	0	\$800	
9. Cost savings from YR 1		Minus \$4,828	
TOTAL	\$ 41,397	\$86,722	\$ 107,136

FINAL REPORT

WTRFC Project # CH-04-400

Project title: Environmental and genetic influences on cherry fruit size
PI: Amy Iezzoni
Organization: Michigan State University, Department of Horticulture
East Lansing, MI 48824, (517)355-5191, iezzoni@msu.edu
Cooperator(s): Matthew Whiting, WSU-IAREC,
Prosser, WA 99350, (509)786-2226, mdwhiting@wsu.edu
Anita Azarenko, Department of Horticulture, Oregon State University
Corvallis, OR 97331, (541)737-5457,
azarenka@science.oregonstate.edu
Jim Olmstead, Michigan State University, Department of Horticulture
East Lansing, MI 48824, (517)355-5191, olmste16@msu.edu
Contract Administrator: Ms. Lorri Busick, busick@msu.edu, (517)355-5191 x 1363

OBJECTIVES

Maximizing fruit size is critical for profitable sweet cherry production. For any given variety, (e.g., Bing), the grower's goal is to achieve the fruit's genetic potential for size by using "proper" management practices. New varieties with the genetic capacity to produce larger fruit, such as Selah, provide an additional means to achieve large fruit size.

Both environmental and genetic methods of fruit size increase have been studied in the past. Currently, great strides are being made in the understanding of the physiological "carrying-capacity" of cherry trees (i.e., optimal leaf to fruit ratios for desired fruit size). Cultural manipulations such as blossom thinning, pruning, irrigation, and fertilizer management are also important methods of achieving large cherry size. However, fruit size continues to be a concern, particularly with the adoption of dwarfing rootstocks and the potential for over-cropping. New varieties released in the past decade, in most cases, have been selected for large fruit size (among other selection criteria) and their fruits are considerably larger than those from standard varieties. Despite these advances, the basic genetic and environmental mechanisms that result in large vs. small fruit are not well understood, thus limiting our ability to maximize the number of consistently large fruit.

Our overall goal was to understand the bases for achieving large fruit size in sweet cherry. Our experiments are designed to provide knowledge that will be used to design future management and genetic improvement strategies that would ultimately result in maximized fruit size in grower orchards. Our objectives were based upon the premise that fruit size is maximized using both optimal cultural practices (environment) and large-fruited varieties (genotype).

The specific objectives of this research were to:

1. (a) Determine the effect of cultural practices, such as crop load manipulation, on cell size and cell number in Bing and Regina cherry. (b) Determine the developmental timing of these differences.
2. (a) Determine the differences in cell size and cell number associated with genetic differences in fruit size using three varieties that differ dramatically in fruit size. (b) Determine the developmental timing of these differences.
3. Determine the quantitative trait loci (QTL) that contribute to large fruit size. This is the first step towards the identification of the major genes controlling fruit size in sweet cherry.

SCHEDULE OF ACCOMPLISHMENTS

End of YEAR 1

- (1) Completion of the comparison of cell size and number from Bing and Regina fruit from crop load treatments.
- (2) Completion of a precise determination of the developmental stage(s) in which differences in cell size and cell number occur in Selah, Emperor Francis (EF), and New York 54 (NY54).
- (3) Initiated the construction of the sweet cherry linkage map.

During the first project year (2004), Bing and Regina fruit from trees adjusted for crop load were sampled from Prosser. Crop load adjustment resulted in significant fruit size differences (see Results and Discussion). Within each variety, the fruit size increase apparent with thinning was due to increases in cell size, not cell number. In contrast, a comparison of fruit from different varieties exhibiting a wide range of fruit sizes confirmed our previous observation that cell number differences are the primary genetic determinant of fruit size. The period between bloom and pit hardening was identified as the developmental period when cell number differences were first apparent (see Results and Discussion). Construction of a sweet cherry linkage map was initiated.

End of YEAR 2

- (1) Completion of a precise determination of the developmental stage(s) in which differences in cell size and cell number occur in Bing and Regina fruit of different sizes subjected to different crop loads.
- (2) Completion of the initial QTL analysis to identify regions containing gene(s) contributing to fruit size in sweet cherry.

During the final project year (2005), Bing, Regina, and Selah fruit with significant fruit size differences were harvested at maturity to confirm the previous year's finding that fruit size increase within genetically identical fruit was due to increases in cell size, not cell number. Additionally, fruit from those varieties were sampled during the developmental period identified in the previous year as most important for cell number increase, thus giving a comparison of both the rate and duration of cell division for varieties with very different final fruit sizes (see Results and Discussion). An initial sweet cherry linkage map was developed as a resource for quantitative trait loci (QTL) mapping of genomic regions important for fruit size in cherry.

SIGNIFICANT FINDINGS AND ACCOMPLISHMENTS

- When comparing different sweet cherry varieties, the most important determinant of final fruit size is the number of cells in the flesh.
- Cell number accumulation within a single variety is remarkably stable over both years and different environments.
- Within the same variety, larger fruit have the same number of cells as smaller fruit, but the cells are larger than those of smaller fruit.
- Differences in flesh cell number among different varieties are not apparent until after bloom.
- When comparing different sweet cherry varieties, both the duration and rate of flesh cell division differs.
- Cell division in the flesh does not continue past pit hardening.
- Fruit size QTL were successfully identified for overall fruit size increase but have yet to be found for cell number increase.

- The results from this two year project were used to successfully obtain a USDA-CSREES-NRI Award of \$400,000 over three years [P.I. Amy Iezzoni; Title: Genomic resources to improve fruit size and quality in cherry].

METHODS

Objective 1 (within genotype): (a) Determine the effect of cultural practices on cell size and cell number in Bing and Regina cherry. (b) Determine the developmental timing of these differences.

Plant material: Samples were collected from mature Bing and Regina trees at WSU-Prosser that were subjected to crop load adjustments and/or exhibited significant within-tree variation for fruit size.

Measurements: Prior to the preparation of tissue sections for microscopy, the quality of each individual fruit was evaluated (i.e., weight, diameter, firmness). Cell number and size was visualized by laser confocal microscopy, taking advantage of resources and equipment available at the Center for Advanced Microscopy at Michigan State University. Images created on the confocal microscope were analyzed using digital image processing software. Fruit sections were created according to protocols described previously (Fig. 1).

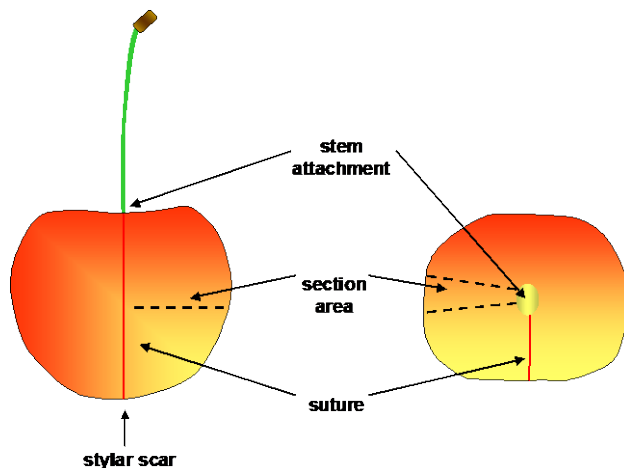


Figure 1. Slide sections were uniformly prepared from the thickest part of the fruit flesh. Radial sections were cut halfway between the point of stem attachment and the stylar scar. Sections consisted of all the flesh from the pit cavity to the epidermal layer.

Objective 2: (a) Determine the differences in cell size and cell number associated with genetic differences in fruit size using three varieties that differ dramatically in fruit size. (b) Determine the developmental timing of these differences.

Plant material: Fruit from three varieties, Selah (~ 12 g), EF (~ 6 g), and NY54 (~ 2 g), were evaluated for differences in fruit cell size and cell number over multiple years. EF and NY54 were used as parents to develop a population designed to identify genes contributing to fruit size by identifying genetic changes associated with domestication.

Measurements: Cell number and cell size measurements were conducted at MSU using the procedures described in Objective 1.

Objective 3: Determine the quantitative trait loci (QTL) that contribute to large fruit size.

Plant material: 200 progeny from reciprocal crosses made in 2001 between EF and NY54 were evaluated and used for development of a molecular marker linkage map. Over 700 additional

progeny from this cross are also planted in Michigan. The larger population will be necessary for future fine mapping and map-based gene cloning.

QTL Analysis: The 200 progeny were scored using “high-throughput” markers (AFLP and SRAP markers) and markers suitable for comparative mapping with other *Prunus* species (SSR markers). In 2005, the first fruit were available for QTL analysis from the fruit size population developed at Michigan State University. Both overall fruit size and cell number measurements were taken from all progeny fruiting in 2005. QTL analyses were performed using QTL Cartographer.

RESULTS AND DISCUSSION

In our comparison of fruit size between a large size cherry (Selah), a medium size cherry (EF) and a small-fruited mazzard cherry (NY54), the difference in final fruit size was primarily due to a difference in cell number and not cell size (Table 1). The nearly 11.5 gram difference in size between NY54 and Selah was due to a 74% increase in the number of flesh cells and only a 24% increase in the size of those cells. Clearly, cell number increase was the most important factor in the increased fruit size of Selah. The characteristic number of cells in the flesh of each variety also proved to be remarkably stable during the course of this experiment, indicating this trait is under strong genetic control (Table 2).

Table 1. Comparison of fruit anatomical and morphological characteristics among Selah, EF, and NY54.

Variety	Fruit ^z wt. (g)	Fruit dia. (mm)	Cell no. (pit to skin)	Avg. cell length (mm)
Selah	12.8 a ^y	26 a	83 a	0.148 b
EF	6.1 b	21 b	47 b	0.168 a
NY54	1.4 c	12 c	27 c	0.136 b

^zAvg. of 25 fruit for weight and diameter, 5 for cell size and number.

^yMean separation in columns by LSD at $P < 0.05$.

Table 2. Yearly comparison of cell number between Selah, EF, and NY54.

Year	NY54	EF	Selah
2003	27 a ^y	47 a	83 a
2004	29 a	41 b	79 a
2005	28 a	38 b	79 a

^yMean separation in columns by LSD at $P < 0.05$.

For the same three varieties, samples taken in 2004 at different fruit developmental stages indicated that differences in cell number approximating the final cell number count were evident by the onset of pit hardening. More importantly, all three varieties sampled at bloom had similar numbers of cells in the ovary wall, tissue destined to become flesh as fruit development proceeds (Table 3). Therefore, the large differences in final cell numbers exhibited in the three varieties happened exclusively during the Stage I fruit development period from bloom to pit hardening. Using this information, an additional set of samples for EF were collected in Michigan, starting at bloom and continuing every 20 growing degree days (base temperature 40 F) until pit hardening occurred. This set of samples further narrowed the time during which fruit cell number increased in EF to a period of 6-10 days after full bloom (Fig. 2). The relatively short duration of this cell division period was surprising, suggesting the basis for final fruit size was determined very early in the fruit developmental period. In 2005, more varieties were evaluated. Samples were collected daily from Selah, Bing, Regina, and NY54 trees at Prosser to determine whether the rate

and/or duration of cell division during this time period differed between varieties that had very different final fruit sizes.

Table 3. Comparison of cell number between Selah, EF, and NY54 at different fruit developmental stages.

Variety	Bloom	Pit harden	Harvest
Selah	24 b ^y	70 a	83 a
EF	17 a	40 b	47 b
NY54	25 b	26 c	27 c

^yMean separation in columns by LSD at $P < 0.05$.

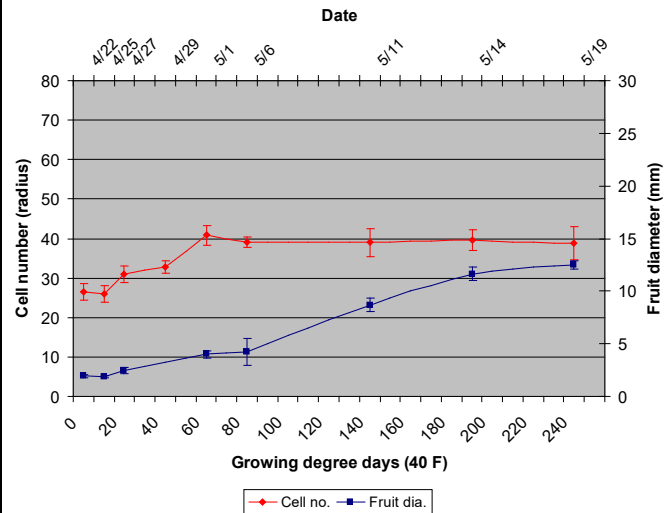


Figure 2. Cell number and fruit diameter increase in EF during Stage I fruit development (Michigan 2004).

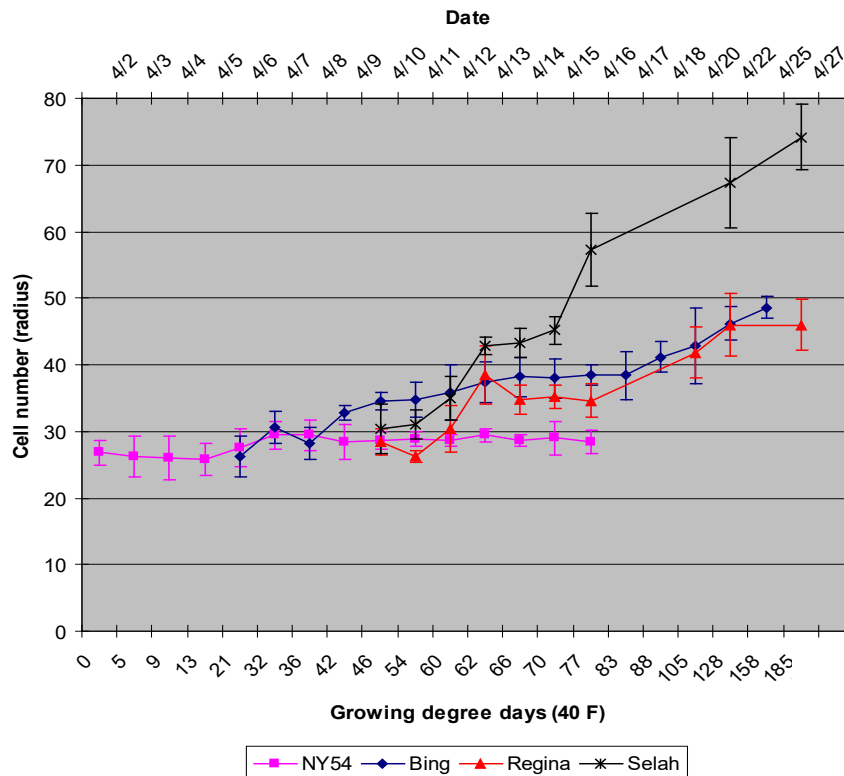


Figure 3. Comparison of cell number increase between NY54, Bing, Regina, and Selah during Stage I fruit development (Washington 2005). Sampling was discontinued when fruit reached cell numbers equivalent to harvest samples.

For these samples, both the duration and rate of cell division differed among varieties (Fig 3). An increase in the number of cells measured in the variety corresponded to an increase in the duration of cell division (Table 4). Selah, the variety with the largest overall fruit size also had the highest rate of cell division during this period.

Table 4. Duration and rate of cell division in NY54, Regina, Bing, and Selah during Stage I fruit development.

Variety	End cell no. pit to skin	GDD accum. when final cell no. reached	Cell division rate (no./GDD)
NY54	28	30	0.06
Regina	45	80	0.22
Bing	47	115	0.19
Selah	79	140	0.34

To better understand the environmental influences on fruit size, fruit of significantly different sizes from the same variety were sampled. Although the samples had a large variation in fruit size, they were genetically identical having been harvested from the same variety. These samples addressed Objective 1, and any cell number or size differences were due to environmental factors and not under genetic control. In 2004, Bing and Regina fruit were harvested from trees that had been thinned to 1 flower bud/spur prior to bloom. Fruit sizes on the thinned Bing trees were nearly two grams larger than the unthinned control at harvest. Thus, the use of crop load adjustment provided a method to generate differences in fruit size within a single variety. However, low crop load on both thinned and unthinned Regina trees resulted in no significant overall fruit size difference between the two treatments. Likewise, spring frost in 2005 prevented random sampling of thinned vs. unthinned treatments for Bing, Regina and Selah from generating significant fruit size differences. In both these cases, large lots of fruit were harvested, and individual fruit were weighed. Two pools of large and small fruit from each variety were created and analyzed. Although no specific treatments were applied to generate fruit size differences, selection of large and small fruit still fit the objective to test environmental differences between fruit from the same variety.

The pertinent question in this experiment was whether fruit size increase in the same variety was due to cell number increase (as indicated in the comparison of Selah, EF, and NY54), or cell size increase. Our results indicate that the large fruit size within a single variety was due to an increase in cell size (Table 5). Both large and small fruit had near identical flesh cell numbers, further illustrating the fact that cell number is under strong genetic control.

The final size of cherry fruit results from both an increase in the number of cells in the fruit and the expansion of those cells. Although expansion of the cell volume contributes greatly to overall fruit size, it is the total number of cells in the fruit flesh that sets the stage for eventual fruit expansion. Our results strongly suggest that the average cell number in the flesh is a genetically controlled trait. Variation in cell number was the most significant and consistent difference between a very small size (NY54) mazzard fruit and the very large sized Selah fruit. There are simply fewer cells available for expansion in NY54 than in Selah. Consistent with our hypothesis that cell number is a strongly genetically controlled trait, analysis of fruit from the

same variety revealed similar cell numbers in the flesh regardless of final fruit size. Because all trees of the same variety are genetically identical, the lack of variation for this trait indicates that variation in fruit size within a single tree is due to environmental influences on cell size.

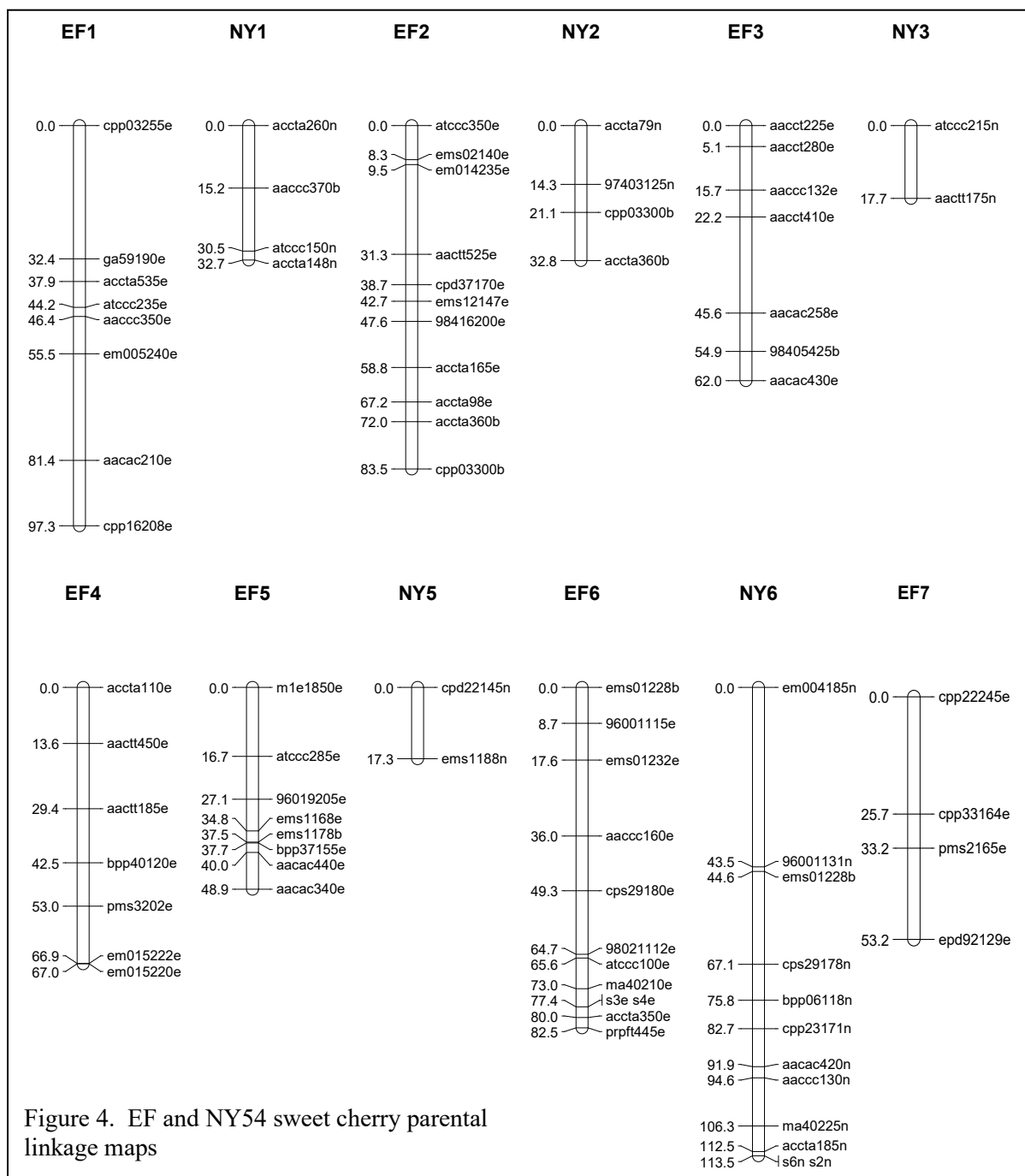
Table 5. Comparison of fruit size characteristics between small and large Bing, Regina, and Selah fruit.

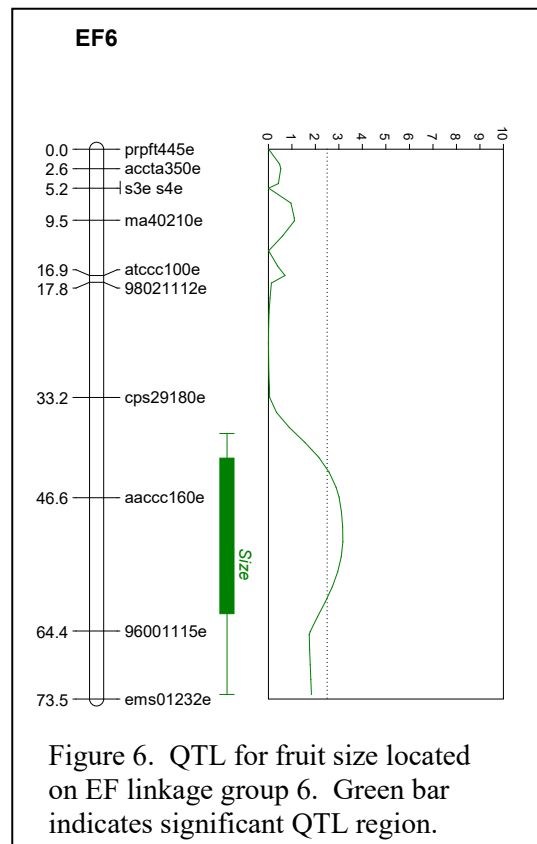
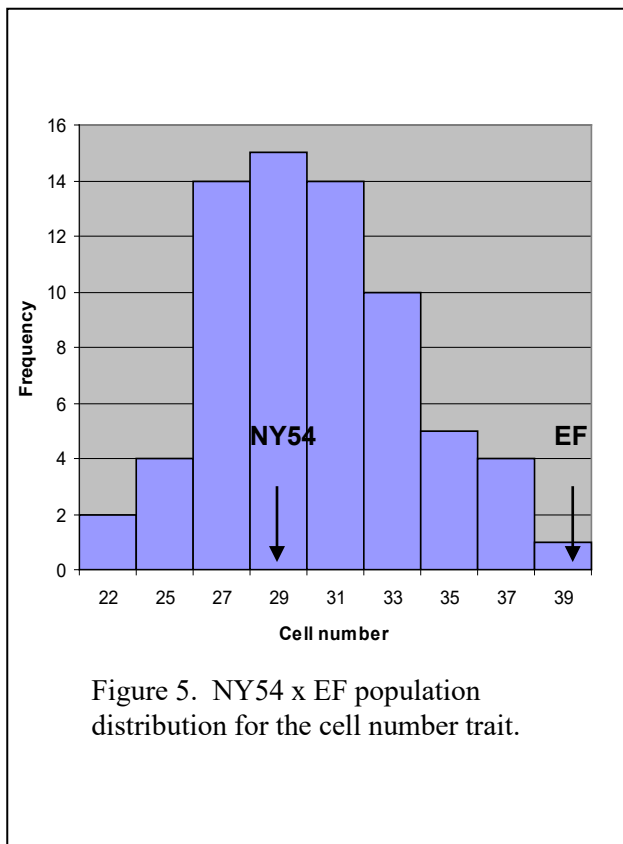
Variety	Treatment	2004			2005		
		Avg. ^z fruit wt. (g)	Cell no. (pit to skin)	Avg. cell length (mm)	Avg. ^z fruit wt. (g)	Cell no. (pit to skin)	Avg. cell length (mm)
Bing	High wt.	9.4***	49 ns	0.196*	11.3***	49 ns	0.208*
Bing	Low wt.	7.6***	48 ns	0.181*	7.5***	48 ns	0.185*
Regina	High wt.	10.3***	46 ns	0.214*	12.4***	47 ns	0.219*
Regina	Low wt.	7.7***	44 ns	0.195*	8.3***	47 ns	0.176*
Selah	High wt.				13.7***	79 ns	0.137 ns
Selah	Low wt.				8.8***	78 ns	0.125 ns

^zMean separation in columns by LSD; n.s. = non significant, *, **, *** = significance at $P < 0.05$, 0.01, 0.001 respectively

Due to low heterozygosity in the NY54 parent, a complete genetic linkage map is not yet available. Currently, 7 and 5 of the 8 total linkage groups for sweet cherry are available for QTL analysis for EF and NY54, respectively. The EF parental map comprises a total distance of 494.4 cM, while the NY54 map only measures 214 cM (Fig. 4). Linkage groups EF1, EF4, EF5, EF6, EF7, and NY5 can currently be aligned with the consensus *Prunus* linkage map available through the Genome Database for Rosaceae (<http://mainlab.clemson.edu/gdr/>). Although there are still several large gaps in both parental maps, these are anticipated to be filled with continued high-throughput AFLP marker development. Map development is ongoing, as the construction of a high density sweet cherry linkage map is one of the goals of the USDA-CSREES-NRI Award.

Fruit were available for 67 of the 200 progeny from the NY54 x EF mapping population. Because of the apparent stability and importance in overall fruit size, analysis efforts were concentrated on the cell number differences among progeny in the population. The population mean for this trait was 31 cells, significantly skewed toward the NY54 parent (Fig. 5). Although the stability of the cell number trait should significantly enhance our ability to locate corresponding QTL, none have yet been identified. This may be due to incomplete coverage of the current linkage map or low numbers of fruiting progeny. Although a QTL for the cell number trait was not found, one significant QTL explaining 23% of the phenotypic variation for fruit diameter was identified on linkage group 6 of the EF parent (Fig. 6). This QTL spans the region occupied by a marker from the AFLP primer combination EcoRI+AA/MseI+CCC.





CONCLUSIONS

The first major outcome of this work is a new understanding of how cell number and cell size contribute to overall fruit size in sweet cherry, and how these parameters are altered given different environmental and genetic influences. In summary, in the set of varieties we have tested, the flowers had approximately the same cell number at bloom. The primary cellular difference between varieties with significant final fruit sizes was an increase in the number of cells in the flesh. However, cell division, and thus final cell numbers was not affected by environmental differences. Surprisingly, cell division in the developing cherry fruit occurred very early in Stage I growth, with larger-fruited varieties undergoing cell division for a longer period of time. This detailed knowledge can be utilized by cherry physiologists to target cultural practices aimed at maximizing fruit size, and geneticists to dissect the genetic control of this important trait.

The second major outcome of this work was the initiation of linkage map construction and QTL analysis for fruit size. This information will be used to design molecular markers for the early identification of selections with large fruit size in the breeding program. Additionally, the data obtained in this project was used to write a proposal that resulted in a \$400,000 USDA-CSREES-NRI Award to not only continue this search for fruit size QTL, but broaden this search to include QTL for other fruit quality traits (sugars, acids, etc.). Therefore no continued funding is requested. Below is an abstract of the NRI Award that began June 1, 2005.

Genomic resources to improve fruit size and quality traits in cherry

Amy Iezzoni (Principal Investigator), Wayne Loesch, Esther van der Knaap*, & Dechun Wang
Michigan State University & *Ohio State University

Fruit size and quality (e.g. firmness, color, sugar etc.) are the most important market driven traits in cherry. For profitable cherry production, growers must achieve sufficient fruit size and quality standards. Unfortunately consistently maximizing fruit size and quality in grower orchards and in new varieties thorough breeding has been difficult because the genetic control of these traits is not well understood and the genetic tools to facilitate trait improvement do not exist. This USDA Award will result in the development of the genomics tools necessary to implement more efficient selection for fruit size and quality traits in breeding programs. Our analyses will also lead to a better understanding of the developmental timing and genetic control of these traits. This opens up the possibility of targeting cultural interventions to enhance or delay a desired fruit characteristic.

BUDGET

Title: Environmental and genetic influences on cherry fruit size

P.I.: Amy Iezzoni
Project Duration: 2 years (2004-2005)
Current Year: 2005
Project Total (2 years): \$50,442

Item	Year 1 (2004)	Year 2 (2005)
Salary ¹	\$16,344	16,998
Wages ²	500	500
User fee – confocal microscope ³	1,500	1,500
Supplies ⁴	4,000	4,000
Travel ⁵	550	550
Plot fees at CHES ⁶	1,000	1,000
Prosser costs ⁷	1,000	1,000
Total	\$24,894	\$25,548

¹Salary for Ph.D. student.

² Wages for student labor to help with sample preparation.

³ Fees for the use of the Confocal Laser Scanning Microscope at MSU's Center for Advanced Microscopy. The user fee is \$15/hr.[<http://www.ceo.msu.edu/Services.htm>]

⁴ Microscopy supplies are budgeted at \$200/yr. Cost of supplies for DNA extraction and marker genotyping is \$3,800/yr. This is based upon supply cost to genotype 190 progeny and the two parents using 25 AFLP primer pairs and 32 SSR primer pairs.

⁵Travel to Prosser at sweet cherry harvest to meet with Matt Whiting relative to the cultural experiments.

⁶Starting in 2004, plot fees are charged at all MSU Horticultural Research Stations. These costs are based upon a fee structure that reflects the cost of standard plot maintenance.

⁷These funds are for to Matt Whiting to cover the cost sampling and overnight shipping.

CONTINUING PROJECT PROPOSAL

YEAR 1/3

Project Title: Sweet cherry dwarfing rootstocks
PI: Amy Iezzoni
Organization: Department of Horticulture, Michigan State University
Co-PI: Matt Whiting (WSU-Prosser)
Contract Administrator: Ms. Lorri Busick, Dept. of Horticulture
MSU, (517) 355-5191, busick@msu.edu

Objective: Identify MSU rootstock selections that may have commercial potential as dwarfing precocious rootstocks for sweet cherry.

Specific Objectives:

1. Complete the planting of the rootstock candidates at the WSU-Prosser and MSU-Clarksville test sites.
2. Identify the most promising rootstock candidates by evaluating tree health, precocity, trunk cross-sectional area, flower density, crop load, fruiting habit and fruit size.
3. Vegetatively propagate the most promising rootstock selections to provide grafted trees for advanced trials at multiple test locations.

Significant findings:

- Plot establishment was completed. The number of MSU rootstock candidates at MSU-Clarksville and WSU-Prosser are 71 and 41, respectively.
- Flower bud and yield data were used to identify promising rootstock candidates. Thirty-five rootstock selections were chosen for advanced testing.
- The number of flower buds on two- and three-year-old wood was determined to be the limiting yield component.

Methods by Objective:

1. *Plot maintenance:* The trees have been and will continue to be managed with minimal pruning and training to permit us to observe any potential rootstock influence on growth habit.

2. *Data collection:* The trees in the MSU and WSU test plots will be evaluated for tree health, structure, survival and trunk cross-sectional area. Reproductive traits to be evaluated include visual estimates of bloom density and crop load based on a scale of 0 to 5. Those selections with a crop load estimate of 2 or higher will be further evaluated to provide a more complete estimate of yield potential. Yield potential/components and fruiting habit of promising rootstock selections will be determined by counting the number of spurs along two- and three-year-old fruiting wood. Two branches will be evaluated from each tree.

3. *Criteria for the identification of superior rootstock selections:* The following criteria will be used: (1) absence of symptoms of graft incompatibility, (2) general tree health (this avoids trees

weakened by over-cropping), (3) mean flower density rating of 2 to 3, (4) acceptable levels of flowering spurs on two- and three-year-old wood, and (5) fruit size equal or larger than that produced on GI 6.

4. *Propagation of advanced selections:* Those rootstock selections that look the most promising will be propagated from softwood cuttings using the following procedures. The mother trees of the selected rootstock candidates will be tested to determine whether they have remained free of PDV and PNRSV. During the week of June 15, softwood cuttings will be taken from the mother trees of the rootstock selections that are planted in the Mother Block at Clarksville. The cuttings will be rooted in 5 inch deep Groove Tube Trays in a 30% peat: 70% perlite mix and placed under mist for 8 weeks. Dying cuttings will be rogued out to prevent spread of saprophytic diseases. Our goal is to obtain a final number of 25 trees per rootstock selection. This will provide enough trees for 5 tree replications at MSU-Clarksville and WSU-Prosser, plus three new locations: The Dalles, OR and the mid-elevation and high-elevation sweet cherry growing regions in WA.

5. *Growing the rootstock liners:* During spring/summer 2005 we conducted a pilot trial to determine the best procedure for producing robust liners. Currently the cuttings rooted in summer 2005 are in the greenhouse, so we can assess the ability of the cuttings to break terminal bud and achieve significant growth in year 1. The outcome of this experiment will determine the strategy/location we use for budding.

6. *Rootstock controls:* Arrange for the purchase of control trees of ‘Bing/GI 6’, ‘Bing/Maxma14’, and ‘Bing/mazzard’

Results and Discussion

Plot establishment: The rootstock evaluation plot at MSU-Clarksville has 30 more rootstock selections under test than the WSU-Prosser plot. This is because we were able to successfully add more trees to the MSU-Clarksville plot in the last year. These trees were grown at Hilltop Nurseries, MI and we personally participated in plant care.

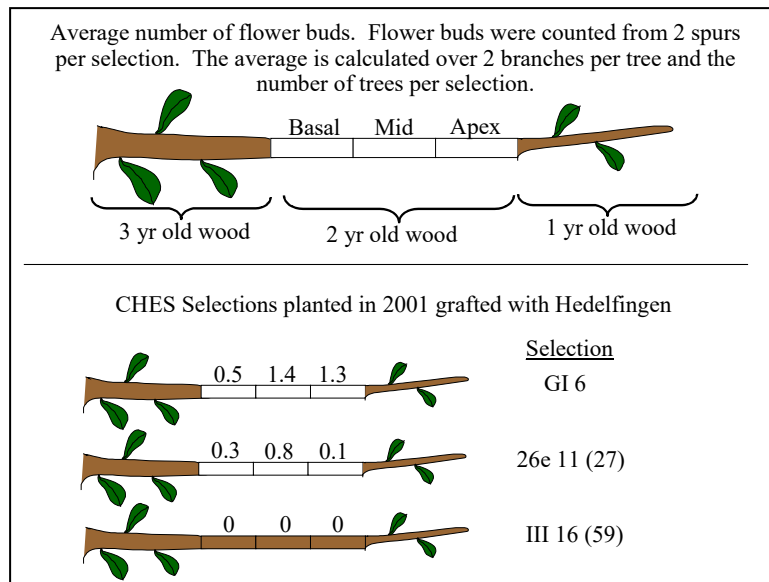
Evaluation: Most of the MSU rootstock selections planted at MSU-Clarksville had mean flower density ratings less than GI 6 (Table 1). None of the rootstocks selections were rated at 4 or higher, ratings indicative of over-cropping. The limiting factor in flower bud density was the number of fruiting spurs, not the number of flower buds per spur.

Table 1. Number of selections grafted with Hedelfingen that received ratings of 0 to 5, based on spur number and flower buds per spur on two-year-old wood. The trees were planted over three years, 2001 to 2003. GI 6 is indicated with an *.

Year\Rating	0 - 0.9	1- 1.9	2 – 2.9	3 – 3.9	4 – 4.9	5
2001	-	14	2	2*	-	-
2002	2	18*	-	1	-	-
2003	8*	-	1	-	-	-

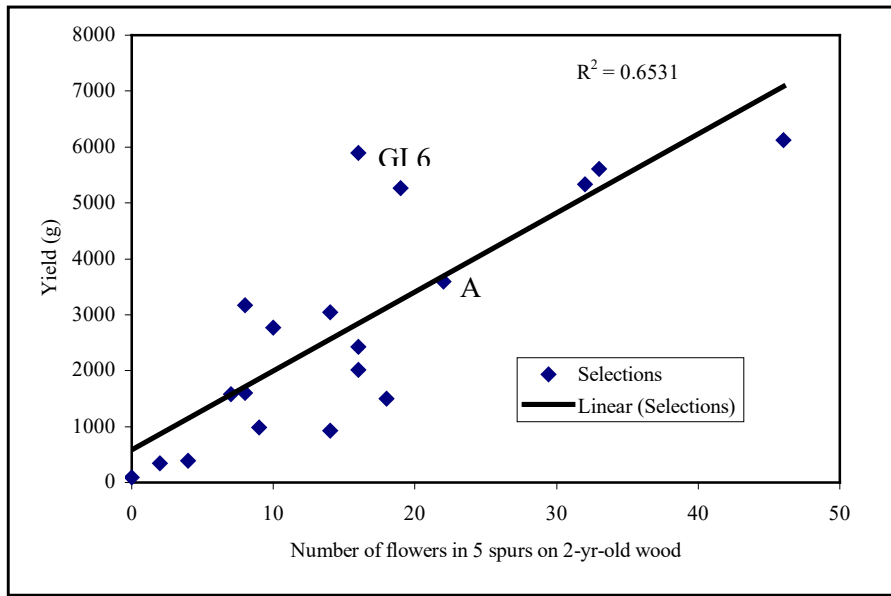
A graphical diagram was used to represent the fruiting capacity of scions grafted onto GI 6 and the rootstock candidates (Fig. 1). This diagram divides the fruiting wood into three sections. Those sections with no fruiting spurs received a value of zero.

Figure 1. Graphical representation of the number of flower buds on two-year-old wood.



The scion data for the MSU rootstocks planted at WSU-Prosser was represented as yield plotted against the number of flowers in 5 spurs (Fig. 2). This statistical separation clearly identifies those selections that confer poor scion productivity. However, the identification of the promising rootstocks is not as straightforward. For example, all of the MSU rootstock selections that resulted in scion yields over 5000 g had mean fruit weights less than the 8.1 g for GI 6 (fruit weight ranged from 6 g to 7.5 g). This suggests that these trees were over-cropped or under an alternate stress. By comparison, rootstock selection “A” resulted in a mean scion fruit size of 9.7 g compared to 8.1 g for fruit from GI 6. Tree yields were 3593 g (~ 8 lbs) and 5892 g (~ 13 lbs), respectively.

Figure 2. Regression analysis of yield (g) versus the number of flowers in 5 spurs on two-year-old wood for Bing grafted onto 18 MSU rootstock selections. The trees were planted in 2002 at WSU-Prosser.



Selections for propagation in 2006: Using the selection criteria outlined in the Materials and Methods and the data obtained, 14 selections were identified for propagation for second testing. Additional data collected in Spring 2006 will be used to confirm or adjust this list. In 2004 and 2005, an additional 21 selections were planted. Spring 2006 will be too early to evaluate flowering and fruiting capacity conferred by these rootstocks. Therefore all of these rootstocks will be re-propagated in 2006. Then, those that do not show promise will be eliminated during the tree budding and plot planting process in future years.

Pilot study: In anticipation of establishing the test sites, we did a pilot study in 2005 to determine if we could grow more robust one-year-old liners (Fig. 3). This requires the liner to break terminal bud and continue growing throughout the summer (Fig. 4).

Over 20 selections were vegetatively propagated in June using standard procedures. The rootstocks were repotted into larger pots in September and placed in a warm greenhouse. Currently this experiment is in progress. Our success or failure in inducing terminal bud break will determine our strategy for tree propagation. The timeline listed below is based on a conservative strategy whereby we are unable to get bud break in August.

Figure 3: Misty room where cuttings were rooted in the pilot study.



Figure 4. (A) Rooted cutting with terminal bud set. (B) Rooted cutting where the terminal bud has started to grow.

A.



B.



Timeline for establishing the Second Test Plots

2006

- Propagate from softwood cuttings the most promising selections for advanced testing.

2007

- Grow the rootstock liners that were produced in 2006.
- In August, chip bud the most promising selections for advanced testing.

2008

- Grow the grafted trees.
- Dig the trees.

2009

- Spring planting at the Test Sites.

Budget

Title: Sweet cherry dwarfing rootstocks
P.I.: Amy Iezzoni
Project duration: 3 years
Current year: 2006
Project total (3 years): \$48,601
Current year request: \$20,718

Year	Year 1 (2005)	Year 2 (2006)	Year 3 (2007)
Total	\$13,408	\$20,718	\$13,935

Budget breakdown:

ITEM	Year 1 (2005)	Year 2 (2006)	Year 3 (2007)
Salaries ¹	\$4,725	\$4,867	\$5,013
Benefits ²	2,183	2,351	2,522
Labor (MSU) ³	1,500	3,000	1,500
Labor (WSU) ⁴	2,000	2,000	2,000
Supplies ⁵	400	1,500	400
Travel ⁶	1,500	2,500	1,500
Tree and freight costs ⁷	100	500	0
Plot costs at MSU ⁸	1,000	1,000	1,000
Greenhouse costs ⁹	0	3,000	0
TOTAL	\$13,408	\$20,718	\$13,935

¹ This represents partial funding (1/8) for technical support to oversee the technical aspects of this project (develop spreadsheets, collect data, and manage, analyze, and summarize the data from the 2 field plots). A yearly salary increase of 3% is included.

² Benefits for YRs 2005, 2006, and 2007 are calculated at 46.2 %, 48.3%, and 50.3%, respectively.

³ Student labor will assist with planting and data collection. The increased labor cost in YR 2 reflects the increased labor needed for virus indexing and vegetative propagation.

⁴ Student labor to assist with data collection in the WSU-Prosser test plot.

⁵ Supplies to include mouse guards, tags and other field supplies. In YR 2, the additional cost is due to the purchase of ELISA kits and propagation supplies (trays, rooting hormone, etc.).

⁶ Travel to WSU for plot evaluation. In YR 2, this includes funds for additional travel to Grand Rapids (2 to 3 times/week) to transport and check on the cuttings.

⁷ The YR 1 request is solely the shipping cost of the trees to be planted at Prosser, as the trees are being donated by Meadow Lake Nurseries. The YR 2 cost is for transport of the cuttings to the propagation nursery. Future budgets will reflect the cost of the trees propagated for the test sites.

⁸ Plot fees are required at all MSU Horticultural Research Stations. These costs are based upon a fee structure that reflects the cost of standard plot maintenance.

⁹ Costs for rental of greenhouse space and grower services at a commercial greenhouse.

CONTINUING PROJECT REPORT
PROJECT NO.: CH-04-412

YEAR 2/3
WSU Project No.: 3355-6202

Project title: Clonal rootstock performance/evaluations

Principal Investigator: Matthew Whiting
Organization: Irrigated Agriculture Research and Extension Center, WSU-Prosser
Address: 24106 N. Bunn Road, Prosser, WA 99350
Phone: (509) 786-9260
E-mail: mdwhiting@wsu.edu

Cooperators: W.E. Howell, NRSP5/IR2 Manager, WSU-Prosser
D.R. Ophardt, Res. Tech. Supervisor, WSU-Prosser

Contract Administrator: Mary Lou Bricker, mdesros@wsu.edu, 509-335-7667 or
Stephanie Brock, sabrook@wsu.edu, 509-786-9224

YR INITIATED: 2004

CURRENT YR: 2005

TERMINATING YR: 2006

OBJECTIVES:

1. Continue evaluation of the NC-140 regional project trial ('Bing' on 17 new rootstocks) established in 1998 for horticultural and physiological evaluations and fruit quality. Projected trial duration is 10 years.
2. Continue evaluating vigor and cropping performance of other orchard trials with key PNW cultivars on various rootstocks
3. Analyze the physiology of interactive rootstock/scion horticultural traits (e.g., canopy leaf area, yield efficiency, precocity, graft compatibility).
4. Establish planting of 2005 NC-140 sweet cherry rootstock trial.

SIGNIFICANT FINDINGS:

- high quality fruit can be grown on precocious, dwarfing rootstocks
- rootstock affected scion vigor, yield, and fruit quality
- rootstock altered 'Bing' fruit maturity (ca. 8 days) and bloom timing (4 days) significantly
- fruit yield was unrelated to tree vigor
- fruit maturity was unrelated to tree vigor
- tree vigor was related negatively to bloom date (i.e., smaller trees bloom earlier than large trees)
- the Gisela series is very precocious/productive
- fruit quality across all rootstocks was excellent (9.6 g – 10.7 g per fruit)
- Mazzard had the third lowest yield (9.4 kg, 20.7 lb/tree)
- the worst quality fruit was harvested from Gisela 209/1 and Edabriz
- no rootstock out-performs Gisela 5 or Gisela 6 in the vigorous – semi-dwarfing categories

- PiKu 1 is less vigorous and more precocious than PiKu 3
- novel crop load management strategies will need to be developed to grow high quality sweet cherries on precocious and dwarfing rootstocks

METHODS:

The 1998 NC-140 plot was planted at WSU-Prosser's Roza Experimental Unit, with 'Bing' as the scion cultivar and 'Van' as the pollenizer, on the German rootstock series Gisela 4 (GI 473/10), Gisela 5 (GI 148/2), Gisela 6 (GI 148/1), Gisela 7 (GI 148/8), GI 195/20, GI 209/1, and GI 318/17; the German rootstock series Weiroot 10, W13, W53, W72, W154, and W158; Edabriz (France); P-50 (Japan); and Mazzard and Mahaleb seedlings as controls. There are 8 replications/rootstock, with guard tree around the plot perimeter, and tree spacing of 19.5 x 19.5 ft (6.0 x 6.0 m) to reduce the potential influence of neighboring trees. Irrigation by microsprinklers and frost protection by wind machine were installed. A duplicate plot was planted for potentially destructive analyses, such as physiological stress treatments. The effects of rootstock on tree yield, vigor, fruit quality, first and full bloom dates, fruit maturity, and senescence and cold acclimation will be documented annually.

A research orchard was planted in 1998 with WSU-Prosser varieties (including Chelan, Cashmere, Benton, Selah, Rainier and Tieton) and elite selections (including 8011-3, 7147-9, and 7903-2) on several Gisela rootstocks (including Gisela 5, 6, 195/20, and 209/1), Mazzard, Mahaleb, and Colt. In this block, tree vigor, fruit yield and quality, and graft compatibility will be monitored. Several of these newly released cultivars (*e.g.*, Chelan, Tieton, Benton, Selah) and advanced selections (*e.g.*, PC 8011-3, PC 7903-2, PC 7147-9) will be subjected to one of two crop load treatments: (1) unthinned control, and (2) 50% removal of blossoms by hand. Tree growth, fruit yield and quality (weight, row-size distribution, soluble solids, and firmness) will be evaluated for each scion grown on Gisela 6, Gisela 5, Gisela 195/20, and Edabriz, where possible.

Another orchard, planted in 2001, will be utilized to evaluate the effects of two new rootstocks (PiKu 1 and PiKu 3) on growth, precocity, fruit quality, and graft compatibility of Celeste, Benton, Selah, Tieton, Regina, Bing, Skeena, Sweetheart, Attika, Rainier, Lapins, Chelan, Summit, Black Gold, White Gold, Glacier, and Sonata.

In a separate trial in cooperation with Amy Iezzoni of MSU, we have planted 21 MSU rootstock selections, totaling 117 trees, in a test plot at the Roza farm. The control rootstock is GI 6 and the scion is Bing with Tieton/GI6 as the pollinator. An additional 243 trees (84 selections) will be planted in 2004. The effects of rootstock genotype on scion growth habit, precocity, and fruit quality will be documented annually.

RESULTS AND DISCUSSION:

1998 NC-140 trial 2005 was the fifth fruiting year (7th leaf) for most of the rootstocks in this trial and we recorded tremendous variability in fruit yield per tree (3.7 – 35.2 kg/tree) (Table 1). Clearly, rootstock has a significant effect on 'Bing' precocity and productivity (Fig. 1). Fourteen of the 17 rootstocks in this trial have improved productivity compared to the industry standard, Mazzard. In this 7th season, many trees have reached full production; most notably so are those in the Gisela series (*e.g.*, Gi 7, Gi 5, Gi 6, Gi 195/20). In 2005, mean yield was 21.0 kg/tree, down about 10% from the previous year. The most productive rootstocks were from the Gisela series: Gi 6 and Gi 7, and Gi 195/20 and W 10 were similar. Each of these rootstocks yielded over 60 lbs of fruit per tree. W 10 and Gi 6 are vigorous rootstocks whereas Gi 7 and Gi 195/20 are semi-vigorous. The least productive rootstocks were unchanged from 2004: W53, P-50, Mazzard, and W154, in order of increasing yield. Trees on each of these rootstocks yielded less than 12 kg (25

lbs) per tree. For W53, by far the most size-controlling of the rootstocks, low yields were due to limited canopy size and therefore inadequate bearing surface. We conclude that, even at a high tree density, this rootstock is too dwarfing to produce commercially acceptable yields. However, for P-50 and Mazzard, low yields may be attributed to excessive vigor and poor floral bud induction. The lack of yield from Mazzard-rooted trees remains a significant concern and its greatest drawback.

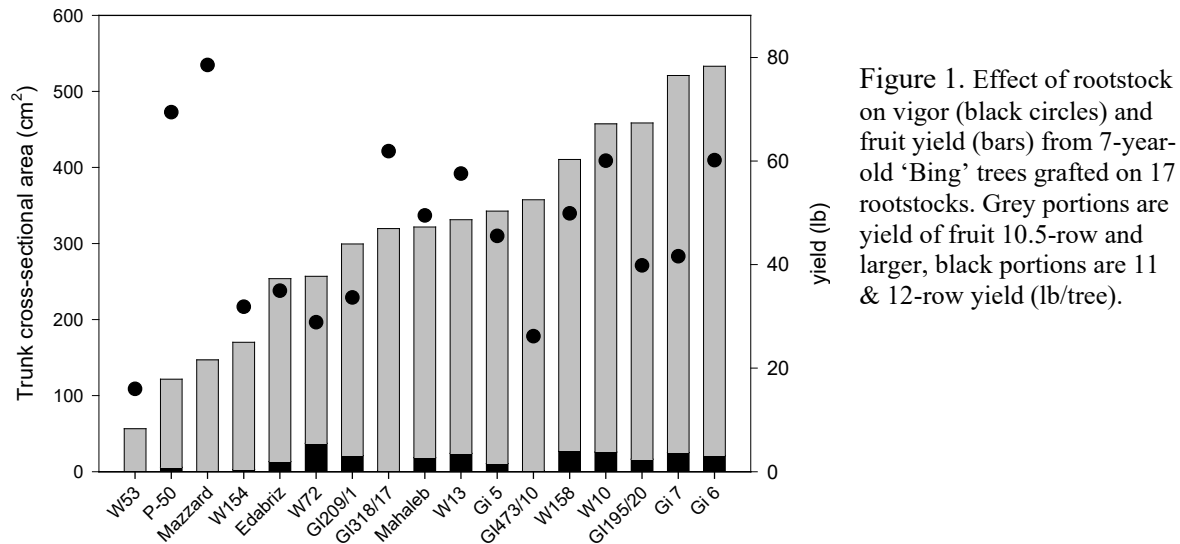


Figure 1. Effect of rootstock on vigor (black circles) and fruit yield (bars) from 7-year-old 'Bing' trees grafted on 17 rootstocks. Grey portions are yield of fruit 10.5-row and larger, black portions are 11 & 12-row yield (lb/tree).

As reported previously, yield and precocity are unrelated to vigor (Fig. 2). By defining high yield as over 20 kg/tree and high vigor as greater than 300 cm² trunk cross-sectional area (TCSA), we can compare and classify rootstocks. Those which possess moderate – high yield and low vigor (i.e., high yield efficiency) may be desirable and appropriate for high density, more efficient plantings. By our analyses, only Gi7, Gi195/20, Gi209/1, and Gi473/10 qualify by falling into the lower right quadrant of Figure 2. In general, most of these rootstocks have also produced lower quality fruit, though in 2005 we harvested excellent quality fruit from trees on these rootstocks. Normally we document a negative relationship between yield efficiency (kg/cm² TCSA) and fruit quality. In 2005 however, there was no significant negative relationship between yield efficiency and fruit quality despite there being a ca. 10-fold difference in yield. Rootstocks in the upper left portion of Fig. 2 are those characterized by low productivity and high vigor. These rootstocks (P-50 and Mazzard) will not provide growers the early returns on investment or size controlling necessary to improve labor efficiency. They are not recommended for 'Bing'.

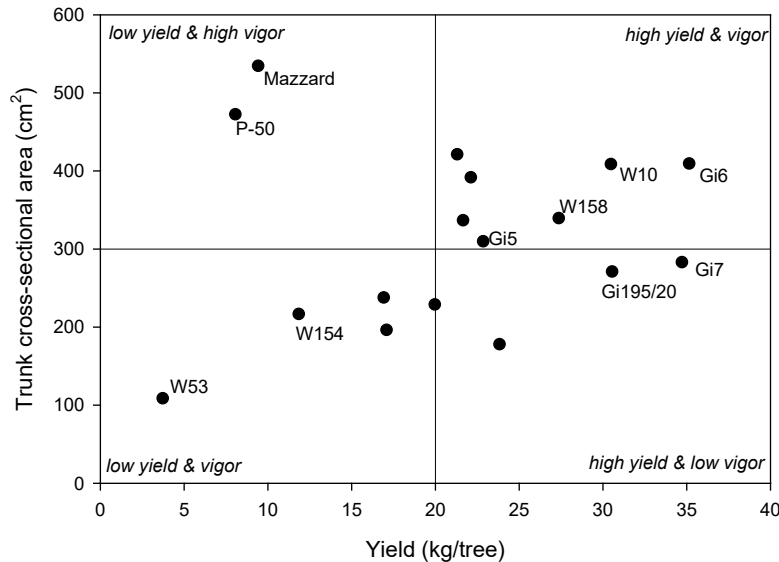


Figure 2. Relationship between yield and vigor (trunk cross-sectional area) of 7-year-old 'Bing' sweet cherry trees grown on 17 rootstocks.

Vigor varied among rootstocks by approximately 5-fold (Figs. 1 & 2). Mazzard is the most vigorous rootstock, W 53 is the least vigorous. Gi 5 and Gi 6 are ca. 58% and 76% of Mazzard, respectively. P-50, Gi 318/17, Gi 6, W10, and W13 are all vigorous (ca. 75%+ of Mazzard). W 158, Mahaleb, Gi 5, Gi 7, and Gi 195/20 were semi-dwarfing (ca. 50 – 65% of Mazzard). Edabriz, Gi 209/1, W 154, W 72, Gi 473/10, and W 53 are dwarfing rootstocks, reducing TCSA to less than 45% of Mazzard. The most promising vigorous rootstock is Gisela 6. It yielded fruit that was 97% 10.5-row and larger (75 lb/tree), 10.5 g, and 258 g/mm. This translates into about 13.5 tons/acre of 10.5-row and larger fruit at 363 trees/acre (8' x 15'). In contrast, Mazzard-rooted trees would yield less than 3 tons/acre of similar quality fruit at 272 trees/acre (10' x 16'), or 3.9 tons/acre at a similar tree density.

Overall, in 2005 fruit quality was outstanding on every rootstock— no rootstock yielded any fruit that was smaller than 12-row, fruit weight ranged from a low of 9.6 g to 10.7 g, soluble solids was 23% on average, and fruit firmness averaged 290 g/mm. Moreover, only one rootstock (W53) yielded *less* than 90% 10.5-row and larger fruit. There was no relationship between yield or yield efficiency and fruit weight or soluble solids but firmness was related negatively to yield efficiency (data not shown). Rootstock is not reported to have a significant, direct effect on fruit firmness. This relationship is likely related to the rootstock's effect on harvest date/fruit maturity. In 2005, we documented an 8-day variation in harvest date based on fruit skin color (data not shown). Bing on Gi 5, Gi 6, and W158 reached optimum harvest maturity on 22 June while fruit on Mazzard did not reach similar maturity until 1 July. Average harvest date for all rootstocks was 24 June. It is not known whether similar discrepancies in harvest maturity would exist for other earlier or later-maturing varieties. However, this result highlights the need to consider rootstock when planning new orchards for a particular harvest season.

Rootstock also affected the date of first and full bloom (data not shown). There was about a four-day (ca. 50 GDD @ base-40) difference between first bloom in Edabriz, Gi 5, W158, and Mahaleb (earliest-blooming, at ca. 680 – 690 GDD) and Mazzard (latest blooming, at 740 GDD). Mazzard-rooted trees were also the last to achieve full bloom, again about 4 days (ca. 70 GDD) later than Gi 5 and Edabriz. At our orchard site, we have not noticed any relationship between first and full bloom dates and frost damage to flowers but in other sites, this may be a concern. We did document in 2005 a significant positive linear relationship ($r^2 = 0.71$) between tree vigor and bloom date (i.e., the more vigorous the tree, the later the bloom).

PiKu trial In 2001 we planted an orchard of 16 scion varieties on both PiKu 1 and PiKu 3 rootstocks. 2005 was this orchard's second year of fruiting for most varieties. On PiKu 3, Bing and Selah were the least productive, though, due to poor fruit set overall, yields were low throughout the orchard (Table 2). BlackGold/PiKu1 was the most productive combination, yielding only 2.8 kg (6 lbs) per tree. Several varieties yielded no fruit on PiKu 3 (data not shown). Again in 2005, PiKu 1 was significantly more precocious, out-yielding trees on PiKu 3 by over 7-fold, though this difference was only ca. 2 lbs/tree. In addition, PiKu 1 remains about 40% less vigorous than PiKu 3. Across all varieties, there were significant, albeit subtle, differences in fruit quality between PiKu 1 and PiKu 3 in 2005. Fruit firmness and soluble solids were about 6% higher on PiKu 1. Tree mortality was similar for both rootstocks – we have documented ca. 10% tree loss. Particularly poor combinations appear to be Attika/PiKu 3 (75% tree death), Glacier on both PiKus (50% tree death), and Lapins/PiKu 1 (50% tree death).

Fruit quality among scion varieties varied considerably in this second year of production (data not shown). Briefly, Summit, Tieton, Attika, Black Gold, and White Gold were among the largest fruit (ca. 10.5 g+ and > 90% 10.5-row and larger) and Sonata, Chelan, and Sweetheart were the smallest (< 9 g/fruit). Sweetheart and Black Gold were the most precocious cultivars, yielding about 2 kg (4.5 lbs) per tree.

Table 2. Effect of rootstock (PiKu 1 and 3) on yield and fruit quality of 3-year-old sweet cherry trees. Data are means of 16 scion varieties. Means followed by the same letters within a column are not significantly different (P>0.05).

Rootstock	Date	Weight (g)	°Brix	Firmness (g/mm)	%<12-row	% 11 & 12-row	% ≥10.5- row	Yield(g)	TCSA(cm ²)
2004									
PiKu 1	6/19	8.0a	22.9a	274b	6a	31a	63a	974a	22.7b
PiKu 3	6/18	8.1a	21.1b	296a	6a	25b	66a	406b	36.9a
LSD		0.4	0.9	19	5	6	8	145	3.6
2005									
PiKu 1	6/26	10.1a	23.8a	347a	1a	8a	91a	1133a	42.2b
PiKu 3	6/28	10.1a	22.4b	327b	1a	11a	88a	152b	72.0a
LSD		0.4	1.1	16	1.7	4	5	245	5.6

Project: Clonal rootstock performance/evaluations
P.I.: Whiting
Project duration: 2004-2006
Current year: 2005
Project total: \$62,523
Current year request: \$21,594

Year	2004	2005	2006
Total	\$19,895	\$21,034	\$21,594

Current year breakdown

Item			
Salaries ¹	6,199	6,301	6,503
Benefits (34% yr 3) ²	1,736	1,953	2,211
Wages ³	6,000	8,000	8,000
Benefits (11% yr 3) ²	960	1,280	880
Equipment ⁴	1,500		
Supplies ⁵	2,000	2,000	2,000
Travel ⁶	1,500	1,500	2,000
Miscellaneous			
Total	\$19,895	\$21,034	\$21,594

¹ One-sixth annual salary for Mr. Efrain Quiroz (Roza orchard manager).

² Benefit rate for year 1 is 28%, 31% for year 2, and 34% for year 3. This increase is due to increase in contribution made by WSU on the behalf of the employee. Time-slip benefit rate for years 1 and 2 is 16%. Year 3 benefit rate is calculated at 11%. The change is due to a change in policy at WSU.

³ Time-slip assistance for harvest, data collection, and fruit quality analyses

⁴ To purchase new computer for fruit quality lab

⁵ Supplies for field work and laboratory analyses

⁶ Travel to plots and vehicle maintenance (@ \$0.485/mile).

Table 1. Effect of rootstock on ‘Bing’ fruit yield, quality, tree size, and yield efficiency. Data within a column followed by different letters are significantly different by LSD ($P < 0.05$).

Rootstock	Yield (kg/tree)	Weight (g)	Firmness (g/mm)	11&12- row (kg)	≥10.5- row (kg)	TCSA (cm ²)	Yield efficiency (kg/cm ²)							
W53	3.7	f	10.7	a	347.1	a	0.0	b	3.8	i	108.7	k	0.037	ghi
P-50	8.1	f	9.8	b	332.2	ab	0.4	b	7.7	hi	472.5	ab	0.017	i
Mazzard	9.4	ef	10.6	ab	315.1	abc	0.0	b	9.8	ghi	534.7	a	0.017	hi
W154	11.8	ef	10.3	ab	313.1	abc	0.2	b	11.2	ghi	216.9	ij	0.055	efgh
Edabriz	16.9	de	10.2	ab	279.9	cde	0.9	ab	16.0	efg	237.9	ghi	0.076	def
W72	17.1	de	10.2	ab	270.3	cde	2.5	a	14.6	fgh	196.3	ij	0.090	bcde
GI209/1	20.0	cde	9.8	b	297.3	bcd	1.4	ab	18.5	defg	228.9	hij	0.093	bcd
GI318/17	21.3	cd	10.7	a	274.8	cde	0.1	b	21.2	cdef	421.2	bc	0.053	fgh
Mahaleb	21.7	cd	10.3	ab	268.4	cde	1.2	ab	20.2	def	336.8	de	0.063	defgh
W13	22.1	bcd	9.9	ab	293.9	bcd	1.6	ab	20.5	cdef	391.7	cd	0.056	efgh
Gi 5	22.9	bcd	10.2	ab	301.0	bc	0.7	ab	22.1	bcdef	309.9	ef	0.077	def
GI473/10	23.8	bcd	9.6	b	294.4	bcd	0.1	b	23.7	bcde	178.1	j	0.136	a
W158	27.4	abc	9.8	b	271.5	cde	1.8	ab	25.6	abcd	339.5	de	0.080	cdef
W10	30.5	ab	10.4	ab	277.9	cde	1.7	ab	28.8	abc	408.8	c	0.073	defg
GI195/20	30.6	ab	10.2	ab	261.4	cde	1.1	ab	29.5	ab	271.1	fgh	0.114	abc
Gi 7	34.7	a	10.3	ab	249.7	e	1.7	ab	33.0	a	283.2	efg	0.123	ab
Gi 6	35.2	a	10.5	ab	257.5	de	1.4	ab	34.1	a	409.5	c	0.086	cdef
lsd	9.1		0.8		43.6		2.0		8.7		58.2		0.036	

CONTINUING PROJECT REPORT

ONGOING PROJECT

TITLE: Cultivars, rootstocks, training systems and fruit quality

PI: Roberto Núñez-Elisea

Organization: OSU, Mid-Columbia Agricultural Research and Ext. Center, Hood River, OR

E-mail: roberto.nunez-elisea@oregonstate.edu

Co-investigators: Anita Azarenko (OSU Horticulture Dept., Corvallis, OR)
Lynn Long (OSU Extension Service, The Dalles, OR)
Mathew Whiting (WSU-IAREC, Prosser, WA)

Cooperators: Jinhe Bai, Xinhua Yin, Lucia Domingo, Pam S. Manning

Contract Administrator: Dorothy Beaton; dorothy.beaton@oregonstate.edu; 541-737-4068

Objectives

- Evaluate new cultivars, rootstocks and training systems for tree and fruit characteristics
- Generate fruit and shoot growth curves to help predict harvest and final fruit size and aid in precision tree management (crop load, irrigation and nutrient management)
- Evaluate fruit quality of promising cultivars and selections
- Develop crop load management strategies to produce large fruit of high quality
- Develop canopy management techniques to control tree size and improve crop quality
- Develop more efficient orchard designs (pollinizer distribution, tree arrangement)

Significant findings for 2005

- *Cultivars and rootstocks.* Cultivars that are showing promise include ‘0900 Ziraat’, ‘Attika’, ‘Early Robin’ and ‘Regina’. Gisela 3 (209/1) continues to show potential for ultra-high density plantings.
- *Fruit growth measurements.* Continuous, automated measurement of ‘Regina’ fruit growth shows potential as a decision making tool regarding irrigation, PGR’s, nutrition and other practices.
- *‘Regina’ harvest maturity.* Delaying ‘Regina’ harvest 5 to 9 days after commercial harvest (July 13) allowed greater color uniformity and higher °Brix, but stem quality faded sooner in storage.
- *Canopy management.* Canopies of 3rd-leaf Sweetheart/Mazzard trees were regenerated by summer pruning in 2004 and 2005. Trees with summer-pruned canopies are denser and significantly smaller than control trees trained as steep leaders.
- *Effect of pollinizer distance on ‘Regina’ yield.* Yields of ‘Regina’ trees dropped greatly with increasing distance from a pollinizer. ‘Sam’ (100% cross-compatible) appears to promote higher yields than ‘Stark’s Gold’ (50% cross-compatible).

Methods

- *Cultivar and rootstock evaluation.* A collection of more than 50 sweet cherry cultivars and selections is under evaluation in The Dalles. Trees are on Mazzard rootstock, trained to steep leaders and are treated with GA₃ every year. An NC-140 rootstock trial involving 15 rootstocks

with 'Bing' as scion and Van as the pollinizer was established in 1998 at Orchard View Farms. Trees are trained to a central leader. Control trees are on *P. mahaleb* rootstock.

- *Fruit growth measurements.* Fruit growth in relation to heat unit accumulation was examined for 'Cristalina' 'Bing', 'Regina' and 'Sweetheart' in Hood River and The Dalles during 2004. In 2005, fruit growth rates of 'Regina' were measured with the aid of electronic sensors programmed to continuously monitor fruit growth at hourly intervals.
- *'Regina' harvest maturity.* 'Regina' trees were harvested July 13 based on average °Brix. Despite acceptable °Brix, a proportion of the crop was still lacking dark red color development. This is common in 'Regina' and is thought to be partly caused by a long flowering period (often near 4 weeks) with sporadic flower-opening peaks, resulting in fruit populations of different ages. A set of 10 trees was harvested on four dates up to 14 days after commercial harvest to allow color development of the latest fruit. Fruit quality was measured at harvest and after storage.
- *Crop load management.* Bloom thinning treatments were applied on heavily flowering 10-yr-old 'Lapins'/Gisela 5 trees during spring 2005 to reduce crop load and increase fruit size. The study was designed to test mixtures of lime sulfur.
- *Canopy regeneration to control tree size.* We regenerated canopies of 3rd-leaf 'Sweetheart'/Mazzard trees by summer pruning during 2004 and 2005. After heading main scaffolds in April 2004, current-season shoots were pruned at 2-week intervals between June 24 and August 9, 2004. In 2005, all new shoots were pruned in late July to produce short shoots and maintain small canopy size. Canopy growth responses, tree size, yields and fruit quality are being compared to control trees of the same age which are being trained a steep leader.
- *Effect of pollinizer distance on 'Regina' yield.* The impact of pollinizer distance and pollinizer cultivar was examined in 2004 and 2005. In 2004 we measured yield of 4th-leaf 'Regina' trees on Gisela 6 rootstock planted at different distances from pollinizer trees. Yields were recorded for individual 'Regina' trees along rows with pollinizers and for trees in adjacent rows without pollinizers. Four different pollinizers, 'Sam', 'Hudson', 'Attika' and 'Stark's Gold' were planted in alternate rows at a ratio of 1:9. However, only 'Sam' trees had adequate bloom in 2004 because the other pollinizers were planted 1-2 years later. Because in 2004 nearly all pollen derived from 'Sam' trees, we had an opportunity to evaluate yields of 'Regina' trees located at different distances from 'Sam' pollinizers. In 2005, 'Stark's Gold' pollinizer trees produced enough bloom to allow a comparison of pollinizer effectiveness with 'Sam'. 'Sam' is 100%, whereas 'Stark's Gold' is 50%, cross-compatible with 'Regina'. Yields of 'Regina' trees immediately adjacent to a 'Sam' or 'Stark's Gold' pollinizer were compared.

Results 2005

- *Cultivar and rootstock evaluation.* Observations for 2005 are summarized in Table 1. It is important to mention that rain occurred close to harvest time, causing various degrees of fruit splitting in orchards throughout the area. Interestingly, some varieties in the collection showed good to very good tolerance to rain, including '0900 Ziraat' and 'Early Robin', and particularly 'Attika' and 'Regina'. 'Bing' showed an estimated 50% of damage due to rain cracking in the variety collection. Additional data can be found at the following link: <http://extension.oregonstate.edu/wasco/horticulture/Research%20Reports/ResearchReports.php> Results of 'Bing' on the best performing rootstocks in the NC-140 project are summarized in Table 2. Highest yields were obtained in Gisela 6, Gisela 5 and Weiroot 158, but highest yield efficiencies (grams of fruit produced per cm² of trunk cross sectional area) were achieved by Gisela 3 and Gisela 5. Gisela 3 produced the smallest trees and had moderate fruit size, so it is worth considering for ultra-high density plantings. Fruit size of Gisela 6 was similar to mahaleb, but with much higher yields. Fruit firmness was moderate to low during 2005 for all rootstocks,

with most trees showing average fruit firmness close to or below the threshold of 250 g/mm (data not shown).

- *Fruit growth measurements.* Micromorphometric measurement (1/1000th of a mm) of 'Regina' fruit growth showed sustained daily fruit expansion for several days after July 13, the date of general harvest. Indeed, fruit harvested 5 or more days after this date had grown up to 1 mm larger during the additional days that fruit remained attached. Measurements showed the immediate response of daily growth rate to temperature and irrigation.
- *'Regina' harvest maturity.* This study explored the question of whether 'Regina' fruit could be left on the tree until uniform color was achieved. At commercial harvest on July 13, 90% of fruit had 17 to 22 °Brix and 34% had °Brix of 19 or higher. Stem quality and firmness were very high, but about 30% of fruit was of a distinctly lighter red color. Taste and texture of all fruit, including the lightest in color and with °Brix of 16 to 18 were excellent. Fruit harvested 5 days after commercial harvest were of a uniform dark mahogany color, 86% had a °Brix of 19 to 22 and 8% had a °Brix higher than 24. Stem retention force was very high (> 500g) for all harvest dates. More than 80% of fruit harvested July 22 and 27 had 22 °Brix or higher. Stems appeared green at harvest but more than 50% were brown within 7 days of storage. In comparison, fruit harvested July 13 retained green stems for more than 30 days in cold storage. Fruit harvested July 27 (14 days after commercial harvest) had 46% of fruit with 24 °Brix or higher, stems of low quality at harvest and 42% of culls. 'Regina' fruit harvested upon reaching uniform dark color on the tree (July 18) had better appearance than fruit harvested July 13 and maintained very good quality during storage. Postharvest quality decreased progressively with later harvest dates.
- *Crop load management.* Treatments were unsuccessful this year due to severe damage caused by frost during flower bud development. Yields were exceedingly low (estimated < 20 lbs per tree) and did not reflect treatment effects. As expected due to the extremely low fruit load, fruit was very large (mostly >30 mm diameter) and of excellent quality, although fruit splitting occurred as a result of rain near harvest time. During 2004 we were successful in reducing crop load and producing larger fruit by combining chemical bloom thinning and manual fruit removal about 3 weeks before harvest. Combined chemical plus manual treatments are planned for 2006.
- *Canopy regeneration to control tree size.* Late summer pruning of 3rd-leaf 'Sweetheart'/Mazzard trees in 2004 produced shorter shoots than early pruning. Light flowering occurred in spring 005 but virtually no fruit set occurred due to poor weather conditions during bloom that prevented adequate pollination. Late summer pruning performed again in 2005 resulted in short shoots during the fall. As a result of summer pruning, canopies of trees pruned for two consecutive years are significantly smaller (about half the height and spread) than canopies of control trees which are being trained as a steep leader (Fig. 1). Summer pruning also stimulated spur formation. Canopy regeneration of young trees by summer pruning appears to be a useful tool to control vigor and increase early production in 'Sweetheart'.
- *Effect of pollinizer distance on 'Regina' yield.* 'Regina' produces fruit of excellent quality but has shown low productivity in Oregon. An important factor in 'Regina' low yields appears to be tree distance from a pollinizer. In 2004, yields of 4th-leaf 'Regina'/Gisela 6 trees were greatly reduced as distance from a 'Sam' pollinizer increased (Fig. 2). Trees immediately adjacent to a pollinizer within the row produced the highest average yields at 12.6 kg/tree. Yields decreased by 27%, 52%, 58% and 59% for trees located 2, 3, 4 and 5 trees (20 ft, 30 ft, 40 ft and 50 ft, respectively) from a pollinizer in the same row. A similar trend was observed in rows without pollinizer trees, although yields were further reduced compared to rows with pollinizer trees. In 2005, average yields in non-pollinizer rows were 37.4 lbs/tree, whereas in rows including both a 'Sam' and 'Stark's Gold' pollinizer yields were 44.8 lbs/tree. Trees located immediately next to a 'Stark's Gold' pollinizer produced 40.3 lbs/tree, whereas those immediately adjacent to a 'Sam' pollinizer produced 51.6 lbs/tree. These results show that 'Regina' yields were notably higher when trees were located next to a pollinizer and that 'Sam' promoted higher yields than 'Stark's Gold'.

Table 1. Fruit characteristics of sweet cherry cultivars and selections under evaluation in The Dalles. Trees are on Mazzard rootstock and trained to steep leaders, unless otherwise indicated. All trees were treated with 25 ppm GA₃ when ‘Bing’ was at straw color.

Cultivar or selection/ rootstock	First bloom	Full bloom	Harvest date	Color		SSC (oBrix)	TA	SSC: TA	Fruit diam (mm)	% 9 to 8 row	% > 8-row	Firmness (g/mm)
				Skin	Flesh							
Chelan			6/14	6	5	20.4	0.68	30	28.1	16	0	511
Early Robin/Gi5	3/18	4/4	6/14			16.8	0.40	42	28.7	40	0	307
Early Robin/Gi5	3/18	4/4	6/16			16.8	0.32	52	28.4	56	0	280
Early Robin/Gi5	3/18	4/4	6/21			17.8	0.36	49	30.7	80	4	268
Tieton#			6/15	6	5	16.2	0.52	31	30.9	76	4	295
Santina/Mzd	3/21	4/7	6/16	6	4	15.9	0.48	33	30.4	68	8	312
Benton#	3/31	4/7	6/22	6	4	22.2	0.88	25	29.6	44	0	308
13S-3-13/Mzd	3/21	4/4	6/22	6	4	17.9	0.76	24	30.3	83	21	191
Sonnet/Mzd	3/24	4/7	6/29	6	5	17.7	0.56	32	30.3	68	0	244
Sam/Gi6	3/31	4/12	6/29	6	4	15.5	0.56	28	27.7	16	0	255
NY304/Gi7	3/31	4/12	6/29	5	4	17.8	0.56	32	29.7	92	8	365
NY412068/Gi6		3/25	6/29	6	5	21.8	0.56	39	29.9	72	16	331
Bing/Gi5		4/3	6/29	5	4	16.6	0.56	30	27.0	8	0	223
Bing/Gi6		4/3	6/29	6	5	19.1	0.72	27	28.4	56	0	252
Bing/Mzd	3/17	4/5	6/29	5	5	20.6	0.80	26	29.6	60	0	263
van/Gi7	3/19	4/4	6/29	6	4	18.1	0.64	28	31.3	80	0	298
Cristalina/Mzd	3/21	4/7	6/29	6	5	17.5	0.44	40	32.2	96	16	280
Sandra Rose/Mzd	3/21	4/6	7/5	6	5	19.5	0.68	29	31.1	80	0	243
Schneider/Mzd	3/28	4/11	7/5	6	5	18.4	0.60	31	29.9	80	0	249
Rainier/Gi7	3/19	4/4	7/7			19.6	0.52	38	31.5	80	24	218
Sonata/Mzd	3/20	4/14	7/7	6	6	19.3	0.82	24	32.6	96	28	340
Attika/Mzd	3/28	4/11	7/7	6	5	17.9	0.64	28	29.1	40	0	277
Sylvia/Mzd	3/28	4/13	7/7	7	6	17.5	0.48	36	30.5	80	0	264
NY270/Gi6	3/19	4/4	7/7	6	4	22.5	0.76	30	30.8	68	4	250
NY252/Gi6	3/21	4/3	7/7			21.6	0.80	27	27.9	28	0	296
0900 Ziraat/Gi5	3/25	4/12	7/7	6	5	20.1	0.52	39	30.5	72	0	210
0900 Ziraat/Gi5	3/25	4/12	7/8	6	5	20.1	0.52	39	26.4	24	0	239
0900 Ziraat/Gi5	3/25	4/12	7/9	6	5	20.1	0.52	39	29.3	76	2	237
NY2131/Gi7	3/21	4/11	7/9			20.9	0.48	44	27.0	0	0	209
NY007/Gi7			7/9			19.8	0.92	22	26.7	0	0	174
Lapins/Mzd		3/31	7/13	6	5	17.4	0.72	24	29.4	52	0	284
Redlac/Mzd	3/28	4/5	7/13	6	5	20.0	0.76	26	30.3	68	0	244
Skeena/Mzd		4/8	7/13	5	5	17.7	0.40	44	32.1	100	8	333
Skeena/Mzd		4/8	7/19	6	6	19.3	0.68	28	31.9	92	20	318
Regina/Mzd	3/31	4/17	7/13	6	4	17.3	0.56	31	29.4	60	0	340
Regina/Mzd	3/31	4/17	7/19	6	6	21.0	0.60	35	30.5	68	0	303
Sweetheart/Gi6		4/2	7/13	5	4	22.5	0.76	30	29.3	40	0	390
Sweetheart/Mzd		4/2	7/19	6	2	22.2	0.68	33	28.7	12	0	310
13S-16-29/Mzd	3/25	4/7	7/19	6	6	20.5	0.80	26	29.5	48	0	363
Symphony/Mzd	3/21	4/2	7/19	5	5	16.3	0.56	29	31.8	92	8	301
13S-42-49/Gi6	3/28	4/7	7/19	6	6	16.1	0.72	22	30.7	88	8	349
NY415311/Gi7	4/6	4/18	7/29	6	5	20.4	0.62	33	26.1	0	0	237
Staccato/Mzd	3/21	4/4	7/29	6	5	18.2	0.64	28	30.9	84	8	334
13S-21-1/Mzd	3/27	4/6	8/4	6	5	19.3	0.75	26	28.5	8	0	312

Table 2 Yield, tree size, yield efficiency and fruit size of best performing rootstocks of the NC-140 sweet cherry rootstock trial. Orchard View Farms, The Dalles, OR. Harvest date: June 22, 2005.

Rootstock	Yield (lbs/tree)	Yield (kg/tree)	TCSA (cm ²)	Yield efficiency (g/cm ² TCSA)	Ave. fruit wt. (g)
Edabriz	90.9 b	41.3 b	129.1 b	319.5 b	8.7 b
Gi 3	86.5 b	39.3 b	94.2 c	416.7 a	8.5 bc
Gi 5	129.5 a	58.8 a	142.6 b	412.3 a	8.1 c
Gi 6	146.5 a	66.5 a	233.0 a	285.4 b	9.1 ab
mahaleb	87.3 b	39.7 b	255.7 a	155.1 d	9.7 a
W 158	127.3 a	57.8 a	206.9 ab	279.3 c	9.0 ab
W 72	86.7 b	39.3 b	115.7 c	340.0 b	8.5 bc



Fig. 1. Canopy regeneration of 3rd-leaf 'Sweetheart'/Mazzard trees by selective summer pruning resulted in compact canopies and trees about 40% smaller (left) than controls trained to a steep leader (right). Compact trees pruned as shown are 6 to 8 ft tall with a canopy spread of about 7 ft and are suitable for planting at high densities (>500 trees/acre). Compact trees of this size are expected to yield about 20 lbs in 2006.

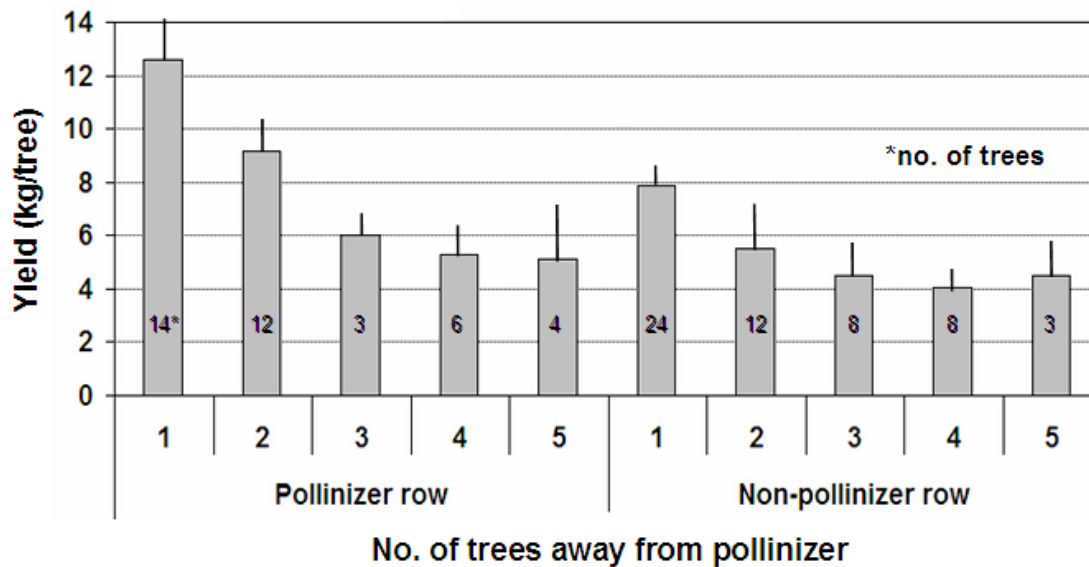


Fig. 2. Yield of 'Regina'/Gisela 6 trees as influenced by proximity to a 'Sam' pollinizer. Each bar is the mean of the number of trees indicated. Vertical lines represent the standard deviation from the mean.

Budget

Title: Cultivars, rootstocks, training systems and fruit quality
 PI: Roberto Núñez-Elisea
 Duration: long-term
 Funding in 2005: \$22,500
 Current year request: \$25,000

	2005	Current year 2006	Year 2007
Original request			
Total			
Current year request			
Salaries-FRA	18,455	15,625	15,625
OPE (60%)	9,045	9,375	9,375
Travel to res. Plots	0	0	0
Total	27,500	25,000	25,000

CONTINUING REPORT

ONGOING PROJECT

Project title: Horticulture management systems for fresh and brine cherries
Principal investigator: Anita Nina Azarenko
Organization: Dept. of Horticulture, Oregon State University
4017 ALS, Corvallis, OR 97331-7304
Research assistant: Annie Chozinski, Department of Horticulture, OSU
Cooperators: Mr. Don Nusom, Nusom Orchards, Gervais, OR
Dr. Frank Kappel, Agriculture Canada, Summerland, BC
Dr. Robert Anderson, Cornell University, Geneva, NY
Contract Administrator: Dorothy Beaton, 100 Strand Agriculture, OSU, Corvallis, OR
97331; Dorothy.beaton@oregonstate.edu; 541-737-3228

Objectives for 2006-2007:

- Identify cherry cultivars suitable for the processing cherry industry (e.g. brine, freezer) and those that may have potential for fresh market production in the Willamette Valley and cooler cherry growing districts.
- Evaluate new potential rootstocks for commercial acceptability in Willamette Valley sweet cherry production systems.
- Refine and test growing degree hour model for fruit growth in main cultivars of dark sweet cherry.
- Begin research to determine if rootstock influences susceptibility of a scion cultivar to *Pseudomonas syringae* pv. *syringae*.

Significant findings and results:

- 1996 *Dark cherry cultivar trial*- Cool temperatures, wind and rain resulted in poor pollination with low to no fruit set in many cultivars. June drop was excessive in those that did set fruit. (Removed)
- 1998 *Blush cherry trial*- June rains caused high cracking (nose/side) in early ripening fresh market selections. Five-year cumulative mean yield per tree was highest for 13N7-39 (harvested 1 week after Bing) and NY8182, fresh and brine cherry respectively and lowest production was NY7690 (Table 1). (Removed)
- 1998 *NC-140 'Bing' rootstock trial*- Mazzard and Mahaleb trees had the least production over 5 years, while Gisela 6 trees was the highest. Highest yield efficiency (YE) was obtained on W53 and the lowest on Mazzard. Gisela 4 (473-10) was most susceptible to bacterial canker with only one tree remaining by 2005. (Removed)
- 2002 *PiKu 1 and 3 trial*- Tree mortality was highest on PiKu 1 rootstocks with bacterial canker symptoms more prevalent on 'Sonata' and 'Black Gold'. All varieties were more fruitful on PiKu 1 with the exception of 'Black Gold' which had no fruit on either rootstock (Table 2).
- 2002 *Top-worked trees* – No significant differences were observed in trunk cross-sectional area (TCSA) for 'Sweetheart', 'Stardust' or 'Royal Ann' trees on Mazzard, Gi 196-4, MXM14 and MXM60. Peak bloom varied slightly between rootstocks within varieties.
- 2003 *Low-budded systems trial trees* – Pruning and training continues for modified central leader, multiple leader and Tatura trellis systems. More variation in both peak bloom and TCSA was apparent in trees trained to multiple-leader.
- 1999 *Interstem trial*- Seventh-leaf yields were significantly higher on high-budded Gisela 5 trees and lowest on trees with an MxM60 interstem. MxM60 low- or high-grafted trees had the largest TCSA. Low- and high grafted Gisela 5 trees had the smallest TCSA and the greatest amount of overgrowth of the graft union. (Removed)
- 2002 *MxM top-worked mechanical harvest trial*- 'Sweetheart' trees on MxM60 had the highest yield (11.2 kg) (Table 3). MxM60 trees had twice the TCSA than MxM14 trees in 2004 however by the end of the current growing season there were no significant differences (96-123 cm²). Fruit quality was consistently good across all rootstocks.

- *2006 variety x rootstock trial* – Trees of ‘Bing’, ‘Tieton’, ‘Sunset Bing’, ‘Sylvia’, ‘Benton’, 13N07-39, ‘Early Robin’, ‘Rainier’, ‘Sweetheart’, and ‘Skeena’ grafted onto Gisela 6, MxM14, Gi196-4 and Mazzard trees were propagated and grown for two orchardists and our program.
- *New blush trial* – trees ordered
- *Pseudomonas trial* – trees ordered
- *Sweet cherry fruit growth curves*- The Growing Degree Hour Model was placed on the IFPnet website and successfully predicted straw and harvest for ‘Bing’ at three locations in The Dalles. ‘Regina’ was harvested later than the model predicted.
- *Bacterial canker tolerance*- Dark cherry cultivars with the least number of symptoms included ‘Sonata’, ‘Sylvia’, and ‘Regina’. A blush cherry genotype with virtually no symptoms is 13N07-39. ‘Sweetheart’ on MXM rootstocks continues to show high tolerance. ‘Sonata’ and ‘White Gold’ on Piku 1 rootstock developed symptoms and died at second leaf. Piku 3 trees show no evidence of *Pseudomonas* symptoms yet.
- *2004 branching trials*- Promalin-painted patches on two-year wood of ‘Bing’ trees induced the greatest amount of shoots (5) above the treatment with the widest angles (79° from trunk). Girdling the trunk then painting also induced many shoots below the girdle with wide angles (52°) and highest shoot length (46cm). However, the leader above the girdle died. Painted buds produced fewest shoots with the shortest lengths.
- *Plant growth regulator effects on stem pull force and fruit quality* – Van: ReTain at 50g a.i. increased stem pull force. Skeena: Fruit size was increased with KT30 from fruit harvested at Matheson’s and Omeg’s. Firmness was higher with ReTain from Matheson. ReTain, Maxcel, and ProVide had the highest stem pull force. Lapins: Fruit size was increased with MaxCel and MaxCel + KT30. Stem pull force and firmness were increased with MaxCel and Promalin.
- *2005 Regina rootstock trial* – Trees were planted at a 9’ X 18’ spacing that were grafted onto Gisela 5, Gisela 6, Gisela 12 and Mazzard.
- *2005 System Fertility Management Trial* – Planted 9’ X 18’ in Corvallis and Hood River, landscape cloth installed, bark and straw amendments applied, pruning and training begun, organic certification acquired. A conservation biology hedge was planted that included hazelnut, ocean spray, elderberry, flowering currant, wild rose, *Compositae* and *Apiaceae* (*Umbelliferaceae*) perennials, *Phaselia* sp., fennel and other re-seeding annuals.

Methods:

- Train, maintain and obtain data on yield, fruit size, tree vigor, bacterial canker tolerance and other relevant data from the existing cherry cultivar trials which include:
2002 Piku 1 and 3 trial (0.20 ha)
2002 ‘Sweetheart’/MxM rootstock trial (0.12 ha).
- Train, maintain and obtain data on overall performance and productivity of trees in the existing blush cultivar/systems trials. Each planting includes four replicates of three trees of each cultivar, rootstock, and training system combination.
Top-worked trees: The rootstocks in the top-worked low-density trial are: Gi196-4, MxM14, MxM60 and Mazzard seedling. ‘Royal Ann’, ‘Sweetheart’ and ‘Stardust’ were top-worked onto these rootstocks. The training systems include: free standing, top-worked trees that are trained to a multiple leader tree and central leader (single multiple bud graft) trees. The rootstocks were planted at 18’ x 18’, in anticipation of mechanical harvest. The total number of trees in this planting are 288 (0.90 ha). Trees were top-worked, pruned and trained in 2003.
Low-budded- The low-budded high density trial includes Gisela 6, Gi196-4, MxM14 and Mazzard rootstocks. The training systems included in this trial are 1) free standing, multiple leader, 2) free standing, central leader trees, and 3) a multiple wire trellis system. This planting was set at 9’ x 16’. The total number of trees planted are 324 trees (0.50 ha). Tree structure is well established.
- Distribute experimental trees to orchardists and establish at the Lewis-Brown farm the cultivar x rootstock trial that includes ‘Bing’, ‘Tieton’, ‘Sunset Bing’, ‘Sylvia’, ‘Benton’, 13N07-39, ‘Early

Robin', 'Rainier', 'Sweetheart', and 'Skeena' grafted onto Gisela 6, Gi196-1, and Mazzard. (0.20 ha)

- Plant new 2006 sweet cherry trial that will contain 'Rainier', 'Regina', 'Skeena', NY113, NY132, NY213, NY288, NY1913, NY7679, NY7690, NY8033, NY8039, and NY9116 Gisela 6 (0.19 ha).
- *Bacterial canker tolerance* will be tested on 20 trees of 'Sweetheart', 'Regina', 'Sylvia' and 'Bing' varieties low-budded onto Mazzard, Gisela 6 and MXM14 rootstocks. Trunks will be injured and sprayed with *Pseudomonas*, held over night in cooler, taken outside for symptom development and assessment.
- Continue testing and refine *growing degree hour* model of cherry fruit growth. Test models over several sites where weather stations are located. Collaborating orchardists will provide peak bloom, straw and harvest dates.
- *Systems Fertility Management* -Identify those ecological soil community management strategies which synchronize soil nutrient availability with tree demand in order to improve long-term farm health, fruit quality, and production in an economic and environmentally beneficent way. A two-tiered approach will be used: a) a comparison of the biological and economic effects of two different methods of organic fertility management during orchard establishment and early production; b) an investigation of mature orchards to evaluate the use of soil community structure as a indicator of the effects of significantly different management practices on orchard health, fruit quality, and productivity. The 'Regina' orchard on Gisela 6 orchard was established in 2005. Geotextile cloth and straw or bark mulch are used in the tree row. Plots are planted in two locations: Lewis-Brown (Corvallis) and MCAREC (Hood River). Trees will be pruned and trained; moisture probes and lysimeters installed; soil quality and moisture, and leaf samples taken and analyzed; and trunk circumference data collected. The USDA competitive grant is covering the cost of all activities and services and supplies with the exception of the plot charges for 0.6ha.
- *Protective cultivation with light spectral management*- Three bi-color (blush) cherry cultivars; 'Early Robin', 'Rainier', and 13N07-39 on Gisela 6 rootstock will be planted in a 3-bay protected culture system. The ripening dates for these cultivars range very early to late in maturity. The system will be constructed in autumn of 2005. Each bay will be 120m (400') long, 8.2m (~28') wide, and 5.2m (17') tall (at the highest point). Three different colored nets of red, blue and pearl will be used to alter light quality. The control will be a standard film. A planting outside of the structure will serve as a commercial comparison. The total planting will be approximately 0.6 ha. The study will be arranged in a randomized complete block design with three replications and 30 trees per plot. The tree density will be approximately 1655 trees/ha (670 trees/acre). Trees will be trained to a central leader system. Black landscape cloth will be used to control weeds. Wilson Irrigation has provided the structure and plastic at cost. Polysac will be reduce the costs of the nets by 50%.

Results: See detailed findings in the following tables.

Table 1. Harvest dates, yield, trunk cross-sectional area (TCSA), yield efficiency (YE), soluble solids (SSC), firmness, fruit size, cracking, and pull force of cultivars and blush selections grafted onto Gisela 5 rootstock and planted in 1998.

Genotype ^z	Harvest date	Yield (kg)	TCSA (cm ²)	YE (kg/ cm ²)	SSC ^y (°Brix)	Fruit size (mm)	Cracks (%)	Firmness (g/mm ²)		Pull force (g)	
								Harvest	Change after 2 weeks in storage	Harvest	Change after 2 weeks in storage
Fresh											
13N7-39	6/29	11.0 _b	118 _{ab}	.09 _c	18.8 _{ab}	28.5 _a	54.7 _b	349 _{def}	.	1360 _a	.
Stardust	6/29	14.3 _a	91 _{fgh}	.16 _a	16.7 _{bc}	25.3 _{cd}	18.0 _{de}	276 _{hij}	-26 _{cde}	981 _e	-113 _{bc}
13N7-32	6/20	1.3 _{gh}	104 _{bcdef}	.01 _{fg}	13.4 _{cde}	25.3 _{cd}	43.3 _c	408 _{bcd}	-21 _a	1230 _b	-280 _{ab}
NY252	6/29	11.4 _b	95 _{defg}	.12 _b	15.9 _{bcd}	24.2 _{cdef}	24.7 _d	325 _{efgh}	-98 _e	820 _g	-303 _d
2N31-19	7/5	11.0 _b	84 _{gh}	.13 _{ab}	16.7 _{bc}	25.6 _{cde}	6.7 _{fghi}	252 _{ij}	+22 _{bcd}	830 _{fg}	-56 _{bc}
Sweetheart	7/11	8.2 _{cd}	98 _{cdefg}	.08 _c	21.5 _a	24.7 _{cde}	0.7 _{hi}	390 _{bcd}	+4 _a	930 _{ef}	-127 _{bc}
NY307	6/20	0.4 _h	110 _{abcde}	.00 _g	11.5 _e	23.8 _{def}	14.0 _{ef}	284 _{ghij}	-17 _{bcd}	850 _{fg}	-83 _{bc}
	6/20	2.0 _{gh}	108 _{abcdef}	.02 _{fg}	17.4 _b	26.0 _{bc}	10.7 _{efg}	238 _j	+13 _{bcd}	1030 _{de}	-216 _{bc}
NY6091											
13S21-14	7/5	4.9 _f	76 _h	.07 _{cd}	17.4 _b	23.6 _{def}	0.0 _i	388 _{bcd}	.	1150 _{bc}	.
NY7690	6/21	0.0 _h	126 _a	.00 _g	18.0 _b	27.5 _{ab}	46.7 _{bc}	413 _{bc}	.	1120 _{cd}	.
Brine											
Royal Ann	6/17	10.4 _{bc}	123 _a	.09 _c	15.6 _{bcd}	22.7 _{fg}	7.3 _{fghi}	303 _{fghi}	.	1170 _{bc}	.
NY8182	6/17	12.2 _{ab}	98 _{cdefg}	.13 _b	12.7 _{de}	22.9 _{ef}	15.3 _{ef}	427 _b	.	1020 _{de}	.
WhiteGold ^x	6/20	5.6 _{ef}	82 _{gh}	.07 _{cd}	17.5 _b	24.1 _{def}	72.7 _a	268 _{hij}	+9 _{bcd}	830 _{fg}	-111 _c
NY9295	6/17	7.9 _{de}	114 _{abc}	.07 _{cd}	15.5 _{bcd}	15.4 _h	3.3 _{ghi}	168 _k	.	1400 _a	.
NY7855	6/20	3.1 _{fg}	92 _{efgh}	.03 _{ef}	16.1 _{bcd}	23.0 _{ef}	9.3 _{efgh}	348 _{defg}	.	1020 _{de}	.
NY518	6/20	5.4 _{ef}	111 _{abcd}	.05 _{de}	17.5 _b	20.8 _g	51.3 _{bc}	561 _a	.	1250 _b	.
	7/5	2.3 _{gh}	89 _{fgh}	.03 _{efg}	18.1 _{ab}	23.2 _{ef}	0.0 _e	361 _{cdef}	-79 _{bc}	1190 _{bc}	-479 _c
13S20-11											
MSD		5.8	19	.03	3.4	1.9	9.0	70	36	100	187

^zMeans separation by Waller-Duncan k-ratio t-test, k-ratio=100.

^yMean of 25 fruit.

^xWhite Gold = NY 13688.

Table 2. Harvest dates, yield, trunk cross-sectional area (TCSA), yield efficiency (YE), soluble solids (SSC), firmness, fruit size, cracking, and pull force of cultivars grafted onto PiKu 1 and 3 rootstocks and planted in 2002.

Genotype	Alleles	Harvest date	Peak bloom	Yield (kg)	TCSA (cm ²)	YE (kg/cm ²)	Fruit size (mm)	SSC ^z (°Brix)	Fruit color (1-7)	Cracks (%)	Firmness (g/mm ²)		Pull force (g)	
											Harvest	Δ ^y	Harvest	Δ
Bing- Piku 1	S ₃ S ₄	6/21	3/28	.71	41.1	.02	26.1	20.0	5.1	48	300	-16b	932	-182
Piku 3			3/26	.05	68.2	.00
Royal Ann- 1	S ₃ S ₄	6/21	3/29	.72	52.8	.01
-3			3/29	.07	84.9	.00
Sonata- 1	S ₃ S ₄ '	6/21	3/21	.29	24.2	.01	23.9	.	6.0	37	313	.	873	.
-3			3/25	.02	54.5	.00
White Gold- 1	S ₃ S ₄ '	6/21	4/2	1.80	27.2	.07	24.7	17.5	.	78	258	-40	1010	-230
-3			4/1	.39	81.0	.00	25.5	16.9	.	74	253	-28	944	-264
Attika- 1	S ₃ S ₆	6/29	4/3	.28	44.7	.00	26.0	19.9	6.0	2	333	.	1185	.
-3			4/1	.16	71.0	.00	25.5	17.1	5.6	3	332	.	1190	.
Black Gold- 1	.	6/29	4/11	.	.	.	24.8	19.8	5.1	17	292	.	1168	.
-3			4/10	.	.	.	26.0	.	5.8	8	251	.	935	.
Lapins- 1	S ₁ S ₄ '	6/29	3/25	.97	62.4	.02	28.4	17.6	5.2	58	306	-85	956	-436
-3			3/24	.05	96.2	.00
Regina- 1	S ₁ S ₃	7/11	4/3	.52	40.5	.01	25.9	24.0	6.7	1	301	-12	1266	-196
-3			4/4	.01	64.6	.00
Skeena- 1	S ₁ S ₄ '	7/11	4/5	.30	63.4	.01	29.1	16.0	6.1	17	378	.	914	.
-3			4/5	.04	75.8	.00
Sweetheart- 1	S ₃ S ₄ '	7/11	3/27	.70	51.8	.01	27.7	16.2	5.2	2	400	6	949	-101
-3			3/26	.05	86.2	.00	28.8	.	.	2	.	.	1030	.

^zMean of 25 fruit.

^yΔ = Change after two weeks.

Table 3. Yield and fruit quality measurements for 2005 'Sweetheart' trees on MXM rootstocks. Fruit harvested on July 8, 2005.

Genotype ^z	Yield (kg)	TCSA ^y (cm ²)	Δ TCSA (cm ²)	YE (kg/cm ²)	Fruit size (mm)	SSC ^x (°Brix)	Fruit color (1-7)	Firmness (g/mm ²)		Pull force (g)	
								Harvest	Δ ^w	Harvest	Δ
MxM60	11.2a	101.8	19.4c	0.12a	25.9ab	20.1	4.7b	379	+72	954	-234
MxM39	8.9a	99.4	41.1abc	0.11a	26.3a	21.6	5.3a	372	+47	926	-175
MxM2	8.8a	97.0	30.0bc	0.11a	26.2a	20.7	4.9ab	370	+20	914	-162
MxM14	4.7b	96.2	52.9ab	0.05b	24.9bc	20.8	4.7b	387	+39	929	-158
MxM46	3.4b	123.9	66.8a	0.03b	24.8c	21.0	5.1ab	389	+33	931	-195
MSD	3.2	ns	27.4	0.05	1.0	ns	0.6	ns	ns	ns	ns

^zMeans separation by Waller-Duncan k-ratio t-test, k-ratio=100.

^yTCSA=trunk cross-sectional area in September 2005.

^xMean of 25 fruit.

^wΔ = Change after two weeks.

Budget

Project title: Horticulture management systems for fresh and brine cherries
Principle Investigators: Anita Nina Azarenko
Project duration: indefinite
Current year: 2006
Current year request: \$54,270

Item	2006
Salaries (0.75 FTE)¹	\$24,375
Benefits (53%)¹	\$13,045
Wages²	\$3,200
Equipment³	\$3,500
Supplies⁴	\$2,850
Travel⁵	\$500
Miscellaneous (plot charges)	\$6,800
Total	\$54,270

¹Salary and benefits for Annie Chozinski, research assistant. Her base salary is \$32,500. The balance of her funding is requested in the “Flowering, pollination and fruit set of ‘Regina’ and ‘Bing’ sweet cherry trees”

²Wages for an undergraduate student: ~400 hrs at \$8.00/hr + \$3.12/mo OPE

³50% of the cost of the tunnel system provided by Wilson Irrigation.

⁴50% of the cost of the colored netting.

⁵Travel includes mileage for travel to and from the Lewis-Brown Research Farm.

CONTINUING PROJECT**YEAR 1/3**

Project title: Alternative nutrient, water and floor management strategies
PI: Xinhua Yin
Organization: Oregon State University - Mid-Columbia Agricultural Research and Extension Center
Address: Hood River, OR 97031-9512
Phone: (541) 386-2030
Fax: (541) 386-1905
Email: xinhua.yin@oregonstate.edu
Cooperators: Jinhe Bai, Post-Harvest Physiologist, OSU-MCAREC
Clark Seavert, Agricultural Economist, OSU-MCAREC
Rita Giuliani, Horticulturist, OSU-MCAREC
Roberto Núñez-Elisea, Horticulturist, OSU-MCAREC
Contract Administrator: Dorothy Beaton, dorothy.beaton@oregonstate.edu, 541-737-3228

Objectives

- 1) Compare drip irrigation with micro sprinkler on water use efficiency, fruit set, quality, storability, and yield of sweet cherry, and grower profitability.
- 2) Examine the effects of fertigation on nitrogen (N) use efficiency, fruit yield, quality, and storability of sweet cherry and grower profitability as compared with broadcast application of dry N fertilizer on soil surface.
- 3) Evaluate the impacts of an integrated drip irrigation and fertigation production system on fruit set, quality, storability, and yield of sweet cherry, grower profitability, and water and N use efficiencies as compared with our current micro sprinkler irrigation plus surface broadcast application of dry N fertilizer system.
- 4) Compare ground cover (straw mulch or fabric) vs. no ground cover, mulch cover vs. fabric cover, and white fabric cover vs. black fabric cover on fruit quality, storability, and yield of sweet cherry, and on soil fertility, soil quality, soil microbial biology, and plant nutrition as well.

Significant Findings

- Drip irrigation consumes only 26% of irrigation water compared with micro sprinkler irrigation.
- Fruit yield under drip irrigation is similar to that under micro sprinkler. However, there is a strong trend of yield increase with straw mulch and fabric covers, particularly with white fabric, relative to no cover.
- Fruit quality including sugar content, firmness, and fruit size does not differ regardless of irrigation and ground cover systems.
- Drip irrigation significantly increases the percentage of marketable fruits by reducing cherry surface pitting compared with micro sprinkler. Fabric cover may also increase the percentage of marketable fruits.
- Fabric cover over the row area of young sweet cherry significantly improves tree N uptake and leaf N content.
- Application of organic fertilizers directly on the top of fabric cover is equally effective as the application of these fertilizers to the beneath of fabric cover.

Methods**Drip Irrigation and Straw Mulch Trial**

A field experiment was initiated on Mel Omeg's orchard at The Dalles, Oregon in 2005. Two irrigation systems (drip irrigation, micro sprinkler irrigation) and four ground management systems [mulch with straw, white fabric cover, black fabric cover, and control (no mulch or fabric cover, but herbicides was used to control weeds)] were evaluated in a split-plot design with four replications.

Soil moisture measurements were taken weekly at the soil depths of 12 inches from May to September. Irrigation scheduling for each treatment was based on soil moisture monitoring, and each plot was irrigated separately. Soil available nutrients at the depth of 12 inches, total nutrient concentrations in leaf, and tree vigor were measured prior to treatment initiation and after harvest. Fruit yield, firmness, size, color, and sugar were determined for each plot. Visual evaluation of fruit surface pitting was conducted after the fruits had been stored in a cold storage room at 33°C for four weeks. Four categories of clear, slightly pitted, pitted, and bruised fruits were used in this evaluation.

IFP Cherry Trial

The IFP experiment was initialized in 2001 on a 3-acre sweet cherry orchard that was planted in April 2001 on a sandy loam soil at the Mid-Columbia Agricultural Research and Extension Center (MCAREC), Hood River, Oregon. Two ground management systems [synthetic fabric cover (an 8-ft wide synthetic fabric cover made of black, woven polypropylenec), control (no cover, but with herbicide applications in the tree row area)] were evaluated. Soil fertility, plant nutrition, and ground management effect on cherry surface pitting were measured in this trial. Other measurements including soil moisture, soil temperature, tree growth, and fruit yield were continuously evaluated by Dr. Roberto Núñez-Elisea at OSU-MCAREC.

Organic Fertilizer Placement Trial

A field experiment was established in 2005 on a 1-acre black fabric-covered adult sweet cherry orchard that was transitioned into organic production in 2003 at MCAREC. Two types of organic fertilizers (fish mill, blood mill) and two placement methods of these fertilizers (broadcast application on the top of fabric cover, broadcast application to the beneath of fabric cover) were evaluated in a split-plot design with four replicates. Soil available nutrients to 12 inches deep, total nutrient concentrations, and tree vigor were measured prior to treatment initiation and after harvest. Chlorophyll content in leaf was measured using a SPAD-502 meter during June, July, and August. Fruit yield was determined for each plot.

Results

Drip Irrigation and Straw Mulch Trial

Differences in soil available N, P, K, Ca, Mg, S, B, Zn, Mn, Cu, pH, or organic matter were primarily negligible between drip irrigation and micro sprinkler or among no cover, straw mulch, black fabric cover, and white fabric cover in August, 2005 (data not presented). However, drip irrigation had slightly lower concentrations of N, P, K, Ca, B, and Mn in leaf than micro sprinkler after harvest (Table 1); which suggests that the uptake of these nutrients by roots may be slightly reduced due to the switch from micro sprinkler to drip irrigation in the first year. Unlike irrigation systems, the four ground cover treatments had similar leaf nutrient concentrations except N (Table 1). The differences of nutrient concentrations in fruit were rarely significant between the two irrigation systems or among the four ground cover treatments (data not presented).

The biggest benefit with drip irrigation was water saving. During the entire season from May to September, drip irrigation consumed only 26% of irrigation water relative to micro sprinkler (Table 2). Compared with no cover, black fabric reduced water use by 8%, and straw mulch and white fabric had a 1 to 3% reduction in water use. Fruit yield with drip irrigation was similar to that under micro sprinkler (Table 2). There was a strong trend of yield increase with straw mulch and fabric covers, particularly with white fabric relative to no cover, although these yield increments were statistically insignificant. Because both irrigation and ground cover treatments were implemented in early May this year, these yield differences may be not fully attributable to the treatment effects alone. Fruit quality including sugar content, firmness, and fruit size did not differ regardless of irrigation or ground cover systems (Table 2). It was interesting that drip irrigation increased marketable fruits (clear + slightly pitted) by approximately five percent (absolute value) via reducing cherry surface

pitting compared with micro sprinkler (Table 3). No benefits were found with straw mulch or fabric covers in reducing fruit pitting relative to no cover.

IFP Cherry Trial

Soil NO_3^- was lower with the covered grids than no cover after harvest in 2005 (Fig. 1). Similar to previous years, differences in soil available P were not significant between the covered and non-covered grids in 2005 (Fig. 2). Covered grids had lower soil available K than no cover in 2005 (Fig. 3). Significant effects of synthetic fabric cover on soil Ca, Mg, S, B, Zn, Mn, Cu, pH, or organic matter were not observed in 2005 (data not presented). Consistent with 2002, 2003, and 2004, leaf N content was 19% greater with the covered than non-covered grids in 2005 (Fig. 4). Because tree size in the covered grids was greater than that in the non-covered grids (data not presented), this suggests that the total N uptake by roots is greatly enhanced due to fabric cover. Similar to 2002, 2003, and 2004, leaf P content was about 20% less with the covered grids than in the non-covered grids in 2005 (Fig. 5). Leaf K was similar for the two treatments in 2005 (Fig. 6). Leaf Ca content was reduced by 10% in 2005 because of fabric cover (Table 4). The decreased leaf P and Ca contents with the covered trees were due to the dilute effects of increased tree growth. The effects of fabric cover on leaf Mg, S, B, Zn, and Mn contents were not significant in 2005 (Table 4). The differences of nutrient concentrations in mature fruit between the two treatments were smaller than those observed in leaf (data not presented).

Fruit quality, such as sugar content, firmness, and fruit size, did not differ between covered and non-covered trees (Table 5). However, fruit pitting evaluation showed that fabric cover maybe could increase the percentage of marketable fruits (clear + slightly pitted fruits) by reducing fruit bruising and pitting problems (Table 5).

Organic Fertilizer Placement Trial

Concentrations of soil available nutrients, such as NO_3^- , P, K, etc. after applying fish mill or blood mill on the top of fabric cover were similar to those following the application of the same fertilizer to the soil surface by removing the fabric cover (data not presented). Neither did leaf nutrient concentrations differ between the two placement methods (data not presented). Leaf chlorophyll content was almost identical between the two placement methods averaged over the two fertilizers (Table 6), which suggests placement method did not affect the availability of applied organic N. Fruit yield in 2005, the first year of experimentation, was not different between the two placement methods (Table 6). It seems there is no need to apply fish mill or blood mill to the beneath of fabric cover. Application of these organic fertilizers directly on the top of fabric cover is equally effective and could save labor.

Budget

Project: Alternative nutrient, water and floor management strategies

PI: Xinhua Yin

Project duration: 2005-2007

Funding in 2005: \$ 18,800

Current year: 2006

Current year request: \$19,053

Year	2005	2006	2007
Total	\$18,800	\$19,053	\$19,311

Budget breakdown

Item			
Salaries ¹	8,500	8,670	8,843

Benefits (49%)	4,165	4,248	4,333
Hourly help ³	2,800	2,800	2,800
Hourly benefits (8%)	224	224	224
Supplies	2,661	2,661	2,661
Travel to field trials	450	450	450
Miscellaneous			
Total	\$18,800	\$19,053	\$19,311

Table 1. Effects of irrigation system and ground cover on leaf nutrient concentrations.

Treatment	N (%)	P (%)	K (%)	Ca (%)	Mg (%)	S (%)	B (ppm)	Zn (ppm)	Mn (ppm)	Cu (ppm)
Micro sprinkler	2.58	0.26	2.64	1.20	0.31	0.14	81.1	17.6	55.2	4.6
Drip irrigation	2.46	0.21	2.36	1.05	0.31	0.14	67.6	15.1	49.1	3.1
Significance	*	*	*	*	ns	ns	*	ns	*	ns
No cover	2.40	0.25	2.60	1.14	0.32	0.14	74.8	17.6	49.0	4.0
Straw mulch	2.38	0.22	2.38	1.19	0.32	0.14	69.7	17.5	51.7	3.6
Black fabric	2.75	0.24	2.55	1.12	0.29	0.15	80.0	14.6	55.3	4.1
White fabric	2.55	0.23	2.46	1.06	0.31	0.14	72.9	15.8	52.7	3.8
Significance	*	ns	ns	ns	ns	ns	ns	ns	ns	ns

* indicates the treatment effect is statistically significant at 5% probability level. Non significant effect is denoted by ns.

Table 2. Effects of irrigation system and ground cover on irrigation water consumption and fruit yield and quality.

Treatment	Water consumption (gallon/tree)	Yield (lbs/tree)	Sugar (°brix)	Firmness (g/mm ²)	Size (mm)
Micro sprinkler	3427.5	49.9	17.3	290	31.1
Drip irrigation	893.5	48.9	17.3	298	30.8
Significance	*	ns	ns	ns	ns
No cover	2226.3	43.9	17.0	305	31.1
Straw mulch	2160.0	49.0	17.5	286	31.1
Black fabric	2042.8	49.3	17.1	299	30.8
White fabric	2213.0	55.3	17.5	287	30.8
Significance	ns	ns	ns	ns	ns

* indicates the treatment effect is statistically significant at 5% probability level. Non significant effect is denoted by ns.

Table 3. Effects of irrigation system and ground cover on fruit surface pitting.

Treatment	Clear	Slightly Pitted	Clear + Slightly Pitted	Pitted	Bruised
	(%)	(%)	(%)	(%)	(%)
Micro sprinkler	70.6	6.5	77.1	17.4	5.5
Drip irrigation	76.2	6.2	82.4	12.6	5.0
Significance	ns	ns	*	ns	ns
No cover	75.8	5.0	80.8	14.4	4.8
Straw mulch	71.5	6.8	78.3	15.8	5.9
Black fabric	74.0	7.1	81.1	14.5	4.4
White fabric	72.3	6.4	78.7	15.4	5.9
Significance	ns	ns	ns	ns	ns

* indicates the treatment effect is statistically significant at 5% probability level. Non significant effect is denoted by ns.

Fig. 1. Soil NO_3^-

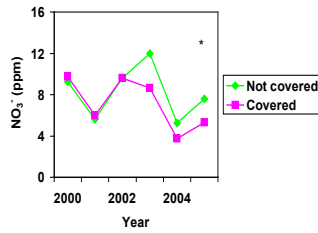


Fig. 2. Soil P

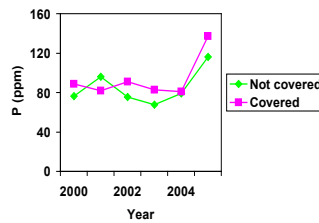


Fig. 3. Soil K

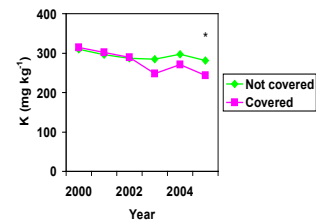


Fig. 4. Leaf N

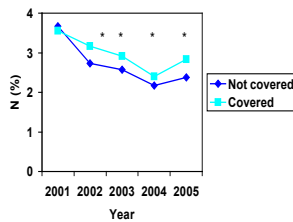


Fig. 5. Leaf P

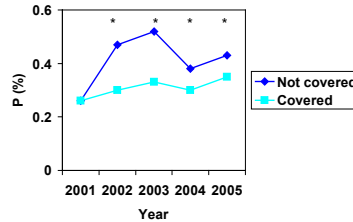


Fig. 6. Leaf K

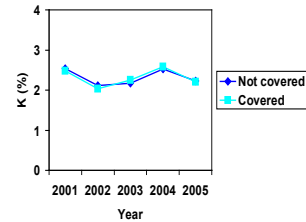


Table 4. Effects of row cover on leaf Ca, Mg, S, B, Zn, Mn, and Cu.

Year	Treatment	Ca (%)	Mg (%)	S (%)	B (ppm)	Zn (ppm)	Mn (ppm)	Cu (ppm)
2001	Not covered	0.95a†	0.24a	0.05a	35.6a	22.8a	48.9a	14.3a
	Covered	0.96a	0.26a	0.05a	35.9a	25.6a	64.0a	13.6a
2002	Not covered	1.55a	0.41a	0.19a	52.4a	24.3a	92.4a	9.1b
	Covered	1.35b	0.31b	0.18a	50.5a	20.0a	88.9a	10.4a
2003	Not covered	1.34a	0.32a	0.09a	52.4a	12.8a	57.4a	8.4a
	Covered	1.19b	0.28b	0.10a	48.6a	13.0a	55.1a	8.6a
2004	Not covered	1.38a	0.32a	0.13a	78.8a	10.4b	50.3a	4.0b
	Covered	1.35a	0.30b	0.13a	77.8a	12.7a	50.9a	4.4a
2005	Not covered	1.53a	0.39a	0.14a	70.9a	12.9a	43.1a	5.9b
	Covered	1.39b	0.37a	0.15a	68.1a	14.8a	45.1a	6.4a

† Values in column within each year followed by the same letter are not significantly different at 5% probability level.

Table 5. Effects of row cover on fruit quality and surface pitting in 2005.

Treatment	Sugar (°brix)	Firmness (g/mm ²)	Size (mm)	Clear (%)	Slightly Pitted (%)	Clear + Slightly Pitted (%)	Pitted (%)	Bruised (%)
Not covered	18.9a	333a	28.5a	81.5a†	4.8a	86.3a	8.4a	5.4a
Covered	17.6a	335a	28.2a	87.5a	4.6a	92.1a	6.2a	1.7a

† Values in column followed by the same letter are not significantly different at 5% probability level.

Table 6. Effects of organic fertilizer types and placement methods on leaf chlorophyll content and fruit yield.

Treatment	Chlorophyll June 17 (µg/cm ²)	Chlorophyll July 8 (µg/cm ²)	Chlorophyll August 4 (µg/cm ²)	Yield (lbs/tree)
Fish mill	40.8	41.8	42.8	21.3
Blood mill	40.8	42.2	42.2	21.6
Significance	ns	ns	ns	ns
On top of fabric cover	40.5	42.1	42.9	22.9
Beneath fabric cover	41.0	41.8	42.0	21.0
Significance	ns	ns	ns	ns

ns indicates the treatment effect is not statistically significant at 5% probability level.

CONTINUING PROJECT REPORT

ONGOING PROJECT

Project # OSCC –3

TITLE: Tree water use, irrigation scheduling and water management systems

PI: Roberto Núñez-Elisea

Organization: OSU, Mid-Columbia Agricultural Research and Ext. Center, Hood River, OR

E-mail: roberto.nunez-elisea@oregonstate.edu

Cooperators: M. Whiting (WSU-IAREC, Prosser, WA)
C. Seavert, X. Yin, J. Bai, L. Caldeira (OSU-MCAREC, Hood River)

Jac le Roux (Irrigation Consultant, Irrinet, The Dalles, OR)

Contract Administrator: Dorothy Beaton; dorothy.beaton@oregonstate.edu; 541-737-4068

Objectives

- To measure soil/plant water status in sweet cherry to determine active root depth, baseline levels of tree water stress and quantify tree water use.
- To evaluate the benefits of irrigation scheduling based on measurements of soil/plant water status vs. a representative grower irrigation method (i.e., calendar schedule).
- To evaluate the benefits of using a woven polypropylene fabric row cover for control of ground vegetation and in water conservation.
- To evaluate the effects of severe drought on growth, production and fruit quality of trees on Mazzard rootstock.
- To evaluate high-efficiency water management strategies (i.e., regulated deficit irrigation, partial rootzone drying) on bearing trees to conserve water and increase fruit quality.

Significant findings for 2005

- Root systems of ‘Regina’/Gisela 6 trees irrigated with micro sprinklers continue to be concentrated at 40 to 60 cm soil depth, as determined by continuous monitoring of water uptake.
- Fifth-leaf ‘Regina’/Gisela 6 trees grown with a woven fabric row cover continued to show greater vigor, branching, canopy spread and higher foliar N content compared to trees growing without row cover. Irrigation based on neutron probe readings helped conserve between 20% and 30% water relative to irrigation applied on a fixed weekly schedule with no reduction in yields or fruit quality.
- Stem water potential readings measuring tree water stress level indicated that greater reductions in water applied (than those determined by soil moisture monitoring) are possible with no negative impact on tree performance.
- Trees growing in row covers had significantly higher yields (a 48% increase) than trees without row covers. Fruit size distribution was similar for trees with or without row covers and whether irrigated according to soil moisture monitoring or a fixed weekly schedule.
- Because of the significantly higher yields obtained with fabric row covers, orchard establishment costs continue to be recovered rapidly for trees with row covers.
- Severe drought imposed on 6th-leaf ‘Lapins’/Mazzard trees by suspending irrigation the entire season caused strong stress and a reduction in tree growth rates; however, trees produced a moderate crop of large fruit of excellent quality.

Methods

Measurement of soil and plant water status

Volumetric soil water content is being measured in several ways: with a neutron probe (model CPN503) with the help of an irrigation specialist and technician (J. le Roux and L. Caldeira), a portable capacitance probe (Sentek Diviner 2000™) or a continuous soil-water monitoring system (Sentek EnviroScan™). Neutron probe measurements were made to a depth of 3 ft. The Diviner 2000™ is manually operated and records data at 10-cm intervals through the soil profile. The EnviroScan™ consists of a network of eight permanent probes, each with four soil sensors at 20, 30, 40, and 60 cm depths. A solar panel and a rechargeable 12-volt battery power the system. Sensors are programmed to collect data at 30-min intervals.

Tree water status is being measured with a pressure bomb (PMS Instruments model 610). Because pressure-bomb measurements reveal the degree of tree water stress, together with measurements of soil moisture status they offer a powerful strategy to increase the efficiency of irrigation management. Excessive stress can be caused by either over- or under-irrigating, and is considered most detrimental during the last 2-4 weeks of sweet cherry fruit growth, preventing fruit from achieving their full size potential. Stem water potentials were measured between 1 pm and 3 pm (period of highest water demand in the day), using leaves previously enclosed in plastic envelopes for at least 2 hours to reduce transpiration and allow leaf equilibration with the water status of the stem, which is highly uniform throughout the tree.

Benefits of a wide polypropylene fabric row cover

A 'Regina'/Gisela 6 orchard planted at the MCAREC in 2001 is being used to evaluate the benefits of an 8-ft wide woven polypropylene row cover for water conservation and weed control. Trees are planted at 18' x 10' (rows x trees; 242 trees/acre) and trained to a central leader. Irrigation scheduling is based on soil water content measurement. Soil moisture content is being measured during the growth season using a portable probe (Diviner 2000, Sentek, Australia). Soil water dynamics were monitored with an EnviroScan system.

Sensors have been installed to record soil and air temperature in covered and non-covered rows to determine the effect of temperature on tree growth. Measurements of stem water potential are being made to determine the effect of row cover on tree water status. Establishment and management costs have been recorded since planting. First commercial harvest of this plot occurred in 2004. Tree vigor, yield and fruit quality were determined.

Assessing the effect of severe drought

A 'Lapins'/Mazzard experimental block planted in 1999 was used for this trial. Trees are planted at 14 x 16 ft in a sandy loam soil with available water content of about 13%. Irrigation was completely suspended in 2005 to impose severe stress and compare responses to adjacent well-irrigated trees. The level of tree water stress was determined by measuring stem water potential periodically throughout the season. Soil water content was monitored with a neutron probe and a portable capacitance sensor (Sentek Diviner 2000™). Observations were made on shoot and trunk growth, bloom, fruit set and fruit quality.

Results 2005

Measurement of soil water content and location of active root system

Irrigation during 2005 was applied from mid-June to late September. Soil water content was consistently higher and around saturation at 60 cm soil depth, gradually decreasing closer to the soil surface. Most water uptake (65% to 85%) of Regina/Gisela 6 trees occurred at 40-60 cm soil depth.

Benefits of a wide polypropylene fabric row cover

As observed in 2004, during 2005 'Regina'/Gisela 6 trees grown with a woven fabric row cover had greater vigor, branching, canopy spread and higher foliar N content compared to trees growing without row cover. Scheduling irrigation according to neutron probe readings helped conserve up to ~30% water relative to irrigation applied on a fixed weekly schedule with no reduction in yields or fruit quality.

Fabric row covers greatly increase yields. Yields in 2005 were 2-3 times greater than in 2004 (Fig. 1). Trees growing in row covers had significantly higher yields (a 48% increase) than trees without row covers (Fig. 1). Fruit size distribution was similar for trees with or without row covers (Fig. 1) and whether irrigated according to soil moisture monitoring or a fixed weekly schedule. Fruit from covered on non-covered trees had excellent firmness (>350 g/mm) regardless of irrigation scheduling method. It is worth noting that more than 80% of the crop consisted of fruit larger than 10-row (of which more than 40% was larger than 9.5-row) and that the price of a 20-lb box of 'Regina' was ~\$45 in mid July, when the experimental plot was harvested. For this reason, establishment costs continue to be recovered rapidly for trees with row covers.

Row covers delay fruit maturity. As in 2004, fruit maturity in row-covered trees was delayed by about 2-3 days in relation to controls, which was attributed to more shading, and possibly higher nitrogen content and greater crop load in trees with row covers.

Row covers promote rapid tree growth. Fifth-leaf 'Regina'/Gisela 6 trees growing in an 8-ft- wide fabric row cover again displayed more vigor and branching than trees without row cover. This response is attributed to the higher moisture content and warmer soil temperature (3 to 4°F) found under the fabric cover. Trees with row cover had a greater canopy spread than those without cover.

Are row covers promoting fruit retention? Trees in row covers had trunks ~20% larger (TCSA) than those without row covers. Since yields were 48% higher with row covers, the increased yield is not fully explained by larger tree size. It seems possible that trees in row covers are capable of retaining more fruit to maturity compared to those without row covers. This hypothesis will be examined during 2006.

Stem water potential was unaffected by cover or irrigation scheduling method. Periodic measurement of stem water potential prior to irrigation showed nearly identical tree water status for trees with and without row covers and did not reveal stress during the irrigation cycle. Maximum stem water potential readings during the critical period of stage III of fruit growth were similar among all treatments.

What is the minimum water required and maximum stress level that sweet cherry trees can tolerate without affecting potential profitability?

Measurement of stem water potential to determine tree water stress level suggests that trees could be irrigated with less water than indicated by soil moisture monitoring with no negative impact on tree performance. Stem water potential readings did not indicate stress at any point during the irrigation cycle. Also, severe drought imposed on 6th-leaf 'Lapins'/Mazzard trees by suspending irrigation the entire season caused moderate stress (-1.7 MPa) during stage III and strong stress (-2.5 MPa) after harvest. Soil water content indicated high soil water deficit at these stem water potential levels. Lack of

irrigation reduced tree growth rates; however, trees produced a moderate crop of large fruit of excellent firmness, soluble solids, flavor and texture. Tolerance to stress would be higher after harvest.

Is it possible that ‘moderate stress’ resulting from minimum irrigation (tentatively, less than 50% of the amount indicated by soil moisture monitoring and ET rates) could adequately sustain tree growth and fruit development? The effects of minimum irrigation on tree growth, production and fruit quality will be examined in 2006 in more detail.

Budget

Project title: Tree water use, irrigation scheduling and water management systems
PI: Roberto Núñez-Elisea
Project duration: Continuous
Funding in 2005: \$22,500
Current year request: \$25,000

	Year 2004	Year 2005	Current Year 2006
Original request			
Total			
Current year request			
Salaries-FRA	16,340	18,455	15,625
OPE (60%)	8,660	9,045	9,375
Travel			
Total	25,000	27,500	25,000

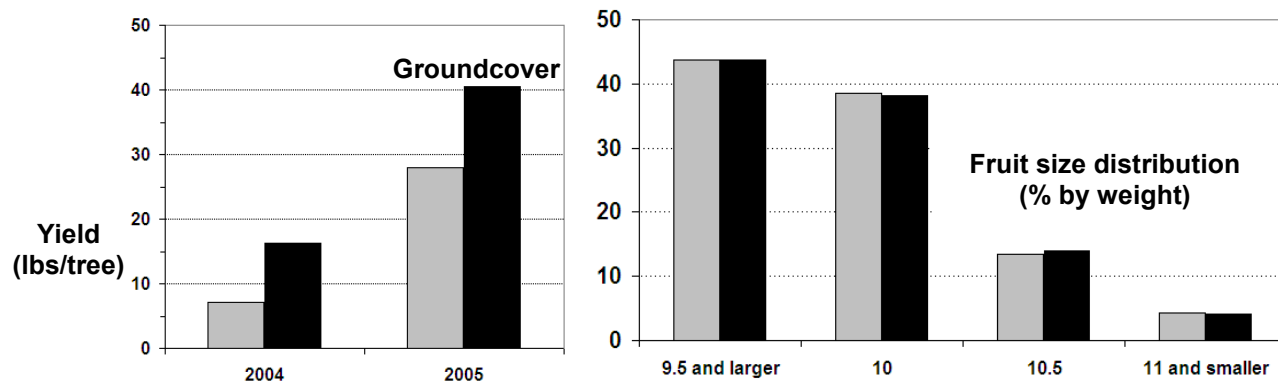


Figure 1. Fabric row covers greatly increased yield of Regina/Gisela 6 in 2004 (131%) and 2005 (48%). Fruit size distribution was unaffected by row covers in 2005. More than 80% of the crop consisted of fruit larger than 10-row in 2005. A 20-lb box of Regina fruit was worth \$45.00 in mid July, when the experimental plot was harvested.

CONTINUING PROJECT REPORT**YEAR 2/3**

PROJECT NO.: CH-04-410 **WSU Project No.:** 3355-5202
Project Title: High density orchard management
Principal Investigator: Matthew Whiting
Organization: Irrigated Agriculture Research and Extension Center, WSU-Prosser
E-mail: mdwhiting@wsu.edu
Co-Investigators: D. Peterson, USDA-ARS, Kearneysville, WV
D.R. Ophardt, Res. Tech. Supervisor, WSU-Prosser
Cooperators: D. Hayden, Pasco, WA
B. Harris, Moxee, WA
D. Allan, Naches, WA
Contract Administrator: Mary Lou Bricker, mdesros@wsu.edu, 509-335-7667 or Stephanie Brock, sabrook@wsu.edu, 509-786-9224

OBJECTIVES:

1. Develop and evaluate novel production systems including specific training/pruning strategies, cultivars and rootstocks that improve labor efficiency and yield excellent quality fruit
2. Develop and refine training strategies that facilitate mechanical harvest and/or platform assist of sweet cherries for the fresh market.
3. Continue to evaluate the effect of Ethephon on fruit quality, maturity, and retention force of different cultivars
4. Model tree vegetative and fruit growth in relation to genetic and environmental factors
5. Identify grower cooperators to participate in Competitive Orchard Systems 2015 and initiate high density research plantings with growers.

SIGNIFICANT FINDINGS:

- Ethephon applications did not elicit a reduction in fruit-pedicle retention force or fruit firmness in all varieties
- there is no consistent reduction in firmness with Ethephon application
- Ethephon did not advance maturity in all varieties
- Bing and Chelan appear better suited for stemfree harvest than Benton and Tieton
- early growth of high density orchards is affected by scion variety, training system, and rootstock
- the relative importance related to tree vigor/growth is: scion variety>training system>rootstock
- Bing and Tieton were the most vigorous, Sweetheart was the least vigorous
- Gisela 12 is ca. 45% more vigorous than Gisela 5
- highest fruit growth rates occurred during stage I of development
- alternating trees from E to W was more productive system than splitting tree into the traditional Y-trellis system

METHODS:

High density orchard management. A new high density orchard was planted in 2003 at about 5' within row spacing and 14' between row spacing for a density of approximately 580 trees per acre. It

is comprised of cultivars that ripen at approximately weekly intervals (Chelan, Tieton, Bing, Skeena, and Sweetheart) on Gisela 5 and Gisela 12 rootstocks. This block is being trained to a y-trellis system in two different ways: (1) trees headed after planting at approximately 20" and alternately tied to opposite sides of the trellis (*i.e.*, three leaders per side in a fan shape) and, (2) trees headed at approximately 30" and split on the trellis (*i.e.*, two leaders, one per side in a central leader shape). The interactions among training method, cultivar, and rootstock will be evaluated. In the first few years, tree growth and precocity data will be collected, including, trunk cross-sectional area, shoot length, number of laterals, flowering, fruit yield and quality.

Vegetative and fruit growth (when present) in this new orchard will be monitored weekly and related to locally recorded environmental data. Solar radiation, relative humidity, wind velocity, soil and air temperature, and soil water content will be continuously and intensively monitored in this orchard by three AgWeatherNet weather stations located approximately 100' apart. The ultimate goal of this experiment is to model reproductive and vegetative development of distinct germplasm to environmental phenomena (*e.g.*, fruit development, harvest date, and full bloom by degree days/heat units).

A new high density (*ca.* 530 trees/acre) of Tieton on Gisela 5 was planted in 2003. In this block, trees will be trained to either a central leader or multiple leader bush system. Growth, precocity and fruit quality will be monitored and compared between systems. This research program has shown that excellent quality fruit can be grown on a variety of training systems. Therefore, the costs associated with production on these various systems may be an important factor in determining their commercial potential. Each different system will be evaluated for labor efficiency by timing harvest and pruning events on a minimum of 50 trees per system.

Mechanical harvest efficiency. Mature Bing trees trained to various systems (*e.g.*, y-trellis, bush, central leader) will be harvested mechanically. Entire rows will be harvested and efficiency will be documented as harvesting time per tree and the number of impacts per tree. In addition, the efficiency of fruit harvest will be evaluated by collecting and weighing: (1) all fruit remaining on the tree (*i.e.*, those fruit not removed by the harvester), (2) all fruit on the ground (*i.e.*, those fruit removed but not collected), and (3) all fruit in bins (*i.e.*, ostensible yield). Quality of fruit subsamples harvested from each system will be evaluated, in comparison to stemless fruit harvested by hand and control fruit (with stems, harvested by hand), by an independent lab (Allan Bros.) for bruising, pitting, mechanical damage, and stem-end tears at the time of harvest and after two weeks in cold air storage.

Ethrel effects. Whole trees will be treated with Ethrel approximately 14 days before harvest. Cultivars to be treated include Chelan, Tieton, Bing, Benton, Lapins, and Selah. The following data will be collected on each of 40 fruit randomly harvested just prior to application and at 2 – 3 day intervals following application until commercial harvest: fruit retention force, fruit weight, soluble solids, firmness, and color. Fruit from treated trees will be compared to fruit from untreated control trees.

RESULTS AND DISCUSSION:

Varietal response to Ethephon Not all varieties responded similarly to the application of Ethephon (Table 1). In 2005, Ethephon reduced pedicel-fruit retention force (PFRF) of Bing, Chelan, and Tieton but not of Benton (Table 1). This differs from results in 2004, when each variety showed significantly reduced PFRF. In 2005, for those affected varieties, the average reduction, measured about two weeks after application, was 35%. Bing and Chelan responded similarly, exhibiting a *ca.* 41% reduction and Tieton was less-affected – PFRF was only 24% lower in Ethephon treated trees. This again contradicts results from 2004 in which Tieton exhibited the greatest reductions in PFRF. Regardless, for no variety did Ethephon reduce PFRF below the target of 400 for ideal removal by the mechanical harvester. For those treated with Ethephon, the lowest values were for Bing at 540 g and the highest were from Benton at 850 g. Our data show that stem retention (% fruit which were removed at the pedicel-spur abscission zone) was high (30% - 90%, Table 1). Even for Bing, at 540g

PFRF, ca. 30% of fruit retained their pedicel. Without Ethephon treatment however, pedicel retention was 90%.

For Bing and Chelan, the reduction in PFRF elicited by Ethephon came with significant reductions in fruit firmness (17% and 20%, respectively) (Table 1). However, both varieties had excellent firmness at harvest despite the reduction. In contrast, Benton and Tieton fruit firmness was unaffected by Ethephon treatment. Fruit firmness was excellent overall – the softest fruit were Benton treated with Ethephon at 326 g/mm. The apparent fruit softening with Ethephon-induced reductions in PFRF is not characteristic of all varieties. Tieton, for example, has consistently shown reductions in PFRF in response to Ethephon treatment without any associated loss of firmness. While the different responses to Ethephon are not fully understood, we can select varieties better-suited for mechanical harvest based on their response to Ethephon. We intend to include new varieties which, without Ethephon application, possess low PFRF (e.g., Skeena, Ambrunes) in the future. It should be noted however, that for no variety did fruit firmness decline to levels which would preclude their being marketed fresh. One assumption that has yet to be tested is that optimum Ethephon application timing is approximately 14 days before predicted harvest date. This target will surely vary with changing environmental conditions, and possibly by variety.

Table 1. Effect of Ethephon on pedicel retention force and fruit quality of 5 varieties. Statistical differences ($P<0.05$) are within a variety, between treatments, and indicated where present.

Variety	Treatment	Pedicel retention force (g)	Firmness (g/mm)	Color*	% Stem retention	Weight (g)
Benton	Ethephon	0.85	326	3.4 b	65	10.9 b
	Control	0.80	354	4.0 a	80	12.6 a
Bing	Ethephon	0.54 b	347 b	4.1 a	30 b	11.5
	Control	0.94 a	416 a	3.5 b	90 a	11.1
Chelan	Ethephon	0.61 b	347 b	4.6 a	33 a	9.0
	Control	0.80 a	433 a	4.0 b	63 b	8.5
Tieton	Ethephon	0.72 b	333	4.0	50	13.9
	Control	1.18 a	332	4.1	40	13.2

*color scale: 0=green, 1=pink, 2=light red, 3=red, 4=dark red, 5=mahogany

Fruit color data are presented as means based on the scale presented in Table 1. Optimum commercial maturity is a score of 4.0. The higher the mean color ranking, the darker, and more mature the fruit. Our data show that in most cases, fruit were picked at or very near optimum commercial maturity. Both Bing and Chelan showed advanced maturity in response to Ethephon. In contrast, Tieton color was unaffected, and color development of Benton was actually delayed by Ethephon (Table 1). Benton fruit weight was also reduced by Ethephon treatment – a result we have not previously observed.

Overall, these results suggest that Bing and Chelan are better suited for potential mechanical harvest than the other varieties we tested. It remains to be seen however if Ethephon will reduce pedicel retention force to below 400g without affecting negatively fruit quality.

Mechanical harvest trial In 2004 we negotiated and signed an agreement with USDA-ARS to transport and house their experimental mechanical harvester in Prosser for a 3-year duration. We will

continue to consult with Dr. Peterson and industry cooperators as we refine orchard systems for maximum harvest efficiency.

In 2005 we did not harvest any fruit with the mechanical harvester. Grower cooperator orchards were not cropped heavily enough and the new orchard at the Roza was similarly unproductive due to poor fruit set. In the past year we have received funding (ca. \$40k) from the IMPACT center at WSU to study the efficiency of the mechanical harvest system, its impact on fruit quality, and consumers' perceptions of stemfree cherries. These projects will complement each other well and lead to a more efficient and rapid analysis of the mechanical harvest system.

High density orchard management In 2005 we continued to refine training concepts to fit the mechanical harvest system and future integration of other mechanization (e.g., platforms). The original concept remains unchanged – develop homogeneous orchard systems comprised of fruiting ‘walls’ rapidly and efficiently while optimizing fruit yield and quality. In the current systems trial we again measured growth and fruiting characteristics of Chelan, Tieton, Bing, Skeena, and Sweetheart on Gisela 5 and Gisela 12 rootstocks. From our ongoing studies of training systems for mechanical or possibly, pedestrian or platform-assisted harvest, we have developed the following principles:

- two single-layer fruiting planes per row (one/side)
- ca. 75 – 80° between planes (each plane \approx 50°- 55° from horizontal)
- each plane consists of vertical fruiting uprights (4 – 7/tree and side though this varies with tree spacing)
- fruiting uprights spaced ca. 18” apart
- horizontal growth is eliminated
- fruiting limbs are renewed below first wire (ca. 28 in)
- upright growth to a height of at least 50 cm (\approx 20 in) above soil

Across all varieties, neither training system or rootstock had any significant effect on fruit quality. Overall, quality was excellent (ca. 93% 10.5-row and larger, 10.2 g per fruit, 350 g/mm firmness). Bing yielded the highest (4.4 kg, 9.8 lb per tree) and Tieton was the least productive (0.6 kg, 1.3 lb per tree) in the trees' first fruiting year (third leaf). The highest yielding combination was Bing trained as alternating trees on Gisela 12 rootstock (5.3 kg, 11.7 lb per tree). This translates into approximately 3.4 tons per acre of fruit that was 91% 10.5-row and larger. We documented significant interaction between variety, rootstock, and training system. For example, Bing was ca. 50% more productive on Gisela 12 vs. Gisela 5 whereas Chelan was twice as productive on Gisela 5 vs. Gisela 12. Alternating trees to the east and west trellises was slightly more precocious system compared to the traditional Y-trellised trees – third leaf yields were 3.1 kg vs. 2.2 kg per tree, respectively. This difference is approximately two-thirds of a ton per acre (1310 lbs) at our tree density and related to the greater growth in the first year in the orchard leading to more two-year-old fruiting wood per tree in trees which were alternating.

Fruit growth rates among varieties were variable (Fig. 1). In general, fruit diameter showed the characteristic double-sigmoid pattern - increasing rapidly in the weeks following fruit set (stage I), followed by a period of minimal expansion (stage II), and a final expansion period prior to harvest. Interestingly, irrespective of variety, highest absolute rates of expansion occurred during stage I. Both Skeena and Tieton exhibited above average stage I growth rates though Chelan were the largest diameter fruit during that stage. This is likely related to the early bloom of Chelan (3 – 4 days before others) and the relatively ‘older’ and therefore, larger fruit. Chelan also exhibited an abbreviated stage II during which expansion rates were higher than those of all other varieties. Sweetheart and Skeena both had a lengthy second stage of development. Moreover, stage III for Sweetheart was also

lengthy during which growth rates were relatively low. In fact, there was very little size increase in the last few weeks leading up to harvest. This may have been due to insufficient supply of photosynthate during stage III because these trees were ostensibly over-cropped, especially on Gisela 5.

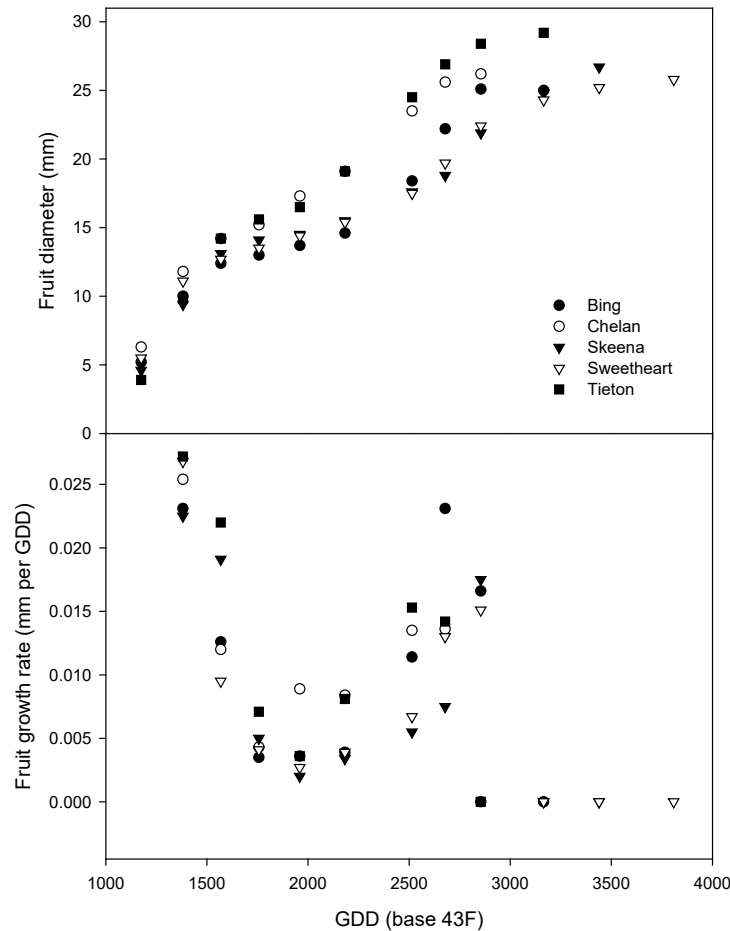


Figure 1. Seasonal trend in fruit equatorial diameter (A) and growth rate (B). Harvest dates: Chelan (7 June), Tieton (18 June), Bing (21 June), Skeena (1 July), Sweetheart (12 July).

Vigor varied among varieties and rootstocks most notably, but also by training system. Tieton was the most vigorous (ca. 65 cm mean shoot length) variety, Skeena was intermediate (ca. 55 cm), and the other three varieties were less vigorous and similar (ca. 47 cm). Gisela 12 was again about 33% more vigorous than Gisela 5 across all varieties. In many cases, particularly for Tieton and Bing on Gisela 12, trees have completely filled their space and renewal pruning will begin this winter. The least vigorous combinations are Sweetheart and Chelan on Gisela 5. Only with hard dormant heading and prudent water, nutrient, and crop load management will these trees fill their space. Clearly, early growth (first – third leaf) is critically important as trees fill their allotted space and develop future bearing surface. These combinations may never exhibit sufficient vigor to become commercially viable. We will follow the relationships between vigor and precocity and productivity closely in the next year as some combinations have filled their space and others have not.

Table 3. Effects of scion variety, rootstock, and training system on yield and vegetative growth of 3rd leaf trees.

Scion	Rootstock	Training system	Yield (kg)	Shoot length (cm)	TCSA (cm ²)
-------	-----------	-----------------	------------	-------------------	-------------------------

Bing	4.4	45.4	45.1
Chelan	1.6	48.5	35.2
Skeena	3.8	54.7	27.7
Sweetheart	3.1	47.0	16.3
Tieton	0.6	65.7	42.1
Gisela 12	2.5	60.2	46.2
Gisela 5	2.8	52.5	24.8
Y-trellis	2.2	57.6	39.4
Alternating	3.1	55.1	31.6

BUDGET

Project: High density orchard management
P.I.: Whiting, M.
Project duration: 2004-2006
Current year: 2006
Project total: \$58,363
Current year request: \$21,094

Year	2004	2005	2006
Total	\$17,895	19,374	21,094

Current year breakdown

Item			
Salaries ¹	6,199	6,301	6,503
Benefits (34% yr 3) ²	1,736	1,953	2,211
Wages ³	6,000	7,000	8,000
Benefits (11% yr 3) ²	960	1,120	880
Equipment			
Supplies ⁴	2,000	2,000	2,000
Travel ⁵	1,000	1,000	1,500
Miscellaneous			
Total	\$17,895	19,374	21,094

¹ One-sixth (or two months) annual salary for Mr. Efrain Quiroz (Roza orchard manager).

² Benefit rate for year 1 is 28%, 31% for year 2, and 34% for year 3. This increase is due to increase in contribution made by WSU on the behalf of the employee. Time-slip benefit rate for years 1 and 2 is 16%. Year 3 benefit rate is calculated at 11%. The change is due to a change in policy at WSU.

³ Time-slip assistance for harvest, data collection, and fruit quality analyses

⁴ Field and laboratory supplies.

⁵ Travel to plots and grower cooperators' orchards (@ \$0.485/mile).

CONTINUING PROJECT REPORT**YEAR 2/3****PROJECT NO.:****CH-04-411****WSU Project No.: 3355-7202****TITLE:**

Characterizing and manipulating sweet cherry source-sink relations

Principal Investigator:

Matthew Whiting

Organization:

Washington State University

Irrigated Agriculture Research and Extension Center

E-mail:

mdwhiting@wsu.edu

Cooperators:

Don Elfving, TFREC, Jim McFerson, WTFRC, Roberto Núñez-Elisea, OSU-MCAREC, Mark Roy, Roy Farms, Larry Cadwell, Benton City

Contract Administrator:Mary Lou Bricker, mdesros@wsu.edu, 509-335-7667 or Stephanie Brock, sabrook@wsu.edu, 509-786-9224**OBJECTIVES:**

1. To develop and evaluate practical strategies for manipulating sweet cherry crop load and maximizing fruit quality.
2. To investigate whole-tree source-sink relations.
3. Investigate the effects of postharvest defoliation on whole-tree physiology and fruit yield and quality.

SIGNIFICANT FINDINGS:

- high quality fruit can be grown on dwarfing, precocious rootstocks with prudent crop load management
- chemical blossom thinners vary in their mode of action and efficacy
- ATS and FOLS are most effective applied to flowers whereas tergitol was more effective applied to leaves
- VOE is not an effective thinning agent
- GA₃ is more inhibiting to flower bud induction than GA₄₊₇
- Yield in the season subsequent to GA₃ application was related negatively and closely to [GA₃]
- gibberellic acid may be an effective crop load management tool for productive orchard systems
- Compared to unpruned trees, summer pruning reduced, by half, whole-canopy NCER
- Summer pruning improved intra-canopy light distribution but had no effect on fruit yield or quality

METHODS:**Objective 1**

Chemical blossom thinning. The efficacy of several blossom thinning agents will be evaluated in multiple locations throughout the PNW. Treatments will be applied in the Yakima valley and Wenatchee region as well as in Hood River/The Dalles. Ammonium thiosulphate (ATS), fish oil + lime sulphur (FO+LS), vegetable oil emulsion (VOE), and tergitol will be applied to entire trees at different rates and timings. Treatments will be applied to heavily cropping Bing and Lapins trees on Gisela 5 at the Roza experimental farm as well as other heavily-cropped trees in grower cooperator orchards. Treatments will be compared for their effect upon floral bud induction (both number of reproductive buds per spur/shoot and floral meristems per bud), fruit set, spur and branch F:LA, and fruit yield and quality. In addition, the tree's physiological response to thinners will be documented by measuring spur leaf gas exchange prior to, and following application, and leaf and shoot expansion rates.

Thinner phytotoxicity will also be evaluated during the winter on trees grown in a greenhouse. Entire potted trees will be sprayed with a wide range of concentrations (0, 1, 2, 4, 8%) of each thinner. *GA to inhibit floral bud induction.* Trees will be treated with GA at varying concentrations (0, 30, 50, and 100 mg a.i./liter) and two stages of flower bud initiation

(roughly equivalent to beginning of stage II and III of existing crop). Treatments will be compared for their effect upon fruit quality during the season of application, floral bud induction (both number of reproductive buds per spur/shoot and floral meristems per bud), return bloom density, spur and branch F:LA, and fruit yield and quality. Initial treatments were applied during summer 2003 and consisted of: 1) Control (no treatment), 2) GA₃ 30 mg a.i./liter (standard program), 3) GA₃ 50 mg a.i./liter, and 4) GA₃ 100 mg a.i./liter. Treatments 3 and 4 were applied as single applications at either the beginning of stage II or stage II, or a double application receiving treatment on both dates.

Objective 2

Potential periods of limiting carbohydrate supply will be investigated by establishing a range of F:LA by thinning fruit buds within Bing trees on Gisela 5, Gisela 6, and Mazzard rootstocks. For each scion/rootstock combination, fruit and shoot growth rates will be monitored weekly and canopy and spur F:LA will be determined at harvest.

Newly released cultivars (*e.g.*, Chelan, Tieton, Benton, Selah) and advanced selections (*e.g.*, PC 8011-3, PC 7903-2, PC 7147-9) from the WSU sweet cherry breeding program planted in 1998 will be subjected to one of two crop load treatments: (1) unthinned control, and (2) 50% removal of blossoms by hand. Tree growth, fruit yield and quality (weight, row-size distribution, soluble solids, and firmness) will be evaluated for each scion grown on Gisela 6, Gisela 5, Gisela 195/20, and Edabriz, where possible.

Objective 3

Summer pruning. The impact of summer pruning on canopy gas exchange, light distribution, growth, and fruit yield and quality in the subsequent season will be studied. Comparisons will be made between trees subjected to summer pruning (not dormant pruned) and dormant pruned control trees. Prior to pruning, canopy LA and light distribution will be measured for each tree. The LA removed from pruning will be collected and measured. In addition, for both treatments, pruned wood will be dried to a constant weight and weighed. Light distribution throughout pruned canopies will be assessed by ceptometer following pruning. In addition, rates of single leaf and whole-canopy gas exchange will be assessed prior to, and following summer pruning. In the dormant season, wood samples will be collected and analyzed for tissue carbon and nitrogen. In the subsequent spring, rates of vegetative growth (*e.g.*, leaves and shoots) growth will be monitored weekly. Tree yield and fruit quality will be determined.

RESULTS AND DISCUSSION:

Blossom thinning

Prosser Roza Trial

In 2005, fruit set of unthinned control trees was surprisingly high (43%) considering the poor weather during bloom. Fruit set was reduced similarly and significantly with applications of ATS, FOLS, and Tergitol, by an average of 45% (Table 1). In contrast, VOE was ineffective at reducing fruit set. Whole-tree yield was statistically unaffected by any thinning treatment due to tree-to-tree variability, though yield from treated trees was numerically lower than untreated by 20% - 40%. We documented slight improvements (ca. + 10%) in fruit soluble solids from ATS, FOLS, and tergitol treatments but no effect from VOE. Fruit firmness was similarly affected – ATS, FOLS and tergitol each improved fruit firmness by ca. 14% but VOE did not. However, individual fruit weight was unaffected by any thinning treatment despite reductions in fruit set (Table 1). Fruit row-size data support the lack of thinning effect on fruit size/weight (Table 2) – every treatment peaked on either 9.5- or 10-row. These data suggest that fruit on unthinned trees were not limited in their development by photoassimilates. ‘Bing’ fruit that average 10 g and peak on 9.5-row are very near their genetic potential – this makes improving fruit quality via thinning impossible. Our crop value data (\$/tree) show that no thinning treatment improved the value compared to unthinned control. This is due to the reductions in fruit set and yield from thinners, without significant improvements in fruit size. Again, this response suggests that fruit from unthinned trees were not limited in their growth by carbohydrate

resources. Indeed, 12.4 kg (27 lb) per tree is not particularly heavily-cropped. We have recorded previously ‘Bing’/ ‘Gisela 5’ yields in this orchard of 20+ kg/tree.

A post-bloom application of 2% FOLS was made in 2005 to investigate the potential for thinning via photosynthetic inhibition. Applications were made at 14 days after full bloom (DAFB) to roughly coincide with the switching from growth supplied by stored resources to being supplied by current season assimilates. In addition, this is a period of high fruit growth rates in early stage I, and therefore, high sink demand. We hypothesize that by reducing assimilate supply at this stage, we may be able to induce resource limitations and fruit drop. Indeed, fruit set (# fruit/100 flowers) was reduced significantly by FOLS applied 14 DAFB (Table 1). This response is likely a result of photosynthetic inhibition from FOLS (Table 3) because pollination/fruit set had already taken place. However, the post-bloom FOLS application was less effective at reducing fruit set than the applications made during bloom. This is likely because post-bloom applications were less phytotoxic compared to applications during bloom (Table 3) and there was no interference with pollination and fruit set – a clear thinning mechanism of bloom applications of FOLS (see discussion below and Table 4). Despite reductions in fruit set, post-bloom FOLS did not affect fruit yield or quality.

Table 1. Effect of chemical blossom thinners applied to ‘Bing’/‘Gisela 5’ trees at 20% and 80% full bloom and 14 DAFB (post-bloom) on fruit set, yield, quality and crop value. Means within column followed by same letter are statistically different ($P < 0.1$).

	Fruit set (%)	Yield (kg)	Brix	Weight (g)	Firmness (g/mm)	Crop value (\$/tree)
During bloom (20% & 80%)						
Control	43.2 a	12.4 a	22.4 b	10.0	305.3 b	37.79
VOE	44.5 a	9.8 a	23.0 b	9.9	316.9 b	32.39
ATS	24.1 b	8.2 a	24.7 a	9.7	358.2 a	26.93
FOLS	26.9 b	7.5 a	24.5 a	9.6	352.5 a	24.28
Tergitol	20.7 b	8.5 a	24.5 a	9.8	347.9 a	28.31
Post-bloom (14 DAFB)						
Control	38.3 a	11.2	23.8	9.2	343	35.3
FOLS	27.3 b	9.6	22.6	9.2	365	30.3

We documented closely the tree’s physiological response to thinners in 2005 to better understand thinning mechanisms. Single leaf net CO₂ exchange rate (NCER) was reduced significantly by every thinning treatment at both bloom and post-bloom application timing (Table 3) though the degree of inhibition and time for recovery varied among treatments (data not shown). Leaf NCER recovery from ATS application occurred the soonest, after approximately 7 days. VOE-treated trees showed recovery of NCER by approximately 14 days after application, while all other treatments recovered after approximately 17 days. Mean NCER throughout the recovery period for the blossom thinners ATS, VOE, tergitol and FOLS was 85%, 72%, 71%, and 67% of the untreated control, respectively (Table 3-1). Therefore, ATS was the least phytotoxic treatment, affecting only a slight reduction in NCER, from which leaves recovered relatively quickly.

Table 2. Effect of chemical blossom thinners on fruit row-size distribution (% per row-size) from ‘Bing’/‘Gisela 5’ trees.

	Row-size category						
	8.5	9	9.5	10	10.5	11	11.5
Control	0	10	42.8	29.7	9.1	5.6	1
VOE	1.3	9.3	46.0	32.7	9.3	1.3	0
ATS	0	9.1	39.7	41.6	7.5	1.5	0.5
FOLS	0.5	7.5	37.7	39.8	9.5	3.5	1.5
Tergitol	0	8.4	49.6	37.6	4.0	0.4	0

Table 3. Overall effect of chemical blossom thinners (applied at 20% and 80% full bloom) and a post bloom thinner (applied 14 d after full bloom) on sweet cherry leaf gas exchange, stomatal conductance, intercellular CO₂ and chlorophyll fluorescence. Letters indicate statistical differences by Duncan analysis of variance test within column and trial ($p = 0.05$).

Bloom Treatment 20% and 80%	NCER ($\mu\text{mol m}^{-2}\text{s}^{-1}$)	g_s ($\text{mmol m}^{-2}\text{s}^{-1}$)	Ci (ppm)	Fo	Fv	$\Phi PSII$	qP
Control	7.0 a	82.9 a	197 b	384 a	1239.8 a	0.505 a	0.765 a
ATS	5.9 b	73.4 ab	215 ab	339 c	1215.8 a	0.513 a	0.752 a
VOE	5.1 bc	75.8 ab	234 a	379 ab	1064.1 b	0.514 a	0.764 a
Tergitol	5.0 bc	73.6 ab	230 a	361 abc	1054.9 b	0.490 a	0.782 a
FOLS	4.7 c	68.5 b	238 a	347 bc	1050.9 b	0.456 a	0.686 a
Post bloom Treatment 2 weeks after bloom							
Control	12.2 a	115.0 a	154 b	400 a	1902.3 a	0.762 a	0.942a
FOLS	9.9 b	97.5 b	165 a	404 a	1835.2 a	0.707 b	0.902b

Interestingly, leaves treated with FOLS 14 days after full bloom showed lower reductions in NCER and a quicker recovery compared to FOLS applications during bloom (Table 3). Leaf NCER was reduced by 19% by post-bloom FOLS whereas the blossom treatments reduced NCER by ca. 33%. In addition, leaf NCER returned to control level by ca. 7 days from the post-bloom application vs. ca. 17 days from the applications during bloom. Young leaves treated with FOLS showed slight leaf marginal burning, whereas leaves treated with the same concentration of FOLS 14 days after full bloom showed no visible signs of damage. These results suggest that leaves become less susceptible to damage with maturity. However, environmental conditions during the bloom application (higher relative humidity) favored chemical absorption over the drier conditions during the post-bloom application (data not shown). Among other factors, it is likely that environmental conditions during early applications accentuated leaf damage, though clearly, leaf ontogeny must also be considered. The post-bloom application of FOLS had no effect on Fo (constant fluorescence) and Fv (variable fluorescence), but significantly lowered (ca. 5 – 8%) quantum efficiency of photosystem II ($\Phi PSII$) (Table 3). Therefore the fraction of energy absorbed by chlorophyll and used in photochemistry was lowered by thinning application. FOLS applied 2 weeks after bloom also reduced photochemical quenching (qP) by approximately 4%. A reduction in qP indicates that the photosystem II reaction centers are closed and thus the efficiency of the entire photosynthetic system is reduced. We will continue to investigate the potential for post-bloom thinning of sweet cherry in 2006.

Table 4. Effect of thinning treatments applied to leaves (not flowers) and flowers (not leaves) on fruit set of ‘Bing’. Letters indicate statistical differences by Duncan analysis of variance test within column ($p < 0.05$). Asterisks indicates significant differences within row.

Treatment	Leaves covered/flowers treated	Leaves treated/flowers covered
Fruit set (% available flowers)		
Control	24.8 ab	34.1 ab
ATS	18.9 b*	42.9 a*
VOE	35.3 a	40.7 a
Tergitol	21.9 ab	19.1 b
FOLS	10.3 b*	21.3 b*

In 2005 we also initiated an experiment to better understand the mechanism by which thinners effect a response. Our previous research and printed reports in other species point to two possibilities – a reduction in tree/spur carbon balance via reductions in net photosynthesis and/or increase in dark respiration, and the interference with pollination and fruit set via causticity to floral structures. Just prior to thinner application, we covered with plastic bags either the entire spur leaf area (flowers exposed) or all flowers (leaves exposed to thinner). We evaluated fruit set near harvest as a percent of available flowers on a spur basis. When only flowers were treated with thinners, fruit set varied by three-fold though no treatment was significantly different from the control. Both ATS and FOLS however were significantly lower fruit set than VOE (Table 4). FOLS reduced fruit set the most, to about 41% of the control. When only leaves were treated with thinning treatments (i.e., flowers were untreated) fruit set of tergitol- and FOLS-treated spurs showed the greatest reductions in fruit set (ca. 40%) and VOE- and ATS-treated spurs showed numerically greater fruit set than the control (Table

4). These contradict previous reports on the inhibition of pollination by VOE by sealing closed the unopened perianth. In addition, it appears that ATS, despite significantly reducing NCER (though it was the least phytotoxic thinning treatment) acts by interfering with pollination. Only for ATS and FOLS was fruit set significantly lower when flowers were treated vs. when leaves were treated (indicated by asterisks in Table 4). This suggests that these thinners are most effective when applied to blossoms rather than leaf tissue. In contrast, tergitol was more effective when applied to leaves, rather than flowers only (44% vs. 12% reduction, respectively). We will repeat this experiment in 2006 with greater replication.

Thinning trials statewide

In 2005 we also conducted thinning trials with industry cooperators WTFRC (Sweetheart/Mazzard), Mary Roy (Lapins/Gisela 5), John Heffron (Rainier/Mahaleb), and Larry Cadwell (Rainier/Gisela 5). Results from each trial will be posted at <http://fruit.prosser.wsu.edu>. Our most promising results were achieved at Heffron's Rainier orchard where FOLS, ATS, and tergitol reduced fruit set similarly and significantly vs. untreated control (Table 6). Similar to our Bing trials, VOE was ineffective. ATS-, FOLS- and tergitol-treated trees yielded fruit that was 68%, 51%, and 36% 9.5-row and larger, respectively vs. only 17% in this size category in the untreated trees. FOLS and ATS increased fruit weight compared to untreated by 17% and 22%, respectively. No treatment significantly impacted fruit color – at least 75% of the fruit had more than 50% of their surface colored.

Table 6. Effect of chemical blossom thinners applied to 'Rainier'/'Gisela 6' trees at 20% and 80% full bloom on fruit set, yield, and quality. Means within column followed by same letter are statistically different ($P < 0.05$).

Treatment	fruit set (%)	%≥9.5-row	%≥10.5-row	%11 & 12-row	weight (g)	% with ≥50% red
Control	65 a	17 b	71 a	27 a	8.1 b	82 a
FOLS	46 b	51 ab	92 a	8 a	9.5 ab	75 a
VOE	70 a	37 ab	83 a	10 a	8.6 ab	87 a
ATS	44 b	68 a	99 a	1 a	9.9 a	90 a
Tergitol	41 b	36 ab	81 a	19 a	8.9 ab	75 a

GA to inhibit floral bud induction We have shown previously that applications of high rates of GA₃ to 7-year-old 'Bing'/'Gisela 1' trees can inhibit the formation of flower buds, reduce yield, and improve fruit quality significantly. In 2004 we conducted an isomer trial on 'Bing'/'Gisela 1' trees to compare the efficacy of GA₃ vs. GA₄₊₇ at reducing return bloom and balancing crop load in the season subsequent to application (i.e., 2005). Every application of GA₃ and GA₄₊₇ in 2004 significantly reduced return bloom and yield in 2005 compared to the control (Table 5). At 100 mg/L, GA₃ and GA₄₊₇ reduced yield by ca. 71% and 34%. At 200 mg/L GA₃ treatment nearly eliminated all flowers with a 95% reduction in yield; GA₄₊₇ was not as inhibiting, reducing yield by 37% (Table 5). No treatment had a positive effect on crop value though GA₃ at 100 mg/L did improve soluble solids (brix) and firmness. Unfortunately, this orchard was not particularly productive – our untreated control trees yielded less than 9 kg (<20 lb). Therefore, fruit growth in untreated trees was not limited by the partitioning of assimilates. Our fruit weight data supports this contention – there was no difference in fruit weight between control trees and those which yielded less than 1 kg. However, this approach to crop load management shows promise and is one we will continue to investigate on heavily-cropped trees and varieties other than Bing.

Table 5. Effect of GA₃ and GA₄₊₇ applications in 2004 on bloom density, fruit yield, and fruit quality in 2005. Treatments are listed by the GA isomer and concentration (mg/L). N = 5, statistical comparisons at alpha = 0.05.

Treatment	Bloom Density (flowers/cm ²)	Yield (kg)	Weight (g)	Brix	Firmness (g/mm)	Crop Value (\$/tree)
Control	29.59 a	8.82 a	10.62 a	22.9 b	307 c	30.41
GA ₃ , 100	10.32 b	2.56 bc	10.06 a	25.9 a	360 a	8.67
GA ₃ , 200	2.21 b	0.48 c	10.53 a	23.9 ab	352 ab	1.59
GA ₄₊₇ , 100	10.08 b	5.80 ab	10.59 a	24.6 ab	325 bc	19.77
GA ₄₊₇ , 200	9.24 c	5.58 ab	10.62 a	24.0 ab	328 bc	19.25

BUDGET

Project:	Characterizing and manipulating sweet cherry source-sink relations
P.I.:	Whiting
Project duration:	2004-2006
Current year:	2005
Project total:	\$83,553
Current year request:	\$25,204

Year	2004	2005	2006
Total	\$29,015	\$29,334	\$25,524

Current year breakdown

Item			
Salaries ¹	6,199	6,301	6,503
Benefits (34% yr 3) ²	1,736	1,953	2,211
Wages (time-slip) ³	13,000	13,000	9,000
Benefits (11% yr 3) ²	2,080	2,080	990
Equipment			
Supplies ⁴	3,000	3,000	3,000
Travel ⁵	3,000	3,000	3,500
Miscellaneous			
Total	\$29,015	\$29,334	\$25,204

¹ One-sixth (or two months) annual salary for Mr. Efrain Quiroz (Roza orchard manager).

² Benefit rate for year 1 is 28%, 31% for year 2, and 34% for year 3. This increase is due to increase in contribution made by WSU on the behalf of the employee. Time-slip benefit rate for years 1 and 2 is 16%. Year 3 benefit rate is calculated at 11%. The change is due to a change in policy at WSU.

³ Time-slip support.

⁴ Includes all thinning and chamber materials (e.g., Mylar, Velcro, PVC) and gas analysis consumables.

⁵ Travel to plots and vehicle maintenance (@ \$0.485/mile).

CONTINUING PROJECT REPORT

YEAR 1/3

Project Title: Understanding N requirements for sweet cherry production (methods assessment)

PIs: P. Millard¹, D. Neilsen², G. H. Neilsen² M. Whiting³

¹ Macaulay Land Use Research Institute, Aberdeen, AB15 8QH, UK

(Tel: +44 1224 318611; email: p.millard@macaulay.ac.uk).

² Pacific Agri-Food Research Institute, Summerland, B.C., Canada V0H 1Z0

(Tel: 250 494 7711, email: neilsend@agr.gc.ca and neilseng@agr.gc.ca).

³ Washington State University, Prosser, WA email: mdwhiting@wsu.edu

Contract Administrator: Denise Neilsen

Objectives:

TECHNIQUE DEVELOPMENT FOR SAP FLOW

1. Test and compare heat balance/heat dissipation probes for sap flow
2. Calibrate in field and green house for transpiration using lysimeters
3. Relate to environmental controls on stomatal closure (temperature, photon flux density)

BASIC UNDERSTANDING OF N REQUIREMENTS FOR CHERRY

4. Determine the contribution of spring remobilized N to total N requirements

The initial proposal (2005) included objectives for the determination of sap composition and N requirements for sweet cherry, based on xylem N flux and the use of ¹⁵N labeled fertilizer. These will not be addressed in this report as funding was targeted to the assessment of sap flow probes. The results of a preliminary experiment on intervening in Fall N withdrawal and spring remobilization to determine their contribution to cherry nutrition and production will be reported.

2006 OBJECTIVES:

5. To assess multiple sensor, heat pulse probes
6. To test the effectiveness of heat dissipation/heat pulse probes to monitor and indicate tree response under field conditions including
 - a. unmulched and mulched cherry trees
 - b. cherry undergoing reduced growing season irrigation
 - c. cherry undergoing postharvest irrigation stress
 - d. cherry undergoing multiple daily irrigation

Significant Findings

1. Heat balance probes provide a more reliable measure of total transpiration (sap flux) than heat dissipation probes
2. Sap flux measured by heat dissipation probes is affected by probe placement in the tree
3. dye tests indicate that sap flow is
 - a. limited to a few outer rings
 - b. affected by the probe insertion
 - c. interrupted by the bud union
4. Measurement with several probes in an individual tree confirms that flow is reduced above the 'rootstock' side of the bud union.
5. Sap flux was sensitive and well correlated to changes in vapour pressure deficit and ET estimates. Decreasing irrigation supply after harvest to 25% atmometer ET reduced sap flux.

6. N was removed from cherry shoot leaves for storage in the framework of the tree between Oct 4 and Nov 1st. Prevention of this normal storage cycle by premature leaf removal dramatically decreased fruit yield the subsequent year. Application of more N-fertilizer could not prevent this from occurring.

METHODS

Sap Flow Probes. There are three major probe types, all of which are based on the measurement of heat transfer in relation to sap movement. Heat balance methods (e.g. Dynagauge) apply a known quantity of heat outside the stem and subtract measured radial and axial conductive losses to calculate the difference due to heat moved convectively by sap. Heat dissipation probes (e.g. Granier) compare temperatures of a constantly heated probe with an unheated reference probe, both of which are inserted into the xylem. The amount of heat dissipated away from the heated probe by convection as a result of sap flow is proportional to the rate of sap flow. Heat pulse probes (e.g. Tranzflo) compare the temperatures of two probes placed radially into the xylem at the same trunk height equidistant from a heater located upstream (lower). A pulse of heat is carried by the sap and deemed to be between the two temperature sensing probes when their temperatures are equal. The rate of sap flow is calculated from the elapsed time and distance between heater and probes. Calculations of flow volume for Granier and Tranzflo probes require an estimate of the area of functional xylem.

Experiment 1. One year old peach trees were planted in the greenhouse and fitted with Dynagage heat balance sap flow gauges as described by Guak et al., 2003. Pots were weighed on a daily basis and sap flux was calculated every 30 min. and summed to give daily total values.

Experiment 2. Granier probes, 1 cm in length, were constructed according to the procedures of James et al., (2003) and installed in one year old Ambrosia/M.9 trees with a Fuji inter stem in the greenhouse. A 1.5 cm circle of bark was removed before probe insertion to prevent errors from phloem transport. Weighing platforms, with a resolution of 10g were constructed using a sheer beam force transducer (Omega Engineering) supported on a steel tripod. Changes in weight and temperature due to sap flow were measured every 10 seconds and averaged each 10 minutes. Calculations of sap velocity and flux were made using Granier's equations (Granier, 1987).

Experiment 3. Xylem tissue functionality was assessed by uptake of a safranin dye solution (Atkinson et al., 2003) by the tree. In the greenhouse, the apple tree stems were cut under water, above the roots and transferred to the dye vessel without exposure to air for 72h. In the field, cherry tree stems were cut at ground level and immediately transferred to water and re cut, before immersion in the dye solution for 72h. The trunks were then sectioned to determine dye flow patterns.

Experiment 4. To assess differential cross-sectional flow, four 1 cm Granier probes were installed in single, 7 year-old Lapins/Gisela.5 cherry tree in the field at different positions around the trunk. Installation procedures were similar to experiment 2 except that a much smaller, 3mm circle of bark was removed. Two of the probes were placed at 30 cm above the graft on the 'rootstock' side and the other two on the opposite side of the tree.

Experiment 5. Granier probes were installed in the trunk at 1 cm above the graft on the opposite side to the rootstock according to the procedures in experiments 2 and 4. Lapins/Gisela.5 trees received post-harvest irrigation at either 100% or 25% of atmometer estimated ET. There were three replications. Vapour pressure deficits were calculated from temperature and relative humidity data collected on-site.

Cherry N nutrition

Experiment 6. An experiment was established in 2004 with 4 treatments, each with 5 single-tree replicates, on young (4th field growing season) Lapins sweet cherry on Gisela 5 rootstock. Treatments included all combinations of 2 N rates (zero and 220 pounds of N per acre) applied as ammonium nitrate (34-0-0) to trees with early (Sept 17th) or no leaf stripping prior to dormancy. The leaf stripping treatment was designed to test the effects of restricting the availability of stored N the next

year. Systematic mid shoot leaf sampling was undertaken throughout 2004-05 and crop yield was also measured in both years.

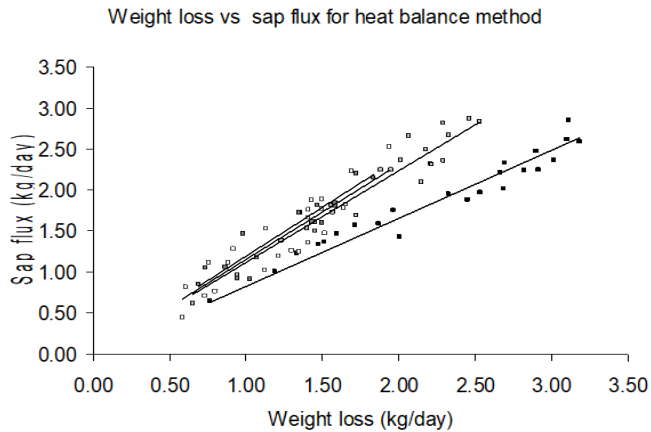


Figure 1. Transpiration estimated from weight loss and sap flow using a heat balance method

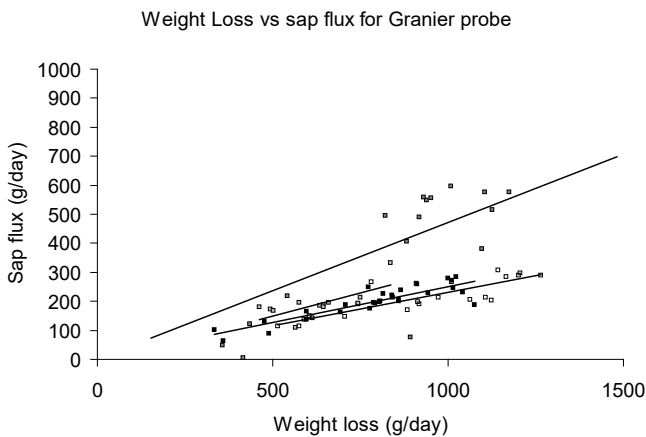


Figure 2. Transpiration estimated from weight loss and sap flow using a heat dissipation probe

(Fig. 3 a) indicates that there is a gap in conduction in the scion on the rootstock side of the bud union (indicated by arrow). In a different tree, the continued effect of the bud union on flow can be seen in the scion at several different heights above the union (Figs. 3 b,c,d). The wound caused by the probe is visible in Fig. 3c, and the effect continues above the probe (Fig. 3d). This is partially due to the relatively large area of bark removed, but Green et al. (2003) have reported on wound effects with heat pulse probes. Note also, that only the outer two rings of the xylem appear to have conducted the dye solution and

RESULTS AND DISCUSSION

Experiment 1. Measurements using a heat balance system with peach trees in the greenhouse indicated an excellent relationship ($R^2 = 0.82$ to $R^2 = 0.94$) between daily weight loss and sap flux (Fig.1). Sap flow estimates ranged from 83-110% of weight loss (slope of line) depending on probe.

Experiment 2. Sap flux measurements in apple trees using the heat dissipation probe (Granier type) were less closely related to weight loss ($R^2 = 0.46$ to $R^2 = 0.78$ (Fig. 2). Fluxes calculated from Granier probe data also underestimated pot weight losses through transpiration as indicated by the low slope of the lines. This raised the issue of how well the probes were measuring flow. Clearwater et al., (1999) found that probe estimates of sap velocity were low if the probe was in contact with a non-conducting part of the xylem and Atkinson et al., (2003) demonstrated that xylem tissue was distorted at the bud union with consequent reductions in hydraulic conductivity.

Experiment 3. The conducting part of the xylem tissue was determined using uptake of a safranin dye/water solution by greenhouse-grown apple trees (Fuji/M.9) and field grown cherry (Lapins/Gisela 5). The sample of bud union tissue for apple

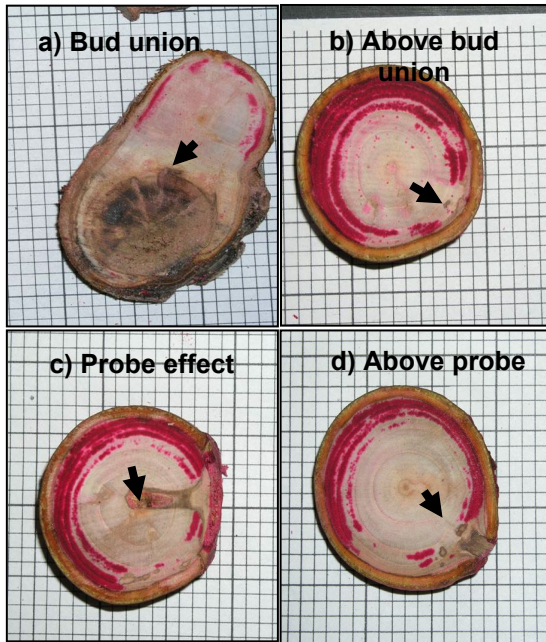


Figure 3. Xylem function in apple using safranin dye solution



Figure 4. Xylem function in cherry using safranin dye solution

that much of the probe is outside of the active xylem. In field grown cherry, the effect of the graft on flow disruption can be seen both within (Figs. 4 a,b) and above (Fig. 4 c) the bud union. Note also that only 2 to 3 of the outer rings have conducted the dye solution.

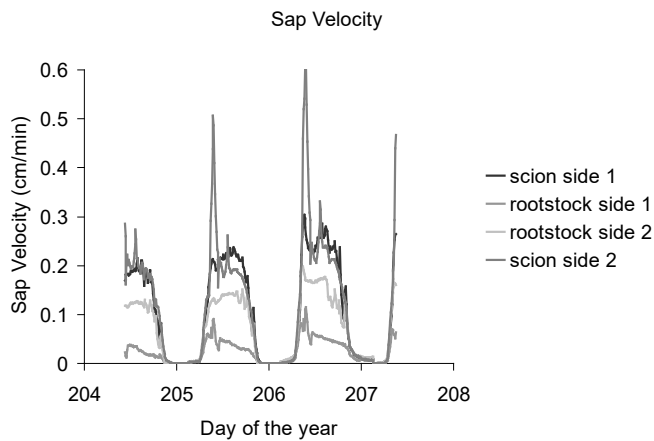


Figure 5. Sap velocity measured by four Granier probes in one tree, 30 cm above the graft.

Experiment 4. The effect of probe placement on flow was examined in a preliminary trial to the field study on cherry. Probes placed on the bud union (rootstock) side of the tree had lower sap velocity than probes placed on the opposite (scion) side (Fig 5). There was a strong diurnal pattern in sap velocity and fluctuations in flow which were likely due to environmental and internal controls on stomatal conductance.

Experiment 5. Total daily sap flow measured for a 35 day period after harvest followed the same patterns as vapour pressure deficit and ET (Fig. 6). Sap flow was less in trees receiving a lower irrigation rate (25% ET) than in trees

receiving 100% ET (Fig. 6). There was a close relationship between ET and sap flux for both irrigation treatments (Fig. 7) corroborating previous findings of our group, when measuring sap flux using a heat balance probe.

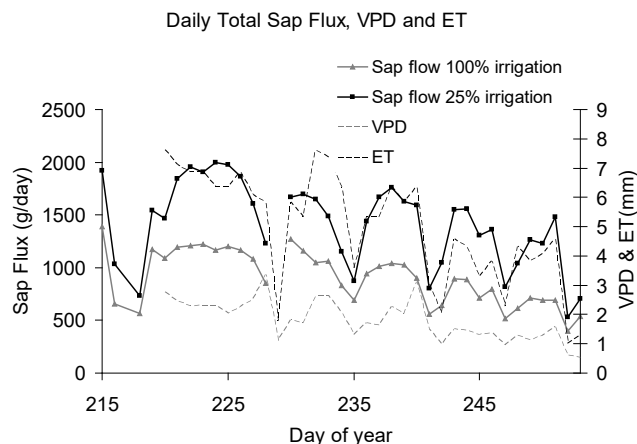


Figure 6. Vapour pressure deficit (VPD), evapo-transpiration (ET) and sap flux in cherry grown at two levels of irrigation

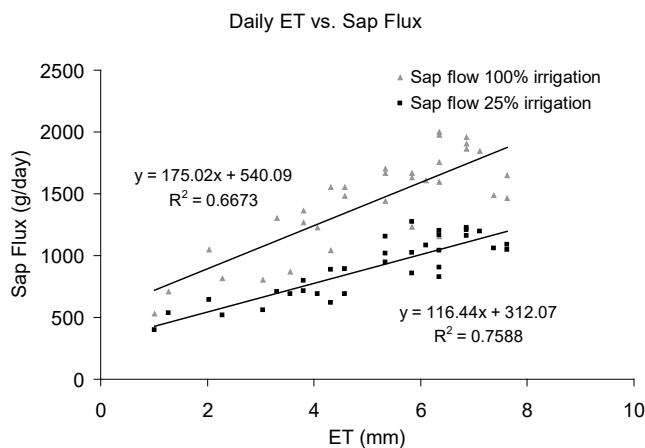


Figure 7. Relationship between sap flux and ET measured with an atmometer for field grown cherries receiving two levels of irrigation

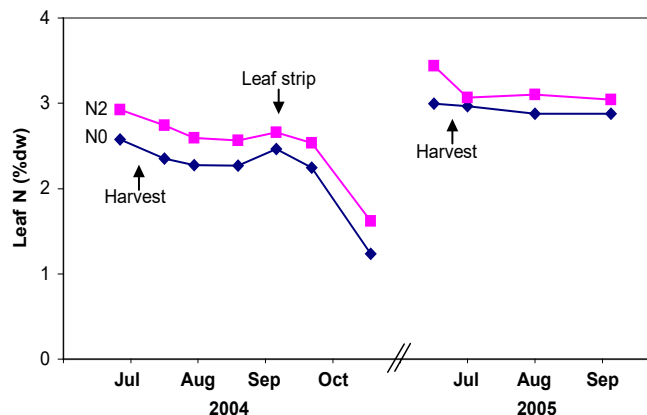


Figure 8. Effect of N application rate and leaf stripping on leaf N concentration of Lapins/Gisela.5 cherry trees

It is evident that heat dissipation probes can measure relative changes in sap flow in response to a variety of factors (VPD, potential ET, water supply to the roots). As such, they are potentially a useful tool in determining the effects of a range of management practices on tree transpiration and indirectly, stomatal function. However, they are more subject to errors than heat balance probes when estimating absolute quantities of sap flux. Some of these errors may be corrected by placement of probes and accurate estimates of the location of xylem conducting tissue. In the next year, it is proposed to evaluate the heat pulse probe, which has the advantage of multiple sensors within one probe. This allows estimates to be made of sap flux over the xylem cross-section, thus potentially eliminating errors due to probe placement outside of the actively conducting rings.

Experiment 6. N treatments affected mid terminal leaf N concentrations with significantly higher leaf N concentration measured 9 of 10 times at N2 during 2004-05 (Fig. 8). There was a pronounced decline in leaf N concentration between October 4th and November 1st, implying this is the critical time period for removal of N from leaves for storage in the woody framework. The Sept 17, 2004 leaf strip treatment was enacted prior to the movement of N from the leaves, thus considerably reducing stored N for these trees.

The leaf stripping experiment did not affect leaf N concentration of the cherry trees in either year (data not shown). However early removal of leaves in autumn dramatically reduced fruit yield the subsequent year (Fig. 9). Yield in 2005 for trees which had premature leaf removal was almost halved while higher rates of N-fertilization minimally affected yield. Leaf N concentration of new year shoot leaves was unaffected by leaf stripping in 2005 and leaf size appeared normal. Size and mass of spur leaves on older fruiting wood was however

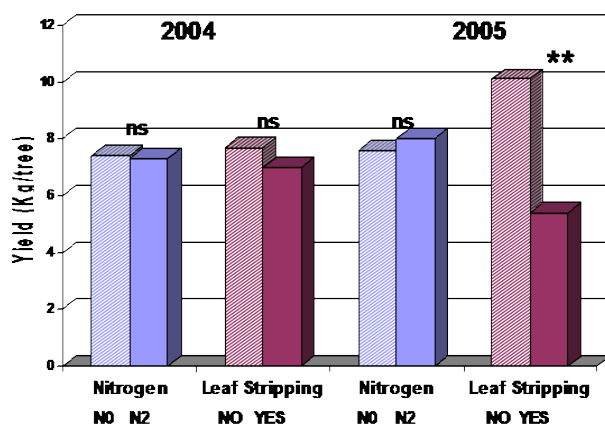


Figure 9. Effect of N application rate and leaf stripping on yield of Lapins/Gisela.5 cherry trees

considerably reduced, leading to reduced fruit yield.

These results imply that normal remobilization and removal of N from leaves in autumn is essential for development of adequate fruit load the following season. These effects are likely to be most pronounced for young trees such as these on dwarfing rootstocks. However factors leading to early defoliation of leaves prior to natural fall including insect damage and early frost are likely to have serious negative consequences to subsequent fruit production. The application of increased amounts of N-fertilizer were unable to

negate the effect.

Literature

- Atkinson, C. J., M. A. Else, L. Taylor and C. J. Dover 2003. Root growth and stem hydraulic conductivity as determinants of growth potential in grafted trees of apple. *J. Exp. Bot.* 54:121-1229.
- Clearwater, M.J., F.C. Meinzer, J.L. Andrade, G. Goldstein and N.M. Holbrook. 1999. Potential errors in measurement of nonuniform sap flow using heat dissipation probes. *Tree Physiol.* 19:681-687.
- Granier, A. 1987. Evaluation of transpiration in a Douglas-fir stand by means of sap flow measurements. *Tree Physiology.* 3:309-320.
- Green, S., B. Clothier and B. Jardine. 2003. Theory and practical application of heat pulse to measure sap flow. *Agron. J.* 95:1371-1379
- James, A.S., M.J., Clearwater, F.C. Meinzer and G. Goldstein. 2002. Heat dissipation of variable length for the measurements of sap flow in trees with deep sapwood. *Tree Physiology.* 22:277-283.

Budget

Project title: Understanding N requirements for sweet cherry production, (methods assessment)

PI: D. Neilsen, G. H. Neilsen, P. Millard, and M. Whiting

Project duration: 2005-2007

Current year: 2006

Project total (3 years): \$15,000

Current year request: \$5,000

Current year breakdown

Item ^z	Year 1 (2005)	Year 2 (2006)	Year 3 (2007)
Technical salaries	4,500	4,500	4,500
Supplies	500	500	500
Total	5,000	5,000	5,000

^z This project has been matched in cash by Agriculture Canada for the 3 year duration (2005-2007) of the project.

CONTINUING PROJECT REPORT

YEAR 1/3

Project title: Flowering, pollination and fruit set of ‘Regina’ and ‘Bing’ sweet cherry trees
Principal investigators: Anita Nina Azarenko and Annie Chozinski
Organization: Dept. of Horticulture, Oregon State University, 4017 ALS, Corvallis, OR 97331-7304
Address: 541-737-9877; azarenka@science.oregonstate.edu
Cooperators: Mr. Don Nusom, Nusom Orchards, Gervais, OR
Mel Omeg, The Dalles, OR

Contract Administrator:

Agricultural Research Foundation, Dorothy Beaton, 100 Strand Agriculture, Oregon State University, Corvallis, OR 97331; Dorothy.beaton@oregonstate.edu; 541-737-3228

Objectives for 2006-2007:

- Determine ovule longevity at different temperatures of ‘Regina’ and ‘Bing’ flowers.
- Compare pollen tube growth rates and fruit set when 2-4 standard pollinizers are used in ‘Regina’ and ‘Bing’ plantings.
- Assess pollen viability of ‘Attika’, ‘Sandra Rose’, ‘Sylvia’, ‘Skeena’, ‘Regina’ and ‘Schneider’s Späte Knorpel’.

Significant findings and results:

- *Growth chamber ovule longevity studies* – At least 20 percent of ‘Bing’ ovules remained viable for 12 days (33 and 217 GDHs) at 5C and 10C. At higher temperatures viability was reduced after 4 days (>540 GDHs). ‘Regina’ declined after 2 days at all temperatures. The approach used in the growth chamber studies with excised twigs needs to be re-evaluated.
- *In situ ovule longevity studies* – Beyond 2100 GDH (10 days after bloom), ovule longevity began to decline (Table 1). After 375 GDH (8 days after bloom), ‘Regina’ ovules began to senesce.
- *Pollen tube growth rates* – On ‘Regina’ stigmas (*in situ*) the combination of ‘Sam’ and ‘Schneider’s Späte Knorpel’ pollen produced tubes which traveled most rapidly through the style after 72 hours (301 GDHs). ‘Sam’ alone was also rapid. ‘Schneider’s Späte Knorpel’ pollen alone had an intermediate growth rate. Pollen tubes of ‘Sylvia’ and ‘Attika’ pollen were the slowest although there was some benefit to their combination (Table 2). ‘Bing’ pollen tube studies using ‘Van’ and ‘Rainier’ pollen were not carried out long enough to see conclusive results.
- *Pollen viability* – After two hours in liquid nutrient media pollen germination was determined highest in ‘Lapins’ and ‘Rainier’ followed by ‘Van’, ‘Sweetheart’, ‘Bing’ and ‘Sam’ (Table 3). ‘Attika’ and ‘Sylvia’ pollen had a meager 6-8 percent viability. ‘Schneider’s Späte Knorpel’ and ‘Regina’ pollen were intermediate (49%).
- *Nectar production* - The amount of nectar produced by sweet cherry flowers in 2005 was not measurable using capillary tubes as collection instruments. Cool temperatures and wind may have contributed to low nectar production.
- *In situ bee activity* – Bees were brought into the fields and weather conditions necessitated liquid glucose bottles to be placed on hives so bees would not starve. Their ability to leave the hive and forage pollen/nectar was inhibited by cold temperatures. Bees were seen dead on blossoms indicating hypothermia in an attempt to feed the brood. Data could not be collected in 2005.

Methods:

- *In situ ovule longevity*- Ovule longevity of 'Regina' and 'Bing' flowers will be determined in two locations, The Dalles and Lewis Brown Farm, over a 14-21 day period. A minimum of three trees will be covered with netting to prevent pollination. Flowers will be removed at daily intervals, placed in a fixative, stained with aniline blue, and observed under a fluorescent microscope. Fluorescence of callose indicates ovule senescence.
- *Pollen tube growth rates*- 'Regina' and 'Bing' flowers will be hand-pollinated with pollen from 2-4 standard pollinizers, alone and in combination. Ten flowers will be collected at 12 hr intervals placed in a fixative, stained with aniline blue, and observed under a fluorescent microscope. The percent of the style traveled by the pollen tube will be measured for each sampling date. Seed will be collected from mature fruit, and then analyzed for s-alleles using molecular markers and PCR technologies. The s-alleles in the seed will indicate the pollen parent. These data will help to identify the most suitable pollinizer(s) and estimate time required for growth of pollen tubes to the base of the style.
- *Pollen viability*- Flowers will be collected and then anthers removed from 'Attika', 'Hudson', 'Regina', 'Sam', 'Sandra Rose', 'Schneider's Späte Knorpel', 'Skeena', and 'Sylvia' flowers obtained from at least two locations in The Dalles and or Hood River. Anthers will be induced to dehisce pollen and pollen will be collected for observing pollen germination and viability. A simple liquid sucrose medium will be used to induce pollen germination. These data will assist us in further identifying the most suitable pollinizers.

Results: See significant findings in the following tables.

Table 1. *In situ* ovule viability of ‘Bing’ and ‘Regina’ in 2005 planted at the Lewis-Brown Farm, Corvallis, Oregon.

Bing				Regina			
Duration Day	Calendar Date	GDHs	% Ovule Viability	Duration Day	Calendar Date	GDHs	% Ovule Viability
0	24-Mar	61	100	0	7-Apr	23	100
2	26-Mar	433	100	2	9-Apr	113	100
4	28-Mar	891	100	4	11-Apr	233	78
6	30-Mar	1210	100	6	13-Apr	298	89
8	1-Apr	1689	88	8	15-Apr	375	89
10	3-Apr	2102	100	10	17-Apr	568	78
12	5-Apr	2466	78	12	19-Apr	661	56
14	7-Apr	3013	78	14	21-Apr	931	44
16	9-Apr	3388	57	16	23-Apr	1418	25

Table 2. *In situ* pollen tube growth in ‘Regina’ and ‘Bing’ pollinated in 10 different combinations.

Pollen source on ‘Regina’ (S ¹ S ³) ^z	% of style traveled by pollen tubes after 72 hrs (GDHs = 301)	Pollen source on ‘Bing’ (S ³ S ⁴)	% of style traveled by pollen tubes after 36 hrs (GDHs = 286)
Sam + Schneider (S ² S ⁴ + S ³ S ¹²)	62 _a	Rainier (S ¹ S ⁴)	12 _a
Sam (S ² S ⁴)	58 _a	Van (S ¹ S ³)	11 _a
Attika + Sylvia (S ³ S ⁶ + S ¹ S ⁴)	57 _{ab}	Van + Rainier	7 _b
Schneider + Sylvia (S ³ S ¹² + S ¹ S ⁴)	52 _{ab}	Control	0 _b
Sam + Sylvia (S ² S ⁴ + S ¹ S ⁴)	50 _{abc}		
Schneider (S ³ S ¹²)	48 _{abc}		
Sam + Attika (S ² S ⁴ + S ³ S ⁶)	35 _{abc}		
Schneider + Attika (S ³ S ¹² + S ³ S ⁶)	33 _{bcd}		
Sylvia (S ¹ S ⁴)	29 _{cd}		
Attika (S ³ S ⁶)	23 _{cd}		
Control	0 _e		
MSD	23		8

^zMeans separation by Waller Duncan k-ratio t-test, k-ratio=100.

Table 3. Pollen viability of ‘Sam’, ‘Schneider Spate Knorpel’, ‘Attika’, and ‘Sylvia’.

Pollen genotype	Viability (%)
Lapins	70 ^a
Rainier	69 ^a
Van	67 ^{ab}
Sweetheart	64 ^{abc}
Bing	63 ^{abc}
Sam	52 ^{bcd}
Schneider	49 ^{cd}
Regina	49 ^{cd}
Skeena	28 ^e
Snyder	15 ^{ef}
Sandra Rose	11 ^f
Attika	8 ^f
Sylvia	6 ^f
MSD	15

^aMeans separation by Waller Duncan k-ratio t-test, k-ratio=100.

Budget

Project title: Flowering, pollination and fruit set of ‘Regina’ and ‘Bing’ sweet cherry trees

Principle Investigators: Anita Nina Azarenko and Annie Chozinski

Submitted to: Agricultural Research Foundation, Oregon Sweet Cherry Commission and the Washington Tree Fruit Research Commission

Project duration: indefinite

Current year: 2006

Current year request: \$17,700

Item	2006
Salaries (0.25 FTE) ¹	\$8,300
Benefits (53%) ¹	\$4,900
Wages ²	\$2,500
Benefits (%)	
Equipment	
Supplies	\$500
Travel ³	\$1,500
Total	\$17,700

¹Salary and benefits for Annie Chozinski, research assistant. Her base salary is \$32,500. The balance of her funding is requested in the “Horticulture management systems for high value fresh and brine cherries”

²Wages for an undergraduate student: ~300 hrs at \$8.00/hr + \$3.12/mo OPE

³Travel includes mileage for travel to and from The Dalles.

CONTINUING PROGRESS REPORT

YEAR 1/3

WTFRC Project # CH-05-508

WSU Project #13C-3655-7299

Project title: Induction of branches in sweet cherry trees in the orchard and nursery

PI: Don C. Elfving, Horticulturist

Organization: WSU Tree Fruit Research and Extension Center, Wenatchee, WA

Cooperators: Matthew D. Whiting, Assistant Horticulturist, WSU-IAREC, Prosser, WA
Dwayne Visser, Agricultural Research Technologist II, WSU-TFREC, Wenatchee, WA

Contract administrator: Mary Lou Bricker (mdesros@wsu.edu) (509) 335-7667; or Tom Kelly (kellytj@wsu.edu) (509) 335-3691

Reporting period: 2005

Objectives:

1. Relate green-tip branching treatment effectiveness to timing of the bioregulator and bark-manipulation treatments in relation to bud development to determine how critical timing is as a factor controlling the branching response.
2. Assess the relation of timing of branching treatments to development of branching, location of branch development on the tree, and number, angle and quality of the lateral branches formed.
3. Assess the effect of cultivar and branch orientation on branching response to bioregulator and bark-manipulation treatments. Examine the potential for combining chemical/physical branch induction with later treatments of ethephon for stimulation of flowering and cropping.
4. Develop new combinations of bioregulator and physical treatments for enhanced cost effectiveness in branch-induction techniques.
5. Determine through longer-term monitoring the benefit in terms of both onset of productivity and sustained productivity of increased lateral branch development during the early years of canopy formation.
6. Explore the opportunity for developing new tree training systems based on the application of effective branch-induction methodology.

Significant findings:

Shoot growth is ending as this is written (September 2005); therefore, final assessments of shoot growth effects in some trials are not yet available.

a. Dormant and green-tip branch induction trials

Six trials were established in 2005 to evaluate the timing of bark injury plus cytokinin/gibberellin paint treatments for branch induction; to test various methods of bark injury to assess how strongly the bark must be injured to secure a satisfactory response to the cytokinin/gibberellin treatment; to evaluate a new cytokinin, thidiazuron, for its potential to induce lateral branching on older wood; and to initiate a small study of application of these branching techniques to tree canopy development in a high-density system.

This year, Perlan (Fine Americas) was used as the cytokinin/gibberellin product for lateral branch induction. Scoring + Perlan in latex paint at 2000 ppm active ingredient (a.i.) before or at green-tip

did not produce strong branching. The best branching this year was observed when scoring + Perlan was applied 2 weeks after green-tip, in sharp contrast to the observations in 2004. The most likely cause for this difference can be attributed to the rather cold temperatures that prevailed after green-tip was attained. This year we observed measurable branch induction when scoring + Perlan paint was applied as late as 4 weeks after green-tip, although the best response occurred at green-tip + 2 weeks. In a related trial, scoring was applied and the Perlan paint applied immediately, after 6 hours or after 24 hours following the scoring cut. There was no difference in branching response due to delaying the application of the Perlan paint after scoring. Last year, the branching response to scoring + Promalin was reduced by half by 2 weeks after green-tip.

One-year-old shoots of young ‘Skeena’ trees were subjected to sanding of the bark, scraping of the bark with a knife or notching of the bark to the cambium, combined with no additional treatment or the contemporaneous application of Perlan 5000 ppm in latex paint. Injuring the bark alone had little effect on branch development. Adding the Perlan in paint had no effect on the sanded sections but was equally effective at branch induction when bark was scraped to remove the epidermis or notched to open a pathway to inner bark tissues. These results indicate that to effectively use a cytokinin-containing product for lateral branch induction in sweet cherry the bark barrier presented by the epidermis must be breached to expose live tissue to the bioregulator. The damage does not have to penetrate to the cambium for the branch-induction process to take place after the bioregulator treatment is applied.

One-year-old shoots of young ‘Lapins’ trees were headed lightly (6-7 inches of terminal wood removed) or left unheaded. Notching the bark or disbudding, whether alone or combined with heading, did not improve the formation of lateral shoots along the one-year-old leader wood. Notching + Perlan paint (5000 ppm) or scoring + Perlan paint were the most effective treatments for branch induction. Disbudding + painting the disbudded area with Perlan was ineffective; the likely reason was that bud removal did not create enough damaged tissue area to permit the entry of sufficient cytokinin to overcome apical dominance.

Two-year-old limb sections of third-leaf ‘Sweetheart’/Mazzard trees were treated at green-tip with applications of thidiazuron (TDZ), a powerful cytokinin. Applications up to 500 ppm in 0.5% v/v Regulaid were made with a paintbrush on the surface of the bark. None of the treatments induced lateral branch development.

Nursery trees of ‘Skeena’/G.6 and ‘Skeena’/Mazzard were planted at 2.5x13 feet in 2005. The Mazzard trees had been headed by the nursery, and the G.6 trees were left unheaded. Trees were either not pruned further in the orchard or they were scored with 2 scoring cuts + Perlan 5000 ppm paint or notched 3-5 times + Perlan 5000 ppm paint. The plan is to repeat these branching treatments to observe canopy development and in particular to examine the possible benefit of the precocious G.6 rootstock on early cropping.

b. Other trials

Lateral branching was induced on one-year wood on third-leaf ‘Skeena’/G.6 trees in 2004 using the scoring + Promalin in latex paint technique. In 2005, those one-year-old lateral shoots bore flower buds and produced fruit (Fig. 1), demonstrating the potential for combining lateral branch induction with a precocious rootstock to increase early yield in sweet cherry. In comparable trees of ‘Skeena’ and other cultivars on Mazzard rootstock in the same orchard, successful branch induction in 2004 was not followed by flowering and fruiting on those induced shoots in 2005.

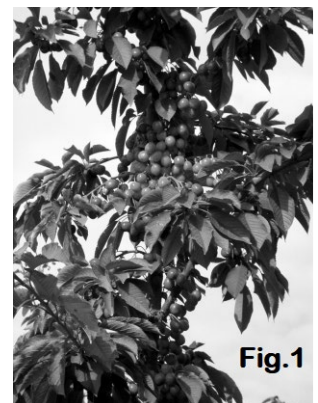


Fig.1

Methods:

Six trials were initiated in the 2005 growing season to test effects of cytokinin-containing bioregulator products on stimulation of lateral branch development in sweet cherry trees at green-tip. These trials were developed as the result of previous observations about the necessity of bark injury as a means to improve the efficiency of cytokinin-induced bud growth stimulation where lateral branching is desired in a limited pruning context.

Results and discussion:

Mixtures of benzyladenine (BA) and gibberellins (such as Promalin or Perlan) have not proven very effective for inducing lateral branching in sweet cherry when painted on buds on shoots at green-tip. Our results have demonstrated that this observation has more to do with lack of uptake or penetration of the bioregulator products with conventional application methods than with a natural lack of responsiveness of the sweet cherry tree to such treatments if the bioregulators are provided a path of entry into the tree. Results from this year's trials demonstrated clearly that it is the outermost layer of the bark that serves as the barrier to entry of cytokinin when this product is applied to the bark. Once that layer has been interrupted, either by scraping or making a cut, the bioregulator treatment will be effective. However, the movement of the cytokinin bioregulator product from the point of entry is limited, so several cuts must be made and treated along the length of any one-year-old shoot in order to assure a well-distributed pattern of lateral shoot development. A new cytokinin, TDZ, was tested in a preliminary trial to determine if this product could induce lateral bud development into shoots on 2-year-old wood. No bark injury was included, and no bud induction was observed.

Acknowledgments:

The assistance and support of the following persons and organizations is gratefully acknowledged: Noel Adkins, Dave Chisholm, Joel Hand, Dr. Chris Ishida, Pete Savage, Tim Scott, Les Woda, Dovex Orchards, Fine Americas, Johnny Appleseed Orchards, Savage Orchards, Scott Orchards, Valent BioSciences, and the Washington Tree Fruit Research Commission.

Budget:

Project title: Induction of branches in sweet cherry trees in the orchard and nursery

PI: Don C. Elfving

Proposed project duration: Three years (2005-2007)

Current year: 2006

Project total (3 years): \$35,367

Current year request: \$11,873

Year	Year 1 (2005)	Year 2 (2006)	Year 3 (2007)
Total	10,242	11,873	13,252

Current year breakdown

Item	Year 1 (2005)	Year 2 (2006)	Year 3 (2007)
Salaries (Technical) ¹	4,200	4,368	4,543
Benefits ² (34% - yrs 2 & 3)	1,302	1,485	1,545
Wages (Time-slip) ¹	1,500	2,000	2,400
Benefits ² (11%-yrs 2 & 3)	240	220	264
Equipment	0	0	0
Supplies ³	500	800	1,000
Travel ⁴	2,000	2,500	3,000
Miscellaneous	500	500	500
Total	10,242	11,873	13,252

¹ Technical and time-slip help to set up trials, apply treatments and collect data as needed.

² Salaries benefit rate for year 1 is 31%. Years 2 and 3 benefit rate is calculated at 34%. This increase is due to an increase in contribution made by WSU on behalf of the employee. Time-slip benefit rate for year 1 is 16%. Benefit rate for years 2 and 3 is calculated at 11%. The change is due a change in policy at WSU.

³ This category includes a variety of miscellaneous supplies, non-capital equipment, consumables, etc. that are needed to carry out the research project. Cell phone charges are allowable under this grant.

⁴ Travel to distant research sites is expensive. These funds will be used to defray costs of vehicular operation, maintenance, and personnel travel costs for travel for Dr. Elfving, Mr. Visser and their employees to research plots in grower-cooperator orchards throughout the south-central and north-central Washington fruit production areas.

CONTINUING PROJECT REPORT

YEAR 2/3

WTFRC Project # CH-04-405

WSU Project #13C-3655-7298

Project title: Bioregulator uses for managing growth, flowering and cropping
PI: Don C. Elfving, Horticulturist
Organization: WSU Tree Fruit Research and Extension Center, Wenatchee, WA

Cooperators: Eugene M. Kupferman, Extension Horticulturist, WSU-TFREC, Wenatchee, WA
James R. McFerson, Horticulturist and Manager, Washington Tree Fruit Research Commission, Wenatchee, WA
Tom Auvil, Horticulturist, Washington Tree Fruit Research Commission, Wenatchee, WA

Matthew D. Whiting, Assistant Horticulturist, WSU-IAREC, Prosser, WA
Tory Schmidt, Agricultural Technician, Washington Tree Fruit Research Commission, Wenatchee, WA

Dwayne B. Visser, Agricultural Research Technologist II, WSU-TFREC, Wenatchee, WA

Contract administrator: Mary Lou Bricker (mdesros@wsu.edu) (509) 335-7667; or Tom Kelly (kellytj@wsu.edu) (509) 335-3691

Reporting Period: 2005

Objectives:

1. Continue to develop improved recommendations for the use of ethephon (Ethrel®, Bayer CropScience) for stimulation of flowering and early fruiting in important sweet cherry cultivars on seedling rootstocks in standard and high-density plantings.
2. Examine the possibility that ethephon treatment for flowering can improve flowering and yield for more than one year after treatment.
3. Explore the potential for use of gibberellic acid (GA₃) as a strategy to reduce flowering the year following treatment on mature sweet cherry trees grown on size-controlling rootstocks where excessive bloom makes crop load control critical for production of fruit of required size and quality.
4. Determine if it is economically feasible to use a single treatment program of GA on cherries to simultaneously obtain both better fruit firmness and quality in the treatment year and also control flowering for the subsequent year as a tool to adjust crop load to benefit fruit quality the next year.
5. Explore in greater detail promising results of preliminary research with SmartFresh® (1-methylcyclopropene, MCP) applied to sweet cherry trees in conjunction with ethephon for loosening fruit for mechanical harvest while reducing negative ethephon effects on fruit quality.
6. If any additional new fruit-abscission products become available, initiate tests for efficacy in loosening sweet cherries while examining effects on fruit quality.

Significant findings:***a. Control of flowering and fruit quality with gibberellic acid***

1. GA₃ and GA₄₊₇ were applied as double applications in 2004 to test control of flowering in 2005 as a strategy for crop quality improvement. Both GA products reduced return bloom in 2005 roughly in proportion to concentration. Applications of 200 ppm of GA₄₊₇ twice reduced return bloom by about 60% while 200 ppm of GA₃ twice was more effective, reducing return bloom by about 90%.
2. Yield in 2005 was reduced in approximate proportion to the reduction in return bloom; the high concentration of GA₃ reduced yield far too much to represent a practical approach.
3. Mean fruit weight was unaffected but fruit firmness was higher in the lower-yielding treatments; brix was not affected by treatment in 2005.
4. Lower yield in 2005 due to reduced bloom was not reflected in an improvement in row-size distribution of the fruit.

b. Ethephon for stimulation of flowering

1. In 2004, ethephon at 300 or 400 ppm was applied three times at 2-week intervals to third-leaf 'Lapins'/Mazzard trees that were vigorous but totally vegetative. There was no evidence of gummosis on any of the treated trees in 2004.
2. Shoot growth was strongly affected by the triple-spray ethephon program. Significant reduction in internode elongation was evident during the period in which ethephon sprays were being applied, but no other response (e.g., lateral branching) was observed.
3. In spring, 2005, some of the trees produced a small amount of bloom. There was no beneficial effect of any of the 2004 ethephon treatments on flowering in 2005.
4. The results of the research with ethephon on Mazzard-rooted cherry trees have been discouraging. The response has varied from the occasional strong promotion of flowering to the more commonly observed minor effect or total lack of effect. Evidently the juvenility factor created by the use of the seedling rootstock is extremely difficult to overcome with a few ethephon sprays. At this point this aspect of this project will be de-emphasized.

c. Fruit loosening and fruit quality effects from ethephon and MCP

1. For the implementation of mechanical harvesting in sweet cherry or to aid hand-harvest of this crop, loosening the fruit from the pedicel must be accomplished by applying ethephon a few weeks before harvest. Unfortunately, ethephon application also accelerates loss of fruit firmness, a key factor in the durability and quality of the fruit after harvest.
2. MCP is an inhibitor of ethylene action. In 2003, spraying cherry trees 2 weeks before harvest with the standard SmartFresh formulation resulted in fruit that was firmer than untreated fruit at harvest; MCP also inhibited the flesh softening otherwise normally associated with ethephon treatment. This exciting development created the impetus for further research.
3. In 2004, application of a sprayable formulation of MCP failed to beneficially affect fruit flesh firmness or other characteristics. It was deemed possible that something in the formulation or the rates of application made the treatments ineffective.
4. In 2005, yet another modification of the sprayable MCP approach was tried in conjunction with ethephon for loosening of sweet cherries. Again, as in 2004, the ethephon program worked as expected but the MCP program did not demonstrate any beneficial effects.

Methods:

One trial from 2004 was carried over in 2005 to evaluate return bloom responses to ethephon applied in 2004. One trial was carried over from 2004 to evaluate the effects of GA on crop load and fruit quality, especially size, the following season. New trials were established in 2005 to 1) examine fruiting and return-bloom responses to applications of gibberellic acid and 2) evaluate effects of MCP and ethephon on fruit loosening and fruit quality for mechanical harvest.

Results and discussion:

GA applied during Stages I and II of sweet cherry fruit development in 2004 reduced flowering in 2005 on 'Bing' trees on G.1 size-controlling rootstock. As one might expect, this effect was dependent on both GA isomer composition and concentration. The GA₃ formulation is considerably more effective at suppressing bloom in sweet cherry than the GA₄₊₇ formulation. The reduction in flowering was accompanied by a proportional reduction in yield that was also concentration-dependent. The data suggest that fruit set was similar on all treatments. In 2005, neither mean fruit size nor brix was affected as yield decreased. The dramatic reduction in both flowering and yield in response to GA applications demonstrates clearly that this tool, if it is to be used effectively, must be employed with considerable care. If the use of GA is to be implemented on a practical scale, it should be considered as only a part of a multi-faceted crop-load management strategy that relies in part on pruning and perhaps in part on bloom thinning or other techniques.

On the other side of the question, using ethephon to stimulate flowering in young trees on seedling rootstocks has not proven to be a reliable strategy. Part of this result may well be due to the specific characteristics of the seed sources used for rootstocks. Since each seed is genetically different, the likelihood is that the degree of juvenility varies slightly from tree to tree, which can easily complicate the goal of uniformly stimulating earlier flowering. So far, the use of higher ethephon concentrations and multiple applications has not proven to be an effective strategy for overcoming the rootstock-induced juvenility effect.

The results this year with MCP were again disappointing. The formulation used this year was an experimental product that was changed from the product used in 2004. On the positive side, trials with sprayable MCP on apple have produced excellent results. It appears possible that application technology may play a major role in the efficacy of sprayable MCP treatments.

Acknowledgments:

The assistance and support of the following persons and organizations is gratefully acknowledged: Tom Auvil, Randy Brown, Mark Bell, Nancy Buchanan, Felipe Castillo, Chris Olsen, Dennis Hayden, Jeff Henry, Dr. Chris Ishida, Heidi Künzel, Dr. Gene Kupferman, Olivia Lenahan, Dr. Jim McFerson, David Ophardt, Chris Sater, Tory Schmidt, Dwayne Visser, Dr. Matt Whiting, Bayer Environmental Science, Brewster Heights Packing, Inc., Hayden Orchards, Valent BioSciences, and the Washington Tree Fruit Research Commission.

Budget:

Project title: Bioregulator uses for managing growth, flowering and cropping and for facilitating mechanical harvesting in sweet cherry

PI: Don C. Elfving

Project duration: 2004-2006 (three years)

Current year: 2006

Project total (3 years): \$42,918

Current year request: \$15,526

Year	Year 1 (2004)	Year 2 (2005)	Year 3 (2006)
Total	13,210	14,182	15,526

Current year breakdown

Item	Year 1 (2004)	Year 2 (2005)	Year 3 (2006)
Salaries (technical) ¹	7,000	7,280	7,571
Benefits ² (34%, yr 3)	1,890	1,966	2,574
Wages (time-slip) ¹	1,000	1,100	1,100
Benefits ² (11% yr 3)	160	176	121
Equipment	0	0	0
Supplies ³	1,160	1,160	1,160
Travel ⁴	2,000	2,500	3,000
Total	13,210	14,182	15,526

¹ Technical and time-slip help is essential to collect the volume of data needed to evaluate growth, flowering, yield, fruit loosening and fruit quality responses to the various bioregulator applications involved.

² Salaries benefit rate for years 1 and 2 is 27%. Year 3 benefit rate is calculated at 34%. This increase is due to increase in contribution made by WSU on the behalf of the employee. Time-slip benefit rate for years 1 and 2 is 16%. Year 3 benefit rate is calculated at 11%. The change is due to a change in policy at WSU.

³ This category includes miscellaneous supplies, non-capital equipment, consumables, repairs, etc. that are needed to carry out the research project. Cell phone charges are allowed under this grant.

⁴ Treatment application and frequent data collection in distant sites, e.g. Pasco, Prosser, Yakima, Brewster, Cashmere, Orondo, Quincy, etc. Includes vehicle lease-to-purchase, operating, repair costs.

Project title: Edible coating to improve storage and marketing quality
PI: Jinhe Bai
Project Staff: Kristi Barckley (Research Assistant) and Debra Laraway (Technician)
Organization: Oregon State University,
Mid-Columbia Agricultural Research and Extension Center
Address, phone, e-mail: 3500 Experiment Station Dr, Hood River, OR 97031
(541) 386-2030 E-mail: Jinhe.bai@oregonstate.edu
Co-PI: Anne Plotto, USDA/ARS, Citrus and Subtropical Products Lab
Winter Haven, FL 33881
Contract Administrator: Beaton, Dorothy A; Dorothy.Beaton@oregonstate.edu;
(541) 737-3228

Objectives (2005)

1. To develop fruit and stem coatings. To develop and evaluate coatings with varying degrees of permeability to water vapor, O₂ and CO₂.
2. To use plant growth regulators and antioxidants to prevent stem shrinkage and stem browning. To add plant growth regulators (gibberellins), antioxidants (ascorbic acid), and minerals (Ca⁺⁺ salts) to edible coatings; to evaluate the additional efficiency of these compounds to extend the postharvest life of cherries.
3. To understand the relationship between the fruit and the stem. After harvest, the fruit may act as a strong sink for the water stored in the stem.
4. Optimization of clamshell container openings to decrease moisture loss of fruit.

Objectives (2006)

1. To develop fruit and stem coatings. To develop and evaluate coatings with varying degrees of permeability to water vapor, O₂ and CO₂.
2. Optimization of clamshell container openings to decrease moisture loss of fruit.
3. Using ethanol release powder to maintain fruit and stem quality.
4. Effect of postharvest handling on antioxidant capacity of cherries.

Significant findings:

- The efficiency of coatings to reduce postharvest moisture loss of cherries is coating formulation and fruit variety dependent.
- Sucrose fatty acid ester was the only coating that significantly improved shininess of cherry fruit.
- Chitosan coatings maintained fruit firmness and stem retention better than control and other coatings.
- Ca propionate dips helped maintain fruit firmness but CaCl₂ did not.
- Peroxyacetic acid, a sanitizer, maintained better fruit quality than control and other dipping treatments.
- For stem coatings, paraffin + polyethylene decreased water loss and browning, decreased stem detaching, and water loss. However, other film forming formulations did not affect stem quality. GA₃ dips slowed down stem browning of 'Bing' cherries.
- An experimental clamshell with smaller openings than the commercial clamshell decreased moisture loss of cherry fruits and doubled the shelf-life.
- An ethanol release powder maintained better stem quality of 'Lapins'.

Methods:

- Coating formulation: Chitosan, candelilla, carnauba, shellac, and paraffin coatings were formulated by OSU-MCAREC & USDA/ARS-Citrus and Subtropical Products Lab for cherry fruit and/or stem coating.
- Commercial cherry coatings from Agricoat, Pace, Deco, and FMC, were also evaluated.
- Other dipping treatments: growth regulators (GA3), antioxidants (ascorbic acid, acetyl cysteine, hexylresorcinols), and sanitizers (peroxyacetic acid and acetic acid) to inhibit stem browning.
- Clamshell development and evaluation: developing new clamshell to improve moisture-retaining property of packaging.
- Ethanol-release pad: A Freund Industrial (Japan) product. The pad was glued on the top lid of clamshells.
- Fruit attributes: Water loss, fruit surface color and gloss, fruit firmness, soluble solids content (SSC) and titratable acidity (TA), antioxidants, stem detachment force, and appearance of stem and fruit were determined during storage at 33 °F with two or three-week intervals. Appearance of stems was scaled using a 5-point scale where 1= clear, 2= <25 % browning, 3= <50% browning, 4= <75 browning, 5= >75% browning of whole stem length. Appearance of fruit was scaled using a 5-point scale where 1= fresh, excellent; 2= fresh, good; 3= fair, market limit; 4= poor; and 5= completely deteriorated.

Results and Discussion

Fruit coating and other dipping treatments: Treatments that showed good results last year were evaluated again this year. After the packing process, fruits were dipped in chitosan coating (dissolved in acetic acid), sucrose fatty acid ester coating, Ca propionate (firming agent), or peroxyacetic acid (sanitizer), respectively, and then stored at 1°C for 2 weeks. Fruit quality was evaluated directly after cold storage and then after one day of being held at room temperature to simulate commercial situation.

Sucrose fatty acid esters, calcium propionate and peroxyacetic acid reduced moisture loss of fruit. However, only peroxyacetic acid significantly improved shelf life of cherries. This contrasts with last years' results, which showed sucrose fatty acid esters and calcium propionate to be promising for cherries. Ca propionate also did not improve stem retention. Ca^{++} has been applied in horticultural crops preharvest and postharvest to improve the postharvest stability of produce (Patten et al., 1983). Our research last season suggested that Ca propionate maintained better stem quality, but further investigation this year does not confirm this.

Peroxyacetic acid extended both stem and fruit life for 2-3 days. The results suggest that quality loss of cherries may be related to microbial contamination, even without visible decay (Table 1).

Stem coating and other dipping treatments: Cherry stems were dipped in chitosan, GA3, paraffin + polyethylene, carnauba or shellac coating/solution, respectively, using a screen system which holds the fruit when stems are in the solution. Paraffin + polyethylene coating decreased water loss and browning, and prevented stem detaching (Table 2). Wax layer from the paraffin and carnauba coatings are clear under microscope. Chitosan caused structural change of the stems (data not shown). We sampled the stems during storage, and will analyze by electron microscopy to see whether coatings effected cell structure. Thus, we will develop coatings based on the stem structure.

Clamshell: We developed a couple of new experimental clamshells. Dimensions of clamshells are confidential pending approval from OSU technology transfer. The capacity of the experimental clamshell was 3.3 pints, as a comparison, a typical commercial clamshell was used which has a capacity of 2.4 pints.

We compared the different clamshells for moisture loss, fruit quality and stem quality of the cherries. We packed ‘Lapins’ cherries in these clamshells and stored them at 33° F for 3 weeks. The critical point at which fruits and vegetables deteriorate due to water loss is at 5%. Water loss of cherries packed in the experimental clamshells did not exceed 5% within 3 weeks of harvest. However, the commercial clamshell reached that critical point after 2 weeks (Fig. 1).

The combination of gases (O₂ and CO₂) in the commercial clamshell is similar to the ambient air because of the huge opening between the top and bottom lids. This opening causes cherry fruits to deteriorate and breakdown after 2-weeks. O₂ is slightly lower and CO₂ slightly higher in the experimental clamshell, which created a favorable environment for the cherries to last up to 3-weeks after harvest. Fruit flavor was not affected by the change in O₂ and CO₂ levels at low and room temperatures.

Bai et al. (1990) reported that Hassaku orange packed in low perforation (0.16%) polyethylene bag had less weight loss, decay and physiological disorders as compared to high perforation (1%) bag.

Ethanol release powder: Ethanol-release powder (Antimold Mild®, Freund Industrial, Japan) allows ethanol vapor to diffuse gradually. It is made from ethanol absorbed onto silica gel that is packed in a special film, laminated with ethylene-vinylacetate and Japanese paper, which regulates ethanol diffusion. The ethanol pad was glued on the top lid of clamshells.

Softening of fruit and browning of stems were retarded by ethanol powder (Fig. 2). Ethanol treatment affects ripening and senescence in some fruit and vegetables (Bai et al., 2004; Plotto et al., 2005; Suzuki et al., 2004). Ethanol vapor treatment of tomato fruit suppressed the climacteric respiratory rise, lycopene synthesis, and chlorophyll breakdown. Ethanol injected into the seed cavity of muskmelon and honeydew inhibited softening. Furthermore, ethanol solution prolonged the vase life of cut carnations by suppressing respiration and transpiration.

In conclusion, cherry fruits had a limited shelf-life (2 weeks) in a commercial clamshell with a big opening between the top and bottom lids. An experimental clamshell, with several smaller openings, decreased moisture loss of cherry fruits significantly, and postponed fruit deterioration without creating anaerobic conditions. The experimental clamshell doubled the shelf life of cherry fruits compared with the commercial clamshell. The experimental clamshell together with ethanol release powder further extended shelf-life of sweet cherries.

Further research will include continuing to explore the best combination of opening ratio of the experimental clamshells and ethanol release powder.

Literature cited:

Bai, J., Abe, K., Kurooka, H. 1990. Effect of harvest maturity, perforation ratio of polyethylene package and storage temperature on quality of Hassaku (*Citrus hassaku* hort. Ex Tanaka) fruit. J. Japan. Soc. Cold Preserv. Food. 16:97—104.

Bai, J., E.A. Baldwin, R. Soliva-Fortuny, J.P. Mattheis and J.K. Brecht. 2004. Effect of pretreatment of intact ‘Gala’ apple with ethanol vapor, heat or 1-methylcyclopropene on quality and shelf life of fresh-cut slices. J. Amer. Soc. Hort. Sci. 129(4):583-593.

Plotto, A., J. Bai, J.A. Narciso, J.K. Brecht, and E.A. Baldwin. 2005. Ethanol vapor prior to processing extends fresh-cut mango shelf-life by decreasing spoilage, but does not always delay ripening. *Postharvest Biol. Technol.* (in press).

Patten, K., Patterson, ME., Kupferman, E. 1983. Reduction of surface pitting in sweet cherries. *Post Harvest Pomology Newsletter*, 1 (2): pp6.

Suzuki, Y., Uji, T., Terai, H. 2004. Inhibition of senescence in broccoli florets with ethanol vapor from alcohol powder. *Postharvest Biol. Technol.* 31: 177-182.

Table 1. Effects of coating and other dipping treatments and ethanol-release pad on fruit and stem quality of 'Lapins' cherries.

Treatment	<u>Weight loss (%)</u>		<u>Respiration (ml kg⁻¹ h⁻¹)</u>		<u>Stem quality index</u>		<u>Fruit quality index</u>	
	Regular	Ethanol	Regular	Ethanol	Regular	Ethanol	Regular	Ethanol
Ca ⁺⁺ + antioxidants.	6.6	5.6	5.8	5.7	0.36	0.57	0.60	0.68
Chitosan	12.5	13.1	6.1	5.7	0.14	0.17	0.27	0.36
Sucrose fatty acid esters	8.8	10.6	6.2	6.7	0.63	0.77	0.51	0.50
Peroxyacetic acid	7.5	10.3	5.9	5.9	0.49	0.79	0.81	0.73
Control	13.1	13.6	5.1	5.1	0.73	0.86	0.64	0.40

Table 2. Effect of stem coatings on stem quality of 'Lapins' cherries. Fruits were packed in commercial clamshells and stored at 33°F for 11 days before evaluation.

Directly after cold storage

Coating	Visible quality index	Water content (%)	<u>Color value</u>		Detachment force (g)
			a*	hue (°)	
Chitosan	0.43	59.0	-1.8	95.4	640
GA3	0.56	55.3	-1.9	96.1	571
parafin+polyethylene	0.74	61.3	-4.5	102.0	697
Carnuba	0.69	57.7	-2.5	98.0	734
Shellac	0.56	60.2	-1.8	96.1	680
Control	0.59	58.5	-3.3	99.5	672

After 24 hrs at 68°F

Chitosan	0.44	57.8	-2.6	97.7	668
GA3	0.60	57.6	-1.4	96.6	648
parafin+polyethylene	0.71	60.8	-3.5	100.0	643
Carnuba	0.53	50.1	-2.2	97.9	602
Shellac	0.78	57.4	-1.5	95.9	600
Control	0.44	56.1	-1.5	95.5	570

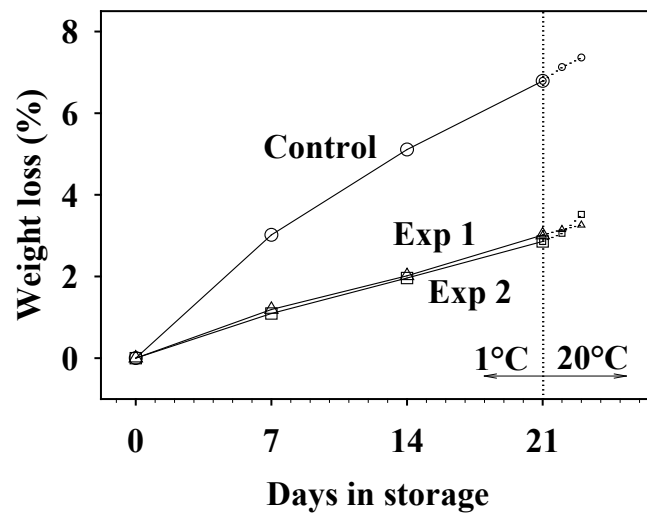


Fig. 1. Effect of clamshell on water loss of sweet cherries 'Lapins' stored at 33°F and then transferred to 68°F.

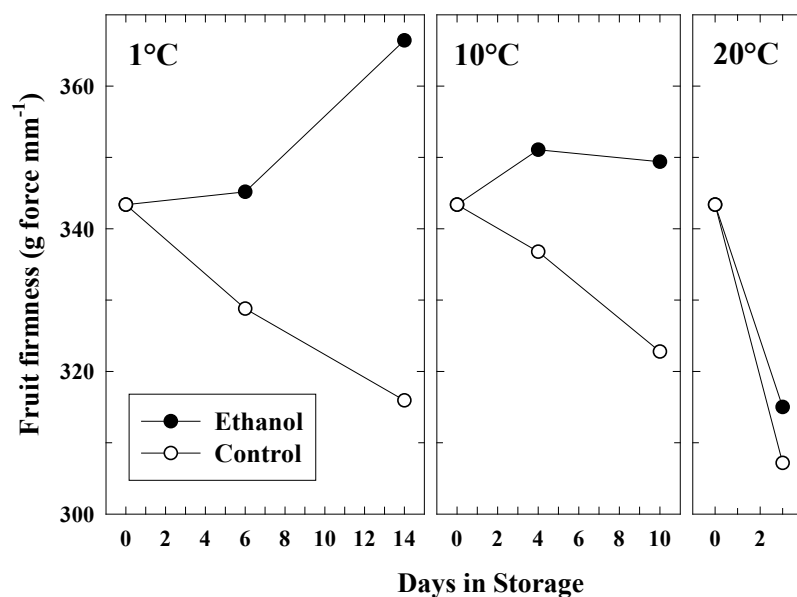


Fig. 2. Effect of ethanol release pad on fruit firmness of 'Lapins' cherries. Fruit were stored at 1°C (commercial ideal temperature), 10°C (abuse cold shelf temperature) and 20°C (abuse shelf and consumer temperature) for up to two weeks.

BUDGET

Project title: Edible coating to improve storage and marketing quality
PI: Jinhe Bai
Project duration: 2004-2006
Current year: 2006
Project total (3 years): \$43,641
Current year request: \$20,943

Item	Year 1 (2004)	Year 2 (2005)	Year 3 (2006)
Salaries ¹		10,200	10,656
Benefits (59%) ¹		4,998	6,287
Wages			
Benefits (%)			
Equipment			
Supplies ²	3,500	3,500	3,000
Travel ³		500	1,000
Miscellaneous			
Total	3,500	19,198	20,943

¹ OSU-MCAREC: \$10,656 – 4-months Faculty Research Assistant time with benefits (59%).

² Supplies include fruits, chemical, and equipment supplies.

³ Travel between Hood River and Winter Haven, FL.

WTFRC Project # CH-05-509

WSU Project #13C-3355-5325

Project title: Improving cherry fruit quality and postharvest shelf life

PI: Larry Schrader

Organization: Tree Fruit Research and Extension Center, Wenatchee, WA 98801

Cooperators: Jianshe Sun, Jun Tian, and Yiping Gong
Tree Fruit Research and Extension Center, Wenatchee, WA 98801

Contract administrator: Mary Lou Bricker (mdesros@wsu.edu) (509) 335-7667; or Tom Kelly (kellytj@wsu.edu) (509) 335-3691

Objectives:

1. Investigate the effects of formulations on stem browning and water loss of cherries after harvest. Study rates and timing for applications of the formulations.
2. Compare water loss in stem-free cherries to water loss in cherries with stems.
3. Apply gibberellic acid at different stages of maturity and study its effect on fruit quality.
4. Study efficacy of GA when tank mixed and applied with RainGard.
5. Conduct microscopic studies to determine anatomical differences among cultivars that differ in their susceptibility to rain cracking.
6. Conduct further studies to improve efficacy of RainGard, cherry cracking suppressant.

Significant findings:

1. Cracking was decreased by 47%, on average, with four weekly applications of RainGard in three orchards in The Dalles, Oregon.
2. For Sweetheart, four RainGard applications at weekly intervals decreased cracking by 38%.
3. With Tieton, four RainGard applications decreased cracking by 35%.
4. Studies with stem-free and normal cherries to decrease water loss and stem browning during storage show promise but need further investigation.

Methods:

- **Objectives 1 and 2:** Two formulations were used initially for these studies. One has been reported to delay stem browning in cherries. The other shows promise of decreasing water loss from cherries after harvest. These two formulations were sprayed on trees and also used for a postharvest treatment. Stem browning and water loss in cold storage were evaluated.
- **Objectives 3 and 4:** Initial experiments to test efficacy were done on single trees in a two-way factorial randomized complete block with four replications. GA was applied alone at different concentrations and also tank mixed with RainGard before application. Applications were made at different intervals prior to maturity.
- **Objective 5:** Digital images were taken with a Nikon SMZ-U dissecting microscope to observe differences in the structure of the stylar scar end of each of several cultivars.
- **Objective 6:** In 2005, thirteen grower/cooperators were selected for efficacy testing of RainGard, a new experimental product to protect cherries from cracking. Locations of these test sites varied widely from Kennewick and Pasco, Washington, on the east to Tonasket, Washington, on the north and to The Dalles, Oregon, on the west. Sufficient rain to cause measurable cracking occurred at only six sites. The

predominant cultivar studied was Bing although at least one trial included Staccato, Sweetheart, Rainier or Tieton. Quality data (fruit weight, color, firmness, soluble solids and titratable acidity) were collected on fruit from 10 of the 13 trials.

All treatments were applied by grower/cooperators. The four treatments for every trial were as follows:

- A. 10% (v/v) RainGard, two applications—at straw color of fruit (or slightly earlier if rain was imminent) and two weeks after the first application;
- B. 5% (v/v) RainGard, two applications with same timing of applications as with treatment A;
- C. 5% RainGard, four weekly applications—first application same as above, and then weekly thereafter;
- D. Untreated control (i.e., no application of RainGard).

Results and discussion:

Objective 1: We compared RainGard and RainStop alone and in combination in three orchards. Cracking occurred in only one orchard, but quality, stem browning and water loss during storage were evaluated in all three. No quality differences were observed except in one trial (Bing) in which soluble solids were higher in the untreated control than in other treatments. In that same Bing trial, stem browning and water loss were significantly higher than at the other two sites. In a trial with Sweetheart, stem browning was highest in the control. The mixture had more stem browning than RainGard or RainStop alone. More work is needed on this issue.

A postharvest test was done with Rainier cherries by dipping them in the treatment for one minute and then transporting them to the lab to record weights and stem browning. Cherries were stored at 34°F for 4 weeks and then evaluated again. Little effect was observed between RainGard, RainStop, a mixture of the two, and the untreated control.

Objective 2: In cold storage, water loss in stem-free Sweetheart cherries as well as stemmed cherries was reduced significantly by RainGard. Results are promising and will be repeated.

Objectives 3 and 4: GA, RainGard alone, and RainGard + GA were applied to Rainier and Bing to determine if GA and RainGard are compatible. No significant differences were observed among the treatments for soluble solids, fruit weight, firmness, titratable acidity or color. More work is planned for next year.

Objective 5: We previously observed that the junction between the stylar scar tissue and the cuticle appears to be open in Bing cherries, partially open in Van and closed in Lapins. “Conductive” tissue appears to be more pronounced in Bing, somewhat less in Van and even less apparent in Lapins. Tieton’s anatomy seems similar to Bing and may account for its susceptibility to cracking. Rainier cherries were also examined in this manner but showed a tight junction between the stylar scar and the cuticle. We have observed that the suture of Rainier cherries is especially susceptible to cracking, but this needs more examination.

Objective 6: To test efficacy, RainGard was applied to Bing cherries in three orchards near The Dalles, Oregon. The mean of all three trials is shown (Fig. 1). Total cracking in all three RainGard treatments was significantly lower than in the untreated control (D). Treatment C (weekly applications) had significantly less cracking than the other three treatments.

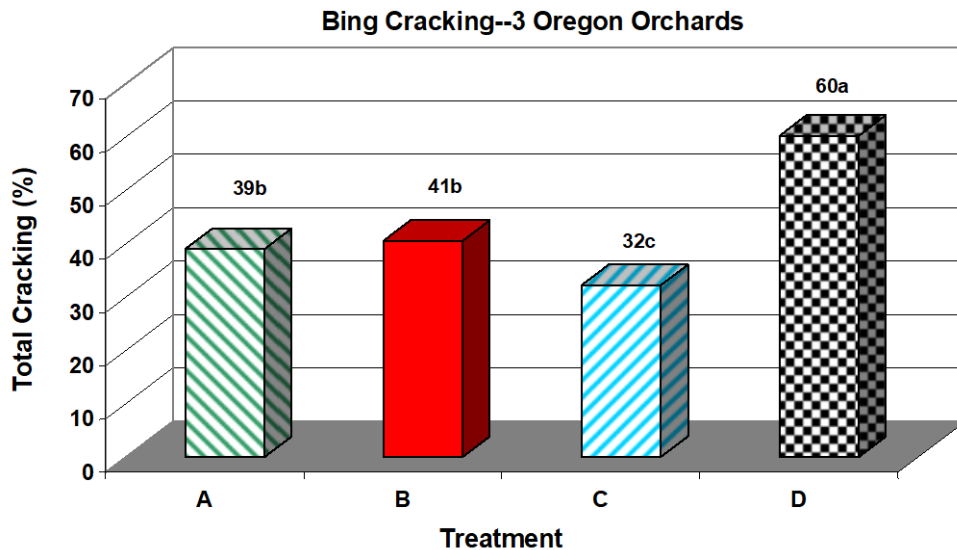


Fig. 1. Total cracking of Bing cherries averaged from three orchards near The Dalles, Oregon. Three RainGard treatments are compared to the untreated control (Treatment D). See methods for description of RainGard treatments. If the number above a bar within the graph is followed by a letter that is different from that above another bar, that bar is significantly different ($P < 0.05$) than the other.

Rainier and Bing were compared in one Oregon orchard, and cracking was significantly higher in Rainier than in Bing with all four treatments (Fig. 2). With Bing and Rainier, all RainGard treatments significantly decreased cracking as compared to the untreated control (D). Cracking in Bing was lowest again in Treatment C.

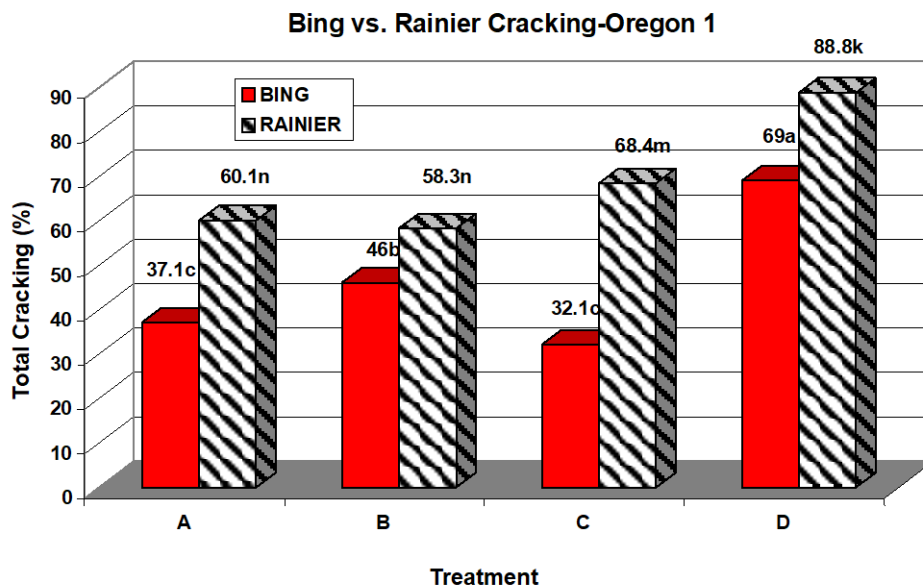


Fig. 2. Comparison of cracking in Bing and Rainier sweet cherries in an Oregon orchard with three RainGard treatments versus an untreated control (D).

With Sweetheart cherries, cracking was significantly lower in Treatment C as compared to other treatments (Fig. 3). With Tieton cherries, cracking was also significantly lower in Treatment C as compared to all other treatments (Fig. 4).

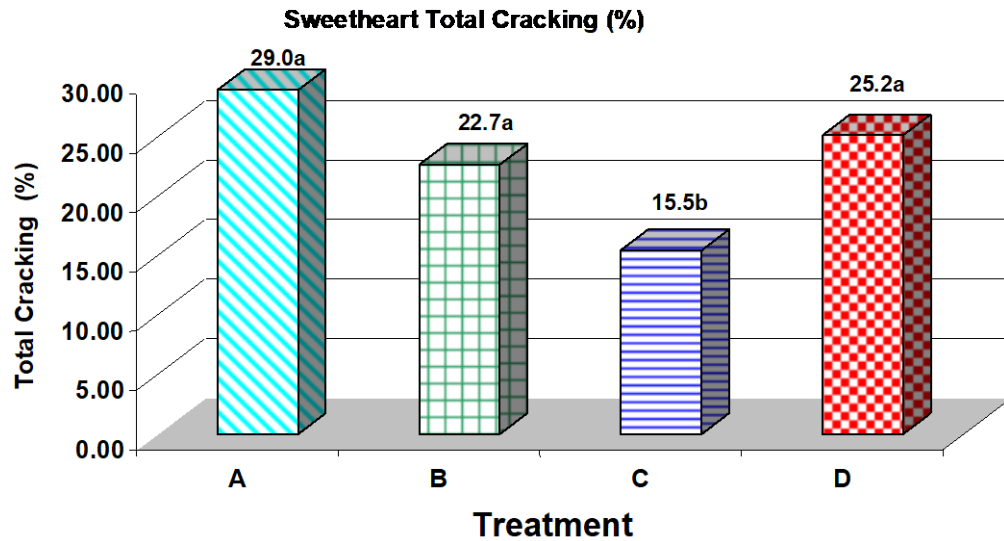


Fig. 3. Total cracking of Sweetheart cherries treated with three RainGard treatments versus an untreated control (D).

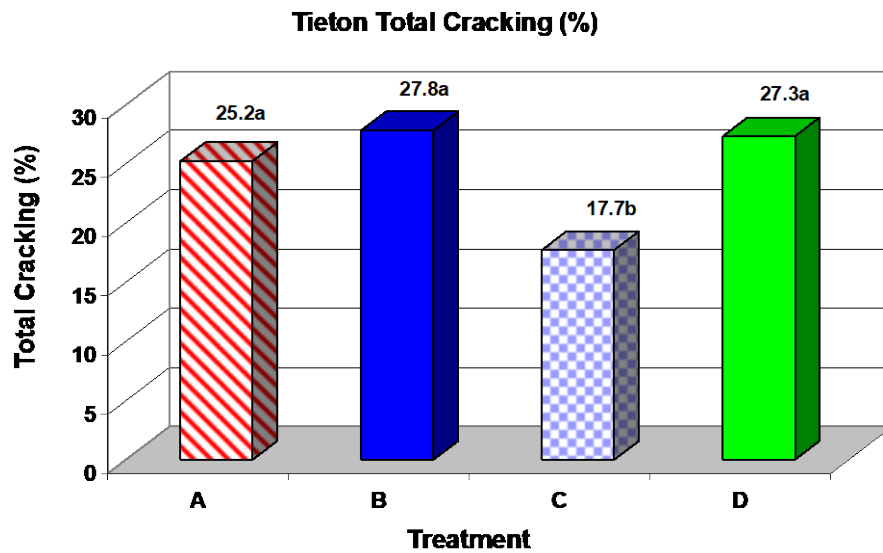


Fig. 4. Total cracking of Tieton cherries treated with three RainGard treatments versus an untreated control (D).

The results indicate that more frequent applications (Treatment C) provided better protection from rain. The surface area of the cherry expands rapidly during the last few weeks of development and within a few days causes the protective film on the cherry to become less effective in protecting from the rain. Weekly RainGard applications maintain the protective film for better protection from rain.

The quality analyses completed on 10 trials showed no appreciable differences among the treatments in any cultivar. This included fruit size, color, titratable acidity, soluble solids and firmness.

Budget:

Project title: Improving cherry fruit quality and postharvest shelf life
PI: Larry Schrader
Project duration: Two years (2005-2006)
Current year: Year 2 (2006)
Project total (2 years): \$41,403
Current year request: \$20,944

Year	Year 1 (2005)	Year 2 (2006)
Total	20,459	20,944

Current year breakdown

Item	Year 1 (2005)	Year 2 (2006)
Salaries ¹	10,608	11,032
Benefits (38%) ²	4,031	4,192
Wages ³	2,000	2,000
Benefits (11%- yr 2) ²	320	220
Equipment		
Supplies ⁴	3,000	3,000
Travel ⁵	500	500
Miscellaneous		
Total	20,459	20,944

¹ Salary requested for a new Associate in Research (25% time).

² Salaries benefit rate for years 1 and 2 is calculated at 38%. Time-slip benefit rate for year 1 is 16%. Benefit rate for year 2 is calculated at 11%. The reduction is due to a change in policy at WSU.

³ Time-slip help to assist with experiments.

⁴ Supplies include chemicals, cell phone charges, other general supplies, and payment for "crop destruct".

⁵ Travel to experimental plots.

NOTE: WSU is including the following information on other funding available for the support of similar research undertaken by the faculty member proposing this research. These resources are listed to identify other support granted for this research and are not included as a commitment of cost-share by the institution.

OTHER FUNDING: FruitGard and Pace International provided over \$21,000 for Objective 6 (efficacy testing of RainGard).

CONTINUING PROJECT REPORT
WTFRC Project: CH-04-406

YEAR 2/3
WSU Project: 3361-4795

Project title: Modeling and managing powdery mildew of sweet cherry
PI: Gary G. Grove and R.A. Spotts
Organizations: Washington State University and Oregon State University
Cooperators: Mike Bush and Tim Smith

Contract Administrator: Mary Lou Bricker (mdesros@wsu.edu; 509-335-7667) or Sharon Taff (staff@wsu.edu; 509-786-2226)

Objectives:

- I. Develop a risk index model (utilizing rainfall, irrigation, temperature, relative humidity, and pathogen presence/activity) for initiating fungicide spray programs and adjusting subsequent spray intervals. Research in 2006 will focus on disease development and spore production over a more broad range of relative humidities at 15-25 C and the elucidation of the effects of high temperatures on powdery mildew colony expansion and spore production.
 - II. Develop means of detecting, identifying, and quantifying airborne propagules of *P. clandestina* early in epidemic progress. Proof of concept was demonstrated in 2004 and 2005. In 2006 this information and other model components will be used to guide the initiation of a fungicide spray program. Results will be compared with a standard phenology-driven program.
 - III. Develop and refine economically viable conventional and organic powdery mildew management programs.
 - IV. Determine the effects of temperature and wetness on acute petroleum oil phytotoxicity. Determine the chronic effects of oils on tree health.
- Objective de-emphasized in 2005 due to insufficient funding:*
- V. Develop baseline sensitivities for resistance-prone compounds. Preliminary studies focused on the DMI fungicides. Future studies will concentrate on Qol and quinoline fungicides.

Significant Findings:

- Investigations on the temperature *range* where the cherry mildew fungus is active on cherry foliage were repeated and expanded in 2005. Disease develops between 10 (50 F) and 27.5 C (81.5 F). Disease did not develop at 7.5 (45.5 F) and 30-35 C (86-95 F).
- Results have been used to form a major component of the Cherry Powdery Mildew Risk Assessment Model. The model *approach* is based on the Gubler-Thomas Grapevine Powdery Mildew Risk Assessment Model but generated using temperature algorithms derived from our controlled-environment research with cherry mildew. This component of the cherry model generates a risk index (*for secondary infection*) value that is used to adjust spray intervals once primary infection has occurred. Values can range from 0 (low risk) to 100 (high risk). Risk indices are calculated daily. The rules for the *beta* version of the model are:

If > 2.5 mm rain occurs (after primary infection), subtract 10 index points

If > 6 hours at 15-25 C (59-77 F) add 20 index points (three *consecutive* days of these conditions are required to initiate the index)

If >6 hours at 27.5 C (81.5 F) then subtract 10 points

If none of the above is true, subtract 10 index points

- The beta version of the model was used to schedule orchard fungicide applications in 2005. The timing of the model-driven sprays was different than the phenology-driven sprays but resulted in the same level of disease control. Mildew severity was 4.7%, 6.1%, and 31.4% in the model, phenology, and unsprayed treatments.
- More in-depth studies of the effects of relative humidity (at the optimum temperature of 20 C (70 F) on disease severity and spore production were conducted in 2005. Disease severity on inoculated leaf disks ranged from 61% - 63% between 50% and 85% relative humidity respectively. Severity levels were 69%, 75%, 87%, 86%, and 87% at 92.5, 96, 97, 98, and 100% RH, respectively.
- Relative humidity had a significant effect on spore production at 20 C (70 F). Spore production ranged from 9.0×10^3 conidia/ml at 50% to 1.4×10^4 at 100% relative humidity. Germination was best described by the equation $(\log) Y = 8.4 + 0.01X$ (where Y = spore production and X = RH at 20 C) with an R^2 of 0.72.
- Cleistothecia (the primary inoculum supply) viability declined from 60% at bud burst to 0% about 1 week after pit hardening. For the second year, the degradation of the ascospore supply required about 155 cumulative degree-days > 10 C (50 F).
- * The PCR assay (using primers developed by the R.A. Spotts group) was found to be extremely sensitive: it detected DNA extracted from 1-5 conidia of *P. clandestina* placed directly into reaction mixtures and from 250-500 conidia placed on glass rods used in air sampling studies.
- * For the second year, the PCR assay facilitated the detection of low levels of *P. clandestina* inoculum in air samples within hours of collection in field studies *prior to disease onset*. Air sampling results also confirmed the presence of ascospores in the orchard when their presence was predicted by the temperature/rainfall (primary infection) component of the model.
- * Oil induced phytotoxicity developed on cherry foliage treated with 2% oil and incubated 2 weeks at 7-10 (45-50 F) and 30 C (86 F), but not at 15 C (59 F), 20 C (70 F), and 25 C (77 F). No phytotoxicity resulted from applications of 1% oil regardless of temperature.
- Fungicide programs comprised of Stylet Oil, Pristine, and Quintec provided excellent control of powdery mildew in the orchard, as did the experimental fungicide V-10118. Severity ranged from a low 3.4% in a Pristine/Quintec alternation program to 31.4% in the untreated control. Severity was 7.0% on trees treated with V-10118. Disease severity was 18.4% in the organic (Stylet Oil and Sonata) program. All fungicide program iterations conformed to the new FRAC guidelines for managing resistance to DMI, QoI, and quinoline fungicides.

Methods:

Objective I. a. Development of risk assessment model. *Effect of temperature and relative humidity on colony expansion and foliar disease severity.* Cherry leaf disks (cv. 'Bing') will be inoculated using a suspension of *P. clandestina* conidia and incubated 21 days at relative humidities of 40-100% at temperatures of 40-95 F (5-35 C). The proportion of disk surface area colonized by powdery mildew will be determined 7, 14, and 21 days after inoculation.

Objective I. b. Development of risk assessment model. *Effect of temperature and relative humidity on spore (conidia) production*. Plants with actively sporulating colonies will be rinsed with water to remove all conidia, placed incubated at 10-35 C (50-95 F) for 14 days at 35, 50, 65, 80, or 90, 95, and 100 % humidity. Sporulation will be assessed 7 and 14 days after inoculation as previously described by Chellemi and Marois (1991).

Objective II. a. Development of risk assessment model. *Meteorological factors affecting spring primary inoculum availability*. A degree-day model of spring ascocarp degradation/ascospore load depletion will be developed using historical and new viability and meteorological data from Washington and Oregon. This equation will be used to generate a predicted value of ascocarp viability at given points in phenological time. Ascocarps will be collected from exfoliating bark and senesced leaves in mildew-infested orchard. Ascospore viability will be assessed using FDA fluorescent vital stain. Validation will be accomplished by mathematically evaluating the relationship between observed and predicted values: actual viability numbers will be regressed on predicted viability numbers. An unbiased predictor should have an intercept of 0 and a slope of 1. Coefficients of determination and standard error about the regression curve(s) will also be used to evaluate the relationship between observed and predicted values.

Objective II. b. Development of risk assessment model. *Verifying pathogen presence and activity. Air sampling studies*. Two air sampling methods will be evaluated in the proposed studies. The first group of studies will be conducted using rotary-impaction air samplers fastened to all-terrain vehicles. ATV's will be driven down orchard rows at weekly intervals. At the conclusion of the sampling period, collection rods will be transported to the lab. DNA will be removed from sampling rods. The second group of studies will utilize rotary-impaction devices operated continuously.

Objective II. b. Use of molecular tools for the timely detection of propagules of the cherry mildew fungus. The identification of trapped propagules will require the use of molecular techniques to identify *P. clausenii* trapped from the orchard air. DNA extractions will be performed by modifying manufacturers instructions using a Bio 101 System FastDNA kit. PCR amplification with universal primers will be performed with Pfu polymerase according the recommended instructions. Amplifications will be performed in a total volume of 25µl using three step cycling. PAmplification products will be run on 1% agarose gel at 120 V for one hour, stained with ethidium bromide, and photographed under UV light. Amplification fragment of expected size is interpreted as a positive result. More detailed information extraction procedures have been published.

Objective III. Various fungicide programs will be evaluated using efficacy and relative input cost as measures of usefulness. Various combinations and rotations of DMI, quinoline, strobilurin (QoI), SAR, oil, whey, and sulfur compounds will be applied to Bing, Rainier, Van, Lapins, or Sweetheart cherries and evaluated for efficacy and phytotoxicity. Compounds will be applied in calendar and weather based management programs. Disease incidence and severity will be determined by randomly selecting five terminal shoots from each plot, and picking five leaves from each terminal starting with the last fully open leaf and working down the shoot for a total of 25 leaves per plot. The percentage of the surface area of the underside of each leaf infected by mildew will be estimated and recorded. Data will be subjected to analysis of variance and means separated according to Fisher's PLSD at $P < 0.05$.

Objective IV. Oil phytotoxicity studies. Cherry seedlings will be treated with various concentrations of narrow range petroleum oils and incubated 14 days at temperatures of 5-35 C. at 75% relative humidity. Phytotoxicity will be evaluated by estimating the amount of necrotic tissue per leaf.

Results and Discussion:

Our vision for improved management of powdery mildew of cherries involves the use of a forecasting model that incorporates components to predict the exhaustion of the overwintered inoculum supply (model component A), primary infection (component B, i.e. ≥ 0.1 " of precipitation at ≥ 50 F), disease pressure once primary infection has occurred (secondary infection risk, component C), and initial presence of *P. clandestina* in the orchard air (component D).

Ascocarp degradation model (component A). Ascocarp (primary inoculum supply) viability at bud burst was about 50%. The primary inoculum supply then gradually declined and was depleted shortly before pit hardening. About 155 cumulative degree-days > 10 C was required to deplete the supply. This model component identifies the period of time over which primary infection (from ascospores) can occur provided adequate moisture and conducive temperatures.

Controlled environment studies. The results of studies on the effects of temperature and relative humidity on foliar infection were used to develop the basic rules (described above) for secondary infection and their subsequent use to generate a disease risk index (model component C). The rules will be used to adjust spray intervals at low (indices of 0-40), moderate (indices of 40-50), and high (indices of 60-100) disease pressures. At this point in time the powdery mildew model is in the experimental or "beta" stage ready for preliminary field-testing and validation. Further improvements will result from in depth studies on the effects of relative humidity temperature on spore production and high temperatures on colony and spore survival.

The results of our controlled environment studies on spore production commenced in 2005. Sporulation at 20 C (70F, the optimum temperature for the colonization of foliage) occurred at relative humidity between 50% and 100%. Multiple regression analyses of the raw data indicated that sporulation on cherry foliage was described by the equation:

$(\log) Y = 8.4 + 0.01X$ (where Y = spore production and X =RH at 20 C (70F)) with an R^2 of 0.72.

The most significant aspect of this is that (although humidity $> 90\%$ favors more abundant sporulation) high humidity is *not* required for spore production.

Field-testing of "beta" version of the new cherry mildew model. See below under "disease management programs".

PCR techniques and air sampling studies. The primers developed by R.A. Spotts were tested for sensitivity for detection of powdery mildew in reaction mixtures and on glass rods used in orchard air sampling studies. The PCR assay was demonstrated to be extremely sensitive, e.g. DNA extracted from 1 and 5 spores placed directly into reaction mixtures was detected 83% and 100% of the time, respectively. The PCR assay consistently detected DNA extracted from 250-500 conidia placed directly on glass rods used for air sampling.

Three different air-sampling techniques (used in conjunction with the PCR assay) were evaluated in

the orchard. The method utilizing a Rotorod air sampler that was operated was the only assay that detected *P. clandestina* in the orchard air early enough to be of practical significance. For the second year, *P. clandestina* was not detected in the orchard air during March early- to mid- April, indicating that “background” DNA from previous epidemics should not result in “false positives”. The initial detection of the fungus in the orchard air in 2005 occurred during a rain event in late-April. The presence of ascospores in the orchard air (which was predicted using component B of the predictive model) during this rain event was confirmed using a Burkard volumetric air sampler. Positives did not occur for the following 13 days but began again 14 days after the initial detection. The resumption of “positives” preceded the appearance of visible symptoms by 3 days. The first signs of powdery mildew were observed in the orchard 17 days after the ascospore release. The air sampling/PCR technique confirmed the presence of the fungus in the orchard throughout the fruiting season. Results of this study should represent the initial step in the incorporation of an inoculum availability component into a cherry powdery mildew risk assessment model. The significance of this component has several potential benefits. The plant disease triangle dictates that any plant disease results from the interaction between host, pathogen, and environment. If the pathogen were absent, even the most disease-conducive weather conditions would not result in an epidemic. Therefore, the application of this technology could serve to delay the initiation of the fungicide program.

Disease management programs. The primary infection (model components A and B) 0.1” precipitation at > 50 F between bud burst and pit hardening) was used to initiate a model-based fungicide program. Subsequent fungicide applications were made at intervals specified by the secondary infection component (component C) of the model (rules outlined above). The model-driven programmed commenced 1 week prior to the industry-standard “phenology” based program. The former program also ended 1-2 weeks prior to the conclusion of the phenology based program with no sacrifice in disease control.

Several organic programs were tested under low- to moderate disease pressure. The best organic program, which consisted of a combination of JMS Stylet Oil and Sonata applications, failed to provide disease control different than the untreated control. V-10118 (an experimental fungicide from Valent, Inc.) provided excellent mildew control. Work on this compound will now proceed through IR-4 to speed the registration process.

Budget:

Project Title: Modeling and managing powdery mildew of cherry

PI: G.G. Grove and R.A. Spotts

Project Duration: 2004-2006

Current Year: 2006

Project Total (3 years): \$125,139

Current Year Request: \$ 43,139

Year	Year 1 (2004)	Year 2 (2005)	Year 3 (2006)
	48,678 (funded: \$41,000)	47,487 (funded: \$41,000)	\$43,139

Current year (2006*) breakdown

Item	Year 1	Year 2	Year 3*
Salaries			
¹ Scientific Assistant	17,520	17,409	19,167
² Benefits 35% (yr 3)	5,606	4,526	6,708
¹ Salaries (hourly labor)	12,000	12,200	10,688
² Benefits 11% (yr 3)	1,952	1,952	1,176
Equipment	-	-	-
³ Supplies	6,700	6,700	1,700
⁴ Travel	4,700	4,700	3,700
Miscellaneous	-	-	-
Total	\$48,678 (funded: \$41,000)	\$47,487 (funded \$41,000)	\$43,139

¹ Salaries: Scientific Assistant Jeff Lunden. Hourly Labor: Oscar Garza.

² Benefit rate for Scientific Assistant in year 1 is estimated at 32%, 26% for year 2 and 35% in year 3. The change in benefit rate over the years is due to changes in the contributions made by WSU on behalf of the employee. Benefit rate for hourly labor is approx. 16% in years 1 and 2 and 11% in year 3. This change is due a change in policy at WSU.

³Supplies consist of PCR reagents, glass rods, Petri plates, anhydrous glycerol, and plot supplies.

⁴Travel - Roza research unit, Tri-Cities region, and Central and Upper Yakima Valley. Includes partial lease on 4 x 4 truck, fuel, and vehicle maintenance charges.

CONTINUING PROJECT REPORT**ONGOING PROJECT****WTFRC Project #:** OSCC-4

Project title: Biology and control of powdery mildew on sweet cherry.
PI: Jill M. Calabro; 3005 Experiment Station Dr;
Hood River, OR 97031; 541-386-2030;
jill.calabro@oregonstate.edu
Co-PI: Robert A. Spotts; 3005; Experiment Station Dr.
Hood River, OR 97031; 541-386-2030
robert.spotts@oregonstate.edu
Organization: OSU Mid-Columbia Agricultural Research and Extension Center
Cooperator: Gary Grove; 24106 N Bunn Rd; Prosser, WA 99350
509-786-9283; grove@wsu.edu
Organization: WSU Prosser Irrigated Agriculture Research and Extension Center
Contract Administrator: Dorothy Beaton; dorothy.beaton@oregonstate.edu; 541-737-4068

Project Objectives:

1. Determine when fruit infection occurs in relation to maturity.
2. Examine the effect of temperature and relative humidity on fruit infection.
3. Develop an early detection method for PM on fruit.
4. Establish a baseline for powdery mildew (PM) resistance to demethylation inhibitors (DMI's) fungicides.
5. Evaluate foliar mildew levels under various management regimes.
6. Study the relationship between powdery mildew infection and pitting.

Schedule of Activities:

Objective	Year			
	2003	2004	2005	2006
1	✓	✓	✓	✓
2			✓	
3	✓	✓		
4			✓	✓
5	✓	✓		
6	✓	✓	✓	✓

Deviations from Original Schedule:

Originally, this project was expected to conclude with the 2005 growing season. Poor fruit set attributed to cold injury and/or inadequate pollination hindered the progress of certain planned experiments. In many cases, studies were entirely discontinued in 2005 due to the lack of fruit. Efforts were unsuccessfully made to find alternative sources of trees and/or fruit, because most of the studies require the use of trees and/or fruit that are not sprayed with fungicides.

2005 Significant Findings:

- ✓ Fruit remain susceptible to PM throughout the growing season, potentially gaining some resistance after reaching 15 °Brix.
- ✓ Powdery mildew is able to effectively infect fruit under a range of temperature and relative humidity combinations.
- ✓ PM resistance to DMI's has been detected in three orchards.
- ✓ Fruit heavily infected with PM are related to significantly more severe pitting.

Methods to Be Employed in 2006:

1. Time of infection of Bing, Lapins, and Sweetheart fruit

Fabric bags made of nylon will be used to cover fruit and exclude mildew spores from full bloom (typically early-mid April) to harvest (typically early July). Bags will be removed from each fruit cluster for a one-week period throughout the growing season so that fruit are exposed to PM spores. Fruit will be assessed for mildew incidence at harvest. Sensors will monitor relative humidity and temperature both within and outside the bags, and an orchard air sampler will collect samples to monitor the daily number of conidia. Three cultivars will be included: Bing, Lapins, and Sweetheart.

In addition to the bagging study, fruit clusters will be inoculated weekly with a spore suspension on five Lapins trees from full bloom to harvest, at which time fruit will be assessed for PM incidence. To minimize natural PM infections, fungicides will be applied biweekly, with the fruit clusters of the study protected with plastic bags. To ensure the effectiveness of the fungicide spray program, foliar PM incidence of five randomly selected border trees will be compared with the trees of the study.

2. DMI fungicide resistance

PM isolates will be collected from orchards in and near Parkdale, OR; Wenatchee, WA; and Yakima, WA and categorized in one of three categories based on the historical use of DMI's as either none/organic, soft, or hard spray program. Actively growing cultures will be maintained on cherry leaves, cultivar Sweetheart, if needed. Leaf disks will be sprayed with the fungicide Elite, Orbit, Procure, Rally, or Rubigan, at 0, 25%, 50%, 100%, or 200% of the labeled rate and then inoculated with PM spores. Fungicide resistance will be rated following an incubation period of 14 days. The procedure will be repeated for each orchard.

3. Effect of mildew on pitting

Fruit with a varying degree of mildew (rated as no = 0%, slight = 1-33%, moderate = 34-66%, severe = 67-100% mildew) will be selected and cooled to either 1°, 4°, or 20° C. A pitting tool will then deliver a standard impact to the shoulder of each fruit. Fruit will be stored for two weeks at either 1° or 4° C and then rated for severity of pitting (1 = none, 2 = slight, 3 = moderate, 4 = severe damage). Twenty-five fruit will be used for each temperature regime/PM infection level

combination. Three cultivars will be included in the study, Bing, Lapins, and Sweetheart. Firmness data, size, color, and °Brix will also be collected for each PM infection level.

Results and Discussion:

1. Time of infection of Sweetheart fruit. Figure 1.

The first conidium was observed on 7 April. The number of conidia detected in the orchard air remained well below levels detected in previous years. In fact, conidia levels declined before harvest, and PM infection on fruit and leaves was less than in previous years.

Studies on cultivars Bing and Lapins were discontinued in mid May, six weeks after the study's initiation, due to poor fruit set at the orchard in MCAREC. Studies with cultivar Sweetheart were conducted as planned, but approximately 40% of the treatments were lost due to poor fruit set in the cemetery block in The Dalles. Remaining fruit were susceptible to PM throughout the growing season. PM incidence was significantly greater on cherry fruit which were never covered by a bag (positive control) than on fruit covered with a bag the entire season (negative control). Fruit infection declined somewhat near the point at which fruit reached 15 °Brix, about 1 ½ weeks before harvest. These studies should be repeated in 2006 and will include Bing, Lapins, and Sweetheart.

2. Effect of temperature and relative humidity on fruit infection. Tables 1 & 2.

Detached green fruit, cultivar Bing, were successfully infected with PM under artificial conditions in the lab. The percent of conidia with hyphae at least twice the length of the conidium after an incubation period of six days was recorded for all twelve temperature – relative humidity combinations. The differences between treatments were not statistically significant, indicating that PM has an excellent fitness for infecting sweet cherry fruit under the temperature and relative humidity conditions common during four major physiological stages of fruit development (full bloom, early pit hardening, pit hardening, and harvest). Only immature, green fruit were inoculated. The highest percentage of spore germination occurred at 18.62 °C, which corresponds to average day time high temperature during pit hardening, usually the last week of May at MCAREC. About 10% of the inoculated fruit developed secondary hyphae and/or an extensive colony of hyphae at the point of inoculation but did not sporulate. The majority of fruit in this category were incubated at 18.62 °C and regardless of relative humidity. Of the 288 total fruit inoculated, only one sporulated; it was incubated at 15.46 °C and 68% relative humidity.

These results contrast a similar study by G. Grove, where spore germination increased with increasing relative humidity. This study found no such relationship. Grove's study, however, defined spore germination as when the germ tube length exceeded the width of the spore, only included one temperature (20 °C), and held the detached fruit for a maximum of 24 hours.

3. DMI resistance.

A leaf disk assay was developed to assess PM resistance to fungicides in the class of demethylation inhibitors (DMI's). Of the ten orchards evaluated, five are suspected as having PM resistance to one or more fungicides. Four orchards, located Hood River, The Dalles, and Prosser showed possible resistance to fenarimol, trade name Rubigan. Two orchards in The Dalles, both with an intensive spray program, have possible resistance to propiconazole (trade name Orbit). One orchard in Hood River is suspected of having resistance to tebuconazole, trade name Elite, and one orchard in The Dalles is suspected of having resistance to myclobutanil, trade name Rally. In accordance with standard fungicide resistance trials in peer-reviewed journals, more data are needed

to determine whether or not an orchard has DMI resistant PM. Studies will be expanded in 2006 to include orchards in Wenatchee, Yakima, Parkdale, Hood River, and The Dalles.

4. Effect of mildew on pitting on Sweetheart fruit. Table 3.

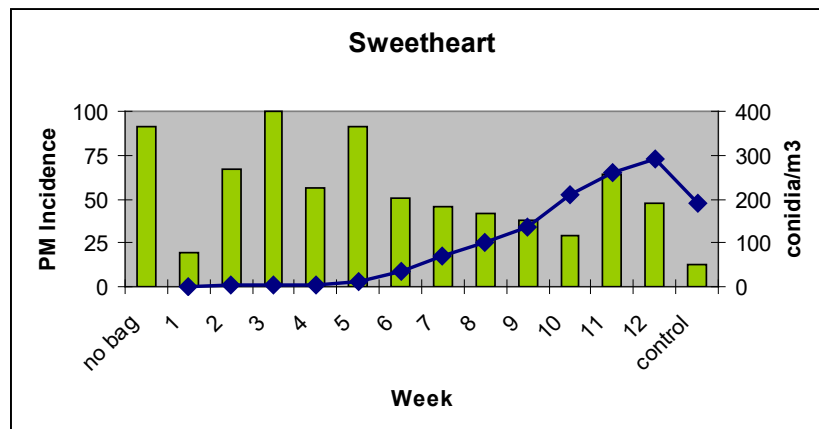
Pitting was significantly more severe on fruit with moderate or heavy PM infections than on fruit with no or light infection. Neither the temperature at the time of impact nor the storage temperature had an effect on pitting severity, which is contrary to 2004 results. In terms of pitting incidence, uninfected fruit had the most number of fruit that did not pit (17%), followed by slightly infected fruit (12%). Of the moderately and severely infected fruit, only 1% in each category did not pit.

Similar to last year, mildewed fruit were less mature than mildew-free fruit, in terms of firmness, size, color, and °Brix. Research by several groups has indicated that sweet cherry fruit become less susceptible to pitting as they mature. Therefore, the relationship between PM infections and pitting is unclear. PM could be directly responsible for increased pitting severity or indirectly by slowing fruit ripening. Overall, pitting damage was less than in 2004. These studies should be repeated in 2006 and will include cultivars Bing and Lapins.

FIGURES & TABLES

Figure 1. Average percent fruit infected, PM incidence, in the bagging study for cultivar Sweetheart. The positive control where fruit were never bagged is indicated by “no bag.” Subsequent weeks correspond to the period of time when fruit were exposed (bag removed) and vulnerable to PM

infection. The negative control where fruit remained covered by a fabric bag the entire season is indicated by “control.” PM incidence ratings were done following harvest. 15 °Brix was recorded between weeks 11 and 12. The number of conidia per cubic meter of air sampled in the orchard is indicated with the line graph.



Temperature		Relative Humidity	% Germinated Conidia
°C	°F		
12.79	55	70	12.25
		79	15.17
		88	11.42
15.46	60	68	9.17

Table 1. Results from the detached fruit inoculation study. The average percent of germinated conidia with hyphae at least twice as long as its conidium, are listed as % Germinated Conidia relative the temperature-relative humidity treatment.

		75	6.50
		83	14.00
18.62	65	68	12.33
		74	22.50
		80	9.25
		70	7.25
23.76	75	75	7.50
Temperature		79	# Fruit with
°C	°F	Relative Humidity	Extensive Hyphae
12.79	55	70	2
		79	2
		88	1
15.46	60	68	3
		75	1
		83	5
18.62	65	68	6
		74	4
		80	2
23.76	75	70	0
		75	1
		79	0

Table 2. The number of fruit with extensive hyphal colonization from the detached fruit inoculation study. A Chi-squared test for homogeneity revealed that extensive, secondary hyphae production is not independent of the temperature – relative humidity treatment.

Injury	Storage	Powdery Mildew Rating
--------	---------	-----------------------

Table 3. The average rating of pit development following a standard impact on Sweetheart fruit with varying degrees of PM infection. A pit rating of 0 = no damage, 1 = slight damage, 2 = moderate damage, and 3 = severe damage. Prior to the injury, fruit were sorted based on their level of

Temp C	Temp C	No PM (a)	Slight PM (a)	Mod. PM (b)	Severe PM (b)
1	1	1.12	1.12	1.64	1.60
1	4	1.08	1.20	1.44	1.60
4	1	0.80	0.84	1.52	1.84
20	1	0.72	0.84	1.56	1.68

PM infection where No PM = no visible PM, Slight PM = 1 – 33% of the fruit surface colonized, Moderate (Mod.) PM = 34 – 66% of the fruit surface colonized, and Severe PM = 67 – 100% fruit surface colonized by PM. Statistical differences in the severity of pitting among the fruit infection levels are indicated by different letters. No differences were evident based on the injury or storage temperature.

BUDGET

WTFRC Project #: OSCC-4

Project title: Biology and control of powdery mildew on sweet cherry.

PI: Jill M. Calabro; 3005 Experiment Station Dr; Hood River, OR 97031; 541-386-2030; jill.calabro@oregonstate.edu

Co-PI: Robert A. Spotts; 3005; Experiment Station Dr; Hood River, OR 97031; 541-386-2030; robert.spotts@oregonstate.edu

Project Duration: 2003-2006

Project Total: \$70,903

Current Year Request: \$18,778

Budget:

Year	2003	2004	2005	2006
Total	16324	17023	18778	18778

Year Breakdown

Item	2003	2004	2005	2006
Salary & Wages	15000	15675	16560	16560
OPE	524	548	1418	1418
Supplies	500	500	500	500
Travel	300	300	300	300
Total	16324	17023	18778	18778

CONTINUING PROJECT REPORT**YEAR 2/3****WTFRC Project #:** CH-04-404**Organization Project #:** 13C-3361-5289**Project Title:** Virus control strategies to assist cherry production.**Principal Investigator:** Ken Eastwell, Associate Plant Pathologist**Organization:** Washington State University – IAREC
24106 North Bunn Road, Prosser, WA 99350
Telephone: (509)786-9385; E-mail: keastwell@wsu.edu**Co-investigators:** Bill Howell, Manager NRSP-5, WSU-Prosser**Cooperators:** Thomas Unruh, YARL, USDA-ARS, Wapato
Narceo Bajet, YARL, USDA-ARS, Wapato
Eileen Perry, Assistant Director, CPAS, WSU-Prosser
Robert Woolley, Dave Wilson Nursery
Ekaterina Riga, Assistant Nematologist, WSU-Prosser
Hui Hou, M.Sc. student, WSU-Prosser
Many growers and fieldmen**Contract Administrator:** Mary Lou Bricker; mdesros@wsu.edu; 509-335-7667**Project Objectives:**

1. Develop progressive strategies to control virus diseases that contribute to the decline of sweet cherry productivity in the Pacific Northwest.

Specific goals for year 2006:

- Confirm the role of pollen in the transmission of *Cherry leafroll virus*
- Explore rootstock selection as a means to minimize tree-to-tree spread of cherry viruses

2. Develop laboratory tests that will make virus testing accessible to growers.

Specific goals for year 2006:

- Serological assays for *Cherry rasp leaf virus* currently in development will be assessed
- Evaluate field application of robust methods to detect various little cherry disease-causing viruses

3. Monitor commercial sweet cherry orchards for emerging virus diseases

Specific goals for year 2006:

- Increase our ability to detect the emerging *Foveavirus* genus of pathogens to assist monitoring efforts in commercial orchards

4. Evaluate the use of remote sensing to identify areas of declining cherry production that may be associated with virus infections.

Specific goals for year 2006:

- Collect spectra on rootstock/variety trial trees that were transplanted to the field in 2005
- Collect thermal imagery of infected orchard trees
- Secure funding for developing prototypes

Significant findings:

- *Cherry leafroll virus* is detected in the pedicels of fruit collected from trees that are not infected with the virus. This suggests significant implications for understanding natural spread of this virus and strategies for its control.
- Root grafting is a significant route of tree-to-tree spread of several important diseases of cherry
- The incidence of Western X disease is increasing throughout WA.
- ELISA was developed for Montmorency stem pitting virus. This proved effective in discriminating in the field samples between this damaging virus and other viruses whose long term consequences are less significant.
- Virus infection may be detected using light reflectance. The silicon detectors required for this technology are inexpensive and can be incorporated into one of several different formats.

Methods to meet proposed objectives:

Objective 1: Develop progressive strategies to control virus diseases that contribute to the decline of sweet cherry productivity in the Pacific Northwest.

We previously confirmed that cherry pollen can carry a significant amount of infectious *Cherry leafroll virus*. Whether this pollen plays a significant role in natural virus-transmission process is still unknown although preliminary evidence suggests that one point of entry for the virus is through blossoms. Pedicels will be collected and analyzed at three times between fruit set and two weeks past commercial maturity. Each pedicel is tested in two parts to provide some indication of virus distribution and movement.

Our research demonstrated that root grafting contributes significantly to the tree-to-tree spread of viruses in cherry orchards, particularly with reference to the spread of *Cherry leafroll virus*. *Cherry raspleaf virus* also enters trees predominantly through the root system, but as a result of nematode feeding rather than solely by root grafting. Since *Cherry raspleaf virus* infects apples, peaches and a number of broadleaf orchard weeds, practical strategies to control the virus once it is established in an orchard are not apparent at this time. The availability of new rootstocks and interstocks has increased options available for cherry orchards – we propose to evaluate rootstocks as a means of providing protection against the soil-borne transmission of *Cherry leafroll virus* and/or *Cherry raspleaf virus*. Candidate rootstocks will be inoculated directly with virus by chip-budding to determine if the rootstock is susceptible to virus infection. A duplicate set of trees will be inoculated above the graft union to reveal any incompatibility or hypersensitive reactions that would lead to tree decline in subsequent seasons. All inoculated trees will be monitored for three years to provide data on virus movement in the various rootstock variety combinations. The aim is to develop specific recommendations for growers wishing to replace diseased trees and minimize the impact of these viruses on orchard profitability and sustainability.

Objective 2: Develop laboratory tests that will make virus testing accessible to growers.

We established that there are two viruses causing little cherry disease in the PNW, but low virus concentration in tissue has hampered the development of economical testing methods. To circumvent this limitation, the coat protein genes of these viruses were isolated and transferred to bacteria that then synthesize relatively large amounts of virus coat protein. This protein is subsequently used for the production of antibodies for enzyme-linked immunosorbent assays (ELISA). Once ELISAs are developed and their reliability confirmed, they will be available for routine, cost-effective testing of orchard trees. These tools will be made readily available to assist in orchard management decisions by providing rapid identification of pathogens. This strategy is also being applied to other viruses known to occur in the PNW including *Cherry raspleaf virus* and *Apple chlorotic leaf spot virus*, both of which can cause serious production problems in cherry.

Isolates of *Cherry raspleaf virus* have been collected from Yakima, Franklin and Chelan counties in Washington and Wasco county in Oregon. Conserved regions of their genomes were identified and used to develop a sensitive molecular assay that has been applied to trees and to nematodes isolated from orchard soil (in cooperation with Dr. Riga). The availability of this sensitive assay opens the door for developing disease management strategies (e.g. virus-resistant and/or nematode resistant rootstocks) that would be economically viable for growers with affected orchards.

Objective 3: Monitor commercial sweet cherry orchards for emerging virus diseases.

Invited visits to cherry blocks provide a good understanding of the disease concerns faced by growers. This orchard-based survey work continues to provide critical awareness of emerging disease problems. Where appropriate, additional biological data is collected during these site visits to help determine the threat each disease poses and strategies to limit their economic impact.

Objective 4: Evaluate the use of remote sensing to identify areas of declining cherry production that may be associated with virus infections.

Two seasons of observations have confirmed the existence of specific spectral changes related to virus status of cherry trees. This information awaits integration into advanced orchard surveillance systems.

Results and discussion:

Objective 1: Develop progressive strategies to control virus diseases that contribute to the decline of sweet cherry productivity in the Pacific Northwest.

There are likely two modes of transmission by which *Cherry leafroll virus* spreads. Within an orchard, root-grafting plays a significant role in tree-to-tree spread. This was illustrated by the number of trees reacting to herbicide treatment after removal of an adjacent herbicide-treated tree. A second mechanism responsible for occasional long distance movement between orchards remains elusive. We have demonstrated the viability of *Cherry leafroll virus* in pollen extracted from cherry flowers and from bees collected in and around cherry orchards. Thus, pollen-mediated transmission of *Cherry leafroll virus* remains a distinct possibility, although the exact mechanism is not known.

During this and previous seasons, we detected *Cherry leafroll virus* in the fruit of trees that are otherwise free of *Cherry leafroll virus* suggesting that virus was being transferred to the bloom of healthy trees via pollen. However, it is unknown if the virus present in the fruit poses a potential risk for infection of the tree. In 2005, fruit was collected from adjacent ‘Van’ and ‘Bing’ trees. Leaf and fruit samples were tested by ELISA and by RT-PCR to determine virus status of each tree considered in this trial. Evidence of virus movement from fruit to the spur was not apparent because virus was not detected in the fruit stems (pedicels). However, significant improvements in virus detection methods this year enabled us to detect *Cherry leafroll virus* in pedicels of fruit from non-infected trees (highlighted boxes in Table I). The variation observed between trees is likely the result of bee scavenging patterns and differences in peak bloom time of individual trees. Although this dataset represents a very small sample size, it allowed us to evaluate procedures and to obtain preliminary data to guide experimental design for the 2006 growing season. The detection of *Cherry leafroll virus* RNA in the fruit stems is highly significant and provides circumstantial support to the hypothesis that this virus could enter an otherwise healthy tree from the blossom.

Table I. *Cherry leafroll virus* is detected in pedicels of fruit collected from non-infected ‘Van’ cherry trees growing adjacent to infected ‘Bing’ trees.

	4 weeks pre-harvest ^a		1 week pre-harvest ^a	
	A ^b	B ^b	A ^b	B ^b
Negative control ^c	0/5	0/5	0/5	0/5
Positive control ^d	1/5	5/5	5/5	5/5
‘Van 1’	0/5	3/5	5/5	5/5
‘Van 2’	0/4	0/4	0/5	0/5
‘Van 3’	0/5	0/5	0/5	0/5
‘Van 4’	0/5	4/5	0/5	0/5

- Results reported as: (number of virus-positive samples)
(number of samples tested)
- “A” is the portion of the stem near the spur, and “B” is near the fruit.
- Negative controls are pedicels from a ‘Van’ tree located 300 ft. from the nearest known source of CLR V.
- Positive controls are pedicels from a ‘Bing’ tree infected with CLR V.

Objective 2: Develop laboratory tests that will make virus testing accessible to growers.

Substantial progress was made in the development of diagnostic reagents for viruses associated with little cherry disease. Antibodies that recognize one of the two coat proteins of *Little cherry virus-2* were produced and are undergoing preliminary evaluation. The second coat protein was recently obtained and is being used in the initial stages of antibody production.

Our studies demonstrated that *Little cherry virus-1* is well established in North America. The extreme sequence variability of this virus thwarted early efforts to isolate the coat protein genes from local isolates. However, using different strategies for isolating this gene, we recently were successful in identifying and sequencing it. This progress will be the basis for developing serological reagents as described above.

Monitoring the dramatic increase in the spread of several leafhopper-transmitted viruses in 2000 led us to predict that the incidence of Western X, a leafhopper transmitted bacteria, would increase dramatically. Regrettably, this has happened, and the incidence of this disease that was so devastating in the 1960's is occurring in new as well as established cherry production areas. Therefore, the needed ability to distinguish between Western X and the two little cherry disease viruses has gained increased significance.

The development of serological reagents for the viruses that cause little cherry disease is an important step in obtaining the tools necessary for good management decisions.

Studies by others identified *Green ring mottle virus* and *Cherry necrotic rusty mottle virus* as members of the *Foveavirus* genus. Our research revealed that cherry rusty mottle, cherry twisted leaf, and Montmorency stem pitting are also caused by viruses of this group. As we accumulate information, it is apparent that there is extensive sequence variability between the different viruses that are associated with these diseases of cherry, but also areas of sequence conservation. This enabled us to develop both broad spectrum and virus-specific molecular assays. Furthermore, we produced antibodies against Montmorency stem pitting virus. An ELISA based on these antibodies is very effective as a diagnostic aid and permits a relatively quick diagnostic answer for growers. This allowed us to discriminate between several viruses that induce symptoms similar to those of Montmorency stem pitting virus at the early stages of infection, and permits the effective and rapid removal of trees infected with this destructive virus as the first step in controlling this disease. *Green ring mottle virus*, which is symptomless in most sweet cherry varieties, does not react with the antiserum that we developed, and thus, its presence does not interfere with efforts to detect disease-causing viruses.

Objective 3: Monitor commercial sweet cherry orchards for emerging virus diseases.

Foveaviruses are emerging as an important group of viruses in cherry production. Many different forms of these viruses are being detected; they are variable both in terms of their molecular properties and in terms of the degree and nature of symptoms they cause. Availability of detection reagents such as those developed for Montmorency stem pitting virus are greatly enhancing our ability to identify and react to virus infections.

Continued surveillance of cherry production areas over the past five years revealed a dramatic increase in the frequency with which Western X disease is encountered. This disease severely impacted WA cherry production in the 1950's and 1960's; careful management of blocks in which the disease occurs is necessary to minimize further impact.

Objective 4: Evaluate the use of remote sensing to identify areas of declining cherry production that may be associated with virus infections.

On 10 June 2005, we collected a spectral dataset in a commercial 'Bing' orchard using techniques described in the 2004 report. Each tree was previously analyzed for the presence of *Cherry leafroll virus* (CLRV), *Prunus necrotic ringspot virus* (PNRSV) and *Prune dwarf virus* (PDV). For this dataset we selected eight trees. Three spur leaves from each tree were collected and two spectral measurements per leaf were obtained. The average spectrum for each infection type is normalized by the average of the healthy leaves (Figure 1); the further the spectrum deviates from 1.00, the greater the difference in reflectance. Hence, this plot emphasizes the wavelengths where the virus-infected plants exhibit the greatest differences. The spectrum for the CLRV infected field trees looks very much like results obtained from lath house trees in 2004. Spectra from PDV and PDV+CLRV trees look similar, but different from the CLRV only leaves. Reflectance values of 582nm, 697nm, 1458nm and 1975nm, as well as derived stress indices of normalized difference vegetation index (NDVI), modified chlorophyll

absorption in reflectance index (MCARI), photosynthetic response index (PRI), water band index (WBI), and red edge vegetation stress index (RVSI) were evaluated. Analysis of Variance (ANOVA) was used to quantify the differences among plants with and without CLRV. Only RVSI could significantly distinguish the non infected leaves from the other infected leaves with an F value of 7.89 (> 0.001). This index was significant in distinguishing non infected leaves from CLRV, CLRV+PDV, and PDV infected leaves at a 95% confidence limit. When only the healthy and CLRV infected leaves were used in the analysis, RVSI again produced the most significant contrast between the infected and non infected leaf measurements with an F value of 18.55 (> 0.001).

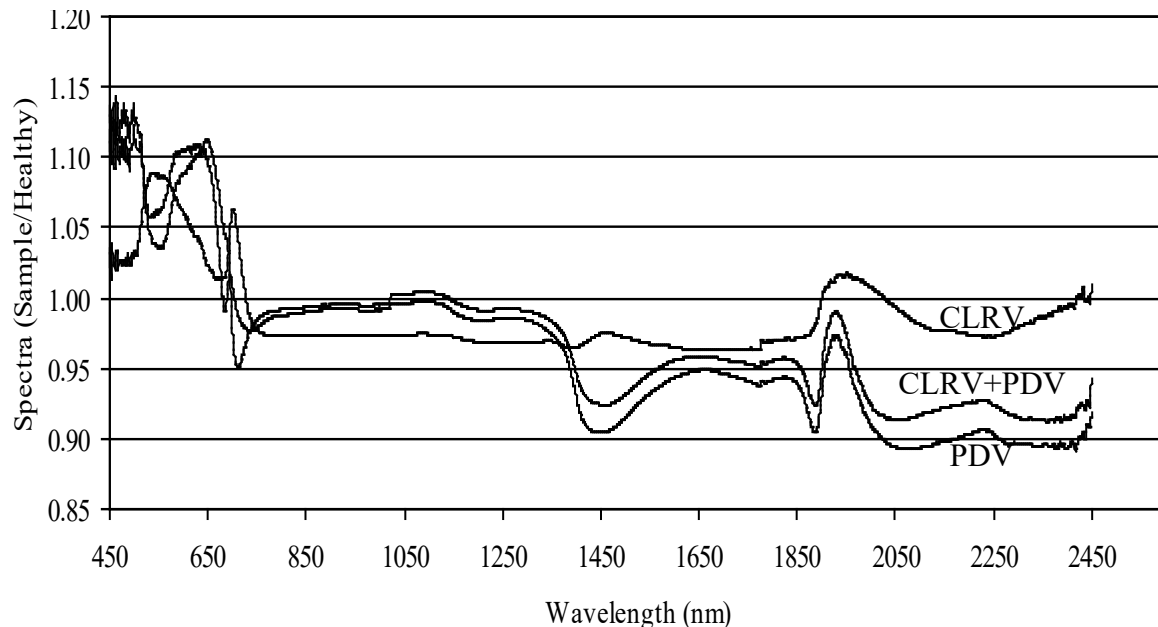


Figure 1. Averaged spectra from CLRV, PDV and CLRV+PDV infected trees in a commercial orchard. The spectra were normalized using the averaged spectrum from healthy trees. Therefore, the graph represents deviation from readings from healthy trees which have the value of 1.00.

On June 22 2005, we used the CPAS multispectral camera from the ground to view trees and leaves in the same commercial orchard. The objective was to determine if the near infrared band was sensitive to differences between the infected and non infected trees, for whole trees down to the leaf level. The camera was set for color infrared viewing in the orchard, and individual frames were acquired and converted to NDVI images using the red and near infrared bands. With multispectral imagery, infected tree appears as darker red, but the observer can be confused by differences in sunlight intensity on the leaves. The NDVI image corrects for these differences, and brighter values indicate healthier canopy. Thus, the difference between CLRV-infected and non-infected trees is more noticeable in the NDVI image. This same type of image analysis was applied at the individual leaf level, and again, the NDVI images enhance the difference between the healthy and infected plant. We feel that these results suggest that a small, portable, lightweight video system sensitive to the red and near infrared bands to produce a real-time NDVI would be of value in locating stressed trees in the orchard. This system could be worn by someone, or attached to a PDA to generate NDVI images.

Future efforts would include thermal infrared viewing (airborne and on ground) to see if there are differences in canopy temperature between healthy and infected trees. We also plan to acquire a second dataset of spectral measurements on the young cherry plants from a trial used to evaluate the differences in variety susceptibility to CLRV. Spectral measurements were made on these trees during 2004, and the plants were moved from the lath house to a field location at IAREC early 2005. However, the plants were severely stressed by late spring and would yield any meaningful spectral measurements this season.

Budget:

Project Title: Virus control strategies to assist cherry production.
Principal Investigator: Ken Eastwell
Project Duration: 2004-2006 (3 years)
Funding history: FY2004 requested: \$29,053 FY2004 received: \$26,616
FY2005 requested: \$31,655 FY2005 received: \$31,655
Current year: 2006
Current Year Request: **\$36,973**
Project Total Request (3 years): **\$97,681**

Item	Year 1 (2004)	Year 2 (2005)	Year 3 (2006)
¹ Salaries	\$14,084	\$14,648	\$17,946
² Benefits	\$4,084	\$5,292	\$7,282
Wages	\$4,000	\$4,000	\$0
Benefits	\$640	\$640	\$0
Equipment	\$0	\$0	\$0
Supplies	\$5,700	\$6,800	\$11,200
Travel	\$500	\$230	\$500
Miscellaneous	\$45	\$45	\$45
Total	\$29,053	\$31,655	\$36,973

¹**Salaries:** Year 3 salaries include 0.25FTE for Greenhouse Attendant. and Research Technologist III.

²**Benefits:** Year 3 benefits are at 43% for Greenhouse Attendant. and 39% for Research Technologist III.

Departure from original budget request: WSU reduced technical support to this program by \$8,000 per year. We are seeking to help off-set this reduction by increasing our original request by one-half (\$4,000) of this amount.

Note: WSU is including the following information on other funding available for the support of similar research undertaken by the faculty member proposing this research. These resources are listed to identify other support granted for this research and are not included as a commitment of cost-share by the institution.

Additional funding sources: Digital imagery for remote sensing and subsequent analyses are provided at no charge to this project by baseline funding to the Center for Precision Agricultural Systems, WSU-Prosser. Fruit tree nurseries, through their Nursery License Surcharge research fund (managed by the Washington State department of Agriculture) provided \$8,900 for the *Cherry rasp leaf virus* genome characterization. Critical support for this research is provided by the National Virus Tested Fruit Tree Program (NRSP-5). A special cooperative agreement with USDA-ARS (Dr. Tom Unruh) provides \$219,000 over the period from 09/25/2003 to 07/31/2008 to investigate vectors and mechanisms of transmission of the little cherry viruses in the PNW and to support the Master's student contributing to this project.

FINAL REPORT

WTFRC Project #CH-04-403

WSU Project #13C-3643-3387

Project title: Biology and management of bark beetles

PI: Jay F. Brunner
Organization: WSU Tree Fruit Research and Extension Center
Address, phone, e-mail: 1100 N. Western Avenue, Wenatchee, WA 98801
(509) 663-8181 ext. 238; jfb@wsu.edu

Co-PIs and affiliations: Tim Smith, WSU Extension, Chelan-Douglas County
Mike Doerr, WSU Tree Fruit Research and Extension Center

Contract administrator: Mary Lou Bricker (mdesros@wsu.edu) (509) 335-7667; or Tom Kelly (kellytj@wsu.edu) (509) 335-3691

Objectives:

1. Identify the bark beetle species attacking pome and stone fruit throughout the state. **(completed)**
2. Develop a clear understanding of the seasonal life history of bark beetles in Washington State and compare that with currently available information. **(completed)**
3. Examine methods of monitoring bark beetles. **(completed)**
4. Develop a means to predict beetle activity at any location. **(partially completed)**
5. Determine the distance bark beetles move from a source to attack healthy orchard trees. **(completed)**
6. Develop a laboratory rearing procedure for bark beetles. **(partially completed)**
7. Determine host suitability of various tree fruits **(completed)** and other deciduous trees **(partially completed)** for bark beetle reproduction. Identify other secondary decomposing beetles found in suspected bark beetle host material. **(not completed)**
8. Develop a bioassay technique for assessing relative toxicity of candidate insecticides. **(completed)** Validate insecticide bioassay results and evaluate new candidate insecticides for efficacy and longevity. **(completed)**
9. Document successful control strategies in heavily infested orchards. **(completed)**

Significant findings:

1. The dominant bark beetle found throughout central Washington was the shothole borer (SHB), *Scolytus rugulosus* Müller (Coleoptera: Scolytidae). An ambrosia beetle (AB), *Xyleborinus saxeseni* Ratzburg (Coleoptera: Scolytidae), was present in high numbers at only one location, a cherry orchard abandoned for several years. At most locations more than one species of scolytids were detected. Species identification was confirmed.
2. It is apparent that two distinct periods of SHB activity occur in Washington, the first beginning in late April and peaking in late May to mid-June. The second begins in mid-July and peaks in late July to early August. The pattern first noted in 2003 was observed again in 2004 and 2005. These data are inconsistent with some reported literature. It appeared that SHB was the species most implicated in damage to healthy trees.
3. Ambrosia beetle (AB) activity was noted throughout the entire growing season. It is likely that 2-3 generations occur each year, but there was overlap between them making clear demarcation of generations difficult. AB activity was first observed in late March, with a second activity period occurring in early June, and a possible third in July and August. Ambrosia beetle activity appeared to be limited to stressed or dying trees and was less likely to cause damage to healthy trees.

4. Traps were useful in identifying peak activity periods, but it was not clear whether they would be useful in setting thresholds for treatments. Yellow sticky traps (unbaited apple maggot traps) seemed to be the most appropriate trap to monitor SHB activity, but ethanol-baited intercept-style traps were necessary to monitor AB activity.
5. Woodpiles of cherry, apple and pear appeared to be the main sources of high beetle densities that caused injury to commercial orchards. However, one population of SHB was found developing on recently cut poplar or cottonwood (*Populus* spp.). Severe injury can result to either stressed or healthy cherry trees adjacent to heavily infested host material.
6. Movement of SHB into live orchards was closely associated with emergence from host material. SHB readily moved distances of 10-50 meters to attack healthy trees on orchard borders but did not move more than 2-3 rows into a healthy orchard.
7. Laboratory rearing of multiple generations on an artificial diet was not successful. Adult SHB survived a short time on a diet, but no reproduction occurred. Adults were reared and collected in large numbers from infested wood returned to the laboratory. Successive generations were then reared on current-year cuttings of apple and cherry.
8. Laboratory host-suitability tests showed variability in SHB development rates. Cherry is known to be a good host for SHB, but sap flow and mold development in rearing arenas limited reproduction. Apple and pear were clearly suitable hosts for SHB with reproductive rates being similar. Development appeared to be accelerated in apple relative to pear.
9. Many insecticides caused mortality of SHB in field-aged bioassays. The pyrethroid Asana was the most active through 21 days after treatment. Guthion and malathion provided good suppression through 14 days. Variable results were noted with the chloronicotinyls Actara and Assail, but there is evidence that this class of insecticide has potential to suppress SHB in the field. Thiodan and possibly Avaunt were also shown to be possible options. Proclaim, Sevin, Success and Carzol all caused mortality but not at levels expected to provide adequate control.
10. Orchard sanitation was the most important factor in contributing to a reduction in SHB densities and damage to live cherry trees. Sanitation involved removing potential host material (e.g. weakened limbs, recent prunings) from the orchard and eliminating any host material outside the orchard. Host material can be “eliminated” by burning wood or brush piles or thoroughly soaking the piles with an effective insecticide delivered by a handgun sprayer. The increased volume of water delivered by handgun applications appeared to be a significant factor in insecticide efficacy.

Methods:

Seasonal life history and monitoring: The seasonal life history of SHB and AB was monitored at 18 locations in north-central Washington over a three-year period. SHB and AB emergence and movement were monitored by rearing adults from infested sources and by trapping adults near the source of infested wood. Infested wood from different locations was collected in the early spring, placed in darkened containers and held under constant temperature conditions. Cages were fitted with glass vials in one side. Emerging beetles (and parasitoids) were attracted to the light. Beetles entering the glass vial were collected 3 times per week. Traps were placed near infested woodpiles outside of orchard blocks and on the orchard border closest to the source. Commercially available intercept-style traps (Lindgren Funnel Trap, 8-funnels, Phero Tech, Inc.; Pane Intercept Trap, IPM Technologies, Inc.) baited with or without ethanol lures were compared to unbaited yellow sticky traps (Pherocon AM, Trécé, Inc.) for their ability to monitor adult emergence from a source and subsequent migration into surrounding orchards. All insects emerging from infested wood in laboratory cages and all insects collected in the intercept-style traps were identified to family by WSU entomologists. Further, all scolytids and associated parasitoids were sent to Dr. Malcom Furniss, (Entomologist *Emeritus*, University of Idaho) for positive identification.

Describe SHB movement from host material to healthy orchards: Orchards that were exhibiting

severe SHB infestation and subsequent damage to healthy cherry trees were identified and monitored for SHB emigration from nearby host material. The host material serving as the source for SHB was determined and then emigration to healthy trees was monitored with yellow sticky traps at the source and the orchard border. Infestation or damage to healthy trees was monitored by visually inspecting trees in a grid-like pattern moving away from the source. Every tree was monitored along the border row and then every row was monitored moving into the orchard until no further SHB damage was noted. Twenty growing shoots were randomly selected from each tree, and the total number of shoots exhibiting wilting or flagging foliage was recorded. SHB emigration and infestation was monitored at four locations in central Washington.

Laboratory rearing of SHB: SHB adults were collected from infested wood as described in the *Seasonal life history and monitoring* section. The majority of adults were collected following peak flight noted in June 2003. Adults were exposed to both drying wood (cherry or apple limbs of >4 inches in diameter, pruned during the previous winter) in a cage and to cups containing an artificial scolytid diet (Southern pine beetle diet # F9761B, Bio-Serv, Inc.). The rearing arenas were examined regularly for SHB feeding activity and any sign of reproduction.

Host suitability: SHB were reared in limb sections of cherry, apple and pear. These limb sections were recently cut pieces of 2- to 5-year-old wood (6" long x 1" diameter) from trees that had received no insecticide treatments. Limb sections were exposed to newly emerged adult SHB in 32 oz. clear plastic deli cups (Prime Source PS232). Twenty-five cups were set up per host, and 5 SHB were added to each cup. The cups were held at 72°F (±2°F) and 16:8 L:D. The wood sections were examined at regular intervals for SHB survival and the emergence of new beetles. Further, SHB sources identified in field monitoring trials were categorized by species/cultivar and estimated age since cutting. Data from these observations were used to describe host material that was suitable for SHB reproduction.

Insecticide efficacy and longevity: In 2003-04, insecticides were evaluated using newly emerged first generation SHB adults. Mature Delicious apple trees at WSU-TFREC were treated with recommended rates of various insecticides. Treated apple branches were collected at 1, 7, 14 and 21 days after treatment (DAT) and returned to the laboratory. Approximately 6" long x 0.5" diameter sections of 2-year-old wood were added to 32 oz. deli cups (Prime Source PS232). Untreated apple branches were used as a control at each evaluation date. Five SHB adults were added to the deli cups, and survival was recorded after 3 days. Five rearing arenas were prepared for each treatment, and each treatment was replicated three times (25 SHB/treatment/DAT). Rearing conditions were 72°F, 16:8 L:D.

Documenting successful control strategies: WSU entomologists worked closely with several growers to identify the source of severe SHB infestations, remove the source, and then document the efficacy of their clean-up efforts. The source of the infestation was determined through visual inspections as well as trapping of adult movement using yellow sticky traps. Sanitation involved cleaning up potential sources of infestation within the orchard by removing pruning cuts from the previous winter, removing weakened branches or limbs from outside orchard sources. "Removal" of outside-orchard sources included insecticide applications or burning of host material.

Results and discussion:

Seasonal life history and monitoring: Several hundred infested fruitwood and ethanol-baited intercept traps for SHB were found throughout Washington was the spruce bark beetle (ver. Malcom Furniss). An ambrosia beetle, *Xyleborus*

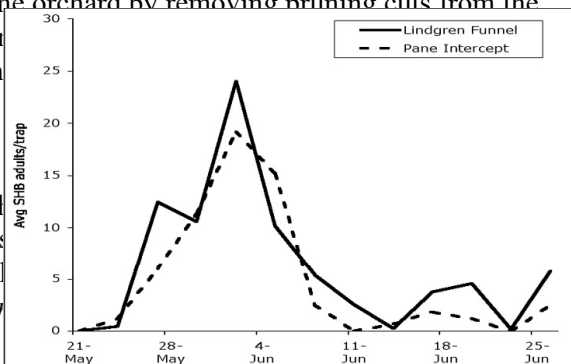


Figure 1: Monitoring SHB with commercially available intercept-style traps baited with an ethanol lure, 2003.

was present in high numbers at only one location, a cherry orchard abandoned for several years. At most locations more than one species of scolytids were detected. A second *Scolytus* sp. (*S. multistriatus*) was found infesting a pile of cherry wood at one location. Cherry is not a reported host for *S. multistriatus*, and in this case *S. multistriatus* infested only the woodpile and was not detected moving into the neighboring cherry orchard. Many other wood decomposing beetles were reared from infested wood. In fact, the majority of beetles collected were associated with dry, dead wood (>18 months old). Buprestids, bostricids and powderpost beetles (Lyctidae) were the primary families of beetles associated with dry, dead wood. SHB and AB appeared to be the primary attackers of weakened trees or recent cuttings (<18 months old). SHB seemed to be the species most implicated in damage to healthy trees, whereas AB were found attacking only stressed or already damaged or weakened trees. Initial observations at the time of collection show that there was fairly significant parasitism (up to 50%) of SHB larvae by *Cheiopachus quadrum* (Hymenoptera: Pteromalidae) (ver. Malcom Furniss).

All traps tested were useful in identifying peak beetle activity periods. There did not appear to be any significant difference in commercially available intercept-style trap types (Fig. 1). Either the Pane Intercept Trap or the Lindgren Funnel Trap was a suitable trapping system to monitor both SHB and AB adult activity. Available ethanol attractants only slightly enhanced trap captures for SHB (Fig. 2). Adult SHB were more highly attracted to the yellow sticky traps than the dark silhouette of the intercept traps (Fig. 3). Although yellow sticky traps seemed to be the most appropriate trap to monitor SHB activity, the ethanol-baited funnel traps were necessary to monitor AB activity.

Two distinct periods of SHB activity occurred in Washington (Fig. 4). SHB activity was first noted in late April or early May and continued through June. The second adult flight was detected in mid- to late July and continued through August and into September. The pattern first noted in 2003 was observed again in 2004 and 2005. Adult SHB were trapped through the entire growing season from initial adult emergence through the end of October. Visual observations of laboratory and field behavior suggested that adult dispersal to suitable hosts and subsequent oviposition occurred shortly after emergence with subsequent activity centered on tending to the maternal galleries.

Traps were useful in identifying peak activity periods of SHB, but it was not clear whether they would be useful in setting thresholds for treatments. We experienced difficulty in locating SHB sources of various population sizes near neighboring cherry orchards that were allowed to remain untreated. However, our observations indicated that if a SHB source was located near a cherry orchard and any significant emergence was detected with yellow traps some control intervention was probably necessary to prevent damage.

AB activity was initially noted in late March or early April in 2004 (Fig. 5). AB activity was detected throughout the entire growing season, with 2 to 3 generations likely. It appeared that a second peak of activity might have occurred in early June, with a possible third generation in July and early August. A similar pattern of summer activity was noted in 2003, but traps were not in place

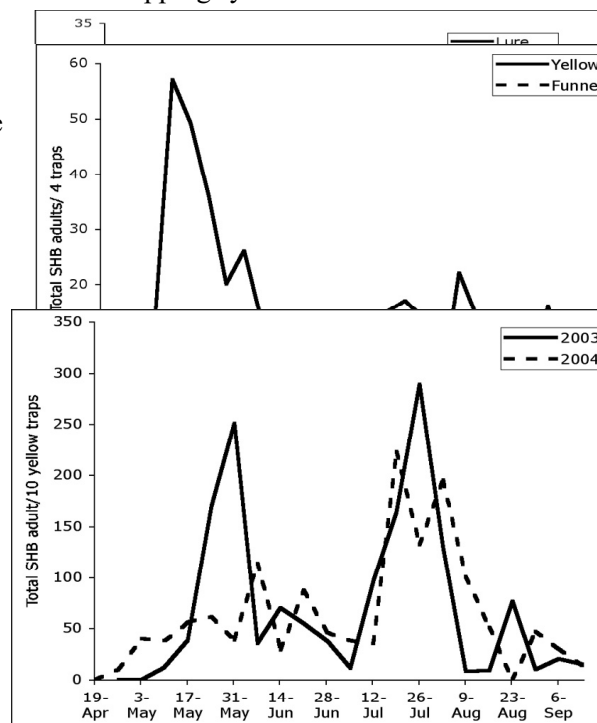


Figure 4: Seasonal life history of SHB, 2003-04.

early enough to detect the first flight.

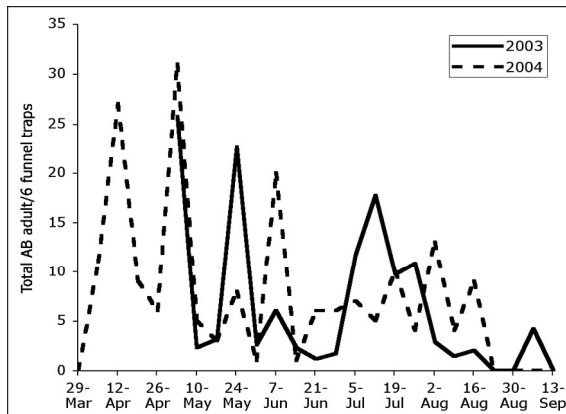


Figure 5: Seasonal life history of AB, 2003-04.

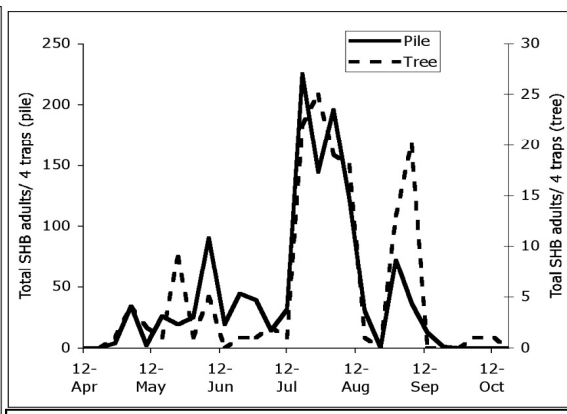


Figure 6: Emigration of SHB from woodpile source to healthy orchard trees, 2004.

Emigration of SHB from source to healthy orchards: Movement of SHB into orchards was closely associated with emergence from host material, generally a pile of recently pruned or cut wood. SHB activity was easier to monitor in the host material than in live trees, as a very large number of adults emerged from a relatively small area. The first impression was that little activity was noted in live orchard trees. However, plotting adult captures in live trees on a second y-axis with a different scale (Fig. 6) showed that relative activity in the host material and live orchard trees was very similar. These data suggested that recently emerged SHB adults were highly dispersive and readily moved distances of 10 to 50 meters from host material to live trees. SHB feeding damage in orchards was most commonly associated with movement from infested sources (Fig. 7). Generally, SHB damage was in close proximity to a source. It appeared that SHB moved readily along a border but did not move more than 2 to 3 rows into a healthy orchard.

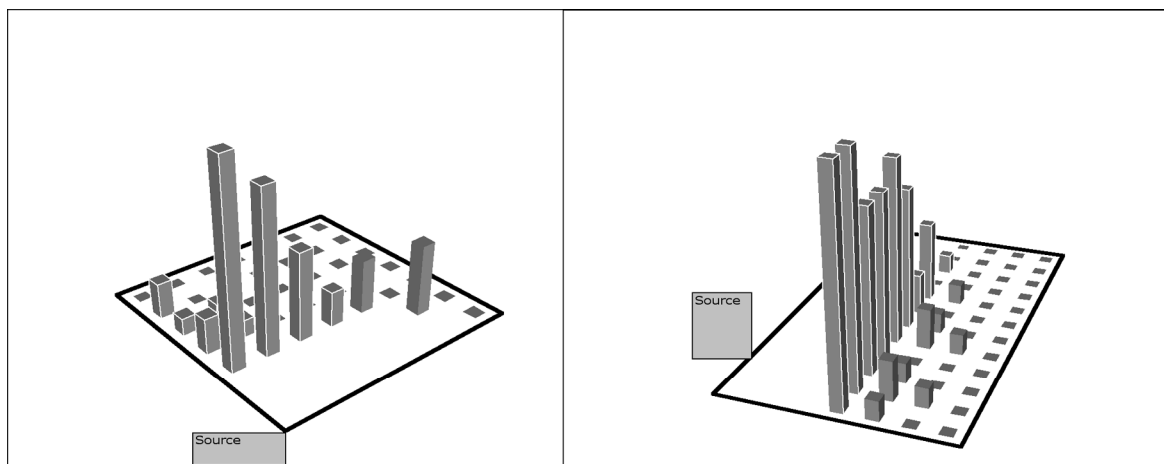


Figure 7: Relative SHB injury levels in orchard trees near an SHB source. Each bar represents a cherry tree sampled in a 20 x 20 ft grid.

Laboratory rearing: Laboratory rearing of multiple generations on an artificial diet was not successful. Adult SHB survived a short time on the diet, but no reproduction occurred. Adults were

reared and collected in large numbers from infested wood returned to the laboratory. Successive generations were then reared on current-year cuttings of apple and cherry. However, in order to rear enough adults to conduct research experiments upon, a large space commitment was necessary. Further, newly emerged SHB adults were highly dispersive and would escape all rearing arenas tested. The most efficient means of rearing SHB may very well be establishing a colony on a pile of recently cut fruitwood and then add new cuttings to the woodpile each year. This would allow for observations of insect behavior under “normal” conditions as well as providing an area where insects could easily be collected.

Host suitability: Host suitability tests showed variability in SHB development and reproductive rates between cherry, apple and pear (Fig. 8). Cherry is known to be an excellent host for SHB development, but copious amounts of sap flow acts to inhibit SHB reproduction, a defensive response in the tree. In our rearing arenas excessive moisture from the cherry cuttings resulted in significant mold development. Mold apparently reduced SHB reproduction in the cherry rearing experiment. The first emergence of new SHB was noted at 50 days in apple, with emergence from an individual arena occurring over a 3 to 4 week period. The first emergence in pear and cherry was noted at 64 days. The emergence pattern in pear was similar to apple but was delayed by 14 days. It was apparent from this study that apple and pear were suitable hosts for SHB with reproductive rates being similar. Development was accelerated in apple relative to pear.

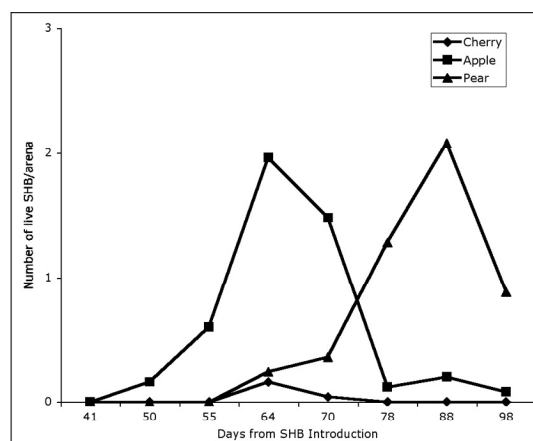


Figure 8: SHB host suitability of various fruitwood types, 2004.

It was generally reported that pome and stone fruits (*Prunus* spp., *Malus* spp., and *Pyrus* spp.) were the primary hosts of SHB. In nearly all of our monitoring trials, fruitwood was discovered to be the source material for the SHB population. This was probably the result of our trials being focused on areas very near orchards. Prunings from the previous winter were most often discovered to be the primary reproductive material. Large pieces of scaffold wood may dry more slowly and provide sufficient habitat for reproduction for a longer period of time, but generally cuttings greater than 18 months old were not suitable for SHB reproduction. We did monitor a recently cut pile of poplar or cottonwood (*Populus* spp.) placed along an orchard border. SHB moved to the cuttings immediately and were able to successfully reproduce on the 3-month old cuttings. Other studies have reported that SHB will occasionally attack ash, elm, and hawthorn, as well as other deciduous trees. It was apparent that at least the potential existed for SHB to colonize many types of deciduous hardwood trees. Thus, efforts focused on locating sources of SHB infestations may need to be directed not only to fruitwood but to other potential hosts as well.

Field-aged bioassays: A preliminary protocol was developed for screening insecticides against SHB adults. The average survival of SHB adults introduced to rearing arenas with untreated sections of wood was above 80% through the 3 days of observation. This level of survival indicates that any significant mortality noted in the rearing arenas could be attributed to pesticide exposure and not to a problem with the protocol.

Table 1. Field-aged bioassay data from candidate insecticide, 2003-04.

Insecticide	Rate (form/a)	Average corrected % mortality-2004 (2003 data in parens)
-------------	------------------	---

		1 DAT	7 DAT	14 DAT	21 DAT
Asana XL	8.0 fl oz	100.0 (100.0)	100.0 (86.1)	100.0 (100.0)	100.0 (77.7)
Actara 25W	4.5 oz	100.0 (91.7)	100.0 (44.4)	100.0 (---)	90.5 (---)
Assail 70W	3.4 oz	100.0 (50.0)	100.0 (30.6)	100.0 (29.9)	95.2 (22.2)
Avaunt 30W	6.0 oz	100.0 (66.7)	100.0 (2.7)	95.2 (0.0)	61.9 (33.3)
Carzol 92SP	1.25 lb	(58.3)	(16.7)	(---)	(---)
Guthion 50W	2.0 lb	100.0 (91.7)	100.0 (44.4)	85.7 (50.0)	42.9 (44.4)
Malathion 50%	1.5 qt	90.9 (100.0)	81.3 (72.2)	61.9 (---)	47.6 (---)
Proclaim 5SG	4.8 oz	50.0	81.3	100.0	66.7
Sevin XLR	1.0 qt	(41.7)	(2.8)	(---)	(---)
Success 2SC	6.0 fl oz	31.8 (75.0)	43.8 (30.5)	52.4 (39.9)	23.8 (11.1)
Thiodan 3E	3.0 qt	(87.5)	(16.7)	(39.4)	(55.6)

Many insecticides caused mortality of SHB in field-aged bioassays (Table 1). The pyrethroid Asana XL (esfenvalerate) was the most active through 21 days after treatment. Guthion (azinphos-methyl) and malathion provided good mortality through 7 days. Variable results were noted with Thiodan (endosulfan), Actara (thiamethoxam), Assail (acetamiprid), Avaunt (indoxacarb), and Proclaim (emamectin benzoate). However, there was evidence to suggest that these insecticides have potential to suppress SHB. Sevin (carbaryl), Success (spinosad) and Carzol (formetanate hydrochloride) all caused mortality but not at levels expected to provide adequate control. The variability of these data may show a weakness in our method (e.g. limited replication), variability based on age of collected adults, or variability among populations. Thus, more insecticide work is necessary to understand the full potential of each insecticide to control SHB under field conditions. Significant mortality was noted with many of these products for up to 7 days, and their repeated use during the growing season may contribute to maintaining SHB populations below damaging levels in most commercial orchards, especially during the first SHB generation when cherry fruit fly sprays are being applied at regular intervals. Cherry orchards may become more susceptible to injury in the postharvest period when insecticide programs for cherry fruit fly and leafroller have ceased and second generation SHB adults are able to move into unprotected orchards.

Orchard sanitation: In the winter of 2003 we monitored a concentrated effort to clean up a large SHB source that had resulted in significant damage to young, healthy cherry trees. Serious damage was noted along the orchard border despite several insecticide applications, including repeat applications of methyl parathion and endosulfan. The source was a firewood pile and a brush pile that was replenished each year with new prunings and not burned. The source was identified in September 2003, and a damage evaluation was made at that time. Damage was high but fairly isolated to the rows adjacent to the source (Fig. 9). During the winter of 2003-04 the orchard was pruned heavily, removing all

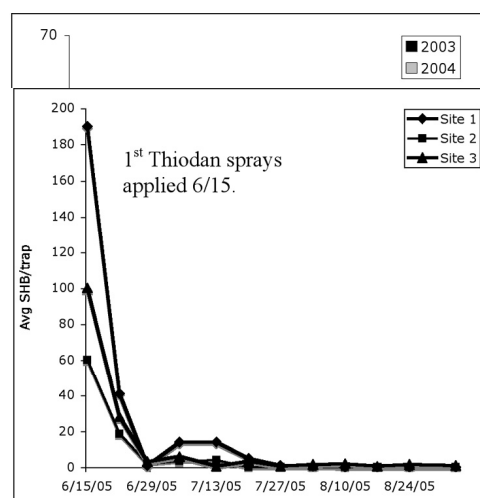


Figure 10: SHB captures from 3 high pressure locations following a sanitation program and an insecticide program targeting host material, 2005.

weakened or damaged branches. Previous year's prunings (2002-03) and current season prunings (2003-04) were returned to the laboratory and placed in emergence cages. Enough wood to fill a 2' x 2' x 1' (L x W x D) emergence cage was collected from each year's prunings. The 2002-03 prunings produced 191 SHB adults, and the 2003-04 prunings produced 6 SHB adults. These data indicated that the majority of SHB were being generated by 1-year-old prunings but also that some reproduction was occurring in the live trees. The grower made a concentrated effort to clean up all source material (firewood and brush piles and prunings) and maintain a clean area near the infested orchard. In 2004 the orchard was monitored with yellow sticky traps, ethanol-baited funnel traps, and visual inspection of damaged shoots. Insecticide applications were going to be timed with increased trap captures. However, only a total of 4 SHB adults and 9 AB adults were trapped in 5 yellow traps and 2 funnel traps over the entire season. No specific SHB insecticide applications were needed in 2004. No SHB damage was noted at any time in the 2004 season (Fig. 9).

In 2005, we identified three locations in Okanogan with easily identified sources of SHB located just outside of cherry blocks. Further, infested limbs and prunings were serving as host material within the cherry blocks. These locations were brought to our attention after first generation adults caused serious damage to orchard borders. Insecticidal control options were limited because one of the blocks was managed organically and the conventional blocks were experiencing damage levels of 50% shoot infestation despite a history of border sprays. Although yellow traps were placed near the end of first generation activity, captures in the first week averaged 116 SHB/trap in the source areas across all three sites (Fig 10). The grower immediately began a concerted effort to remove all possible host material from within the orchard. This involved picking up the previous winter's cuttings and pruning out all weakened branches or limbs. This host material was added to the source material located outside the orchard. The grower then started an intensive spray program targeting the woodpiles. Thiodan was applied by handgun on a 10-to 14-day retreatment interval for the rest of the season. Care was taken to thoroughly soak the entire woodpile. Although the new host material was added to the woodpiles, no second-generation activity was noted at any of the three sites.

Our experience with SHB management indicated that orchard sanitation was the most important factor in contributing to a reduction in SHB densities and damage to live cherry trees. Sanitation programs must include removing potential host material (weakened limbs, recent prunings) from within the orchard and eliminating any host material outside the orchard. Host material can be "eliminated" by burning the wood or thoroughly soaking the wood with an effective insecticide delivered by a handgun sprayer. The increased volume of water delivered by handgun applications was an important factor in insecticide efficacy.

Budget:

Project title: Biology and management of bark beetles

PI: Jay F. Brunner

Project duration: 2 years

Current year: 2004-05

Project total (2 year): \$32,000

Year	Year 1 (2004)	Year 2 (2005)
Total	16,000	16,000

Current year breakdown

Item	Year 1 (2004)	Year 2 (2005)
Salaries ¹	4,178	4,345

Benefits (29%)	1,212	1,260
Wages ²	7,000	7,000
Benefits (16%)	1,120	1,120
Equipment	0	0
Supplies ³	1,000	1,000
Travel ⁴	1,490	1,275
Miscellaneous	0	0
Total	16,000	16,000

¹ Mike Doerr – one month salary – conduct bioassays and supervise rearing.

² One person for four months to conduct field sampling.

³ Supplies include rearing materials, traps and preservation materials for beetles. Cell phone charges are allowed under this grant.

⁴ Travel – one vehicle for 3-4 months at \$350/month plus fuel to collect field samples.

Final Report
WTFRC Project# CCAB

Project Title: Use of surfactants to remove surface pests

P. I.: James D. Hansen

Organization: USDA-ARS, Wapato, WA

Address: 5230 Konnowac Pass Road, Wapato, WA, 98951,
(509) 454-6573 jimbob@yarl.ars.usda.gov

Co-PI's: none

Contract Administrator: Carolyn Yager, 509-454-6575, cyager@yarl.ars.usda.gov

Objectives:

The overall objective of this program is to determine the removal and lethal effects of food grade silicone-based materials on surface pests of sweet cherries that are suitable for commercial packing lines. Efficacious parameters to be determined are:

1. **Identification of best material.** Specific formulations of emulsions and defoamers that best remove or kill will be determined for a range of surface arthropods.
2. **Exposure duration.** Time-efficacy curves will be calculated to determine the most efficacious dip duration.
3. **Physical methods.** A shower will be used to dispense the best surfactant and compared to efficacy of submersions of the same material. In other tests, the bath material will be circulated within dip bins.
4. **Surface arthropods examined.**
 - a. Grape mealybug, *Pseudococcus maritimus* (Ehrhorn) (Homoptera: Pseudococcidae).
 - b. Obliquebanded leafroller (OBLR), *Choristoneura rosaceana* (Harris) (Lepidoptera: Tortricidae).
 - c. Western flower thrips, *Frankliniella occidentalis* (Pergande) (Thysanoptera: Thripidae).
 - d. Twospotted spider mite, *Teranychus urticae* Koch (Acari: Tetranychidae).
 - e. Tydeid mites, spp. (Acari: Tydeidae).

Significant findings:

- The waxy coverings of grape mealybugs and the silk shelters of OBLR larvae dissolved more thoroughly with increased concentrations of sodium hypochlorite (bleach) (Fig. 1 and 2).
- The waxy coverings of mealybugs also dissolved more with increased exposure to bleach (Fig. 1).
- Water baths with bleach, followed by water showers effectively removed OBLR larvae (Fig. 3).
- Tydeid mites were difficult to expunge, but showers aided in their removal (Fig. 4).
- Water and water with surfactant were efficacious in removing two-spotted spider mites (Fig. 5).
- The “360” formulation of the silicone emulsion surfactant was more effective in removing western flower thrips than the “335” formulation (Fig. 6).

Methods:

Experimental Design. Test materials were examined as dips. Efficacy was determined by submerging specific arthropods for predetermined durations. No dips were longer than 5 minutes. Applicable life stages of selected surface arthropods were examined. A spider mite colony and an OBLR colony at YARL were used as sources for the test subjects, and tydeid mites were field collected on grape leaves. Mealybugs from an established colony at YARL represented generic surface insects. To test efficacy, a known number of arthropods (live and dead) were placed on cherry fruits, then recounted (live and dead) after treatment.

Specific objectives were obtained by the following procedures.

1. **Identification of best material.** Food grade silicone-based surfactants were examined at different concentrations. Physical properties of the materials, such as foaminess and residue formation, were noted.
2. **Exposure duration.** Dip efficacy was tested at 2, 3, and 5 min. Longer durations would not be practical for the packing line.
3. **Physical methods.** A shower system, previously built to test hot water systems, was used to wash surface arthropods with ambient water. A pump system was used to test circulating baths within dip baths.

Results and discussion:

The two surfactants that were examined are food grade polydimethyl silicone emulsions. The “360” formulation has 60% active ingredients (ai) whereas the “335” has 35% ai. Both chemicals are rated for indirect food contact at the recommended 1% formulation rate and both materials were shown not to cause fruit damage to cherries in recent tests conducted at UC-Davis.

Mealybugs are covered with a waxy protective material and OBLR larvae reside within protective silk shelters (Fig. 1 and 2). Both the wax and the silk can be dissolved by using bleach at concentrations that do not damage cherry fruits. The OBLR has become a phytosanitation problem in California exports when the silk shelters become attached to cherry fruits. Elimination of the silk will aid in larval removal. Further studies will examine the use of showers to wash away bare OBLR larvae.

The OBLR larvae used in the shower tests were more active than the ones used in the bleach bath tests above and inclined to move off the fruits. However, the water showers effectively removed those remaining when used after a bleach bath (Fig. 3). In this test, the showers were the same duration as the preceding baths. This procedure shows promise and additional refinements are needed to make the methods compatible with commercial operations.

The tydeid mites were the most difficult to remove (Fig. 4). They are very small and hide in tight places. Unlike the other arthropods we examined, these mites were treated on grape leaves, their natural host. This was necessary because of the numbers needed to conduct tests and these mites are uncommon in cherry fruits from the Pacific Northwest. The surfactant baths alone were not sufficient for their removal. The showers increased the efficiency in removal, but additional experimentation is needed to develop procedures that can be used on cherry fruits.

Spider mites have also been a phytosanitation problem for exported cherries. Our studies indicate that water alone will remove a third of the pretreatment population, but the rate of removal is increased with a surfactant (Fig. 5). The two formulations we examined worked equally well, with a slight edge going to 360 EFG.

The 360 EFG surfactant was significantly effective in removing western flower thrips when exposed to 3 min or less (Fig. 6). Thrips are sometimes a phytosanitation problem, but not as severe as mites or OBLR. The numbers were examined were from natural infestations and were lower than

the artificial infestations of the other arthropods. Yet, our study indicates that methods for OBLR removal are compatible to thrips elimination.

Water baths, particularly those with bleach, were effective in removing protective waxes and silks of the surface pests. Incorporating a shower also increased removal efficacy. Additional studies will be conducted to improve these removal techniques and to test a combination surfactant with bleach. Further research will be conducted using immature apples to represent cherry fruits during the winter months.

Budget:

Project title: Use of surfactants to remove surface pests
PI: James D. Hansen
Project duration: 2005
Current year: 2005
Project total (1 year): \$8,700
Current year request: \$8,700

Item	Year 1 (2005)
Wages ¹	7665
Benefits	767
Supplies ²	268
Total	8,700

¹ GS-5, with 10% benefits, for 10 weeks .

² Supplies: Maintenance of arthropod colonies; routine laboratory equipment (towels, holding containers, vials, etc.); purchase of fruits.

YARL Contribution:

1. Materials and equipment
 - a. holding cages and environmental rooms for test arthropods
 - b. chemicals to be evaluated
 - c. test facilities
 - d. shower unit and circulating dip baths
 - e. temperature probes and data acquisition
2. Transportation for obtaining fruits
3. Labor: GS-8 lead technician to supervise activities

Acknowledgements:

Funding for this project came, in part, from the California Cherry Advisor Board (CCAB). I thank the following for their support and participation: M. Heidt (ARS-Wapato), M. Watkins (ARS-Wapato), D. Pearson (ARS-Wapato), J. Culbertson (CCAB-Lodi), and B. Mitcham (UC-Davis). The surfactants were provided by Ivanhoe Industries (Mundelein, IL). T. Unruh (ARS-Wapato) provided the initial OBLR, and L. Wright (WSU-Prosser) provided the tydeid mites.

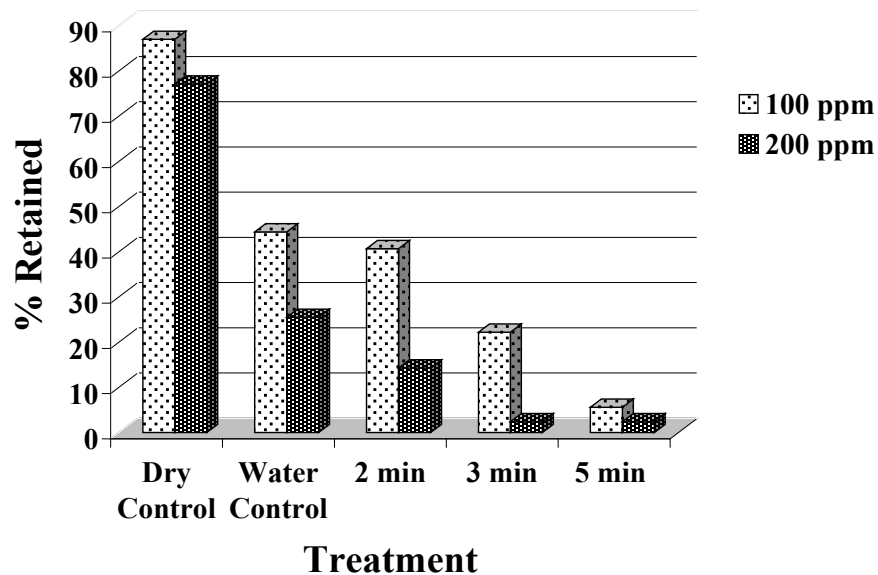


Fig. 1. Retention of grape mealybugs exposed to two bleach concentrations for three time periods.

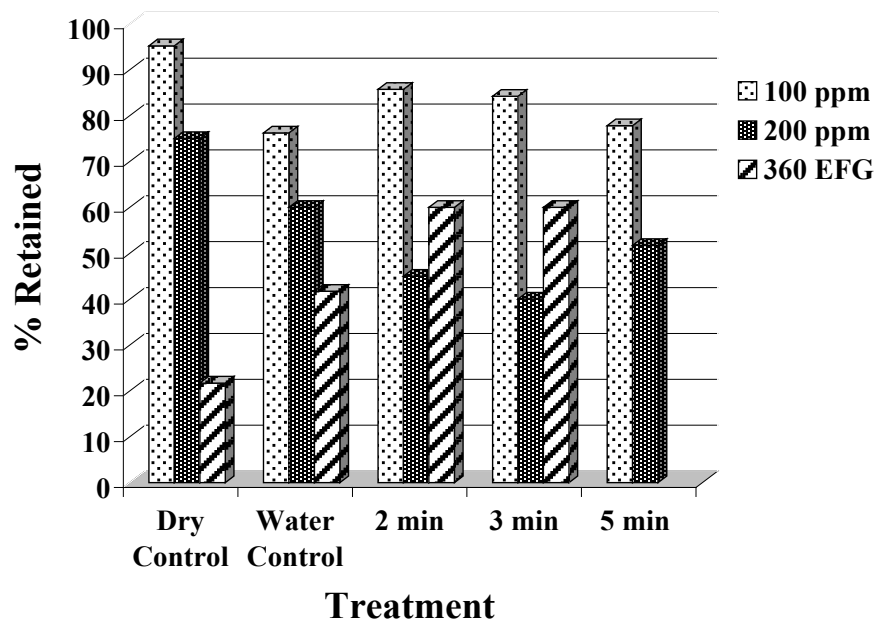


Fig. 2. Retention of OBLR larvae exposed to different bleach concentrations or 1% 360 EFG surfactant for three time periods.

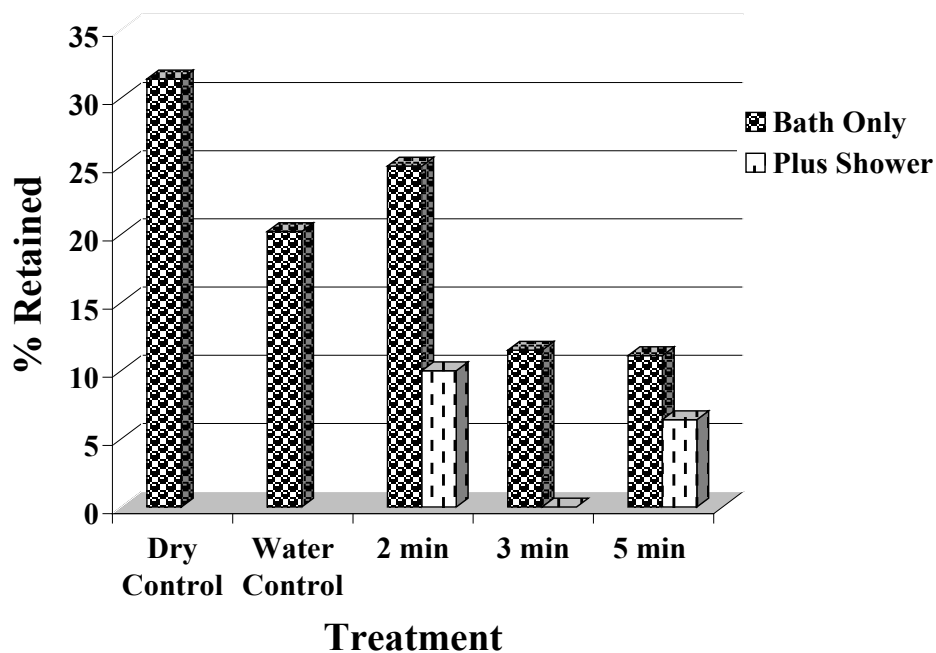


Fig. 3. Retention of OBLR larvae exposed to 100 ppm bleach concentrations in baths for three time periods, with some followed by water showers at the same durations as their respective baths.

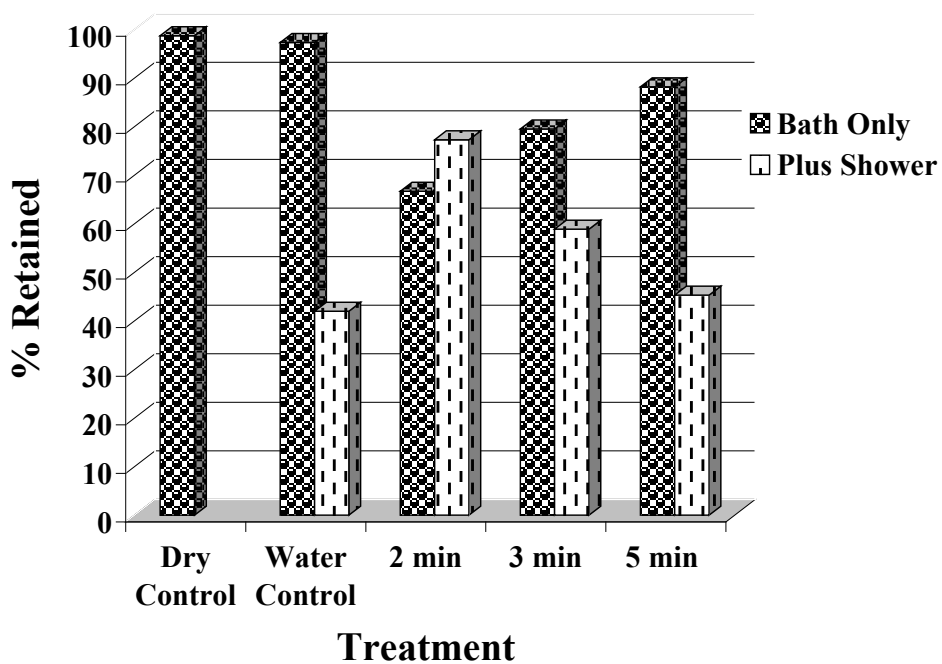


Fig. 4. Retention of tydeid mites exposed to 1% 360 EFG surfactant in baths for three time periods, with some followed by water showers at the same durations as their respective baths.

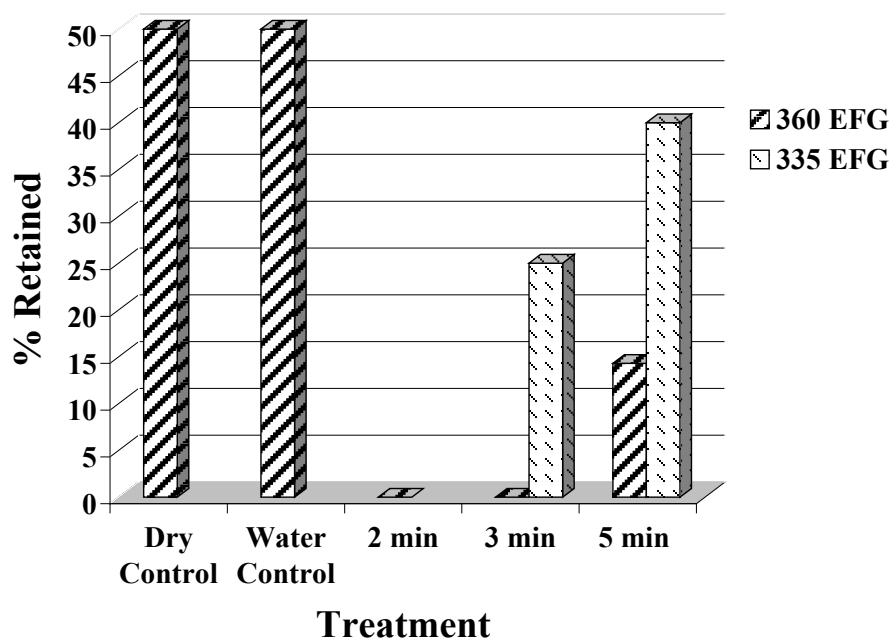


Fig. 5. Retention of western flower thrips exposed to three time periods of two surfactant formulations.

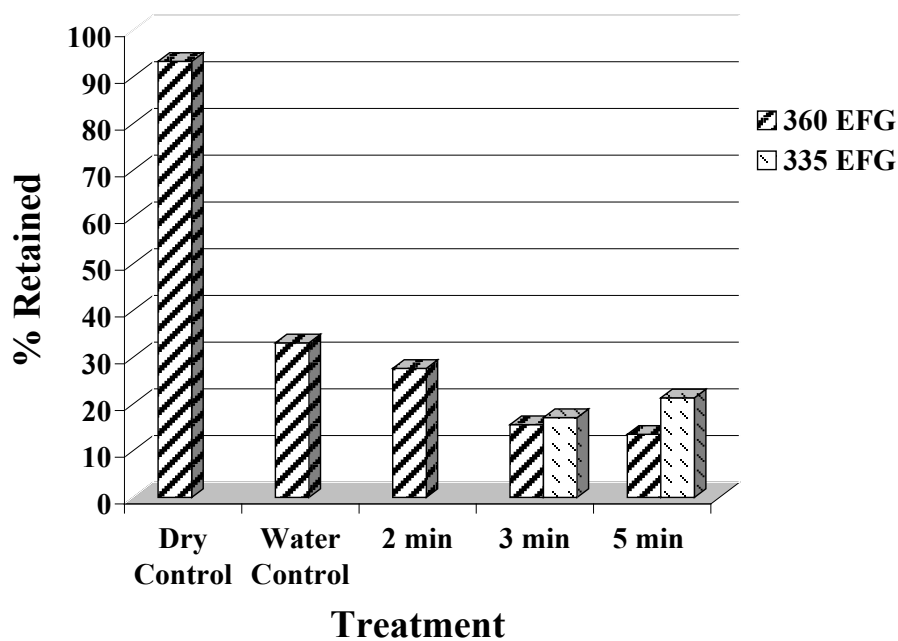


Fig. 6. Retention of twospotted spider mites exposed to three time periods of two surfactant formulations.

CONTINUING PROJECT REPORT
WTFRC Project # CH-04-401

YEAR 2/3

Project title: Evaluation of insecticide effects on the biology of cherry fruit flies
PI: Wee Yee
Organization: USDA-ARS
Cooperators: Many homeowners in central and western Washington
Contract Administrator: Pete Landolt, e-mail: landolt@yarl.ars.usda.gov
phone #: (509) 454-6570.

Objectives (2004-2006):

- 1) Determine effects of commercial insecticides on mortality, sublethal responses, behavioral responses, and translaminar effects.
- 2) Determine relative importance of contact or ingestion mechanisms of kill.
- 3) Determine residual activity of insecticide formulations in the field.
- 4) Determine effects of insecticide formulations on larval infestations in the field.

Goals and Activities and Anticipated Accomplishments for 2006:

- 1) Complete studies on behavioral responses to insecticide-treated cherries, preference for treated versus untreated cherries.
- 2) Determine feeding times on insecticide droplets.
- 3) Determine effects on egg production of flies treated with sublethal amounts of insecticides.
- 4) Further define translaminar effects on cherries with various stages of fly eggs and larvae during early, mid, and late season cherries, test effects of new insecticides such as Avaunt (indoxacarb) and Actara (thiamethoxam) on mortality, biology of flies.

Significant findings in 2005:

- Entrust and GF-120 (spinosad insecticides) were the most effective insecticides and caused 100% mortality within 1-4 days, followed by Provado (imidacloprid) and Calypso (thiacloprid).
- Calypso had the highest ovicidal (egg-killing) and larval-killing activity.
- All insecticides reduced larval emergence from cherries, with Provado most effective.
- Spinosad in GF-120 and Entrust had equally high contact and oral activity; Provado and Calypso had higher oral than contact activity.
- GF-120 had residual activity of 14 days; Entrust, Provado, and Calypso had no activity after 14 days of aging.

Methods for 2006:

1) *Complete studies on behavioral responses to insecticide-treated cherries; preference for treated versus untreated fruit.* Experiments using the label rates of and label directions for the following were conducted: (1) Entrust (organic formulation of spinosad, no bait), (2) GF-120 (spinosad mixed with bait containing sugar, protein, attractants), (3) Provado (imidacloprid), Calypso (thiacloprid). Other insecticides such as Avaunt (indoxacarb, an oxadiazine) and Actara (thiamethoxam, a neonicotinoid) will also be tested. An untreated control was included. These products will be tested without sugar (except GF-120).

Cherries will be sprayed with insecticides and residues will be allowed to dry. Flies will be released inside cages with the treated cherries. In one test, there will be a choice of treated versus

untreated cherries. In the other test, there will either treated or non-treated cherries. Groups of 20 or 10 flies will be exposed to one treatment per pint-size container. Tests will be conducted over 3 weeks. A new set of cherries will be introduced into containers every three days. Numbers of eggs in the cherries will be counted. If insecticides are repellent, there should be more eggs in control cherries.

2) *Feeding times on insecticide droplets.* Observations will be made of flies exposed to droplets of insecticide solution with and without 20% sucrose. Solutions will be GF-120, Entrust, Provado, and Avaunt, and Actara. Flies will be weighed before and after exposures; percent responding will be determined. The numbers of flies approaching the droplets but not feeding will also recorded. Mortality will be recorded at 1, 3, and 7 days post exposure. Repellent effects (avoidance of drops) will be recorded.

3) *Effects sublethal amounts of insecticides on egg production of flies.* For each treatment, egg production of flies exposed to small amounts of insecticide on the dorsa or ingested will be determined by allowing flies to lay eggs into untreated cherries at days 10-14. Surviving flies exposed previously to insecticides will be paired with non-exposed mates to observe for mating and egg production.

4) *Further define translaminar effects on cherries with various stages of fly eggs and larvae.* Cherries will be collected during early, middle and late season in Richland and Kennewick and treated with insecticides. These correspond to periods when cherries have mostly eggs, young larvae, and old larvae, respectively. Samples of cherries from each period will be opened and preserved in EtOH to determine the stages inside the fruits at the time of treatments. Applications of Entrust, GF-120, Provado, Calypso, and Avaunt ? (high label rates) will be made.

2005 Results and Discussion:

1) *Determine mortality and translaminar effects.* Entrust and GF-120 (spinosad insecticides) were the most effective insecticides against adults and caused 100% mortality within 1-4 days, followed by Provado (imidacloprid) and Calypso (thiacloprid). This pattern changed when immature stages were studied. Calypso had the highest ovicidal (egg-killing) activity and highest activity against the early instar larvae (Table 1). The late instars were resistant to all insecticides in one test, but equally susceptible to all of them in a second test (Table 2). Larvae that survived and pupated were not affected by the insecticides. When infested cherries were sprayed with all insecticides, there was no evidence the large larvae were killed by insecticides, based in dissections of fruit. However, larval emergence rates were reduced, indicating movement of all materials into the cherries. The materials either prevented egg hatch or killed a small percentage of the small larvae. The results suggest that sprays applied on unpicked fruit after harvest may reduce populations.

2) *Determine relative importance of contact and ingestion mechanism of kill.* Contrary to expectations, spinosad in GF-120 and Entrust had equally high contact and oral activity; Provado and Calypso had higher oral than contact activity (Table 4). Although it is possible the materials may have spread down the thorax of flies and caused the flies to feed on the materials, the fact is that flies did not need to directly ingest the materials to suffer high mortality. Thus sprays of GF-120 and the other insecticides may affect control through both contact and ingestion of materials.

3) *Determine residual activity of insecticide formulations in the field.* There were clear effects of aging insecticides and baits and of different insecticides on fly mortality (Table 5). Entrust lost effectiveness when aged over the 14 days, whereas GF-120 did not lose any effectiveness over 14

days. Provado lost effectiveness after only 3 d of aging. Calypso did not lose any effectiveness over 14 d of aging, but it also was ineffective, causing lower mortality than any of the other materials (Table 5). At 1 DAE, mortality within 0-d old residues ranked as follows: GF-120=Entrust>Provado>Calypso. With 3-d old residues, the ranking was GF-120>Entrust=Provado=Calypso. With 7- and 14-d old residues, GF-120 caused greater mortality than all other materials, with Calypso least effective. These same rankings were generally seen at 3 and 7 DAE, even though there was increased overall mortality, including in the controls. Significantly, GF-120 was the only material that caused 100% mortality by 7 d after exposure when aged 0, 3, and 14 d (Table 5). The results clearly suggest that GF-120 has the longest residual activity when there is little precipitation. It is possible that the bait component of GF-120 (sugars, ammonium acetate, oil, etc.) protected the spinosad from degradation or that it concentrated the spinosad on the leaves. Entrust possibly was more easily washed off or subjected to degradation by the sun's rays. Under ideal conditions, it appears that GF-120 can be applied every 14 days and remain effective. The others need to be applied every 7 days.

Significance to the Industry and Potential Economic Benefits: The effectiveness of various newer insecticides on the biology and control of cherry fruit flies clearly differ. The results indicate that the choice of newer insecticides had significant impacts on fly control, which is increasingly important as older insecticides begin to be phased out. The use of the most effective insecticide reduces the risk of infestations in orchards and of bins being rejected at the packinghouse. Less frequent use of insecticides through a thorough understanding of the effects of insecticides on different life stages of the fly may save spray costs. The continued use of one material, including spinosad, may potentially result in resistance, if not in fruit flies, then perhaps in other, non-targeted, currently pests on cherries such as leafrollers. Thus, we should always be ready with alternative or backup insecticide chemistries and not wait and be unprepared when such problems arise.

Table 1. Effects of insecticides and insecticide exposure time on percent egg hatch (\pm SE) of cherry fruit flies after 14 d in the laboratory

Treatment	15-s Insecticide Exposure	Continuous Insecticide Exposure
Control	41.0 \pm 5.8b	42.4 \pm 9.0b
Entrust	37.0 \pm 6.9b	0.0 \pm 0.0a
Provado	25.0 \pm 4.7b	0.0 \pm 0.0a
Calypso	3.3 \pm 1.6a	0.0 \pm 0.0a

15-s exposure: six replicates of 50 eggs; continuous exposure: five replicates of 50 eggs each. Means followed by the same letter inside parentheses within days are not significantly different (LSD test, $P > 0.05$).

Table 2. Effects of treating small and large larvae with insecticides on percent mortality (\pm SE) of larvae and pupae of cherry fruit flies after 2 d

Treatment	Small Larvae (2-3 mm long)	
	% Larvae Dead	Larval Lengths (mm) ^a
Control	6.9 \pm 4.5a	3.5 \pm 0.1a
Entrust	69.7 \pm 5.0b	3.3 \pm 0.2 a
Provado	77.3 \pm 6.1b	3.0 \pm 0.1 a
Calypso	91.0 \pm 5.6c	3.1 \pm 0.3 a

<u>Large Larvae (5.5-7 mm long) Test 1</u>			
Treatment	% Larvae Dead	% Larvae Alive	% Pupae
Control	35.0 ± 15.5a	30.0 ± 14.7a	35.0 ± 2.9ab
Entrust	72.5 ± 11.1a	10.0 ± 5.8a	20.1 ± 7.1a
Provado	37.5 ± 11.1a	20.0 ± 7.1a	42.5 ± 8.5b
Calypso	32.5 ± 6.3a	2.5 ± 2.5a	65.0 ± 6.5c

<u>Large Larvae (5.5-7 mm long) Test 2</u>			
Treatment	% Larvae Dead	% Larvae Alive	% Pupae
Control	38.0 ± 8.0a	22.0 ± 8.6a	40.0 ± 4.5b
Entrust	90.0 ± 3.2 b	4.0 ± 2.4 b	6.6 ± 2.4a
Provado	72.4 ± 11.2b	4.0 ± 2.4 b	23.6 ± 10.3b
Calypso	77.3 ± 4.1b	0.0 ± 0.0 b	22.7 ± 4.1b

Five (small larvae, large larvae, test 2) or four replicates (large larvae, test 1) of 10 larvae each. Means followed by the same letter inside parentheses within dates are not significantly different (LSD test, $P > 0.05$).

^aMeasured 48 h after end of experiment and storage in water.

Table 3. Effect of spraying cherries with insecticides and bait on larval mortality (\pm SE) and numbers of larvae (\pm SE) of cherry fruit flies emerging from cherries collected in Kennewick, WA, 2005

Treatment	<u>Dead Larvae/10 Fruit^a</u>		<u>Live Larvae/10 Fruit^a</u>	
	No.	Length (mm) ^b	No.	Length (mm)
Control	0.2 ± 0.2a	6.8	12.6 ± 1.5b	4.9 ± 0.4a
Entrust	1.0 ± 1.0a	4.5	12.0 ± 1.8b	4.2 ± 0.3a
GF-120	0.8 ± 0.5a	4.6 ± 0.6	11.4 ± 2.9b	4.2 ± 0.4a
Provado	1.2 ± 0.4 a	3.5 ± 1.7	6.4 ± 0.5a	3.8 ± 0.2a
Calypso	0.8 ± 0.4a	2.1 ± 0.4	6.4 ± 0.7a	4.9 ± 0.3a

Treatment	<u>No. Pupae/100 Fruit</u>			% Pupae Dead
	Days 1-15	Days 16-30	30-Day Total	
Control	130.8 ± 12.5b	44.4 ± 5.8d	175.2 ± 10.8d	36.9 ± 3.2a
Entrust	70.4 ± 10.3a	33.3 ± 6.9cd	103.4 ± 11.6b	39.6 ± 6.4a
GF-120	66.0 ± 5.5a	18.6 ± 3.9bc	84.6 ± 7.9ab	40.4 ± 6.1a
Provado	58.2 ± 13.6a	6.0 ± 3.6a	64.2 ± 11.2a	42.3 ± 11.1a
Calypso	89.0 ± 15.7a	12.0 ± 7.4ab	101.0 ± 16.7b	39.4 ± 6.6a

Five replicates of the control and each treatment.

^aAt 8 days post-treatment; one application only.

Means followed by the same letter inside parentheses within dates are not significantly different (LSD test, $P > 0.05$).

^bTwo few to analyze statistically.

Table 4. Effects of contact with and ingestion of insecticides on mean days survived (\pm SE) post treatment^a by single adult cherry fruit flies

Treatment	N	Contact with Water or Insecticide ^b		Ingestion of Water or Insecticide ^c		N	Males	N	Females
		Males	N	Females	N				
Water	17	19.1 ± 3.0a(b)	25	15.5 ± 2.4a(b)	17	16.1 ± 3.0a(b)	18	17.3 ± 3.1a(b)	
Entrust	18	4.9 ± 2.2a (a)	20	2.8 ± 1.4a(a)	15	1.0 ± 0.0a(a)	17	1.2 ± 0.2a(a)	
Provado	15	7.5 ± 2.4a(ac)	18	10.2 ± 2.4a(b)	17	4.3 ± 2.1a(a)	18	7.0 ± 2.4a(a)	
Calypso	16	13.3 ± 3.1b(bc)	18	12.3 ± 2.9b(b)	17	6.1 ± 2.3 ab(a)	19	4.3 ± 2.1a(a)	

Water and all treatments contained 20% sucrose.

^aFollowed for maximum of 30 d; flies 3-7 d old at treatment.

^b2 μ l drop applied on top of thorax.

^c2 μ l drop exposed to flies; all flies fed, but drop not always entirely consumed.

Means followed by the same letter within rows outside parentheses are not significantly different (LSD test, $P > 0.05$).

Means followed by the same letter within columns inside parentheses are not significantly different (LSD test, $P > 0.05$).

Table 5. Effects of field-aged insecticide and bait residues on leaves (6-20 June 2005) on mean cumulative percent mortality (\pm SE) of adult cherry fruit fly in the laboratory at 1, 3, and 7 days after exposure (DAE)

	Age of Residues on Cherry Leaves at Initial Exposure to Flies			
	0 d (fresh)	3 d	7 d	14 d
1 DAE				
Control	3.3 \pm 2.0 a(a)	1.7 \pm 1.7a(a)	3.5 \pm 2.1a(a)	1.7 \pm 1.7a(a)
Entrust	70.0 \pm 8.2c(c)	26.0 \pm 13.8b(b)	14.6 \pm 6.5ab(a)	0.0 \pm 0.0a(a)
GF-120	78.3 \pm 10.1a(c)	72.4 \pm 9.5a(c)	70.1 \pm 12.8a(b)	79.4 \pm 6.9a(c)
Provado	34.2 \pm 6.6b(b)	17.0 \pm 4.5b(b)	23.0 \pm 9.4b(a)	5.0 \pm 3.3a(ab)
Calypso	8.3 \pm 4.6a(a)	3.5 \pm 2.1a(ab)	8.8 \pm 5.6a(a)	13.3 \pm 5.0a(b)
3 DAE				
Control	10.1 \pm 1.6a(a)	3.3 \pm 2.0 a(a)	8.6 \pm 2.6a(ab)	3.3 \pm 2.0a(a)
Entrust	95.0 \pm 3.3b(c)	76.7 \pm 19.4b(c)	65.0 \pm 11.0b(c)	8.3 \pm 3.7a(ab)
GF-120	98.3 \pm 1.7a(c)	100.0 \pm 0.0a(c)	91.1 \pm 7.1a(d)	96.9 \pm 3.1a(c)
Provado	63.0 \pm 8.4c(b)	42.3 \pm 10.2bc(b)	32.3 \pm 11.4ab(b)	11.7 \pm 6.2a(ab)
Calypso	16.7 \pm 5.3a(a)	19.3 \pm 3.2a(ab)	8.8 \pm 5.6a(a)	26.7 \pm 8.1a(b)
7 DAE				
Control	23.0 \pm 3.0a(a)	17.6 \pm 2.7a(a)	20.4 \pm 9.6a(a)	15.0 \pm 4.9a(a)
Entrust	98.3 \pm 1.7b(bc)	88.3 \pm 11.7b(c)	98.3 \pm 1.7b(c)	17.2 \pm 7.5a(b)
Gf-120	100.0 \pm 0.0a(c)	100.0 \pm 0.0a(c)	98.2 \pm 1.8a(c)	100.0 \pm 0.0a(a)
Provado	91.7 \pm 4.6c(b)	57.4 \pm 11.1b(b)	41.0 \pm 9.7ab(b)	23.3 \pm 6.7a(a)
Calypso	31.7 \pm 7.2a(a)	35.1 \pm 7.6a(ab)	23.8 \pm 4.2a(ab)	35.0 \pm 10.7a(a)

Five replicates of 12 flies each (8 males, 4 females).

Means followed by the same letter within rows outside parentheses are not significantly different (LSD test, $P > 0.05$).

Means followed by the same letter within columns inside parentheses are not significantly different (LSD test, $P > 0.05$).

Budget:**Project title:** Evaluation of insecticide effects on the biology of cherry fruit flies**PI:** Wee Yee**Project duration:** 2004-2006**Current year:** 2006**Project total (3 years):** \$81,000**Current year request:** \$27,000

Item	Year 1 (2004)	Year 2 (2005)	Year 3 (2006)
Salaries	22,050 ^a	22,050 ^a	22,050^a
Benefits	2,450 ^a	2,450 ^a	2,450^a
Wages	0	0	0
Benefits	0	0	0
Equipment	1,000	1,000	1,000
Supplies	1,000	1,000	1,000
Travel	0	0	0
Miscellaneous	500	500	500
Total	27,000	27,000	27,000

^aOne GS-3 (6,000), one GS-5 (18,500)

CONTINUING PROJECT REPORT
WTFRC Project # CH-04-402

YEAR 2/3

Project title: Fly feeding ecology and food-based lures and baits
PI: Wee Yee
Organization: USDA-ARS
Co-PI(s) and affiliations (s): Pete Landolt, USDA-ARS
Cooperator (s): Dr. Carol Lauzon, California State University; various homeowners with cherry trees
Contract Administrator: Pete Landolt, e-mail: landolt@yarl.ars.usda.gov; phone #: (509) 454-6570.

Objectives (2004-2006):

- (1) Identify foods of western cherry fruit flies in nature.
- (2) Determine when the flies feed, both daily and seasonally, and how much sugar and protein flies feed on in nature.
- (3) Determine the most attractive protein and sugar baits in the laboratory and field; baits that stimulate highest feeding.

Goals and Activities and Anticipated Accomplishments for 2006:

- 1) Continue studies on foods of flies; identification of substrates most likely used by flies.
- 2) Continue studies on protein feeding; determination of amounts of sugar and protein in the environment and in flies.
- 3) Identification of more attractive and longer-lasting baits; determination of most effective bait sprays.

Significant findings in 2005:

- Nearly all feeding occurred on leaf surfaces, with grazing behaviors most common; most nutrients appear to be obtained from leaf surfaces during early season. Honeydew and leaf nectaries are not or rare sources of sugars for flies.
- Flies exposed to leaves only survived on average 1 day more than controls, suggesting low but some nutrient acquisition.
- Main protein or nitrogen source probably bacteria on leaf surfaces, with bird droppings a minor source.
- In the laboratory, only hungry flies responded to three protein baits: GF-120, Nulure, and Mazoferm. All were equally attractive to hungry flies.
- In the field, flies were equally attracted to water, GF-120, Nulure, and Mazoferm, suggesting the baits have little attraction even at close range in nature.
- Mazoferm and NuLure sprays with spinosad and GF-120 sprays reduced larval infestations of cherries compared with controls, but did not eliminate them.

Methods for 2006:

(1) *Continue studies on foods of flies; identification of substrates most likely used by flies.* Foods used by flies in nature and whether these foods change over the season will be determined. Identification of foods may lead to improved attractants to use in bait sprays. The hypothesis is that flies feed primarily on nutrients (leachates) on surfaces of cherry leaves, but these nutrients change over the season. Flies can obtain all their required nutrients within a cherry tree. The design of the experiment will consist of four independent variables – season, food type, fly sex, and fly physiological state (0-6 d old, and ≥ 14 d old; older flies are more likely mated) and two main response variables – fly longevity and fecundity. Each tree will be considered a replicate. Cherry leaves of various stages over the season will be either washed with methanol or unwashed and exposed to flies inside cages in the laboratory. Mortality of flies will be determined daily. If survival

is prolonged, fecundity will be determined by exposing flies to artificial wax cherries, which will act as oviposition substrates. Representative flies will be analyzed for sugar, protein, and fat content using Anthrone and other standard biochemical tests. Nutrients on leaves will be identified and quantified using HPLC. In a separate test, potential fly food sources such as cherry leaves, cherry fruit, aphid honeydew on leaves, bird droppings, pollen on leaves, and combinations of these (in particular, cherry fruit and bird droppings and honeydew and bird droppings) will be collected over the season and exposed to 0-6 and ≥ 14 old flies inside pint- or one gallon-size cartons. The role of bacteria in fly nutrition will also be studied by exposing flies to diets with and without bacteria (collaboration with Dr. Carol Lauzon). Survival and fecundity of flies exposed to each treatment will be determined.

A second study will be conducted to determine the movement of flies within localized areas to test the hypothesis that flies find their food through indiscriminant feeding on leaves because few point sources of food are available on cherry trees. This may improve the way bait sprays are applied to trees. The design will consist of two independent variables - season and behavioral activity category, which includes feeding, walking, mating, and resting. The percentages of flies engaged in activities will be calculated, with each of three trees considered a replicate. Fly feeding and movement patterns will be determined by watching individual flies on trees within pre-determined marked areas of 1 square meter on the tree periphery. The amounts of times spent by flies within these areas up to 15 min will be recorded. At least 20 flies will be followed two or three times a week on three trees during the season. Following flies will be challenging, but detailed studies of this sort indicate flies can be seen easily in some trees. If too many flies are lost by 20 min, the length of time followed will be reduced (starting with 15, 10, down to 5 min) or numbers of flies followed increased, or both. Feeding activities will be either grazing on leaf surfaces or feeding on specific visible food substances such as cherry juice, bird droppings, or exudates from leaf nectaries.

(2) *Continue studies on protein feeding; determination of amounts of sugar and protein in the environment and in flies.* Methods will be similar to the first part of (1) above. Protein will be removed from cherry leaves using water and detergent and analyzed using Bradford or Lowry reagent and a spectrophotometer. The nitrogen concentrations will be converted into protein.

(3) *Identification of more attractive and longer-lasting baits; determination of most effective bait sprays.* In the laboratory, three available commercial baits will be further tested: GF-120, Mazoferm (a corn extract), NuLure, and various formulations of yeast compounds. New bait formulations with various known proportions of proteins, ammonia compounds, sugars, and cherry fruit volatiles (aldehydes, alcohols, and esters) will also be tested against the most effective baits. Sun-blocking agents will be added to baits and tested. Baits will be sprayed on leaves and aged for 1, 2, and 3 weeks. Leaves with the residues will then be exposed to flies in laboratory assays.

In the field, studies to determine the attractiveness of various food baits for flies. These baits will eventually be mixed with small amounts of insecticides that will be ingested and will kill the flies. Hypotheses to be tested are that flies are most attracted to baits that produce high ammonia release and that high bait volumes are needed for attraction, and that flies find baits (as with natural food) mostly through indiscriminate grazing. During the last 4 seasons in central Washington, enough trees with high fly infestations have been located to make this project feasible. The general design will consist of up to five baits plus a control in the field (not in cages). Additional treatments will include the baits plus sugar and ammonium carbonate. The two main independent variables are season and bait type. Each of three trees will be considered a replicate. Studies will also be conducted to determine the effect of bait sprays on fly responses and on larval infestations in cherries. The percentages of flies attracted to the baits will be calculated, with each of three trees considered a replicate.

Baits at 2,500 μ l, 5,000 μ l, or higher volumes per 5 -10 cherry leaves on large trees, with each treatment about 1 m apart. An ammonium hydroxide lure will be used as the known attractant

and an unbaited branch will be used as the control. Numbers of flies that arrive within 30 and 15 cm of the baits and that feed on them will be recorded every 2 min over 30-min periods. Based on the results, specific baits will be chosen for further tests on fly movements and behaviors around baits on leaves. The movement of individual flies within a 1 square m area will be recorded over a 30-min period, similar to the methods in objective 1 above, except that baits will be present. The most effective baits will be applied as sprays on isolated cherry trees in homeowners' yards 5 days after first fly emergence in May. In central Washington, there are few abandoned cherry orchards with fly populations, so studies need to be conducted with these isolated trees. The numbers of flies seen feeding, resting, and mating on treated and control trees during 6-min searches will be recorded twice a week throughout the season. Fruit will be collected at the end of the season and the infestation levels determined through rearing of larvae.

2005 Results and Discussion:

(1) *Identify foods of western cherry fruit flies in nature.* Nearly all feeding occurred on leaves, with grazing behavior most common (Table 1). Most nutrients appeared to be obtained from leaf surfaces during early season. Honeydew and leaf nectaries appear not to be main sources of sugars for flies. The extensive grazing behaviors on leaf surfaces are consistent with the fact that flies exposed to leaves only survived on average 1 day more than controls, suggesting low but some nutrient acquisition. The feeding behaviors in the field, at least during early season, most likely reflect the low concentrations of food, forcing flies to move over large areas in trees and increasing the likelihood they will encounter bait spray droplets. The main protein or nitrogen source is probably bacteria on leaf surfaces, although bird droppings are also a source. Field observations indicated flies fed on cherry juice and bird droppings, but not frequently.

(2) *Determine when the flies feed, both daily and seasonally, and how much sugar and protein flies feed on in nature.* Observations indicate flies feed mostly in the morning, although evening activity is difficult to document because few flies are seen during later time of the day. Feeding and responses to baits were low during the middle of the day, especially on warm days. During these times, males were mostly on fruit, not feeding. Females, if on leaves, appeared to rest the majority of the time. Chemical analyses are being conducted to quantify nutrient uptake by flies in nature. Flies frozen at -80 C are awaiting analyses.

(3) *Determine the most attractive protein and sugar baits in the laboratory and field; baits that stimulate highest feeding.* In the laboratory, only flies deprived of protein responded to three protein baits: GF-120, Nulure, and Mazoferm (Table 2, only GF-120 shown, responses to others similar). All were equally attractive. Females and males responded similarly. Results suggest that unless flies are deprived, they will not respond to baits that are close to them. There was some indication that protein-deprived flies sometimes stayed nearer GF-120 droplets longer than protein-fed flies even when though they did not feed. Young and old flies behaved similarly, even though it was expected that older flies would be more responsive due to longer deprivation periods.

Consistent with laboratory results, in the field, flies were equally attracted to water, GF-120, Nulure, and Mazoferm, suggesting the baits have little attraction even at close range. This was true when baits were sprayed on leaves or placed on leaves within 3 cm of the flies (Tables 3 and 4). Careful observations suggested flies "stumbled" onto baits while grazing on the leaf surfaces. This is supported by the fact that water and baits were visited equally by flies. Another interesting observation was that flies seemed visually stimulated by the drops. Upon placement of a water or bait drop (which is nearly black in the case of Nulure), some flies would rapidly walk to the drop as if to investigate. This may be related to the flies' aggressive responses to other flies as competitors for food sources on leaves, or as competitors for territories. In other cases, there were no responses for 5 min durations even when the bait was only 1 cm away. These flies often would be on the undersides of leaves, resting. Flies on the surfaces of leaves appeared more likely to engage in feeding (grazing behaviors). Data seemed to support the contention that flies generally feed on the leaf tops and rest on the undersides.

(4) *Effects of bait sprays on fly control.* In a spray test in Yakima, weekly Mazoferm and NuLure sprays with spinosad and GF-120 sprays did not reduce numbers of adult flies. Numbers on most trees were high. However, the sprays did reduce larval infestations of cherries compared with controls, although they did not eliminate them (Table 5). Explanations for the lack of complete control include immigrating flies that may have infested experimental trees, lack of attractiveness of baits, and sufficient food quantities in trees that may have resulted in populations of flies that were not hungry and thus non-responsive, as suggested in the laboratory and field behavioral observations. Also, because of low fruit numbers in most trees, flies may have congregated on the few fruit, resulting in relatively high infestations across treatments. The method of infestation evaluation, rearing larvae in fruit, may lead to different conclusions than the use of the brown-sugar flotation method, which misses eggs. By holding fruit for 30 days, eggs that were in harvested fruit developed into larvae. Despite this, the results suggest that different spray baits are equally effective because none of them is especially attractive and that flies are essentially encountering them through chance encounters. The one consistent factor that results in effective kill is spinosad, which is highly toxic to the flies.

Significance to the Industry and Potential Economic Benefits: It is important to identify the natural foods of flies because they may compete with bait sprays intended to control the flies; also, it may lead to identification of attractants that can be incorporated into baits. The early results with Mazoferm, GF-120, and NuLure suggest flies find baits through normal (wide-ranging) foraging behavior rather than by a strong directed orientation or hopping flights towards odors, which seems to be how flies respond to ammonia lures. They also suggest several food-based baits are as effective as the GF-120 bait, as long as spray coverage is sufficiently high to allow flies to find them through normal foraging. If inexpensive alternatives to existing baits can be developed, they may reduce costs to growers, who need to spray baits weekly during the cherry season. Use of effective and long-lasting baits may help reduce spray frequencies and will help reduce costs associated with finding larvae in fruit.

Table 1. Observations of feeding activity on undefined and defined matter (\pm SE) of *Rhagoletis indifferens* on leaves and fruit in Zillah, WA, 2005

		Feeding on Leaves					
		% On Undefined Matter			% On Defined Matter ^a		
5/19-6/12	N	Grazing	% Time ^b	Nectary	Cherry Juice	Honeydew	Bird Feces
Females	77	37.7	20.4 \pm 3.8	1.3	3.9	0	1.3
Males	74	21.6	10.0 \pm 2.5	1.4	4.1	0	0
		Feeding on Fruit					
		% On Undefined Matter			% On Defined Matter ^a		
5/19-6/12	N	Grazing ^a	% Time ^b	Cherry Juice	Honeydew	Bird Feces	
Females	28	14.3	20.1 \pm 10.7	3.6	0	0	
Males	99	1.0	7.6	0	0	0	

Flies observed for maximum of 10 min; only observations > 0.5 min included.

^aPercent of observed flies feeding.

^bOf those that fed.

Table 2. Effects of feeding history on numbers and durations of feeding and non-feeding events (\pm SE) of grouped cherry fruit fly on or near sugar and GF-120 droplets on artificial leaves over 60 min.

Feeding Events: 3-5 day old Flies		
Feeding	No. Feeds/Fly	Total Feed Duration/Fly (min)

History	Bait	Female	Male	Female	Male
S	Sugar	0.17 ± 0.07a	0.10 ± 0.05a	0.46 ± 0.20b	0.17 ± 0.10a
S	GF-120	0.53 ± 0.11b	0.30 ± 0.10b	0.65 ± 0.13b	0.54 ± 0.17b
Y + S	Sugar	0 ± 0a	0 ± 0a	0 ± 0a	0 ± 0a
Y + S	GF-120	0 ± 0a	0 ± 0a	0 ± 0a	0 ± 0a

Feeding Events: 14-16 day old Flies

S	Sugar	0.17 ± 0.09	0.07 ± 0.04	0.31 ± 0.17	0.16 ± 0.12
S	GF-120	0.27 ± 0.11	0.17 ± 0.07	0.64 ± 0.30	0.50 ± 0.29
Y + S	Sugar	0 ± 0	0 ± 0	0 ± 0	0 ± 0
Y + S	GF-120	0 ± 0	0 ± 0	0 ± 0	0 ± 0

Non-Feeding Events: 3-5 day old Flies

Feeding	No. Visits on Leaf/Fly		Total Duration on Leaf/Fly (min)		
History	Bait	Female	Male	Female	Male
S	Sugar	0.63 ± 0.24a	0.83 ± 0.19a	3.57 ± 1.79a	3.69 ± 1.27a
S	GF-120	1.03 ± 0.24a	0.67 ± 0.20a	6.36 ± 1.64ab	2.70 ± 0.80a
Y+ S	Sugar	0.50 ± 0.17a	0.43 ± 0.13a	3.19 ± 1.42a	2.17 ± 0.64a
Y+ S	GF-120	1.07 ± 0.17a	0.77 ± 0.07a	8.87 ± 1.36b	3.79 ± 0.72a

Non-Feeding Events: 14-16 day old Flies

S	Sugar	1.17 ± 0.24	0.83 ± 0.19	6.50 ± 1.75	5.83 ± 1.54
S	GF-120	1.13 ± 0.27	1.07 ± 0.22	11.15 ± 2.64	10.75 ± 3.16
Y + S	Sugar	0.47 ± 0.13	0.43 ± 0.13	3.91 ± 1.12	2.85 ± 1.56
Y + S	GF-120	0.53 ± 0.09	0.40 ± 0.11	4.54 ± 1.49	3.75 ± 1.63

Feeding history: S: 5% sugar solution; Y+S: yeast + sugar: mixed dry 20% yeast extract and 80% sugar.

10 replicates per treatment and sugar control; each replicate consisted of three females and three males.

Means followed by the same letter within columns and age group are not significantly different (ANOVA, Fisher's LSD Test, $P > 0.05$).

Table 3. Effects of bait sprays applied on leaves of cherry trees on presence of cherry fruit flies near spray droplets and feeding

100 ul – No. Flies near (<15 cm) of feeding + SE							No. Flies
Date	N	Water	GF-120	Nulure	Mazoferm	Seen + SE	
24 May	5	0.0 ± 0.0	0.30 ± 0.17	0.03 ± 0.02	0.0 ± 0.0	6.4 ± 2.1	
27 May	5	0.0 ± 0.0	0.19 ± 0.13	0.0 ± 0.0	0.18 ± 0.11	13.6 ± 3.2	
31 May	3	0.0 ± 0.0	0.0 ± 0.0	0.23 ± 0.20	0.0 ± 0.0	9.0 ± 3.5	
10 ml – No. Flies near (<15 cm) solutions ± SE							No. Flies
Date	N	Water	GF-120	Nulure	Mazoferm	Seen + SE	
7 June	3	0.63 ± 0.36	0.13 ± 0.0	0.08 ± 0.08	0.08 ± 0.08	22.0 ± 1.2	
10 June	3	0.17 ± 0.08	0.67 ± 0.23	0.0 ± 0.0	0.10 ± 0.08	22.3 ± 3.8	
14 June	3	0.13 ± 0.13	0.04 ± 0.02	0.04 ± 0.02	0.21 ± 0.11	14.3 ± 0.9	

Table 4. Effects of placing bait droplets near cherry fruit flies on leaves on fly feeding responses ± SE on cherry trees, May-June 2005

Bait	Sex	N	% Response	No. Times Fed	Total Time	Mean Time/Feed
Water	F	25	48.0	0.72 ± 0.19	0.07 ± 0.02	0.06 ± 0.02
			0.05 ± 0.02			
GF-120	F	27	48.1	1.15 ± 0.31	0.62 ± 0.19	0.34 ± 0.10
			0.50 ± 0.19	0.22 ± 0.08		
Nulure	F	20	35.0	1.20 ± 0.44	0.19 ± 0.10	0.06 ± 0.03
			0.09 ± 0.06	0.06 ± 0.04		
Mazoferm	F	23	47.8	1.04 ± 0.47	0.23 ± 0.12	0.12 ± 0.03
	M	20	25.0	0.30 ± 0.13	0.03 ± 0.01	0.02 ± 0.01

Feeding responses pooled from 11 d of observations.

Table 5. Effects of bait sprays on adult numbers of cherry fruit flies and larval numbers \pm SE from cherries in Yakima, 2005

Treatment	N	No. Adults/Trap ^a	No. Larvae/fruit	% Reduction	No. Fruit Picked
Control	8	$257.2 \pm 133.1a$	$0.907 \pm 0.137a$	-----	214.6 ± 21.9
GF-120	4	$16.0 \pm 6.5a$	$0.501 \pm 0.218ab$	45	73.8 ± 29.0
Nulure	5	$49.0 \pm 15.2a$	$0.412 \pm 0.103b$	55	233.4 ± 45.4
Mazoferm	5	$53.0 \pm 29.1a$	$0.125 \pm 0.062b$	86	273.2 ± 80.0

^aCumulative numbers over 34 d.

Means within columns followed by the same letter are not significantly different (ANOVA, LSD test, $P > 0.05$).

Project title: Fly feeding ecology and food-based lures and baits

PI: Wee Yee

Project duration: 2004-2006

Current year: 2004

Project total (3 years): \$62,174

Current year request: \$20,087

Item	Year 1 (2004)	Year 2 (2005)	Year 3 (2006)
Salaries	18,000 ^a	18,000 ^a	18,000 ^a
Benefits	2,000 ^a	2,000 ^a	2,000 ^a
Wages	0	0	0
Benefits	0	0	0
Equipment	0	0	0
Supplies	2,000	87	87
Travel	0	0	0
Miscellaneous	0	0	0
Total	22,000	20,087	20,087

^aOne GS-3 (\$6,000), one GS-5, part time (\$14,000)

Project title: Cherry fruit fly control options

PI: Timothy J. Smith

Organization: WSU Extension, North Central Washington

Address, phone, e-mail: 300 Palouse, Wenatchee, WA 98801
(509) 667-6540; smithtj@wsu.edu

Research Assistant: Esteban Gutierrez, East Wenatchee.

Introduction and Justification

Cherry fruit fly was identified as the top priority in the TFRC Cherry Research Committee yearly priority setting sessions. The objective of this project has been to develop safe and highly effective new control material options, as the carbamate and organophosphate class insecticides available at the inception of this work were (and continue to be) under regulatory pressure, and no alternative methods and chemistries were likely to be registered soon.

Significant Results Summary:

Objective 1: Identify new conventional and organic cherry fruit fly control products and methods.

- ! Twelve products have been included in the trials.
- ! Several other promising products remain to be tested.

Objective 2: Assess efficacy of new insecticides and control methods for cherry fruit fly.

- ! Most of the candidate products were quite effective, especially when applied at “moderate” or “full” proposed label rates and at 7 or 10 day spray intervals. Rate and interval data will be used for future label directions.
- ! This project first recognized and demonstrated the efficacy of GF-120 Bait as a Cherry Fruit Fly control. Early adoption of this control method is **saving the PNW Cherry growers about \$1 million each year** in labor, application and material costs.
- ! Three products were identified as alternatives to dimethoate as post-harvest “clean-up” sprays.
- ! Organic growers are now fully able to control this pest with the GF-120 bait and/or Entrust. One commonly recommended organic product, azadirachtin (neem) was proven ineffective.

Objective 3: Work with industry toward the registration of effective new CFF control products.

- ! This project has added eight products shown to be effective cherry fruit fly control materials.
- ! Impending registration of the new products tested in this project will lead to availability of new chemistries that are highly effective and have fewer negative effects on the foliage, environmental concerns, and labor issues than many current material choices.

Results and Discussion:

Objective 1, Identification of Candidate Products: Products included in this project during the 2003-05 trials included Assail, Calypso, Azadirachtin, Provado, Success, Entrust, GF-120NF Bait,, Pyganic, an unmentionable insect growth regulator, and three very promising numbered products. Eight of the products tested had not been tested in the field for effect on cherry fruit fly when first included in this project. Some interesting products remain untested, usually due to lack of current interest on the part of the registrant. New options are being included each year.

Objective 2, Efficacy Trials: Most tested products controlled CFF very well at moderate or full rates applied at 7 to 10 day intervals. Lower rates often showed some slight failure rate at ten day intervals, and most products became less effective when applied at 14 day intervals, even with full standard rates. This interval and rate information will be used during the development of use directions for these products, and during educational programs.

Spinosad was proven as an effective CFF control active ingredient during earlier work first by the PI and then by others. Entrust, an organically acceptable formula of sprayable spinosad was shown effective during this project. The GF-120 NF bait was first (2002) shown to be an option as a cherry fruit fly control material and method through this project. Application of insecticidal bait is a new practice to Pacific Northwest tree fruit producers, so research and educational efforts were closely linked. Numerous presentations and publications gave the cherry growers opportunity to become aware of this material and its' potential. *Use in the first two years of registration has saved Washington cherry growers about \$1,160,000* in labor, machinery and material costs, and economic benefits will continue at about \$1 million per season at current use levels. Adoption of this new technology has essentially eliminated a serious and increasing problem with cherry fruit fly in organic orchards. It has also enhanced the conventional growers' ability to treat their orchards in a timely manner, despite wind. Use of the product increased by 360 percent in 2005 vs. 2004, and acceptance of this technology is expected to increase at a slower rate as the more skeptical growers gain confidence in its efficacy. Applicator exposure to products with potential to inhibit cholinesterase was reduced by about 6,600 hours during May, June and July of 2005

Three control materials were tested for effect on cherry fruit fly larvae inside the fruit, for possible alternatives for post-harvest dimethoate. Sections of a single most-highly infested tree were sprayed at the time that the third instar larvae were starting to cut emergence holes in the fruit skin. The fruit was harvested 24 hours after treatment, and then suspended at room temperature over sand. The number of larvae that emerged were counted. All three alternatives appeared quite effective, though further research is required. See table 4.

Provado, Assail and Calypso controlled black cherry aphid (*Myzus cerasi*) when used at rates and application timings intended for cherry fruit fly control.

Azadirachtin (neem) was proven not effective as a cff control. It has been recommended to organic growers for this purpose for many years. Data could be interpreted that the product had some suppressive effect, as the degree of fruit infestation on treated trees was lower than would be expected on untreated trees supporting similar high numbers of adults.

A previously untested insect growth regulator was very suppressive of larval infestation. Adults were apparently unaffected by the product, and a trap captured over 100 adults during the four weeks of treatment. This level of adult infestation would normally lead to 60 -100% fruit infestation.

Table 1. Details of 2005 Trials (Not reported in previous project updates):

Treatment	Trees / Sites	Days Interval Spray	Flies / Trap 2005	Fruit Sample Number	Larvae Found in Fruit
Calypso SC 480, 3 oz/A 1st treatment, Carbaryl 4 pints/A second, Calypso 3 oz/A third treatment, Success 4 oz 4th. Treatment, and GF-120 BAIT weekly during and after harvest	3 / 3	3 @ 10 3 @ 7	3 11 55	1000 1000 1000	0 0 0
Calypso SC 480, 4 oz/A 1st treatment, Carbaryl 4 pints/A second, Calypso 4 oz/A third treatment, Success 4 oz 4th. Treatment, and GF-120 BAIT weekly during and after harvest	3 / 3	3 @ 10 3 @ 7	3 55 27	1000 1000 1000	0 0 0
Calypso SC 480, 6 oz/A 1st treatment, Carbaryl 4 pints/A second, Calypso 6 oz/A third treatment, Success 4 oz/A 4th treatment + GF-120 BAIT weekly during and after harvest	3 / 3	3 @ 10 3 @ 7	3 55 27	1000 1000 1000	0 0 5*
Provado 1.6F, 6 oz/A 1st. Treatment, Carbaryl 4 pints/A second, Provado 6 oz/A third treatment, Success 4 oz/A 4th treatment + GF-120 BAIT weekly during and after harvest.	18 / 4	3 @ 10 3 @ 7	55 27 11 35	1000 1000 1000 1000	0 0 0 0
An Insect Growth Regulator	1 / 1	10	101	1000	11**
Assail 30SG, 2.5 oz / A 10 day	3 / 3	10	21 5 55	1000 1000 1000	0 0 0
Assail 30SG, 2.5 oz / A 14 day	2 / 2	14	34 18	1000 1000	5 1
Assail 30SG, 4.0 oz / A 14-day	3 / 3	14	31 31 18	1000 1000 1000	3 4 3
Numbered Product X 4.5 oz. / A	4 / 4	7	3 5 55 34	1000 1000 1000 1000	0 0 0 0
Numbered Product X 6.0 oz. / A	4 / 4	7	3 5 55 34	1000 1000 1000 1000	0 0 0 0

Table 1, Continued. Treatment	Trees / Sites	Days Interval Spray	Flies / Trap 2005	Fruit Sample Number	Larvae Found in Fruit
Numbered Product Y 1 oz / A + 0.5% Oil	3 / 3	10	89 5 55	1000 1000 1000	0 0 0
Numbered Product Y 2 oz / A + 0.5% Oil	3 / 3	10	55 74 34	1000 1000 1000	0 0 0
Numbered Product Y 3 oz / A + 0.5% Oil	3 / 3	10	117 55 10	1000 1000 1000	5** 1 0
Numbered Product Y 4 oz / A + 0.5% Oil	3 / 3	10	3 55 74	1000 1000 1000	0 0 0
Numbered Product Y 2 oz / A No Oil	3 / 3	10	55 26 61	1000 1000 1000	2 4 4
Numbered Product Z 10 fl oz/ A	3 / 3	10	55 74 34	1000 1000 1000	0 0 0
Entrust 1.9 oz / A	5 / 5	7	3 5 55 26 34	1000 1000 1000 1000 1000	0 0 0 0 0
Untreated Check Trees	5 / 5	na	265 565 87 238 150	1000 1000 1000 1000 250	447 497 303 540 339

Notes:

* One interval of 12 days between sprays may have caused control difficulty.

**With this number of adults on the trap, would normally expect near 100% infestation.

Table 2. Summary of Previous Trials:

	Year	Trees / sites	Flies / Trap Prior Year*	Flies / Trap Treated Year	Total Fruit Inspected	Total Larvae Found
Untreated Checks	na	12	< 20	144	8065	2428
Provado	1999	8/1	150+	14	800	0
Provado	2003	4/1	21	1	800	0
Provado	2004	6/2	50+	40	1000	0
Calypso	2003	21/6	25	4	4600	0
Calypso	2004	29/11	150+	93	5050	9 **
Assail	2002	7/1	50+	4	900	0
Assail	2003	24/6	39	28	3600	0
Assail	2004	24/9	100+	59	5200	1 **
Product Y	2002	5/1	30	3	800	0
Stylet Oil	1999	4/1	100+	16	800	6

* "Failures" are due to research intentions. The rate was too low, or interval too long, or both.

Table 3. Organic CFF Control Product Summary:

	Year	Trees / sites	Flies / Trap Prior Year*	Flies / Trap Treated Year	Total Fruit Inspected	Total Larvae Found
Untreated Checks	na	12	< 20	144	8065	2428
Aza-Direct	2004	12/6	50+	55	2000	102
GF-120NF	2002	4/1	50+	4	500	0
GF-120NF	2003	22/6	59	11	2500	1 **
GF-120NF	2004	29/10	73	16	6400	1 **
GF-120NF	2005	32/13	57	3	12,000	0
Success	1997	1/1	50+	7	500	0
Success	1998	9/1	100+	14	3200	13 ***
Success	1999	25/2	150+	13	2500	0
Success	2002	2/1	50+	4	500	0
Entrust	2003	10/3	29	5	2400	0
Entrust	2005	5/5	110	25	5000	0

* Average trap catch year prior to first treatment, if data available.

** Note: The single larva was found in fruit sample taken from multiple tree, highly infested sites. (Example: 200 cherry fruit fly adults were captured on one trap, 15 trees). No larvae were found in fruit in second year or third year of treatment on these bait-treated sites.

***Control failures in this replicated plot were probably due to treatment contamination by mature females from heavily infested untreated “check” trees within 50 feet. Some treatments were low rates at 10 day intervals.

Post-harvest Treatments:

Provado, Assail and Calypso applied on a date that would have been “post-harvest,” led to excellent control of large 3rd instar CFF larvae in fruit that had an average of 136 percent larvae emergence from fruit in the non-treated check. All products tested appear to be very acceptable replacements for dimethoate, the only product currently recommended for controlling larvae in fruit remaining on harvested trees. This may give products with this chemistry a great advantage as a pre-harvest product, as application may control newly hatching eggs or larvae that may have slipped through earlier control programs. At this time, dimethoate is not a popular choice, as it sometimes causes leaf marking and drop. While this post-infestation effect seems relatively certain, it is so significant that more extensive trials to document this effect on larvae inside the fruit should be carried out.

Table 4. Post harvest “Clean-up” Spray Options:

Product	Rate	Fruit Sample	Larvae Emerged
Dimethoate 267	64 oz./400 gal./A	250	1
Provado 1.6F	8 oz./400 gal./A	250	0
Calypso SC 480	8 oz./400 gal./A	250	20
Assail 30 SG	8 oz./400 gal./A	250	8
Untreated	0	250	339

Objective 3, New Registrations: There are four relatively new, effective products available for management of cherry fruit fly. (Success, Provado, Entrust and GF-120 NF Bait). It is likely that an additional two will be labeled in 2006 or 2007 (Assail and Calypso), greatly aided by this project. Two numbered products included in this research program (unique chemistries) are projected to be registered by 2008 - 2010; another two promising new materials (a numbered product and an insect growth regulator) will require continued research prior to adoption.

Organic growers now have organically acceptable, effective material choices for management of cherry fruit fly, a pest that was nearly out of control in 2003. Other organic options are being considered.

Methods and materials:

Several small cff infested sweet cherry orchards were used as sites for replicated trials. In total, 275 infested cherry trees on 83 sites have been included in this project over the past three seasons. Each of these sites consisted of sweet cherry trees that were documented or reported as infested with fruit fly, and volunteered by the owners as test subjects, with a signed agreement that the fruit could not be consumed if treated with unregistered products. In return, the cherry tree owner was assured a “clean” tree the next year, and often had trees treated with registered products available for immediate consumption.

Isolated abandoned cherry trees were left as unsprayed “checks” to document the relationship between prior season trap count and current season trap catch and fruit infestation. One of the unsprayed trees developed 136 percent infestation in 2005, a new local record. (Three hundred thirty-nine cff larvae were taken from 250 fruit.)

All test sites were monitored with the standard Trece baited AM yellow sticky 9 x 11 inch traps to document adult presence on the trial trees and potential infestation of fruit. Spraying usually greatly reduced trap catch, but did not eliminate adults until the second year of effective treatment. This was expected, as some CFF pupae remain in the soil for two seasons.

The trial applications began in mid- to late May, when the first adult was trapped in the area. Sprays were applied at 7, 10 or 14 day intervals from that date until the normal harvest maturity, which occurred during the last ten days of June. Usually, a total of six 7-day, four 10-day, or three 14-day treatments were applied during this time. At harvest-time, a 250 – 1000 cherry sample was collected from each replicate and placed into cold storage. At the suggestion of professional entomologists, all replicates were sampled at the 1000 fruit level in 2005, to better insure an accurate assessment of infestation level. The sites were usually treated with GF-120 Bait for 2 -3 weeks after harvest.

The test fruit was checked for larva with the Washington State Department of Agriculture standard brown sugar solution method for the detection of CFF larvae in large batches of fruit. In this extraction technique, cherries are crushed carefully, then place in a solution of seven pounds of brown sugar dissolved in five gallons of water. The specific gravity of CFF larvae is less than that of the solution, which causes them to float to the surface of the cherry/syrup mixture. The light colored larvae are relatively easy to observe floating on the dark surface, even when they are in their first instar. This method assured that large numbers of fruit could be sampled, assuring detection of even low numbers of small larvae. Larvae were easily detected in fruit taken from untreated check trees. Some samples of fruit were also suspended on a grate over sand to check for naturally emerging larvae. This larva detection method did not appear significantly more accurate than a carefully run brown sugar solution larval extraction technique.

Application: All materials except the bait were applied with a backpack air-blast/mist sprayer in about 100 gallons water per acre. Post harvest treatments were applied at about 400 gallons per acre (drip). The GF-120NF bait was applied in orchards with a 12 volt, electric pump, auxiliary sprayer strapped to a “four-wheel” All Terrain Vehicle. Two adjustable-angle D2 disc nozzles (no cores) were used to direct streams of the bait/water mix across the upper 1/3 of the tree. Calibration trials proved that 20 fluid ounces of the bait could be mixed 1:4 with water, and then applied to one side of each tree (on alternate row middles) at 6.5 to 7 mph through D2 nozzles. Application took about 2.5 to 3 minutes per acre. The side of the trees treated was alternated weekly, but the row ends and outside rows were treated every week. The “backyard” tree bait sites were treated with a 1:3 bait to water mix applied with hand-held “window washer” squirt bottles adjusted to apply a solid stream of mixture. Rate per acre was adjusted by varying the amount of mixture that was applied relative to the size of each test tree. Bait was re-applied after significant rainfalls. Heavy dew would likely dissolve the bait speckles, possibly leading to control failures, but heavy dew is rare in North Central Washington, so was not monitored.

Discussion:

All products, rates and timings were tested under pest population conditions far in excess of what would be expected in commercial orchards. As adults emerge daily during the season, spraying usually greatly reduces, but does not prevent adult trap catch on infested trees. However, effective control products protect the fruit from larval infestation by controlling adults prior to their maturation

and egg deposition. Most of the treatments greatly reduced or eliminated infestation.

Full rates were used at seven day intervals in the first year of efficacy trials. If the product appeared promising, the subsequent seasons work included rate and interval assessment, in search of the “failure point.” Under the severe test conditions, some rates and intervals failed to completely control cherry fruit fly. The fact that a few larvae were found in these sub-optimal treatments should be considered results of a successful research effort, rather than an indication that the product will not be effective when used as directed.

Low numbers of larvae were more often found when rates were dropped to 0.33 to 0.5 of proposed full label rates and spray intervals were increased to 10 or 14 days. Lower or moderate rates seem to work well with 7 day spray intervals. Moderate rates appeared to be effective at 7 or 10 day intervals. However, even highest rates of an otherwise effective product failed to fully prevent larval infestation when spray intervals were increased to 14 days.

Under the severe test conditions, GF-120 bait treatment “failed” twice out of the 35 site treatment years. (Treatment year = one site treated for one season.) The fact that two larvae were found out of 18,750 cherries crushed from these 35 treatment sites might be considered incidental results of a successful research effort under highly unusual infestation pressures, rather than an indication that the product will not be effective when used as directed under normal pest pressure situations. In both cases that larvae were found after bait treatment, the treatment sites can only be described as abnormal. The treatment trees were very tall, interwoven, and the fruit was infested at 50 - 100% the year prior to first treatment. In one case, the trees were removed after the first season, but the other, site 04-9, provides a great case study. On this “failure site,” the three infested trees are 45-55 feet tall, and 100% of the fruit was infested and unharvested in 2003. Evaluation of 2003 trap catch by the local pest board was suspended after over 50 adults were captured the first week of trap deployment. During 2004, the first season of bait treatment, 205 adults were captured on a single trap, despite a 40 oz./A rate of GF-120, applied weekly. However, after one season of treatment, the larval infestation fell from the reported 100 percent to 0.25 percent (1 larva in 250 fruit). In 2005, the second season of treatment, the trees were treated at the 20 oz./A rate weekly. Adult capture fell from 205 to 2 for the season, and no larvae were found in a 1000 fruit sample.

In 2005 no larvae were found in 12,000 fruit sampled from twelve bait-treated sites. The only control product applied to these previously highly infested sites was GF-120 bait. Similar untreated check trees developed larva counts ranging from 30 to 136 percent.

Other effects:

Despite as many as five weekly applications at higher than necessary rates, no treatment in this project has resulted in leaf marking, yellowing or shedding, fruit marking, or excessive mite flare-ups leading to significant leaf damage. Some moderate leaf symptoms induced by mite feeding were observable by late summer on some of the trees treated with up to five weekly applications of Provado, Assail, and Calypso.

Many of the candidate products have not yet been tested on all common sweet cherry varieties, so potential for leaf drop sensitivity in some varieties, or marking of light colored cherries is unknown.

Continuing Project Report**Year 1/3**

Project Title: Managing soil ecology to improve soil and tree function
PI: Anita Azarenko

No report submitted