



2014 Research Proposals

and

2013 Research Reports

WRRC Board of Directors - with term expiration date, December 1 . . .

<u>Year</u>	<u>Seat</u>		<u>Year</u>	<u>Seat</u>	
10	1	Vacant - District #2	13	8	Kyle Haugen
13	2	Adam Enfield, President Lynden	15	9	Erin Thoeny Woodland
14	3	Kristie Clark, Treasurer Lynden	14	10	Vacant - District #4
14	4	Ralph Minaker Everson	WSDA	11	Ted Maxwell Olympia
15	5	George Hoffman Ridgefield	WRRC Office		
15	6	Glenn Sakuma Burlington	Henry Bierlink, Executive Director <i>henry@red-raspberry.org</i>		
09	7	Vacant - District #2	Michelle Hungerford, Office Manager 1796 Front Street, Lynden, WA 98264 (360) 354-8767		

2014 Production Research Priorities

#1 priorities

- Develop cultivars that are summer bearing, high yielding, winter hardy, machine-harvestable, disease resistant, virus resistant and have superior processed fruit quality
- Understanding soil ecology and soil borne pathogens and their effects on plant health and crop yields.
- Fruit rot including pre harvest, post harvest, and/or shelf life.
- Soil fumigation techniques and alternatives to control soil pathogens, nematodes, and weeds.
- Management options for control of the Spotted Wing Drosophila

#2 priorities

- Root weevils
- Labor saving cultural practices including mechanical pruning and tying techniques.
- Nutrient/Irrigation management
- Viruses/crumbly fruit, pollination
- Mite management

#3 priorities

- Vertebrate pest management
- Product and Production Certification Systems - food safety & security, standards, traceability
- Maximum Residue Limits (MRL) – residue decline curves
- Weed management
- Foliar & Cane diseases – i.e. spur blight, yellow rust, cane blight, etc.

Of Note:

- Pest Management as it affects Pollinators
- Season extension: improve viability of fresh marketing

2014 WRRRC Production Research Budget Worksheet

PAGE	PROJECT TITLE	RESEARCHER (S)	2013 FUNDED	REQUEST of WRRC	Other Sources	WSCPR Applied	Total Project	WRRC Approved	WRRC %
5	Genomic Infrastructure	Finn	\$1,000	\$1,000	\$1,590,000		\$1,591,000		0%
10	Cooperative Raspberry Cultivar Development	Finn	\$5,000	\$6,500	\$500		\$7,000		93%
23	Red Raspberry Cultivar Development	Dossett	\$10,000	\$12,000			\$12,000		100%
33	Raspberry Breeding, Genetics, Clone Evaluation, Machine Harvest Trials	Moore	\$67,000	\$70,000	\$19,835		\$89,835		78%
41	Tracking the movement of RBDV in Red Raspberry from pollination to systemic infection using Real-time RT PCR	Moore, Lanning		\$8,750			\$8,750		100%
44	Evaluating the Feasibility of Coordinated Regional on-farm Trials of Advanced Raspberry Selections	Peerbolt	\$8,500	\$11,200			\$11,200		100%
49	Use of a mycoinsecticide targeting novel SWD preimaginal life stages and potential synergism with spinosad	Tanigoshi	\$5,000	\$10,865			\$10,865		100%
57	Quantifying Root Weevil Impacts and Post Treatment Root Recovery	Jacoby, Tanigoshi		\$13,918		\$12,268	\$26,186		53%
63	Determining the ability of Spotted-Wing Drosophila to overwinter in weedy blackberry	Miller, Tanigoshi		\$3,774		\$4,000	\$7,774		49%
66	Testing herbicides for weed control in red raspberries	Miller	\$4,600	\$4,807		\$4,250	\$9,057		53%
68	Integration of Factors to Improve Soil Health	Benedict	\$7,500	\$5,787			\$5,787		100%
74	Mobile App for Pest Scouting and Record Keeping	Benedict		\$11,230	\$11,230		\$22,460		50%
80	Incidence and Detection of <i>Verticillium dahliae</i> in Red Raspberry Production Fields	Weiland, Benedict	\$10,000	\$13,582			\$13,582		100%
88	Characterizing the soil ecology of red raspberry produced under different production regimes	Zazada, Dunlap		\$17,925			\$17,925		100%
96	Fine-tuning Vydate applications in red raspberry for <i>Pratylenchus penetrans</i> control	Zasada, Walters	\$9,000	\$5,883			\$5,883		100%
103	Effectors, strategy to control plant-parasitic nematodes	Elling	\$7,000	\$13,872	\$290,000		\$303,872		5%
110	Biology and control of <i>Botrytis</i> fruit rot	Peever		\$21,996	\$44,992	\$21,996	\$88,984		25%
117	Evaluating soil fumigation alternatives in Washington raspberry fields	Walters		\$8,095			\$8,095		100%
121	Evaluating fungicide efficacy for <i>Botrytis</i> control	Walters		\$9,624			\$9,624		100%
125	Management of Fungicide Resistant <i>Botrytis</i> in Red Raspberry	Schreiber	\$16,000	\$10,000		\$15,000	\$25,000		40%

129	Red Raspberry trellising demonstration plot for development of automation technologies	Karkee, Tarara	\$10,000	\$72,007	\$65,725		\$137,732		52%
136	Exploring opportunities for developing value-added dried products from raspberries.	Girish Ganjyal		\$18,365			\$18,365		100%
145	Developing edible coatings for raspberries use in muffins	Girish Ganjyal		\$12,661			\$12,661		100%
149	Processed fruit quality of newly released raspberry selections	Ross		\$11,010			\$11,010		100%
159	Innovative Packaging Technologies To Enhance The Safety And The Quality Of Fresh Raspberry	Gianfagna, Yam (Rutgers)		\$6,777	\$9,943		\$16,720		41%
163	Ultraviolet light (UV-C) treatment for improving berry safety	Sablani		\$13,695			\$13,695		100%
175	Connecting Raspberry Breeding to Crop Markets through Use of Molecular, Economic and Consumer Science	Daniels	\$1,000						
176	Trap and bait testing for SWD in NW & SW Washington Raspberries	Burrows Peerbolt	\$3,700						
177	Evaluation of resistance to <i>Verticillium dahliae</i> and <i>Phytophthora rubi</i> in red raspberry	Moore Weiland	\$6,600						
	Physiologist reserve	DeVetter		\$20,000			\$20,000		100%
	Future Projects						\$0		
	Total Production Research		\$171,900	\$415,323	\$2,032,225	\$57,514	\$2,505,062	\$0	
	Research Related	WRRRC expenses	\$5,000	\$5,000			\$5,000	\$5,000	
	Small Fruit Center fee		\$2,500	\$2,500			\$2,500	\$2,500	
	TOTAL		\$179,400	\$422,823	\$2,032,225	\$57,514	\$2,512,562	\$7,500	

2014 Proposed Research Budget

\$262,007

2013 Research reports follow the 2014 proposals for each researcher.

Application Cover Sheet

Application Date: November 11, 2010

Name of Applicant Organization/Company:

USDA-ARS, HCRL; 3420 NW Orchard Ave. Corvallis, OR 97330

USDA-ARS, NCGR; 33447 Peoria Rd., Corvallis, OR 97333

Principal Investigators:

Co-Project Directors

Chad Finn, USDA-ARS Research Geneticist

Nahla Bassil, USDA-ARS Research Geneticist

Jungmin Lee, USDA-ARS Research Chemist

Jill Bushakra, USDA-ARS Post Doc

Co PIs

Courtney Weber, Cornell Univ.

Gina Fernandez, NC State Univ.

Penny Perkins-Veazie, NC State Univ.

Joe Scheerens, Ohio State University

Emily Rhoades, Ohio State University

Robert Agunga, Ohio State University

Todd Mockler, Oregon State University

Other collaborators

Julie Graham, Scottish Crop Research Institute

Feli Fernandez-Fernandez, East Malling Research

Song Joong Yung, Chonbuk National University

Commercial growers

Project title:

Support of SCRI Proposal “Developing the Genomic Infrastructure for Breeding Improved Black Raspberries”

Year Initiated (current year) 2011 Current Year **2013 Terminating Year 2015**

Funding

Total amount requested: \$1,000/yr, \$5,000 for 5 years

Our SCRI grant application was successful and we are receiving \$1,590,717 to accomplish the goals set out in the proposal from 2011-2015. The grant was officially awarded October 1, 2011. The Washington Red Raspberry Commission had committed \$1,000/year to this project if we were successful. We have also sought supporting funding from the North American Raspberry and Blackberry Association and the Oregon Raspberry and Blackberry Commission.

Title of project: Support of SCRI Proposal “Developing the Genomic Infrastructure for Breeding Improved Black Raspberries”

Year Initiated __2011____ Current Year 2012____ Terminating Year _2015__

Brief description of project (<200 words) describing objectives and expected outcome.

Specialty Crop Research Initiative Grants are a major source of funding for berry research. We recently received a \$1.59 million dollar grant entitled "Developing the Genomic Infrastructure for Breeding Improved Black Raspberries".

This proposal seeks to advance and streamline efforts to identify a variety of traits of interest to growers and consumers in black raspberry germplasm, and then integrate them into breeding programs with the goal of developing new disease resistant cultivars that satisfy the demands of the marketplace while adding to the sustainability and profitability of the industry. A major focus of this project is to develop, and make available, genomic tools such as linkage maps, ESTs, SNP and SSR markers for use in raspberry breeding.

How does this tie into red raspberries in Washington? Black raspberries have historically been a source of valuable traits (e.g. disease and insect resistance, fruit firmness) in red raspberry. A great deal of what we learn will be applicable to red raspberry and the genomic information will be useful for the successful SCRI planning grant “Roadmap Development for U.S. Raspberry Producers: Forging Links Between New Tools for Breeding Programs and Crop Markets” and for the full project that we will submit next year.

Justification and Background: (Issue you plan to address, why, 400 words maximum)

The Specialty Crop Research Initiative Grants have become a major source of funding for small fruit research. Last year we submitted a proposal called Developing the Genomic Infrastructure for Breeding Improved Black Raspberries that was successful. Within these grants we are expected to have a 50% match. We have significant commercial and academic matching funding but feel strongly that is important to ask for other funds that while only a small portion of the \$1.74 million we have in matching shows an industries willingness to contribute. We asked for support last year and the WRRRC said they would provide a \$1000 match.

This proposal seeks to advance and streamline efforts to identify a variety of traits of interest to growers and consumers in black raspberry germplasm, and then integrate them into breeding programs with the goal of developing new disease resistant cultivars that satisfy the demands of the marketplace while adding to the sustainability and profitability of the industry. A major focus of this project is to develop, and make available, genomic tools such as linkage maps, ESTs, SNP and SSR markers for use in black and red raspberry breeding.

How does this tie into red raspberries in Washington? Black raspberries have historically been a source of valuable traits (e.g. disease and insect resistance, fruit firmness) in red raspberry breeding. We have characterized a great diversity of black raspberry germplasm and most importantly have identified 4 new sources of raspberry aphid resistance. If we can develop markers for traits such as these sources of aphid resistance, we can then fairly easily move them into red raspberry. We expect that a great deal of what we learn will be applicable to red raspberry and objective 5 clearly points to this. We also expect that the genomic information we

learn will be useful for the project “Roadmap Development for U.S. Raspberry Producers: Forging Links Between New Tools for Breeding Programs and Crop Markets” that was successfully submitted as a planning grant and the full project that they will submit one year from now.

Relationship to WRRRC Research priority(s):

Our objectives for raspberry breeding most closely align with a #1 Commission Priority as we are trying to develop cultivars “that are summer bearing high-yielding, winter hardy, machine harvestable, disease and virus resistant and have superior processed fruit quality. The traits we identify and the tools we develop will be useful in developing improved red raspberry cultivars.

Objectives:

The overall goal of this proposal is to develop and make available genomic tools for the improvement of black and red raspberry (*Rubus occidentalis*, and *R. idaeus*, respectively, subgenus *Idaeobatus*) and begin the application of these tools in using wild black raspberry germplasm for crop improvement. Specifically:

- 1) Transcriptome sequencing and high throughput genomic sequencing.
- 2) Developing molecular markers from genomic and EST sequences.
- 3) Studying genotype by environment interaction on specific traits of interest in crosses involving diverse wild black raspberry germplasm.
- 4) Using molecular markers for mapping specific traits of interest in crosses involving diverse wild black raspberry germplasm.
- 5) Evaluate transferability of SSR markers developed in black raspberry to red raspberry.
- 6) Better understanding of consumer preferences for market expansion.
- 7) Delivering research results and training in molecular breeding to the industry, breeders, and students through a multifaceted outreach program.

Procedures (<400 words):

I would be delighted to share the detailed procedures with the WRRRC if they feel it would be useful. I sent a copy with this proposal to the WRRRC office if someone is interested in reading the proposal.

Basically, we are growing out mapping populations from controlled crosses and they have been planted in multiple research and commercial settings. We will evaluate the plants for observable plant, fruit, and phenological traits and then tie this information to their genotype. In this process, we will develop markers for a wide variety of traits that will facilitate black and red raspberry breeding especially for traits that are not easily observable (e.g. aphid resistance) or so that you can stack more than once source of resistance into a genotype.

Anticipated Benefits and Information Transfer:

We will develop markers that will be useful in marker assisted red and black raspberry breeding. This will hopefully open up new opportunities and sources of variability for developing improved red and black raspberry cultivars.

Budget:

For each year 2011-2015

Salaries: Student labor (GS-2)	\$1,000
Total per year	\$1,000
Total for 2011-2015	\$5,000

Washington Red Raspberry Commission Progress Report

Title: Support of SCRI Proposal “Developing the Genomic Infrastructure for Breeding Improved Black Raspberries”

Personnel:

Co-Project Directors

Jill Bushakra, USDA-ARS, Post Doc; Chad Finn, USDA-ARS Research Geneticist; Nahla Bassil, USDA-ARS Research Geneticist; Jungmin Lee, USDA-ARS Research Chemist
Commercial growers in Oregon, Washington, North Carolina, and New York

Reporting Period: 2013

Accomplishments:

We completed propagation of the mapping populations and either shipped them to locations in the eastern US or planted them at our location and the commercial grower locations in the Pacific Northwest. All populations established well with almost no plant losses. We began the discussion on phenotyping protocols. The USDA-ARS NCGR group isolated DNA from parents and each individual in the mapping populations. The OSU CGRB generated transcriptome sequences from leaves, stems, canes, green berries, red berries, and ripe berries of ‘Jewel’. These RNA sequences will be assembled next and then used to develop additional markers to populate the black raspberry linkage map. We’ve constructed a preliminary genetic linkage map and are using this to further assemble the draft genome. We have presented our results at several international and regional conferences. Our summer intern assisted in the collection and analysis of several fruit and growth traits at the Corvallis location and screened more than 200 markers for potential use in the mapping project.

Results:

We are on track. Research plots have been established and lab work is progressing. We are gathering phenotypic data on each individual plant at the different locations.

Publications:

Bassil, N.V., B. Gilmore, K. Hummer, M. Dossett, T. Mockler, S. Filichkin, M. Peterson, C.E.

Finn, J. Lee, G. Fernandez, P. Perkins-Veazie, C. Weber R. Agunga, E. Rhodes, J.C.

Scheerens, K. Lewers, J. Graham, F. Fernández-Fernández, and S.J. Yun. 201X. Genetic and genomic resources in black raspberry *Acta Hort.* Submitted (5/30/2102).

Bassil, N., M. Dossett, B. Gilmore, T. Mockler, S. Filichkin, M. Peterson, K. Lewers, and C.

Finn. 2012. Developing genomic resources in black raspberry. Sixth Rosaceous Genomics Conference, Trento, Italy. (Abstract).

Bushakra, J., N. Bassil, M. Dossett, B. Gilmore, T. Mockler, D. Bryant, S. Filichkin, J. Weiland,

M. Peterson, C. Bradish, G. Fernandez, K. Lewers, J. Graham, and C. Finn. 2012. Black raspberry genomic resource development. *Plant and Animal Genomics*, San Diego, CA. (Abstract).

Bushakra, J.M., N. Bassil, M. Dossett, T. Mockler, D. Bryant, M. Peterson, C.E. Finn. 2013.

Developing a genotyping by sequencing protocol for linkage map construction in black raspberry. American Society of Horticultural Scientists Annual Meeting, Palm Desert, CA (Abstract).

Project Title: Cooperative raspberry cultivar development program

PI: Chad Finn,
USDA-ARS, HCRL
Research Geneticist
541-738-4037
Chad.finn@ars.usda.gov
3420 NW Orchard Ave.
Corvallis, OR 97330

Cooperators: Pat Moore, WSU
Michael Dossett Agriculture and Agri-Foods Canada

Year Initiated __2013__ **Current Year** 2014-2015__ **Terminating Year** _Continuing__

Total **Project Request:** Ongoing. Year 1: \$6,500

Other funding sources:

Current pending and support form attached

I receive and apply for funding each year with Bernadine Strik from the Oregon Raspberry and Blackberry Commission towards the cooperative raspberry and blackberry breeding program. This funding is complementary not duplicative.

I have received significant funding from the USDA-ARS, Northwest Center for Small Fruit Research and the USDA-ARS Plant Exploration Office towards our black raspberry research. We will receive matching funds from the North American Raspberry and Blackberry Association (\$3,000) and from WRRC (\$1,000) for matching funding on a black raspberry SCRI grant. While a different crop, red and black raspberry are closely related and their genetics have a lot in common and we hope that much of what we learn about aphid resistance and verticillium tolerance in black raspberry will carry over to red raspberry.

Description describing objectives and specific outcomes

The Northwest is one of the most important berry production regions in the world. This success is due to a combination of an outstanding location, top notch growers, and a strong history of industry driven research. The USDA-ARS berry breeding programs in Corvallis have a long history of developing cultivars that are commercially viable. New cultivars that are high yielding, machine harvestable, and that produce very high quality fruit are essential for the long term viability of the industry. Cultivars that replace or complement the current standards, primarily 'Meeker' would help towards that goal. The breeding programs in the region have a long history of cooperation exchanging parents, seedlings, and ideas and thoroughly testing and evaluating each other's selections. Cultivars developed by these integrated programs will benefit the entire industry in the northwest. The specific objectives include developing:

- Cultivars for the Pacific Northwest in cooperation with Agriculture and Agri-Food Canada and Washington State University that are summer bearing high-yielding, winter hardy, machine harvestable, disease and virus resistant and have superior processed fruit quality (#1 Priority).
- Fresh market cultivars will be pursued that provide season extension: improve viability of fresh marketing through floriculture or primocane fruiting types (Of Note Priority).

Relationship to WRRRC Research Priorities.

The objectives tie directly to the following priorities:

- Develop cultivars that are summer bearing, high yielding, winter hardy, machine-harvestable, disease resistant, virus resistant and have superior processed fruit quality
- Season extension: improve viability of fresh marketing

Ideally new cultivars will have improved pest resistance and so this work ties indirectly to the following priorities:

- Fruit rot including pre harvest, postharvest, and/or shelf life.
- Viruses/crumble fruit
- Foliar & Cane Diseases – i.e. spur blight, yellow rust, cane blight, etc.

Objectives:

- To develop cultivars for the Pacific Northwest in cooperation with Agriculture and Agri-Food Canada and Washington State University that are summer bearing high-yielding, winter hardy, machine harvestable, disease and virus resistant and have superior processed fruit quality (#1 Commission Research Priority).
- New fresh market cultivars will be pursued that provide season extension: improve viability of fresh marketing through floriculture or primocane fruiting types (Of Note Priority).
- To develop cultivars using new germplasm that are more vigorous and that may be grown using reduced applications of nutrients and irrigation (#2 Priority) and that are less reliant on soil fumigation (#1 Priority).

Procedures:

This is an ongoing project where cultivars and current selections serve as the basis for generating new populations from which new selections can be made, tested, and either released as a new cultivar or serve as a parent for further generations. All of the steps are taking place every year i.e. crossing, growing seedlings, selecting, propagating for testing, and testing.

Thirty to forty crosses will be done each year. Seedling populations are grown and evaluated in Corvallis, Ore. Selections are made and propagated for testing at the Oregon State University - North Willamette Research and Extension Center (Aurora, Ore.). Washington State University and Agriculture and Agri-Food Canada selections, in addition to the USDA-ARS selections, that looked outstanding as a seedling or that have performed well in other trials, are planted in replicated trials (4, 3 plant replications). Selections that we are less sure of are generally planted in smaller observation trials (single, 3 plant plot). Fruit from replicated and observation plots are

harvested and weighed, and plants and fruit are subjectively evaluated as well for vigor, disease tolerance, winter hardiness, spines, ease of removal, color, firmness, and flavor.

Fruit from the best selections are processed after harvest for evaluation in the off season.

Selections that look promising are propagated for grower trials, machine harvest trials, and for evaluation trials at other locations in Washington and B.C. This usually involves cleaning up the selections in tissue culture and then working with nurseries to generate plants for trials.

While not directly related to red raspberry at first glance, our efforts in black raspberry, which are supported by separate funding, have the potential to positively impact red raspberry. While much is specific to black raspberry, our work on aphid resistance should have applications for red raspberry. We have screened populations from across the eastern US for resistance to raspberry aphid, which is a major vector for several viruses. To this point we have identified four sources of resistance and are in the process of studying these sources further and of developing molecular markers that can be used to more efficiently select for this trait in the breeding program. We have also identified sources of verticillium resistance in this material while Meeker was susceptible. These sources can be moved into red raspberry relatively easily if there are molecular markers to facilitate identifying genotypes with resistance.

Anticipated Benefits and Information Transfer:

This breeding program will develop new raspberry cultivars that either are improvements over the current standards or that will complement current standards. In addition, the information generated on advanced selections from the WSU and B.C. programs will be made available and aid in making decisions on the commercial suitability of their materials.

Results of all trials will be made available to the industry to help them make decisions in their operations.

Budget:

Amount allocated by Commission for previous year: \$ 4,000

Funds from the USDA-ARS will be used to provide technician support and the bulk of the funding of the overall breeding project.

Salaries: Student labor (GS-2)	\$3,500
Operations (goods & services)	500
Travel ¹	1,000
Other: "Land use charge" (\$3,500/acre)	2,000
Total	\$7,000

¹To visit Puyallup, Lynden, and/or grower trials and field days in Washington

Current & Pending Support					
Chad Finn					
Name(List PI #1 first)	Supporting Agency and Project #	Total \$ Amount	Effective and Expiration Dates	% of Time Committed	Title of Project
Current:					
Finn, C.E.	North American Raspberry and Blackberry Assoc.	\$3,000	7/2013-6/2014	1	Funds towards industry matching on SCRI grant "Developing the Genomic Infrastructure for Breeding Improved Black Raspberries"
Finn, C.E.	Oregon Raspberry and Blackberry Commission	\$1,000	7/2013-6/2014	1	Funds towards industry matching on SCRI grant "Developing the Genomic Infrastructure for Breeding Improved Black Raspberries"
Finn, C.E. and J. Lee	Northwest Center for Small Fruit Research	\$32,500	7/2012-6/2014	2	Extending RosBREED in the Pacific Northwest for strawberry processing traits
Finn, C.E.	Oregon Blueberry Commission	\$13,068	7/2013-6/2015	2	Developing PNW Cultivars That May Resist Blueberry Shock Virus
Strik, BC, and Finn, C.E.	Oregon Blueberry Commission	\$17,000	7/2013-6/2014	2	Cooperative Breeding Program- Blueberries
Strik, B.C. and C.E. Finn	Oregon Raspberry and Blackberry Commission	\$23,000	7/2013-6/2014	2	Production System/Physiology Research and Cooperative Breeding Program- Raspberries and Blackberries
Strik, B.C. and C.E. Finn	Oregon Strawberry Commission	\$12,000	7/2013-6/2014	2	Cooperative Breeding Program - Strawberries
Finn, C.E., N.V. Bassil, J. Lee, G. Fernandez, P. Perkins-Veazie, C. Weber, T. Mockler, R. Agunga, E. Rhoades, J.C. Scheerens, W. Yang, K. Lewers, J. Graham, F. Fernández Fernández, S.J. Yun.	USDA Specialty Crop Research Initiative	\$1,590,717	10/1/2011-9/30/2015	10	Developing the Genomic Infrastructure for Breeding Improved Black Raspberries

Finn, C.E.	Washington Blueberry Commission	\$8,400	7/2013-6/2014	2	Blueberry breeding-Cultivar and Selection evaluation
Finn, C.E.	Washington Blueberry Commission	\$13,068	7/2013-6/2015	2	Developing PNW Cultivars That May Resist Blueberry Shock Virus
Finn, C.E.	Washington Strawberry Commission	\$3,220	7/2013-6/2014	2	USDA-ARS Cooperative Strawberry Breeding Program
Finn, C.E.	Washington Red Raspberry Commission	\$5,000	7/2013-6/2014	2	Cooperative raspberry cultivar development program.
Finn, C.E.	Washington Red Raspberry Commission	\$1,000	7/2013-6/2014	1	Funds towards industry matching on SCRI grant "Developing the Genomic Infrastructure for Breeding Improved Black Raspberries"
Oregon Raspberry and Blackberry Commission (McKenzie, C. et al)	Oregon Department of Agriculture 2013 Specialty Crop Block Grant	\$55,407	7/2013-6/2014	2	Enhancing domestic sales of processed NW caneberries as an ingredient to food manufacturers/food service through educational outreach to targeted product/menu development specialists
Name(List PI #1 first)					
Pending:					
Finn, C.E.	Washington Red Raspberry Commission	\$7,000	7/2014-6/2015	2	Cooperative raspberry cultivar development program.
Finn, C.E.	Washington Red Raspberry Commission	\$1,000	7/2014-6/2015	1	Funds towards industry matching on SCRI grant "Developing the Genomic Infrastructure for Breeding Improved Black Raspberries"

Project No:**Title:** Cooperative raspberry cultivar development program**Personnel:** Chad Finn, Research Geneticist

USDA-ARS, HCRL; 3420 NW Orchard Ave. Corvallis, OR 97330

Reporting Period: 2013

Accomplishments: Our goal is develop raspberry cultivars that either are improvements over the current standards or that will complement them. In addition, the information generated on advanced selections from the WSU and BC programs will be made available and aid in making decisions on the commercial suitability of their materials. ‘Vintage’ has been released. ‘Lewis’, ORUS 1142-1, ORUS 3705-2, ORUS 3959-3 are in grower trials in Washington as floricane fruiterers and ORUS 4090-1 and ORUS 4090-2 are propagated for trial as primocane fruiterers. Five selections have been propagated for planting in machine harvest trial in Lynden. We have 77 floricane fruiting and 41 primocane fruiting red raspberry selections from our crosses in trial, in addition to numerous WSU and BC selections (Table 1). ORUS 3239-1, ORUS 3696-1, ORUS 3700-2, and ORUS 3722-1 were identified as having excellent root rot resistance in Puyallup. Of these, ORUS 3722-1 has excellent commercial potential and is being propagated for Washington machine harvest trial; it also has an RBDV resistant parent. We made 39 red raspberry selections (31 floricane, 8 primocane).

Results: Thirty two red raspberry and 14 black raspberry crosses were attempted in spring 2013. A new seedling field was established containing red raspberry, black raspberry and blackberry seedlings. We made 39 red raspberry selections (31 floricane, 8 primocane). The selections were mostly made as potential cultivars however several are germplasm selections with a wild *R. innominatus* parent in their background. We have been working with Asian germplasm for several generations and it is now nearly cultivar quality with some parental material displaying good root rot tolerance; ORUS 3229-1 is an example of this as it has vigor on heavy soils, high yields, easy to harvest... but is yellow and a bit rough. We hope this material will be useful to our program as well as to Pat Moore’s and Michael Dossett’s. Table RY1 lists the genotypes that were harvested in 2013 or will be harvested in 2014. Presented in Tables RY2-RY5 are the results from 2013. ORUS 4090-1 and ORUS 4090-2 have been propagated for grower trial in addition to the others listed previously- ORUS 1142-1 and ‘Lewis’. ORUS 3696-1 and ORUS 3722-1 were identified as having excellent root rot resistance in Puyallup. ORUS 3696-1 has excellent potential as a parent, while ORUS 3722-1 has good commercial potential and is being propagated for Washington machine harvest trial; it also has an RBDV resistant parent.

While not directly related to red raspberry at first glance, our efforts in black raspberry have identified verticillium wilt resistance and aphid resistance (that should translate into virus resistance for the aphid transmitted viruses). While verticillium has not been a problem in the past there was some concern raised that there may be more problems with verticillium than we were aware. If these sources of resistance hold up they can be moved into red raspberry especially if there are molecular markers to identify genotypes with resistance.

Publications: Received notification that ‘Vintage’ patent will go through. Until a new cultivar is released and the notice published in a scientific journal, results from our trial are mostly presented informally in Commission reports and oral presentations.

Appendices

Table RY1. Red raspberry genotypes in the ground for evaluation in 2013 and/or 2014.

<i>Floricanne Fruiterers</i>				<i>Primocane Fruiterers</i>		
ORUS 1025-10	ORUS 3718-1	ORUS 4178-1	WSU 1447	Cascade Bounty	ORUS 3698-2	ORUS 4289-5
ORUS 1041-1	ORUS 3718-2	ORUS 4179-1	WSU 1499	Cascade Delight	ORUS 3736-1	ORUS 4291-1
ORUS 3229-1	ORUS 3722-1	ORUS 4179-3	WSU 1507/1539	Chemainus	ORUS 3736-2	ORUS 4384-1
ORUS 3234-1	ORUS 3722-1	ORUS 4183-2	WSU 1511	Lewis	ORUS 3983-3	ORUS 4386-1
ORUS 3519-1	ORUS 3722-2	ORUS 4283-1	WSU 1660	Meeker	ORUS 4086-1	ORUS 4388-1
ORUS 3523-1	ORUS 3723-1	ORUS 4283-2	WSU 1738	Oregon 1030	ORUS 4086-2	ORUS 4388-2
ORUS 3525-1	ORUS 3736-3	ORUS 4283-3	WSU 1750	Royalty	ORUS 4086-3	ORUS 4388-3
ORUS 3528-1	ORUS 3767-1	ORUS 4284-1	WSU 1792	Rudi	ORUS 4090-1	ORUS 4389-1
ORUS 3533-1	ORUS 3767-2	ORUS 4284-2	WSU 1794	Saanich	ORUS 4090-2	ORUS 4486-1
ORUS 3533-2	ORUS 3767-3	ORUS 4284-3	WSU 1912	TulaMagic (Frutafri)	ORUS 4097-1	ORUS 4487-1
ORUS 3534-1	ORUS 3771-1	ORUS 4285-1	WSU 1914	Tulameen	ORUS 4097-2	ORUS 4487-2
ORUS 3535-1	ORUS 3776-1	ORUS 4371-1	WSU 1948	Ukee	ORUS 4097-3	ORUS 4487-3
ORUS 3539-2	ORUS 3958-1	ORUS 4371-2	WSU 1964	Wakefield	ORUS 4097-4	ORUS 4494-1
ORUS 3696-1	ORUS 3959-1	ORUS 4371-3	WSU 1996		ORUS 4097-5	ORUS 4494-2
ORUS 3698-1	ORUS 3959-2	ORUS 4371-4	WSU 2010		ORUS 4098-1	ORUS 4495-1
ORUS 3702-1	ORUS 3959-3	ORUS 4371-5	WSU 2011		ORUS 4098-2	
ORUS 3702-2	ORUS 3961-1	ORUS 4373-1	WSU 2029		ORUS 4099-1	NY 02-57
ORUS 3702-3	ORUS 3973-2	ORUS 4375-1	WSU 2068		ORUS 4099-2	NY 05-44
ORUS 3702-4	ORUS 4075-1	ORUS 4380-1	WSU 2075		ORUS 4280-1	
ORUS 3705-1	ORUS 4076-1	ORUS 4380-2			ORUS 4280-2	Anne
ORUS 3705-2	ORUS 4080-1	ORUS 4380-3	BC 1-16-38		ORUS 4280-3	Crimson Giant
ORUS 3707-1	ORUS 4089-1	ORUS 4380-4	BC 3-14-12		ORUS 4285-1	Crimson Night
ORUS 3707-2	ORUS 4089-2	ORUS 4465-1	BC 92-9-15		ORUS 4287-1	Double Gold
ORUS 3711-1	ORUS 4094-1		BC 93-6-30		ORUS 4289-1	Heritage
ORUS 3711-2	ORUS 4095-1		BC 96-22R-55		ORUS 4289-3	TulaMagic (Frutafri)
ORUS 3713-1	ORUS 4097-4				ORUS 4289-4	Vintage

Table RY2. Mean yield and berry size in 2012-13 for floricanne fruiting raspberry genotypes at OSU-NWREC planted in 2010.

Genotype	Berry size (g) 2012-13 ^z	Yield (tons·a ⁻¹)		
		2012	2013	2012-13
2012	4.3 a			5.66
2013	4.1 a			2.99
<i>Replicated</i>				
Meeker	3.6 b	5.67 a	3.85 a	4.76
WSU 1507	4.8 a	5.65 a	2.14 b	3.89
<i>Observation</i>				
ORUS 3961-1	5.3	3.43	4.47	3.95
ORUS 3707-1 ^y	4.0	5.54	1.78	3.66
ORUS 3959-1	4.1	5.39	1.91	3.65
ORUS 3713-1 ^x	4.2	3.87	3.36	3.61
ORUS 3722-1	5.6	4.37	2.45	3.41
ORUS 3767-3	3.8	3.49	3.00	3.24
ORUS 3958-1	4.3	3.66	2.00	2.83
ORUS 3771-1	4.3	2.87	2.68	2.78
ORUS 3718-1 ^w	4.9	-	1.07	-

^z Mean separation within columns by LSD, $p \leq 0.05$.

^y $\frac{1}{4}$ *Rubus corchorifolius*, $\frac{3}{4}$ cultivated red raspberry.

^x $\frac{1}{8}$ *R. coreanus*, $\frac{7}{8}$ cultivated red raspberry.

^w $\frac{1}{8}$ *R. parvifolius*, $\frac{7}{8}$ cultivated red raspberry.

Table RY3. Mean yield and berry size in 2013 for floricanes fruiting red raspberry genotypes in replicated and observation trials at OSU-NWREC planted in 2011.

Genotype	Berry size (g) ^z	Yield (tons·a ⁻¹)
<i>Replicated</i>		
ORUS 3229-1 ^y	3.5 d-f	4.76 a
WSU 1660	3.3 f	4.24 ab
Meeker	3.4 ef	3.38 bc
ORUS 3234-1 ^y	3.9 cd	3.35 bc
WSU 1792	5.3 a	3.16 c
ORUS 3959-3	4.9 b	3.08 c
WSU 1750	3.4 ef	2.92 c
Ukee	3.8 c-e	2.89 c
WSU 1511	4.0 c	2.82 cd
WSU 1948	3.8 c-e	2.74 cd
WSU 1912	2.9 g	1.86 de
BC 96-22R55	4.5 b	1.55 e
<i>Nonreplicated</i>		
WSU 1507	4.5	3.43
ORUS 4179-1 ^x	2.8	3.21
WSU 1738	3.3	2.51
WSU 1447	4.5	2.00

^z Mean separation within columns by LSD, $p \leq 0.05$.

^y $\frac{1}{8}$ *R. coreanus*, $\frac{7}{8}$ cultivated red raspberry.

^x $\frac{1}{2}$ *R. occidentalis*, $\frac{1}{2}$ *R. idaeus*.

Table RY4. Mean yield and berry size in 2011-2013 for primocane fruiting raspberry genotypes at OSU-NWREC planted in 2010.

	Type	Berry size (g)	Yield (tons·acre ⁻¹)			
			2011	2012	2013	2011-13
2011		3.4 a				3.58 c
2012		3.4 a				1.57 a
2013		3.1 a				1.98 b
<i>Replicated</i>						
ORUS 4090-1	Red	3.5 a	2.50 a	4.12 a	2.64 a	3.09 a
Heritage	Red	2.9 b	0.83 c	3.73 a	1.71 a	2.09 b
ORUS 4090-2	Red	3.4 a	1.38 b	2.88 b	1.57 a	1.95 b
<i>Observation</i>						
ORUS 4097-2	Red	3.4	2.09	3.65	2.30	2.68
ORUS 4097-1	Red	4.3	1.48	3.68	2.42	2.52
ORUS 3736-1	Purp.	6.1	1.06	0.53	1.07	0.89

Mean separation within columns by LSD, $p \leq 0.05$.

Table RY5. Mean yield and berry size in 2012-13 for primocane fruiting red raspberry genotypes at OSU-NWREC planted in 2011.

	Berry size (g)	Yield (tons·a ⁻¹)		
		2012	2013	2012-13
2012	3.5 a			2.22 a
2013	3.5 a			1.94 a
<i>Replicated</i>				
ORUS 4097-1	4.0 a	2.12 a	2.26 a	2.19 a
ORUS 4097-5	3.0 b	2.05 a	2.13 a	2.09 a
ORUS 4287-1	3.7 a	2.49 a	1.43 b	1.96 a
<i>Nonreplicated</i>				
ORUS 4097-3	3.9	5.17	2.41	3.79
ORUS 4280-1	3.6	2.40	2.04	2.22
ORUS 4289-1	2.4	1.35	1.26	1.30
ORUS 4289-2	2.9	1.37	1.10	1.24
ORUS 4097-4	3.4	1.44	0.96	1.20
ORUS 4098-1	2.6	1.43	0.88	1.16
Vintage	3.1	-	1.34	-

Mean separation within columns by LSD, $p \leq 0.05$.

Table RY6. Mean yield and berry size in 2013 for primocane fruiting red raspberry genotypes at OSU-NWREC planted in 2012.

Genotype	Berry size (g)	Yield (tons·a ⁻¹)
<i>Replicated</i>		
ORUS 4289-4	2.3 c	2.20 a
Heritage	2.6 b	2.11 a
Crimson Giant	4.7 a	2.03 a
ORUS 4289-3	2.4 bc	1.35 a
ORUS 4289-1	2.3 c	1.29 a
<i>Nonreplicated</i>		
ORUS 4289-5	2.3	2.28
ORUS 4388-1	4.6	2.24
Crimson Night	3.1	2.01
NY 02-57	2.5	1.96
NY 05-44	2.8	0.96
Double Gold	3.1	0.65
Niwot (black rasp)	2.7	0.46

Mean separation within columns by LSD, $p \leq 0.05$.

Table RY7. Ripening season for floricanе fruiting red raspberry genotypes at OSU-NWREC. Planted in 2010-11 and harvested 2012-13.

Genotype	Year planted	Harvest season			No. years in mean	Rep/ Obsv.
		5%	50%	95%		
ORUS 3771-1	2010	15-Jun	22-Jun	29-Jun	2	Obsv
ORUS 4179-1	2011	11-Jun	25-Jun	9-Jul	1	Obsv.
ORUS 3767-2	2010	15-Jun	25-Jun	25-Jun	2	Obsv
ORUS 3707-1	2010	18-Jun	25-Jun	2-Jul	2	Obsv
ORUS 3767-3	2010	15-Jun	29-Jun	13-Jul	2	Obsv
ORUS 3722-1	2010	22-Jun	29-Jun	13-Jul	2	Obsv
WSU 1447	2011	18-Jun	2-Jul	16-Jul	1	Obsv.
WSU 1507	2011	18-Jun	2-Jul	16-Jul	1	Obsv.
WSU 1511	2011	18-Jun	2-Jul	16-Jul	1	Rep
WSU 1660	2011	18-Jun	2-Jul	16-Jul	1	Rep
WSU 1912	2011	18-Jun	2-Jul	16-Jul	1	Rep
BC 96-22R-55	2011	25-Jun	2-Jul	16-Jul	1	Rep
ORUS 4080-1	2011	2-Jul	2-Jul	30-Jul	1	Obsv.
WSU 1507	2010	22-Jun	2-Jul	23-Jul	2	Rep
ORUS 3958-1	2010	22-Jun	6-Jul	20-Jul	2	Obsv
Meeker	2011	25-Jun	9-Jul	16-Jul	1	Rep
ORUS 3229-1	2011	25-Jun	9-Jul	23-Jul	1	Rep
Ukee	2011	25-Jun	9-Jul	23-Jul	1	Rep
WSU 1750	2011	25-Jun	9-Jul	23-Jul	1	Rep
WSU 1792	2011	25-Jun	9-Jul	23-Jul	1	Rep
WSU 1948	2011	25-Jun	9-Jul	23-Jul	1	Rep
ORUS 3234-1	2011	25-Jun	9-Jul	30-Jul	1	Rep
ORUS 3718-1	2010	2-Jul	9-Jul	23-Jul	1	Obsv
ORUS 3713-1	2010	22-Jun	9-Jul	23-Jul	2	Obsv
ORUS 3959-3	2010	29-Jun	9-Jul	27-Jul	2	Obsv
ORUS 3961-1	2010	22-Jun	13-Jul	27-Jul	2	Obsv
Meeker	2010	25-Jun	13-Jul	23-Jul	2	Rep
ORUS 3959-1	2010	29-Jun	13-Jul	23-Jul	2	Obsv
ORUS 3959-3	2011	2-Jul	16-Jul	30-Jul	1	Rep
WSU 1738	2011	9-Jul	16-Jul	23-Jul	1	Obsv.

Table RY8. Ripening season for primocane fruiting red raspberry genotypes at OSU-NWREC. Planted in 2010-12 and harvested 2011-13.

Genotype	Year planted	Harvest season			No. years in mean	Rep/ Obsv.
		5%	50%	95%		
Vintage	2011	30-Jul	6-Aug	27-Aug	1	Obsv.
ORUS 4097-4	2011	3-Aug	10-Aug	17-Aug	2	Obsv.
ORUS 3736-2	2010	5-Aug	12-Aug	28-Aug	3	Obsv.
ORUS 4289-5	2012	13-Aug	13-Aug	27-Aug	1	Obsv.
ORUS 4289-4	2012	13-Aug	13-Aug	27-Aug	1	Rep
ORUS 4289-2	2011	3-Aug	13-Aug	27-Aug	2	Obsv.
ORUS 4280-1	2011	3-Aug	17-Aug	3-Sep	2	Obsv.
ORUS 4287-1	2011	6-Aug	17-Aug	14-Sep	2	Rep
NY 02-57	2012	13-Aug	20-Aug	3-Sep	1	Obsv.
ORUS 4289-1	2012	13-Aug	20-Aug	3-Sep	1	Rep
ORUS 4289-3	2012	13-Aug	20-Aug	3-Sep	1	Rep
Heritage12	2012	13-Aug	20-Aug	10-Sep	1	Rep
ORUS 4098-1	2011	17-Aug	24-Aug	10-Sep	2	Obsv.
NY 05-44	2012	13-Aug	27-Aug	3-Sep	1	Obsv.
ORUS 4388-1	2012	13-Aug	27-Aug	3-Sep	1	Obsv.
Crimson Night	2012	13-Aug	27-Aug	17-Sep	1	Obsv.
ORUS 4097-1	2011	6-Aug	27-Aug	28-Sep	2	Rep
ORUS 4289-1	2011	13-Aug	27-Aug	14-Sep	2	Obsv.
ORUS 4097-3	2011	13-Aug	27-Aug	17-Sep	2	Obsv.
ORUS 4090-2	2010	12-Aug	28-Aug	16-Sep	3	Rep
ORUS 4090-1	2010	9-Aug	30-Aug	18-Sep	3	Rep
ORUS 4097-2	2010	12-Aug	30-Aug	18-Sep	3	Obsv.
Crimson Giant	2012	13-Aug	3-Sep	24-Sep	1	Rep
Double Gold	2012	20-Aug	3-Sep	17-Sep	1	Obsv.
Niwot (black rasp)	2012	20-Aug	3-Sep	24-Sep	1	Obsv.
ORUS 4097-5	2011	6-Aug	3-Sep	1-Oct	2	Rep
Heritage10	2010	19-Aug	4-Sep	23-Sep	3	Rep
ORUS 4097-1	2010	16-Aug	6-Sep	27-Sep	3	Obsv.

2013 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

Continuing Project Proposal

Proposed Duration: (3 years)

Project Title: Red Raspberry Cultivar Development

PI: Michael Dossett

Organization: Agriculture and Agri-Food Canada

Title: Visiting Fellow

Phone: (604) 796-2221 ext. 1284

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Cooperators:

Pat Moore, WSU Puyallup
Chad Finn, USDA-ARS, Corvallis
Nahla Bassil, USDA-ARS, Corvallis
Tom Forge, Nematology/Plant Pathology AAFC
Andrew Jamieson, Berry Breeder AAFC Kentville NS

Year Initiated 2013 **Current Year** 2014 **Terminating Year** 2015

Total Project Request: **Year 1** \$12000 **Year 2** \$12000 **Year 3** \$12000

Other funding sources:

This is part of a larger project to support berry breeding in British Columbia in the near term until longer-term solutions can be found after the program was cut by the federal government in 2012. In January 2013, we applied for funding through AAFC's Growing Forward II Initiative. In August 2013, we received word that the project was accepted for funding, though we are still waiting for a formal public announcement before we can openly discuss it. As such, information that the project was funded is still regarded as confidential. This proposal leverages industry dollars 1:3 and all money received from Washington berry commissions in addition to BC industry associations is applied to this effort. More information regarding the budget is outlined later in the proposal.

Description: This project is to support the continued effort to breed raspberry cultivars adapted to the PNW. Chemical pest control measures are becoming increasingly unavailable, making genetic resistance and tolerance more important. Breeding for resistance, yield, and fruit quality is the most sustainable way to address industry needs and ensure long-term competitiveness. We will continue to cross and select from a diverse gene pool and evaluate previous selections with the following specific objectives:

- Develop red raspberry cultivars and elite germplasm, stressing suitability for machine harvest, fruit quality, as well as resistance to root rot, RBDV and other diseases
- Develop red raspberry cultivars and elite habit that is suitable for machine harvesting and produces high yields of superior fruit quality and fruit rot resistance.
- Identify and select raspberries with dark red fruit for processing that also exhibit characteristics that are suited for IQF processing
- Identify and incorporate new sources of resistance to aphids, spider mites, and other insect pests.

- Continue development and testing of molecular tools to speed up the process of selecting and identifying parents and seedlings in the program with durable disease resistance and outstanding quality traits.

Justification and Background: (400 words maximum)

The red raspberry industry is facing more challenges than ever with increased production costs and increased pressures on prices from the global marketplace. Genetic improvement is one of the most sustainable ways for the raspberry industry to maintain its competitive edge in the long-term. Improved quality, yield, and resistance to pests and diseases to help alleviate these problems are realistic and achievable goals that will benefit raspberry producers in Washington State.

The BC breeding program has a long history of producing cultivars with excellent fruit quality characteristics and has been making steady progress in recent years to combine this with improved resistance to *Phytophthora* root rot and RBDV. In 2012, we expanded our efforts to identify machine-harvestability in our selections by contracting with a local grower to machine harvest our replicated plots. This effort was so successful we expanded it to additional plots and evaluation of seedlings in 2013. We plan to continue this, because we believe this is the fastest way to identify selections with merit and weed out selections that lack potential for the majority of PNW growers. In April 2012, AAFC announced that it was cutting support for the program with the expectation that local industries pick up the slack. This proposal aims to keep the program running and continue the improvement and evaluation of germplasm in which the Washington and BC raspberry industries have invested heavily over the past several years.

While there are currently raspberry breeding efforts in Washington and Oregon, each program has its strengths and weaknesses inherent in the germplasm base and breeding lines they have established through their history. One of the strengths of the BC program is the firmness and quality of its selections. We will continue to collaborate and exchange information and selections with the programs in Washington and Oregon so that promising material gets evaluated in as many test locations as possible and so that we can continue to combine efforts to complement the strengths of each program. Over the next few years, AAFC has verbally committed to providing office and lab space in support of the continuation of this program, as well as limited field space and staff support. While this means that the cost of continuing to staff and run the program has risen dramatically, this project will ensure that the investments of time and money already made towards the program will not be lost and that this effort can continue.

Relationship to WRRRC Research Priority(s):

This project directly addresses the WRRRC #1 priority to develop cultivars that are summer bearing, high yielding, winter hardy, machine-harvestable, disease resistant, virus resistant and have superior processed fruit quality

Objectives:

Each of the specific objectives listed above will be attempted during the project period and each is an ongoing process that will be addressed in this funding year and in future funding years. While many inferior plants can be identified and eliminated in the early stages of the process, selections must be tested rigorously over a period of several years by the project staff and producers before they can be recommended for release and commercialization.

Procedures:

The breeding program is an ongoing project that continually makes new crosses and selections each year with the objective of developing new cultivars to support the raspberry industry. The current expectation is that the next DIAP program will be for a period of five years, at which time alternative measures will need to be in place for the continuation of the program. The program operates on a cycle such that all activities in this project occur at some point in the season of every year. This includes:

- Making new crosses - emphasizing combining parents with machine harvestability and resistance to RBDV and root rot
- Planting new seedling fields from previous year's crosses for future evaluation
- Selection of mature seedling plantings with an emphasis on fruit quality and machine-harvestability
- Establish replicated trials of selections to assess machine-harvestability, quality, and yield
- Test field plantings for RBDV to establish which selections are susceptible and which may be resistant
- Screen selections in replicated trials for root rot resistance in the greenhouse to establish potential for resistance
- Propagate promising selections for further trial at our substation and on producers' fields.
- Conduct collaborative research and testing with USDA-ARS in Corvallis, WSU, AAFC, and elsewhere.

A specific part of this project with more definite timelines is the development and evaluation of molecular genetics tools to identify markers for insect and disease resistance as well as other traits. This is in collaboration with Pat Moore, and Nahla Bassil, testing new markers, and then validating those markers across breeding populations to assess their utility. The first stage of this work (marker identification) has begun. We are currently in the process of screening markers in two populations that segregate for different sources of root rot resistance, a newly identified source of RBDV resistance, and three sources of aphid resistance (one broken, two unbroken). Basic linkage maps are essentially complete, but we are actively adding markers to these maps to increase their resolution and the ability to identify markers tightly linked to traits of interest. The populations have already been screened for aphid resistance. Screening for root rot resistance has started in the greenhouse and will continue over the next few winters in addition to planting in a field with heavy pressure in Puyallup, WA. Testing for RBDV infection will be an ongoing process.

Anticipated Benefits and Information Transfer: (100 words maximum)

Specific benefits that will result from this project include:

- Continued development of new cultivars and selections that will provide alternatives for producers with high fruit quality and improved yield and resistance to pests and diseases.
- Continued development of technologies that will assist this and other breeding programs to more efficiently select promising genotypes in the future.

Results will be transferred to users through regular presentations at field days, and local meetings such as the LMHIA Short Course and the Washington Small Fruit Conference with information on new releases and selections available for testing.

Budget: Indirect or overhead costs are not allowed unless specifically authorized by the Board

	2013	2014	2015
Salaries^{1/}	\$	\$	\$
Time-Slip	\$10000	\$10000	\$10000
Operations (goods & services)	\$2000	\$2000	\$2000
Travel^{2/}	\$	\$	\$
Meetings	\$	\$	\$
Other	\$	\$	\$
Equipment^{3/}	\$	\$	\$
Benefits^{4/}	\$	\$	\$
Total	\$12000	\$12000	\$12000

The costs we are asking WRRC to support represent approximately 28% of the red raspberry portion of the industry contribution needed for the next cycle of funding. We have allocated this primarily to student labor for field planting, plot maintenance, and harvest, as well as some operational costs towards contracting for mechanical harvesting of plots (\$100/hour for machine and driver). Hiring students for the summer period costs approximately \$10,000/student. With the leveraged support, the budget we are proposing to WRRC will cover the cost of contracting the machine harvester and hiring a summer crew of four students (May 1 – August 30) to work on planting and maintaining plots (weeding, some pruning, trellis building and take down, etc.) before and after the fruiting period as well as harvesting/weighing fruit from the plots during the period from late June to early August. All other project costs including travel, supplies, scientist salary, overhead, etc., will be coming from dollars contributed by BC industry associations.

Budget Justification

^{1/}Specify type of position and FTE.

^{2/}Provide brief justification for travel requested. All travel must directly benefit project. Travel for professional development should come from other sources. If you request travel to meetings, state how it benefits project.

^{3/}Justify equipment funding requests. Indicate what you plan to buy, how the equipment will be used, and how the purchase will benefit the growers. Include attempt to work cooperatively with others on equipment use and purchase.

^{4/}Included here are tuition, medical aid, and health insurance for Graduate Research Assistants, as well as regular benefits for salaries and time-slip employees.

Other funding sources:

All amounts are requested funds for the coming fiscal year and are pending final approval. Funding covers all costs for the salary for the scientist, hiring student labor, materials and supplies, land rental for plot space in Abbotsford (raspberries and strawberries), travel and contract costs for mechanical harvesting, winter pruning/tying, and some land prep.

Raspberries (50% Effort, \$43,315 needed):

BC Raspberry Industry Development Council	\$31,315
Washington Red Raspberry Commission	\$12,000

Blueberries (40% Effort, \$33,847 needed):

BC Blueberry Council	\$28,847
Washington Blueberry Commission	\$5,000

Strawberries (10% Effort, \$5,926 needed):

BC Strawberry Growers Association	\$3,326
Washington Strawberry Commission	\$2,600

Government of Canada – Federal Matching Dollars: \$249,263

Title: Red Raspberry Cultivar Development

Personnel: Chaim Kempler, Research Scientist (retired), Michael Dossett, Visiting Fellow
Brian Harding (retired), Gosia Zdanowic and Georgia Kliever, Technicians
Agriculture and Agri-Food Canada, Pacific Agri-Food Research Centre, PO Box 1000, 6947 #7
Hwy. Agassiz, BC, Canada, V0M 1A0, Michael.Dossett@agr.gc.ca Tel: 604-796-2221 ext.1284
Fax: 604-796-0359

Reporting Period: 2012-2013

Summary:

In 2013, the PARC program evaluated three machine-harvested and one hand-harvested trial plots at the Clearbrook substation. Despite difficulties early and late in the harvest period, a number of selections with superior machine-harvestability were identified. In addition, we evaluated roughly 11,500 raspberry seedlings and made new 190 selections. Roughly 65 of the 81 crosses performed in 2013 were geared toward combining improved root rot resistance and resistance to raspberry bushy dwarf virus with the best yielding selections that had superior machine-harvest quality from trials. Germplasm development work continued with trials aimed at inferring heritability of firmness, crumbliness, and yield associated with mechanical harvesting, as well as identifying and incorporating new sources of insect and disease resistance and developing markers for more efficient selection of resistance in the future. The BC Raspberry Industry Development Council, BC Blueberry Council, and BC Strawberry Growers' Association are actively trying to find a long-term solution to keep the program operating, though it is entirely dependent on soft money at this time.

Machine-harvested Trials

Three machine-harvested yield trials were run at the Clearbrook substation in 2013. The first was an unreplicated trial of 112 10-plant plots and the other two were replicated trials of ~60 3-plant plots which had been hand-harvested in previous years. Due to complications with funding and obtaining needed permission for operation of the harvester at the Clearbrook site, our first machine-harvest was not until July 8, about a week and a half later than needed to evaluate many of the earlier ripening selections. Equipment problems with the harvester meant that the last machine-harvest was on July 26, about a week or so before the end of the season. Despite missing yield data from these early and late picks, we were able to make good evaluations on fruit crumbliness and machine-harvestability of the plots and this is really the most important part of the evaluations from the machine-harvest trials.

Evaluation of the large unreplicated machine-harvest plot confirmed observations on the baby crop last year for many of the selections. Most of the selections identified last year as having good machine-harvest quality were again observed to have excellent quality this year, although generally fruit was softer and crumblier from additional exposure to *Raspberry bushy dwarf virus* (RBDV) and higher pressure from Spotted-winged Drosophila. Overall yields are reflective of the trends observed last year, but a number of higher-yielding selections from last season had more modest yields this year. Unfortunately, the most interesting selections from this trial from a quality standpoint have no resistance to root-rot and only a few have potential resistance to RBDV. Two selections are the exception to this, BC 5-11-1 (BC 90-19-34 x WSU 1112) has been identified in the greenhouse for resistance to root rot and may prove to have tolerance in the field and BC 6-64-75 (Cascade Delight x Saanich) has potential for some root rot tolerance based on its pedigree. This field was removed in fall 2013 and the best selections from this trial have been propagated and were planted in machine-harvest trials in Lynden in fall 2012 and 2013. A full summary of yield, fruit size, and season for all three trials is available upon request.

Selections of particular interest from the 2010-planted machine harvest trial at

Clearbrook:

BC 5-11-1: (BC 90-19-34 x WSU 1112) Nice dark fruit with good flavor and medium firmness. Machine-harvests very nicely. Resistant to root rot. Was slow to establish and did not yield particularly well in 2012, though 2013 yields were close to Chemainus.

BC 5-34-24: (WSU 1112 x Chemainus) Fruit looked very nice in the machine harvest trial in 2012 and 2013. High quality and attractive with color that is borderline, but probably OK. Yields were slightly higher than Chemainus or Saanich. Greenhouse root rot screening gave intermediate results, so not strongly resistant, but may hold up OK on some marginal sites. This selection has dual-purpose potential and may have a place in the fresh market.

BC 6-22-44: ([Qualicum x Willamette] x Saanich) Absolutely beautiful fruit, machine-picked very well in 2013 and 2012 with yields about 20-30% higher than Chemainus and Saanich. Flavor is only OK. This was probably the most impressive fruit in 2012 and 2013 in our machine-harvest trials, but the plants don't look particularly healthy or aggressive. Based on pedigree, will not have root rot resistance.

BC 6-22-35: ([Qualicum x Willamette] x Saanich) Yielded 50% higher than Chemainus or Saanich in 2012, but yields were off noticeably in 2013. Fruit is very nice quality with OK flavor and a dusty appearance. Based on pedigree, will not have root rot resistance.

BC 6-27-41: ([Qualicum x Willamette] x Saanich) Exceptionally high yielding with about 50% higher yields than Saanich or Chemainus in 2012 and also very high in 2013. Fruit is very attractive and machine-harvests extremely well. Color may be borderline and flavor is only questionable to OK (similar to Saanich). Unfortunately, it looks likely to be susceptible to root rot. Plants were planted at the WRRC machine-harvest trial in Lynden in 2012.

BC 6-50-41: (Saanich x Canby): Dark fruit machine-harvests very cleanly and with about 30% higher yields than Chemainus or Saanich in 2012 and very strong yields in 2013 as well. Fruit holds shape well and looks good in a machine-harvest tray, but may be on the soft side? Flavor is OK to good. Both parents sensitive to root rot.

BC 6-50-44: (Saanich x Canby): Very similar to BC 6-50-41 but with berries slightly smaller. Dark fruit that machine-harvests very well with about 30% higher yields than Chemainus or Saanich in 2012 and very strong yields in 2013 as well.

BC 6-64-75: (Cascade Delight x Saanich) Very high yielding (~35% higher than Chemainus or Saanich in 2012) with nice large good quality fruit. Some crumbly fruit in 2013 seems to correlate with individual plants which tested positive to RBDV, otherwise fruit is very nice, but possibly too light for processing. Might have a spot in the fresh market, and may have some root rot tolerance from Cascade Delight.

Hand-harvested Yield Trials:

A replicated yield trial planted in 2011 was harvested for the first time in 2013. The main point of interest for this trial is that it is the first replicated trial planting with the advanced selection BC 92-9-15. While quality was a major issue in the early season fruit due to rain nearly every day at the start of the season, the volume of fruit appeared to be comparable to Malahat. This selection is also firmer than Malahat and has very good flavor. It has consistently held up much better to root disease pressures in grower fields than Malahat and is expected to replace this cultivar. BC 92-9-15 is expected to be named later this winter at the LMHIA Short Course in Abbotsford. Summary results from this trial are available upon request.

Other work:

The program made a big push to combine machine-harvestability with better tolerance to root rot and resistance to RBDV with a larger than average crossing block, of which ~65 crosses were devoted to combining these traits in elite selections. This will be planted in a replicated planting to inheritance of machine-harvestability and yield in the seedling populations.

We evaluated approximately 11,500 seedlings this year, about 2.5 – 3 times the average number

for the program. The backlog was due in part to lack of space to plant seedlings in 2009 and 2010, as well as a seed exchange with Harvey Hall in 2010. About half of the seedlings were evaluated at least once over the course of the season from the mechanical harvester and seedlings were selected from the harvester which appeared to give good fruit quality as it came down the belt. Of the 190 selections made in 2013, 60 were from the seed exchange, while the remaining 130 came from BC crosses in 2008, 2009, and 2010. There are many exciting traits in this material ranging from season extension to high-yielding plants and extremely firm large chunky fruit that looks like it will mechanically harvest. Perhaps even more exciting is the potential for combining these traits with disease resistance. Of the 190 selections made in 2013, 123 have at least one parent that was resistance to RBDV, and 75 have two resistant parents. In addition, 96 of these selections had at least one parent that had some degree of tolerance to *Phytophthora* root rot, while both parents of 34 of these selections were resistant to root rot. Thirty of these selections combined parents with root rot and RBDV resistance.

Germplasm Development

Linkage Mapping:

Since 2012, the program has been working with two genetic linkage mapping populations which segregate for different sources of aphid resistance, root rot resistance, and resistance to *Raspberry bushy dwarf virus* in addition to other interesting traits. This effort continued in 2013. At present, we have basic linkage maps constructed with a combination of previously published markers from red raspberry, new markers developed by the program in 2012. We are in the process of adding more markers to this map from markers developed in-house through sequencing in red raspberry and from the soon-to-be-released draft of the black raspberry genome sequence. This last group of markers should allow better comparison between red and black raspberry genomes and aid in transferring specific traits of interest between the two species. All the seedlings in these populations have undergone an initial screen for aphid resistance from 3 different aphid biotypes and in 2013 we began screening individuals for root rot resistance in the greenhouse. We are very close to having tentative markers/map positions for 2 aphid resistance genes and for RBDV resistance. In addition, we will be validating a marker which appears to be associated with resistance to *Phytophthora* root rot, as we screen more individuals for this disease. The plan is to plant all of these seedlings in the field in Abbotsford in 2014 as well as in a replicated root-rot field trial in Puyallup, WA, where Pat Moore screens selections for field resistance to this disease.

Root Rot Screens:

In 2013, the breeding program continued to refine the process of screening selections and germplasm for resistance to *Phytophthora* root rot in the greenhouse. To date, more than 300 selections and about 40 different wild seedling populations have been screened for root rot resistance. In late 2012 and early 2013, we began using quantitative PCR to help validate results from these screens and learn more about the process. One thing we have learned is that PCR allows us to more objectively evaluate relative resistance of selections as compared to visual assessment of damage to roots. This may be, in part, related to contamination in the greenhouse from *Pythium* and other pathogens which can cause darkening of roots without *Phytophthora* infection. PCR results are very closely tied to pedigree, with the most susceptible selections tested being derivatives of Glen Moy through Qualicum and/or Cowichan. Cowichan, Rudi, and BC 3-14-12 were all found to be extremely sensitive to infection, showing symptoms and plant death more quickly than what had been regarded as our susceptible control, Malahat. Another thing we learned is that Malahat is not as susceptible to *Phytophthora* as previously thought, with degree of susceptibility being very similar to its parent, Meeker. Susceptibility of Malahat in the field may be more related to a complex of *Phytophthora* and susceptibility to other soil-borne pathogens weakening plants. In support of this, our tests found that Malahat died in less than two weeks from saturated field soils in which PCR tests revealed that this infection was due to

Pythium rather than *Phytophthora*.

Germplasm enhancement:

In 2013, the program continued germplasm enhancement by planting seedlings of crosses and backcrosses with several different wild *Rubus* species including *R. phoenicolasius*, *R. lasiostylus*, *R. niveus*, *R. occidentalis*, and *R. spectabilis*. The most interesting of these are the crosses derived from *R. lasiostylus*, which feature very large and very firm fruit, resistance to root rot, excellent vigor, and high productivity. This material is generally very light-colored (*lasiostylus* has yellowish fruit) and requires at least another generation to be commercially viable. The crosses derived from salmonberry (*R. spectabilis*) are also very interesting, with some blooming as many as three weeks before Malahat and other early raspberry cultivars. While this is still a long-ways from leading to new varieties, it has the potential to have strong resistance to root rot in addition to moving the raspberry season for fresh fruit significantly earlier.

Wild *Rubus idaeus*:

Since the late 1970s, the program has been active in acquiring and evaluating wild red raspberry germplasm. Starting in 2011 and continuing in 2013, we renewed these efforts acquiring wild germplasm from repositories in the US and Canada and actively collecting and soliciting collections from collaborators. The main goal of this work is to identify new sources of variation for resistance root and cane diseases, foliar diseases, insects, and viruses. In 2013, we screened just over 40 wild populations for resistance to root rot in the greenhouse and planted ~700 individuals representing ~60 populations from across North America in the field for further evaluation. Derivatives from this material could be viable for commercial production in as few as 2-3 generations. One example of the outcome of this ongoing work is illustrated below.

Fig. 1 BC 10-3-9 is one example of the germplasm enhancement ongoing in the breeding program to help sustain long-term breeding progress. It is a very productive half-wild raspberry selection with large (4-5g) soft fruit, vigorous canes, and a new source of aphid resistance. It is expected to have excellent root rot resistance and be a good breeding parent.



Publications:

We anticipate additional publications to follow in the coming months as the manuscripts are currently in the process of being written.

Dossett, M., C. Kempler, and H. Daubeny. 2013. BC 90-19-34 and BC 93-16-43 Red Raspberries. *HortScience*. 85:664-667.

Current & Pending Support

Instructions:					
1. Record information for active and pending projects. 2. All current research to which principal investigator(s) and other senior personnel have committed a portion of their time must be listed whether or not salary for the person(s) involved is included in the budgets of the various projects. 3. Provide analogous information for all proposed research which is being considered by, or which will be submitted in the near future to, other possible sponsors.					
Name (List PI #1 first)	Supporting Agency and Project #	Total \$ Amount	Effective and Expiration Dates	% of Time Committed	Title of Project
Michael Dossett	Current: AAFC, WWRC, RIDC, LMHIA	\$801,266	April 1, 2013 – March 31, 2018	50%	Current funding comes from AAFC’s Growing Forward II Initiative in the form of a proposal with two sections, “Berry Cultivar Development” and “Berry Germplasm Development.” In this initiative, industry dollars are matched 1:3 with Federal government support. Since this is an umbrella project, I have broken down portions and time commitments by commodity for illustrative purposes. While we received word about this funding in August 2013, a formal public announcement has not yet been made, so this information is still confidential. Red Raspberry Breeding for the Pacific Northwest
	AAFC, BCBC, LMHIA	\$641,012	April 1, 2013 – March 31, 2018	40%	Blueberry Breeding for the Pacific Northwest
	AAFC, WSC, BCSGA, LMHIA	\$160,253	April 1, 2013 – March 31, 2018	10%	Evaluating Strawberry Cultivars and Germplasm for BC and Northern Washington

PROJECT: 13C-3755-5641

TITLE: Red Raspberry Breeding, Genetics and Clone Evaluation

CURRENT YEAR: 2014

Proposed Duration: continuing

PI: Patrick P. Moore, Scientist

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2606 W Pioneer

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Co-PI: Wendy Hoashi-Erhardt, Scientific Assistant

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Project Request: \$70,000 for 2014-2015

Other funding sources: USDA/ARS Northwest Center for Small Fruits Research
\$19,835 for 2013-2014 for both raspberry and strawberry breeding

Description: The program will develop new red raspberry cultivars for use by commercial growers in Washington. Using traditional breeding methods, the program will produce seedling populations, make selections from the populations and evaluate the selections. Selections will be evaluated for adaptation to machine harvestability by planting selections with cooperating growers. Promising selections will be propagated for grower trials and superior selections will be released as new cultivars. Specific traits to incorporate into new cultivars are high yield, machine harvestability, raspberry bushy dwarf virus (RBDV) resistance and root rot tolerance with superior processed fruit quality.

Justification and Background: The Pacific Northwest (PNW) breeding programs have been important in developing cultivars that are the basis for the industry in the PNW. New cultivars are needed that are more productive, machine harvestable, RBDV resistant and tolerant to root rot while maintaining fruit quality. Replacement cultivars for 'Willamette' and 'Meeker' and new cultivars that extend the season are needed. With 99+% of the Washington production used for processing, new cultivars need to be machine harvestable.

There has been a history of cooperation between the breeding programs in Oregon, British Columbia, and Washington and material from other programs evaluated. This cooperation needs to continue as cultivars developed by these programs will be of value to the entire PNW raspberry industry.

Relationship to WRRRC Research Priorities: This project addresses a first-tier priority of the WRRRC: Develop cultivars that are summer bearing, high yielding, winter hardy, machine-harvestable, disease resistant, virus resistant and have superior processed fruit quality

OBJECTIVE: Develop summer fruiting red raspberry cultivars with improved yields and fruit quality, and resistance to root rot and raspberry bushy dwarf virus. Selections adapted to machine harvesting or fresh marketing will be identified and tested further.

Procedures: This is an ongoing project that depends on continuity of effort. New crosses will be made each year, new seedling plantings established, new selections made among previously

established seedling plantings, and selections made in previous years evaluated.

1. Crosses will be made for summer fruiting cultivar development. Primary criteria for selecting parents will be machine harvestability, RBDV resistance, root rot tolerance, yield and flavor.
2. Seed from the crosses made in 2013 will be sown in 2013-2014. The goal will be to plant 108 plants for each cross.
3. Selections will be made among the seedlings planted in 2011 and 2012. Seedlings will be subjectively evaluated for yield, flavor, color, ease of harvest, freedom from pests, appearance, harvest season and growth form. Based on these observations, seedlings will be selected for propagation and further evaluation. Typically, the best 1% or less of a seedling population will be selected.
4. The selected seedlings will be propagated for testing. Shoots will be collected and placed into tissue culture. Selections that are not successfully established in tissue culture will be propagated by root cuttings, grown in the greenhouse and then propagated by tissue culture.
5. Eight plants of each selection will be planted in a grower planting for machine harvesting evaluation. Three plants of each selection will also be planted at WSU Puyallup in observation plots.
6. The machine harvesting trials established in 2011 and 2012 will be harvested in 2014. Evaluations will be made multiple times through the harvest season.
7. Samples of fruit from promising selections will be collected and analyzed for soluble sugars, pH, titratable acidity and anthocyanin content.
8. Selections that appear to machine harvest well will be planted in a second machine harvesting trial, in replicated plantings at WSU Puyallup for collection of hand harvest data and screened for root rot tolerance and RBDV resistance (if potentially resistant based on parentage).
9. The replicated plantings established in 2011 and 2012 at WSU Puyallup will be hand harvested for yield, fruit weight, fruit rot and fruit firmness.
10. Selections identified in machine harvest trials and other evaluations as having potential for release as a new cultivar will be propagated for grower trials in plantings sufficient to evaluate for suitability for IQF use.

ANTICIPATED BENEFITS AND INFORMATION TRANSFER:

This program will develop new raspberry cultivars that are more productive or more pest resistant. The emphasis of the program is on developing machine harvestable cultivars. Such cultivars may result from crosses made this year or may already be under evaluation. When a superior selection is identified and adequately tested, it may be released as a new cultivar and be available for commercial plantings. Promising selections and new cultivars will be displayed at field days. Presentations will be made on breeding program activities at grower meetings.

PROPOSED BUDGET:

Funds from the Northwest Center for Small Fruit Research and support provided by WSU Agriculture Research Center will be used to provide partial technical support for the program.

The funds requested will be used for technical support, timeslip labor; field, greenhouse, and laboratory supplies; and travel to research plots and to grower meetings to present results of research. The proposed budget also includes \$3,000 for farm service fees.

Budget:	2013-2014	2014-2015
00 Salaries	2,075	8,614
Scientific Assistant (0.10 FTE)		
Ag Res Tech 2 (0.10 FTE)		
Ag Res Tech 1 (0.10 FTE)		
01 Timeslip Labor	37,562	28,500
03 Service and Supplies	19,910	19,309 ¹
04 Travel	4,000	4,000 ²
07 Benefits		
SA, ART2, ART1		6,814
Timeslip	3,606	2,765
Ag Res Tech 2	847	
Total	\$68,000	\$70,000

¹ Includes: Field, greenhouse, and laboratory supplies; \$3,000 for farm service fees and \$13,000 for expenses for the following test plantings for evaluation of raspberry selections.

Maintenance and harvest of test plantings

Machine harvesting trial established in 2011 – Honcoop Farms \$3,000

Machine harvesting trial established in 2012 – Honcoop Farms \$3,000

Maintenance of test plantings

Machine harvesting trial established in 2013 – Curt Maberry Farms \$3,000

Establishment and maintenance of new test planting

Machine harvesting trial to be established in 2014

 Will work with the WRRC to identify a suitable grower for the
 2014 machine harvesting trial \$4,000

² Travel to research plots and to grower meetings to present results of research

Current Support

Name (List PI #1 first)	Supporting Agency and Project #	Total \$ Amount	Effective and Expiration Dates	Title of Project
Moore, P.P. and Hoashi- Erhardt	Northwest Center for Small Fruit Research	\$19,835	2013-2014	Small Fruit Breeding in the Pacific Northwest
Moore, P.P. and Hoashi- Erhardt	Washington Red Raspberry Commission	\$68,000	2013-2014	Red Raspberry Breeding, Genetics and Clone Evaluation
Moore, P.P. and Hoashi- Erhardt	Washington Strawberry Commission	\$24,000	2013-2014	Genetic Improvement of Strawberry
Moore, P.P. and Hoashi- Erhardt	Oregon Raspberry and Blackberry Commission	\$4,000	2013-2014	Genetic Improvement of Raspberry
Moore, P.P. and Hoashi- Erhardt	Oregon Strawberry Commission	\$4,000	2013-2014	Genetic Improvement of Strawberry

Pending Support

Name (List PI #1 first)	Supporting Agency and Project #	Total \$ Amount	Effective and Expiration Dates	Title of Project
Moore, P.P. and Hoashi-Erhardt	Washington Red Raspberry Commission	\$70,000	2014-2015	Red Raspberry Breeding, Genetics and Clone Evaluation
Moore, P.P., K.K. Lanning and R.R. Martin	Washington Red Raspberry Commission	\$8,750	2014-2015	Tracking the movement of RBDV
Moore, P.P. and Hoashi-Erhardt	Washington Strawberry Commission	\$37,000	2013-2014	Genetic Improvement of Strawberry

Project: 13C-3755-5641
Title: Red Raspberry Breeding, Genetics and Clone Evaluation
Personnel: Patrick P. Moore, Scientist, Washington State University Puyallup Research and Extension Center
Wendy Hoashi-Erhardt, Scientific Assistant, WSU Puyallup

Reporting Period: 2013

Accomplishments: *Advanced Selections/ new release.* A selection has been identified that machine harvests well, is root rot tolerant and raspberry bushy dwarf virus (RBDV) resistant. Yields at WSU Puyallup and in machine harvesting trials have been good. Release of this selection as a new cultivar has been approved by the college cultivar release committee.

Four selections were harvested in 2013 that appeared very promising and have been sent to a propagator for increase for grower trials in 2014. One of these selections was machine harvested in 2013 by a grower in a small planting. The fruit released very easily and the fruit was firm after harvest. The other three selections were machine harvested in machine harvesting trials. One selection was harvested in 2012 and 2013 and looked good in both harvest seasons with good yields. The color is an excellent processing color. The other two selections were harvested for the first time in 2013. The fruit released very easily and the fruit was very firm. Both of the last two selections were made as the seedlings were being machine harvested at a grower field.

Crosses/seedlings/selections. 72 crosses were made for cultivar development with 69 crosses with at least one parent testing RBDV resistant. Ten additional crosses were made for germplasm development or studies. Selections were only made in a portion of the 2011 seedling field because of poor growth. This planting will be kept for an additional year and the seedlings evaluated a second year. Twenty-nine selections were made for cultivar development and 12 for germplasm development. Thirty-seven of the 41 selections have at least one RBDV resistant parent. **WSU 1507** was a parent of 31 of the 41 selections.

Machine Harvesting Trials. A new machine harvesting trial was planted in Lynden with 67 WSU selections, 25 BC selections, 5 ORUS selections and **Meeker**, **Willamette** and **Lewis** for reference. There were 19 selections in the 2010 and 2011 plantings that machine harvested well and showed potential. All of these selections were either planted in new machine harvesting plots in 2013 or will be propagated for planting in 2014 for further evaluation. The most promising of these selections will be planted with a cooperating grower for additional evaluation.

Selection Trial Puyallup. The 2010 and 2011 replicated plantings at Puyallup were hand harvested in 2013. In the 2011 planting, **Rudi** was the earliest ripening with a midpoint of harvest of June 29, two days before **Willamette** (Table 1). **Ukee** and **WSU 1738** were the latest ripening three days after **Meeker**. The 2011 planting will be harvested again in 2014. The replicated planting established in 2010 was hand harvested for the second time in 2013 (Table 2). **WSU 2115** was the earliest ripening with a midpoint of harvest that was 4 days ahead of **Willamette** and 17 days earlier than **Meeker**. **WSU 2115** had large fruit in both harvest seasons and was among the firmest. **WSU 1439** had the second greatest yield in 2013 with large, firm fruit. The midpoint of harvest for **WSU 1439** was a week later than **Meeker**. In 2013, **WSU 2029** was harvested for the first time on July 15 and the last harvest was August 20 with a midpoint of harvest of July 31. In comparison, **Meeker** had a midpoint of harvest of July 13.

Publications:

Machine Harvesting Open House. Lynden, WA. July 11, 2013.

Finn, C.E., P.P. Moore, B.C. Strik, B.M. Yorgey and R.R. Martin. 2013. 'Vintage' Red Raspberry. HortScience 48:1184-188.

Villamor, R.R., Daniels, C.H., Moore, P.P. and Ross, C.F. Preference Mapping of Frozen and Fresh Raspberries. 2013. Journal of Food Science 78: S911-S919.

Ward, J.A., J. Bhangoo, F. Fernandez-Fernandez, P. Moore, J.D. Swanson, R. Viola, R. Velasco, N. Bassil, C.A. Weber and D.J. Sargent. 2013. Saturated linkage map construction in *Rubus idaeus* using genotyping by sequencing and genome-independent imputation. BMC Genomics 14:2 <http://www.biomedcentral.com/1471-2164/14/2>

Moore, P.P., & Hoashi-Erhardt, W.K. 2012. Comparative Response of Red Raspberry Cultivars to Phytophthora Root Rot. Journal of the American Pomological Society. 66, 28-33.

Table 1. 2013 harvest of 2011 planted raspberries, Puyallup, WA

	Yield (t/a)		Fruit weight (g)		Fruit rot (%)		Fruit firmness (g)		Midpoint of harvest	
Meeker	10.5	a	3.6	de	4.9	b	159	a-c	7/12	a-e
BC3-14-12	10.4	a	4.7	ab	11.1	a	165	ab	7/12	a-e
Rudi	10.1	ab	4.0	cd	9.0	ab	160	a-c	6/29	f
WSU 1660	9.7	ab	3.2	e	4.5	b	159	a-c	7/10	c-e
Ukee	9.0	a-c	3.5	de	3.7	b	165	ab	7/15	a
C Bounty	8.8	a-c	3.5	de	7.1	ab	149	c	7/9	de
WSU 1948	8.7	a-c	3.2	e	3.9	b	156	a-c	7/14	a-c
WSU 1738	8.2	a-c	3.5	de	6.9	ab	160	a-c	7/15	ab
WSU 1912	7.3	a-c	3.0	e	3.6	b	162	a-c	7/10	c-e
Willamette	6.5	bc	3.6	de	8.1	ab	152	bc	7/2	f
BC96-22R-55	6.5	bc	4.2	bc	3.3	b	167	a	7/13	a-d
WSU 1792	6.5	bc	5.3	a	12.8	a	168	a	7/11	b-e
WSU 1447	5.2	c	3.9	cd	8.9	ab	159	a-c	7/8	e
	8.2		3.8		6.7		160		7/10	

Three replications of 3 plant plots.

Means followed by the same letter within a column are not significantly different using Tukey's Studentized Range (HSD) Test, P=0.05.

Table 2. 2012-13 harvest of 2010 planted raspberries, Puyallup, WA.

Selection	Yield (t/a)			Fruit weight (g)		Fruit rot (%)		Fruit firmness (g)		Midpoint of harvest	
	2013	2012	Total	2013	2012	2013	2012	2013	2012	2013	2012
Meeker	11.0 a	12.8 ab	23.8 a	3.4 cd	3.6 cd	4.6 cd	6.5 a	160 ab	62 bc	7/13 cd	7/18 b
WSU 1439	10.3 a	-	-	6.4 a	-	4.9 cd	-	171 a	-	7/20 b	-
WSU 2029	10.3 a	-	-	4.0 bc	-	1.3 d	-	147 bc	-	7/31 a	-
C Bounty	10.2 a	10.1 a-c	20.3 ab	3.8 bc	4.0 cd	5.5 c	7.9 a	156 ab	60 c	7/11 cd	7/13 c
WSU 1511	9.8 a	8.8 bc	18.5 ab	4.0 bc	4.2 c	6.9 bc	6.4 a	155 ab	62 bc	7/15 bc	7/20 ab
WSU 1741	9.5 a	10.3 a-c	19.7 ab	2.9 d	3.3 d	5.5 c	5.6 a	155 ab	69 bc	7/9 cd	7/12 c
WSU 2115	7.6 ab	14.4 a	22.0 a	5.7 a	6.6 a	10.3 ab	6.6 a	171 a	90 a	6/26 e	7/6 d
WSU 1750	7.6 ab	7.1 c	14.7 bc	3.5 cd	3.8 cd	3.5 cd	6.4 a	164 ab	63 bc	7/15 bc	7/22 a
WSU 1626	5.1 bc	6.7 c	11.9 c	4.4 b	5.1 b	5.8 c	5.2 a	158 ab	70 bc	6/30 e	7/11 c
Willamette	2.0 c	8.8 bc	10.8 c	3.0 d	3.8 cd	11.2 a	6.3 a	134 c	75 b	6/30 e	7/5 d
	8.3	9.9	17.7	4.1	4.3	5.9	6.4	157	69	7/11	7/13

Three replications of 3 plant plots.

Means followed by the same letter within a column are not significantly different using Tukey's Studentized Range (HSD) Test, P=0.05.

2014 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

New Project Proposal

Proposed Duration: 1 year

Project Title: Tracking the movement of RBDV in Red Raspberry from pollination to systemic infection using Real-time RT PCR.

PI: Patrick Moore, PhD

Organization: Washington State
University

Title: Scientist

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Address: 2606 W. Pioneer Way

City/State/Zip: Puyallup, WA 98372

Co-PI: Kara Lanning

Organization: Washington State University

Title: PhD Candidate

Phone: 253-445-4653

Email: kara.lanning@email.wsu.edu

Address: 2606 W. Pioneer Way

City/State/Zip: Puyallup, WA 98372

Cooperator: Bob Martin, PhD USDA-ARS, Corvallis, OR.

Year Initiated 2014 **Current Year** 2014 **Terminating Year** 2014

Total Project Request: Year 1 \$8,750.00

Other funding sources: *Patents*

Notes: Kara Lanning's doctoral program is currently funded by patent royalties which covers her tuition, graduate student stipend and general costs associated with her research.

Description:

Raspberry bushy dwarf virus (RBDV) is a highly prevalent pollen-borne virus that significantly impacts the Pacific Northwest. While the majority of RBDV research is directed towards breeding immune cultivars, little work has been done to understand how the virus interacts with susceptible cultivars and with other viruses. The main objective of this project is to replicate work that was done at WSU Puyallup in Spring 2013, confirming the observed time course of RBDV infection from pollination to systemic infection. Confirmation of our preliminary results will allow us to assess the possibility of using early pruning as a method to limit field spread of the disease. The results of this work will be distributed through academic publications and in grower meetings.

Justification and Background:

Raspberry bushy dwarf virus (RBDV) is the most prevalent virus that affects *Rubus* species worldwide (Martin, 2002). RBDV is a pollen-borne virus that is commonly associated with drupelet abortion and has been reported to cause yield reductions up to 40%. RBDV can also result in leaf yellowing, decreased cane number and vigor (Martin, 2002). As RBDV is pollen-borne, the time frame of possible infection is limited to the time of bloom in any given year.

It was recently discovered that RBDV and *Raspberry leaf mottle virus* (RLMV) have a synergistic relationship and together are the cause of raspberry crumbly fruit disease (Quito, 2012). Both viruses are highly prevalent in Washington State. In a mixed infection, RLMV increases the RBDV concentration by 400 fold, therefore significantly increasing disease severity compared to the symptomatology observed in a single RBDV infection.

Current research efforts have been primarily directed towards breeding for RBDV resistance. Little work has been done to elucidate the plant pathogen interactions of RBDV and its host, along with other strategies to possibly curtail field spread. To address this gap of knowledge, a pollination experiment was designed and conducted at WSU Puyallup in 2013 to identify the time course of RBDV infection and to assess the possibility of early pruning as a strategy to decrease field spread of RBDV.

For our preliminary work, susceptible and resistant cultivars were placed in a screenhouse together with RBDV infected plants that served as an inoculum source for pollen transmission. At time of bloom, a bumblebee hive was placed in the screenhouse to mimic natural pollination. Two weeks post-bloom, both the hive and the infected plants were removed from the screenhouse. At specific time points post-bloom plant tissues were destructively collected and analyzed for RBDV using real time RT-PCR; an efficient, quantitative detection method that is highly sensitive for RBDV. Preliminary results indicate that RBDV moves very quickly through the red raspberry plant. Further work is needed to confirm the observed time course of disease development and to rule out any possibility of contamination in the samples.

Relationship to WRRRC Research Priority(s):

Viruses/crumbly fruit is listed as a #2 priority for the Washington Red Raspberry Commission.

Objectives:

Objective 1: Track the time course of RBDV from pollination to systemic infection in Red Raspberry.

Objective 2: Ascertain if early pruning could be an effective strategy to limit the field spread of RBDV.

Procedures:

The anticipated length of this project is one year.

Disease free ‘Meeker’ and ‘Willamette’ plants will be kept and maintained in a screenhouse. In spring 2014, infected plants will be placed in the screenhouse to serve as a source of RBDV infected pollen the transmission studies. Experimental plants will be randomly placed between the infected plants. Additional controls will be incorporated in the experimental design to rule out pollen contamination on the plant surfaces. All experimental plants will be monitored for the onset of bloom. At time of bloom, a bumblebee hive will be placed in the screenhouse. Any blossoms that bloom prior to the introduction of the hive will be removed. The hive and inoculum plants will be removed from the screenhouse two weeks post-bloom. At this time, plants will be randomly selected for destructive sampling.

Plant tissues that will be collected include: the receptacle, peduncle, mid fruiting lateral, mid florican and the base of the florican. All tissues will be processed carefully to limit any possible cross contamination with infective RBDV pollen or material. Total RNA will be extracted from all collected samples. All RNA extractions will then be evaluated for RBDV using real time RT-PCR.

Sampling will continue weekly until RBDV is detected in all tissue types. Once RBDV is detected at the base of the cane, primocane tissues will be collected and evaluated for the presence of RBDV. All plants will be maintained and evaluated for RBDV infection through the end of the 2014 fruiting season. All plants will be tested for RBDV infection by ELISA in spring 2015.

Anticipated Benefits and Information Transfer:

This research will directly benefit growers by identifying the time course of RBDV infection from time of bloom to systemic infection. It will also add to information on the interaction between raspberry plants and the virus. The results of our study will allow us to determine if early pruning could help circumvent the field spread of RBDV by removing fruiting canes before the virus moves into the root system.

The results of this study will be published in an academic journal and presented at grower meetings.

References:

R.R Martin. 2002. Virus Diseases of *Rubus* and Strategies for Their Control. Acta Hort 585:265-270.

D. F. Quito-Avila, R.R Martin. 2012. Real-time RT-PCR for detection of *Raspberry bushy dwarf virus*, *Raspberry leaf mottle virus* and characterizing synergistic interaction in mixed infections. J. Virol. Methods 179:38-44.

Budget:

	2014
Operations (goods & services) ¹	\$8,750.00
Total	\$8,750.00

Budget Justification

¹The requested funds will cover the cost of molecular laboratory reagents and materials, including the supplies for RNA extraction, real-time RT-PCR reactions, disposable laboratory plastics, and genetic confirmation of test plants and amplicons of RT-PCR products.

2014 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

Project Proposal

Proposed Duration: (1year)

Project Title: Evaluating the Feasibility of Coordinated Regional on-farm Trials of Advanced Raspberry Selections—Third Year

PI:

Tom Peerbolt

Organization: Peerbolt Crop Management

Title: Co-owner and Senior Researcher

Phone: 503-289-7287

Email: tom@peerbolt.com

Address: 5261 North Princeton St.

City/State/Zip: Portland, OR 97203

Co PIs

Chad E. Finn – USDA-ARS-HCRU, Corvallis, OR

Patrick Moore – Washington State University, Puyallup, WA

Julie Enfield – Northwest Plants/Enfield Farms, Lynden, WA

Year Initiated 2012 **Current Year** 2013 **Terminating Year** 2014

Total Project Request: \$11,200

Other funding sources:

Agency Name: Northwest Center for Small Fruit Research

Amt. Requested/Awarded: \$32,554

Notes: This is a similar project that will allow us to also test black raspberries and fresh market caneberries of all types and disseminate testing information over a larger geographical area and to a larger audience.

Description: Organize and put in place a pilot network of regional on-farm grower trials for evaluating raspberry advanced selections issuing from the USDA-ARS/OSU breeding program in Corvallis and the WSU breeding program in Puyallup. The goal is to combine public and private resources in ways that would accelerate the commercialization of our genetic resources. This request is for the third year of this project. Over the first two years the grower/cooperator network has been developed; one & two year old plantings have been established; the infrastructure has been created for collecting, recording and distributing trial information. The 2014 season will be the third season funding will support the first full implementation of the project.

Justification and Background:

The northwest raspberry breeding programs have been a cornerstone of the industry's success. Their ability to produce cultivars of commercial value is crucial to continued success. Global competition is increasing and public funding for these programs at our land grant institutions is under increasing budget constraints.

This program could strengthen the breeding programs by:

- Giving support to the existing research station based field trials by adding a strong, natural link that would improve the present method of sending advanced selections on to the propagators to be multiplied for grower trials.
- Decreasing the time needed to evaluate the commercial potential of selections.
- Increasing the industry-wide knowledge of new releases potential before they are released.
- Increasing the breeding programs and industry's ability to effectively manage our genetic resources using intellectual property tools (e.g. plant patenting and plant breeders rights) by having information on a cultivar's potential well in advance of its release and patenting.

This program could support the growers by:

- Improving the quality and quantity of information they have for business planning.
- Currently, advanced selections are tested and new cultivars are released based on limited knowledge of their overall commercial potential and viability under various northwest growing conditions. This system forces the grower to either make a decision to plant a new cultivar based on inadequate data, or delay a decision for years until an adequate track record has reduced the risk level.
- Providing new communication links between the growers, nurseries and plant breeders.
- Allowing growers to actively participate in selection evaluations within established protocols and without needing to invest their own resources to pay for the plants and all the planting costs.

This program could strengthen the propagators and wholesale nurseries by:

- Improving their decision-making methods and reducing their risk.
- The present system puts the propagators/wholesale nurseries in the position of guessing how many of which selections and new releases to produce. This has led to economic losses to the nurseries caused by over and/or under production of material. It has created a disincentive for the wholesale nurseries to make available or test new products.
- Providing them with objective evaluations of new material under a variety of growing conditions to pass on to potential customers.

Relationship to WRRC Research Priority(s): Priority 1 Develop cultivars that are summer bearing, high yielding, winter hardy, machine-harvestable, disease resistant, virus resistant and have superior processed fruit quality

Objectives:

- Organize and put in place a pilot network of regional on-farm grower trials for evaluating raspberry advanced selections issuing from the USDA-ARS/OSU breeding program in Corvallis and the WSU breeding program in Puyallup.
 - Place trials on farms located in a variety of regional growing conditions. This network would connect growers, commodity commission contractors, wholesale nursery propagators, public small fruit breeders, and small fruit researchers for the purposes of:
 - 1) Improving the quality and breadth of information available on advanced selections,
 - 2) Improving the efficiency of this information's distribution to the grower/processor base.
- The overall goal of the project is to combine public and private resources in ways that would accelerate the commercialization of our genetic resources. All objectives are included in 2013.

Procedures:**Review of initial project guidelines**

- Tissue culture plants will be used.
- Maximum of 5 red raspberry selections (processed, but could include some fresh selections).
- Minimum of 3 grower sites per selection per year.
- Site guidelines would be representative of the major northwest growing regions including:
 - At least two sites in Northern Washington and one in SW Washington or Oregon.
- Maximum number of plants per selection per trial of machine harvested raspberries would be 1000 plants to produce enough fruit for processing potential. This could be considerably less depending on site and consensus of participants as to the size trial needed.
- Minimum number of plants could be as low as 10 for a fresh market or hand-picked trial.

Year three (2014) procedures

- Establish new plantings following procedures similar to those used in years one and two (2012 & 2013).
- Spring evaluation will be made of plant health/winter damage of the year one and two plantings.
- Evaluations will be made of the first full harvest of the year one (2012) plantings.
- Evaluations will be made of the initial (baby) harvest of the year two (2013) plantings.
- Evaluations will be made in the fall to determination whether to continue for another year's data of the year one plantings.
- Advisory group will be communicating as needed to coordinate activities.
- Administrator will be giving periodic updates to participants. Disseminating and archiving information as needed.

Grower/cooperator arrangements

- Testing agreements would be created and approved by WSU (or WSURF) and by USDA.
- Growers would sign testing agreements that would include: on-site visits by other growers and researchers (arranged and agreed to in advanced); participation in the evaluation process; and a testing agreement which includes a prohibition of any on-farm propagation of advanced selections.

Anticipated Benefits and Information Transfer:

- The anticipated benefit to the breeding program, growers, propagators and wholesale nurseries are detailed above in the Justification section of this proposal.
- The results be transferred to users by the Administrator will be giving periodic updates to Washington red raspberry growers and the industry. Disseminating and archiving information as needed through meeting presentations, newsletters and production of summary 'fact sheets'.

References: none.

Budget:

	2013	2014	2015
Salaries ^{1/}	\$5,000	\$	\$
Travel ^{2/}	\$2,200	\$	\$
Outreach ^{3/}	\$1,500	\$	\$
Other (Propagator payments) ^{4/}	\$2,500	\$	\$
Total	\$11,200	\$	\$

Budget Justification

^{1/}Specify type of position and FTE. Administrator of project at 10% FTE

^{2/}Provide brief justification for travel requested. Travel and related expenses to meet with growers and propagators, deliver plants, check plantings, attend meetings and workshops.

^{3/}Outreach will be accomplished by giving periodic updates to Washington red raspberry growers and the industry. Disseminating and archiving information as needed through meeting presentations, newsletters and production of summary 'fact sheets'

^{4/}These funds will be paid out by the Commission from invoices from the propagators.

Washington Red Raspberry Commission

Progress Report 2013 (second year)

Title: Regional On-farm Trials of Advanced Raspberry Selections

Personnel:

PI: Tom Peerbolt –Peerbolt Crop Management.

Co PIs: Chad Finn – USDA-ARS; Pat Moore – WSU; Julie Enfield – Northwest Plants

Reporting Period: 2013

Accomplishments:

Advanced selections/varieties planted in 2012 & maintained in 2013: WSU 1507, WSU 1912, WSU 1948, Rudi.

Advanced selections/varieties planted in 2013: BC 92-9-15, Lewis.

Participating growers & selections in their trials 2013:

- **Ralph Minaker** 250 Lewis (Whatcom County, Everson area).
- **Rob Dhaliwal** 250 Lewis, 250 BC 92-9-15 (Whatcom County, South Lynden area).
- **Enfield Farms** 250 Lewis (Whatcom County, West Lynden area).
- **Sakuma Farms** 12 Lewis, 50 BC 92-9-15 (Skagit County, Mt. Vernon area).
- **Don Sturm Farm:** 100 Lewis, 100 BC 92-9-15 (North Willamette Valley, Oregon).

Results:

- Over the first two years the grower/cooperator network has been developed; one & two year old plantings have been established; the infrastructure has been created for collecting, recording and distributing trial information. The 2014 season will be the third season funding will support the first full implementation of the project with mature harvests to evaluate of the 2012 plantings, continued observation of one year old plantings (2013) and establishment of new selection plantings in 2014.
- One of the 2012 plantings (Hoffman Farms) was inadvertently tilled under. One of the risks of onfarm trials. The replication of plantings will allow us to collect valid observational data even with the loss of this one trial.
- Initial fruit observations of the 2012 & 2013 plantings were made in July of 2013. Some fruit was bearing available for comparisons of characteristics and grower comments were recorded and will be shared in presentations and newsletters as appropriate.
- The project is on course with the entire infrastructure in place both on farms and in the methods of collecting and distributing information.
- This next season will allow us to fully evaluate the overall efficacy of this project as far as real costs and potential value to the industry.

2014 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

New Project Proposal

Proposed Duration: 1 year

Project Title: Use of a mycoinsecticide targeting novel SWD preimaginal life stages and potential synergism with spinosad

PI: Lynell K. Tanigoshi

Organization: Washington State University

WSU Entomology Professor

(360) 848-6152

tanigosh@wsu.edu

Address: WSU-NWREC, 16650 State Route 536, Mount Vernon, WA 98273-4768

Co-PI: Beverly Gerdeman

Washington State University

WSU Entomology Research Associate

(360) 848-6145

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Year Initiated 2014

Current Year 2014

Terminating Year 2014

Total Project Request: \$10,865

Other Funding Sources: Seeking funding from NARF small fruit and Washington State Commission on Pesticide Registration.

Description: The addition of mycoinsecticides to conventional pesticides has been found to be synergistic and increase susceptibility of insects to the insecticide (Ericsson et al. 2007, Farenhorst et al. 2010, Sharififard et al. 2011). We propose to test this potential synergy against spotted wing drosophila, *Drosophila suzukii* (Matsumura), SWD, in red raspberry with foliar applied tank mixes of Entrust[®] SC, plus mycoinsecticides. In addition a soil-applied mycoinsecticide will test the potential of mycoinsecticides targeting novel SWD preimaginal life stages, the late 3rd instar and puparium. These field experiments will be applied to a late season red raspberry field at WSU NWREC.

Justification and Background:

Washington State is the #1 producer of raspberries in the US. Red Raspberry is highly susceptible to economic damage from SWD, one of the few and most injurious direct pests of raspberries. SWD infested berries are unmarketable and without effective insecticides and methods to control the pest, berry production will continue to decline. There is a desperate need for multiple options that effectively control spotted wing drosophila in both conventional and organic production systems. Entrust is the only OMRI listed product for organic growers with reported residual activity for managing SWD, therefore identifying methods to increase efficacy are a high priority.

The spinosad biopesticide, Entrust[®] SC is classified as IRAC Group 5 insecticide. While laboratory topical bioassays indicate good efficacy, field residual results have been disappointing. In western Washington, raspberry field residuals range from 1 - 7 days, resulting in grower confusion. This confusion has reduced grower confidence in Entrust and contributed to a risky, calendar-spray approach, alternating pyrethroids with organophosphates.

Synergistic interaction is a combination of stressors resulting in a greater effect than expected from cumulative independent exposures. A unique synergy between fungal pathogens and insecticides has been identified. Insects succumb to fungal pathogens when the spores attach to the insect, germinate and grow into the cuticle, taking 3–14 days (Farenhorst et al. 2010). Normally, such an extended lethal time would be too risky for a dangerous pest like SWD but days before they die, insects become “zombies” no longer an economic threat. Tank-mixing fungal pathogens with insecticides diverts the insects’ insecticide detoxifying enzymes toward the fungal toxins, reducing the insect’s immune response and decreasing the lethal time. In Africa, tank-mixing a fungal pathogen renewed efficacy against resistant mosquitoes, indicating potential for resistance reversal (Farenhorst 2010). Biopesticides are exempt from MRL tolerances therefore would not pose any market risk.

SWD is currently managed with foliar adulticides. Third instar larvae drop from berries and pupate in the soil (personal observation) rendering late instar larvae and pupae potentially amenable to attack by soil entomopathogenic fungi. Targeting late instar larval/pupal stages would provide an additional means of control until now ignored. The soil is the only site where three of the four life stages are present. As high tunnel production of red raspberry increases, increased humidity and UV protection will enhance use of mycoinsecticides. The topic of synergists to enhance control of SWD has not been investigated to our knowledge.

Relationship to WRRRC Research Priority: #1 priority, Management Options for Control of SWD.

Objectives:

- Investigate the efficacy of a soil-applied mycoinsecticide against SWD preimaginal stages.
- Investigate efficacy of a foliar tank mix against SWD
- Investigate synergistic activity with tank-mixed spinosad + mycoinsecticide applied as a foliar SWD adulticide.



Fig. 1. Funnel directing the fall of SWD larvae inside a bucket rim trap.

Procedures:

The field trial will take place in a late season 0.17 acre red raspberry field of mixed red/yellow raspberries located at WSU NWREC. Individual plots will extend 30 feet from post to post, separated by a buffer of a single post-length and staggered from row-to-row. Foliar applications will be tank-mixed and applied with a CO2 backpack sprayer and soil applications will be applied in a 2-foot band to the soil beneath the canes on each side of the row, using a single flat-fan nozzle at 40psi. Each replicate will have two funnels and bucket rim traps. These traps consist of a funnel suspended beneath ripe berry clusters 1-3 feet above a bucket rim pushed into the soil to concentrate and confine larvae dropping from the fruit and improve chances of recovery (Fig. 1).

Treatments:

1. Foliar application - Entrust SC + *Metarhizium anisopliae*
2. Foliar application - Field rate of Entrust SC
3. Foliar application of *M. anisopliae*
4. Soil drench application *M. anisopliae*
5. Foliar Application – Entrust SC + *Beauveria. bassiana*
6. Foliar application of *B. bassiana*
7. Soil drench application *B. bassiana*
8. Untreated

Five days following the mycoinsecticide treatment, insect-proof nets will be stretched over each bucket rim for 5 additional days. Emerging adults will be collected every 3 days for two weeks. Captured adults will be placed in drosophila culture vials with standard diet and incubated 7-10 days at high humidity to promote sporulation. After 1 week, a 2 cm layer of soil will be removed from inside the rims and sieved to recover puparia and larvae. All larvae and pupae will be incubated at high humidity in Petri dishes, inside a plastic box with moistened paper towel to promote sporulation, which should be observed within 5 days.

Efficacy of foliar Entrust/mycoinsecticide applications will be determined through weekly random berry samples and insect vacuum counts. Berry samples will be placed into a mesh-covered box and held for adult emergence. Weekly insect vacuum samples of each treatment will be taken and SWD will be separated, counted and incubated encouraging sporulation to determine infection rates.

Anticipated Benefits and information Transfer: This study is anticipated to improve field performance and confidence in the spinosad biopesticide, Entrust SC, for control of SWD, reinstating a valuable IRM management tool. The study is also expected to determine if preimaginal stages could be affected with a soil application of a mycoinsecticide. Specific benefits include:

- Improved field efficacy of spinosad through addition of a mycoinsecticide.
- Reduction in lethal time for mycoinsecticides.
- Prevention of resistance development to spinosad through addition of a mycoinsecticide.
- Evaluation of a novel life stage target in SWD, preimaginal stages, creating a multiple control approach.

Results will be presented at local and regional grower meetings, Whatcom County Ag Monthly Newsletter and on the WSU NWREC Entomology website.

References:

- Ericsson, J., J. Kabaluk, M. Goettel and J. Myers. 2007. Spinosad interacts synergistically with the insect pathogen *Metarhizium anisopliae* against the exotic wireworms *Agriotes lineatus* and *Agriotes obscurus* (Coleoptera: Elateridae). J. Econ. Entomol. 100 (1): 31-38.
- Farenhorst, M., B. Knols, M. Thomas, A. Howard, W. Takken, M. Rowland and R. N'Guessan. 2010. Synergy in efficacy of fungal entomopathogens and permethrin against West African insecticide-resistant *Anopheles gambiae* mosquitoes. PLoS ONE 5(8): e12081. Doi: 10.1371/journal.pone.0012081

Sharififard, M., M. Mossadegh, B. Vazirianzadeh and A. Zarei-Mahmoudabadi. 2011. Interactions between entomopathogenic fungus *Metarhizium anisopliae* and sublethal doses of spinosad for control of house fly, *Musca domestica*. Iran J. Arthropod-Borne Dis. 5(1): 28-36.

Budget:

	2014
Salaries ^{1/}	\$7,645
Time-Slip	\$
Operations (goods & services)	\$200
Travel ^{2/}	\$
Meetings	\$
Other	\$
Equipment ^{3/}	\$
Benefits ^{4/}	\$3,020
Total	\$10,865

Budget Justification

^{1/}1 month salary at 100 FTE for Research Associate \$4,582 and 1 month salary at 100 FTE for Research Technician \$3,063.

^{4/}1 month for Research Associate \$1,677 (36.6%) and 1 month for Research Technician \$1,343 (43.86%).

Current and Pending Support

Name (List PI #1 first)	Supporting Agency and Project #	Total \$ Amount	Effective and Expiration Dates	% of Time Committed	Title of Project
Lynell K. Tanigoshi	NIFA-SCRI/UC Davis	\$214,986.00	2011-2015	20	Biology and Management of Spotted Wing Drosophila on Small and Stone Fruits
Lynell K. Tanigoshi	WA Red Raspberry Commission	\$5,000	2015	5	Management impacts of a unilateral insecticide tactic to control spotted wing drosophila, <i>Drosophila suzukii</i> (Matsumura) and the resurgence of economic populations of spider mites and affects on their phytoseiid mite predators.
Lynell K. Tanigoshi	WA Strawberry Commission	\$4,800	2015	5	Revisiting the chemical management of root weevils and SWD in strawberry.
Lynell K. Tanigoshi	WA Blueberry Commission	\$9,921	2015	5	Spotted wing drosophila management and its impact on MRLs and resistance management.
Lynell K. Tanigoshi	WA St. Comm. Pesticide Reg.	\$9,921	2013	5	Spotted wing drosophila management and its impact on MRLs and resistance management.
Lynell K. Tanigoshi	NW Agr. Res. Foundation	\$21,587	2013	10	Alternative chemistries and application methods for cabbage maggot and other select pests of cabbage seed production.
Wei Yang	OR Blueberry Commission	\$7,366 (WSU)	2013	5	Effects of delivery methods on insecticide efficacy against spotted wing drosophila (<i>Drosophila suzukii</i>) in commercial blueberries.

Washington Red Raspberry Commission Progress Report

Project No.: 13C-3443-5370 (1 year Termination Report)

Title: Management impacts of a unilateral insecticide tactic to control spotted wing drosophila, *Drosophila suzukii* (Matsumura) and the resurgence of economic populations of spider mites and affects on their phytoseiids mite predators

Personnel: **Lynell K. Tanigoshi**, Entomologist¹
Beverly S. Gerdeman, Research Associate, Entomology¹
G. Hollis Spitler, Agricultural Research Technician¹
¹WSU-Mount Vernon NWREC

Accomplishments:

SWD. A persistent calendar spray is critical for managing a pest with cryptic life stages such as SWD, providing coverage as new waves of adults emerge from protected pupation sites in the soil. Due to the short red raspberry season, SWD can be managed effectively with pyrethroid/OP rotational partners. SWD field mortality is observed to rise as the season progresses and multiple sprays overlap. This cumulative layering of dissipating residues at weekly intervals enhances the effect of individual insecticides.

Root weevils. Topical mode of entry of our registered weevicides demonstrated more rapid mortality compared with contact/ingestion-stomach mode of entry. It is common to observe recovery of intoxicated root weevils, however these walking *zombie* weevils cease laying eggs, or lethargic/moribund and certainly don't feed on foliage compared with the untreated check weevils. Growers should not retreat but know your treatments are effective and usually moribund and exposed adults will die within 24 hrs if exposed to the sun.

Spider mites. Although growers should be vigilant against potential spider mite flare-ups from the heavy use of pyrethroids for control of SWD, no reports of sudden mite outbreaks have been observed thus far. Red raspberry growers currently have an effective miticide arsenal with at least 6 different mode of action chemistries.

Results:

SWD. Field and lab efficacy trials indicate the insecticides registered for spotted wing drosophila (SWD) will all provide economic control of adults for 5-8 days between treatment rotations during harvest. Our testing of rotational combinations of 3 different mode of action insecticides (i.e., pyrethroids, OPs, spinosyns) underscored the impact of cumulative exposure of SWD to a season-long calendar spray program. This strict cover spray regimen results in an increase in mortality rates as the season progresses from layer after layer of residues, mainly sequestered on the foliage. Be it the result of synergy, if not, even sublethal levels may stimulate efficacy of subsequent sprays. Lab bioassays of adult females on field aged residues of Danitol showed residual longevity of >90% at 14 days posttreatment. This knowledge of days of residual activity for each effective adulticide will enable a berry grower to plan a sequence of 4-5 season long rotations at 7-8 days based on his market(s) and enhancing the activity of less

residual insecticides such as the OPs and spinosyns with Danitol with his 7 day treatment interval.

Root weevils. Topical exposure of pyrethroids to the clay colored root weevil resulted in mortality within 1 day after exposure (1 DAT). Contact residual and stomach poison activity via feeding on foliage showed long exposure was required for complete mortality. Weevils treated with pyrethroids Asana and Mustang Maxx displayed considerable recovery as well as moribund adults to 6-9 DAT where mortality was 100 to 93% respectively. If these recovered/moribund adults were removed from further field exposure, they would eventually die without have laid eggs as well. In a field environment where the entire field has been treated, the weevils would continue to be exposed by encountering field rates of dried residues or ingested treated foliage so the continued exposure to the residual longevity of these pyrethroids should be enough to provide complete mortality within 6-9 DAT.

Spider mites. Because of the heavy reliance on pyrethroid chemistries, growers are concerned about spider mite population flare-up through reproduction stimulation. To date, there is no evidence for late season mite outbreaks resulting in premature defoliation and reduction of berry quality. We tested 6 different MOA registered acaricides on early season yellow spider mite (YSM) populations that consisted mostly of adults and eggs. At 6, 9 and 14 DAT, all of the acaricides were significantly lower than the untreated check. The acaricide arsenal for spider mites is in good stead should YSM or twospotted spider mite (TSSM) select for resistance or reproductive stimulation due to pyrethroid disruption of the phytoseiid predatory mites, *Neoseiulus fallacis*.

MRL guidelines. The attached caneberry rotational partner table (Table 1) focuses on those registered insecticides with MRLs equal to or greater than the US MRLs, thus providing a list of insecticides with favorable MRLs for the 4 target markets, Japan, Taiwan, S. Korea and China. This list was developed based on grower requests and can be amended on a yearly basis to reflect any changes, however users are urged to check the USDA FAS database prior to application of any chemical <http://www.mridatabase.com/>. A rational list of effective insecticides for use in berries has been lacking. This table has been well received by growers wishing a list that narrows all the registered insecticides down to those with favorable MRLs that still have efficacy against SWD.

Publications

Tanigoshi, L. K., B. S. Gerdeman and G. H. Spitler. 2012. Yellow spider mite control in red raspberry, 2012. *Arthropod Management Tests*, Vol. 37.

Tanigoshi, L. K., B. S. Gerdeman, G. H. Spitler and W. Q. Wang. 2013. Field evaluation of micro-sprinklers to control spotted wing drosophila in blueberry. 72nd Annual Pacific Northwest Insect Management Conference, Portland, OR, 7-8 January 2013, pp. 99-102.

Tanigoshi, L. K., G. H. Spitler and B. S. Gerdeman. 2013. Field efficacy of several labeled and experimental insecticides for spotted wing drosophila control in blueberry. 72nd Annual Pacific Northwest Insect Management Conference, Portland, OR, 7-8 January 2013, pp. 105-107.

Tanigoshi, L. K., G. H. Spitler and B. S. Gerdeman. 2013. Evaluation of several labeled and experimental insecticides for root weevil control in small fruits, 2012. 72nd Annual Pacific Northwest Insect Management Conference, Portland, OR, 7-8 January 2013, 108-109.

Gerdeman, B. S., L. K. Tanigoshi and G. H. Spitler. 2013. Reproductive readiness of spotted wing drosophila. 72nd Annual Pacific Northwest Insect Management Conference, Portland, OR, 7-8 January 2013, pp. 103-104.

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Gerdeman B. S., L. K. Tanigoshi and G. Hollis Spitler. 2013. Illumination: A technique for observing SWD eggs. Whatcom AgMonthly, September.

Gerdeman B. S., L. K. Tanigoshi and G. Hollis Spitler. 2013. 2013 berry rotational partners and select MRLs. Whatcom AgMonthly, September.

Table 1. Caneberry SWD Rotational Partners 6/18/2013

Market	Rotational Partners		MRL	PHI DAYS	REI Hours	Residual Activity	Yearly Maximum Applications	Retreatment Interval Days	Comments	
Japan	3A Pyrethroid Pyrethrins	Pyganic pyrethrins	US 1 Japan 1	0	12	0			Virtually no residual. Kills only by direct contact with insects.	Japan has 2 highly effective classes of insecticides available for rotation – pyrethroids and an organophosphate. Spinosyns provide a third rotational partner. While 3 neonicotinoids are an option, they are poor adulticides. This list contains a reasonable array of rotational options making Japan the most tolerant target market for raspberry exports.
		Brigade 2EC bifenthrin	US 1 Japan 1	3	12	5-7	5	7	May stimulate mite populations.	
		Asana XL esfenvalerate	US 1 Japan 1	7	12	5-7	4		7 day PHI makes Asana difficult to use. Don't apply Asana within 7 days of pollination.	
	1B Organophosphate	malathion	US 8 Japan 8	1	12	3-5	3	5	An effective chemistry for SWD.	
	5 Spinosyns	Entrust 2SC, Success spinosad	US 0.7 Japan 0.7	1	4	~1	6	5	Good topical performance but minimal foliar activity.	
	4A Neonicotinoid	Assail 30SG acetamiprid	US 1.6 Japan 1.6	1	12	1-3	5	7	Neonics are poor adulticides but they suppress oviposition and an application can simultaneously control aphids.	
		Provado 1.6F imidacloprid	US 2.5 Japan 4	3	12	1-3	5	7		
Actara thiamethoxam		US 0.35 Japan 0.5	3	12	1-3	3	7			
Korea	3A Pyrethroid	Mustang Max zeta-cypermethrin	US 0.8 Korea 2	1	12	5-7	6	7	Pyrethroids are highly effective against SWD but Brigade can result in high mite populations.	Mustang Max is a highly effective pyrethroid but the only rotational partner is a slow-acting, poor adulticide, neonicotinoid, thiamethoxam.
		Brigade 2EC bifenthrin	US 1 Korea 1	3	12	5-7	5	7		
		Pyganic pyrethrins	US 1 Korea 1	0	12					
	4A Neonicotinoid	Actara thiamethoxam	US 0.35 Korea 1	3	12	1-3	3	7	Neonics are poor adulticides but they suppress oviposition and an application can simultaneously control aphids.	
Taiwan	3A Pyrethroid	Mustang Max zeta-cypermethrin	US .8 Taiwan 2	1	12	5-7	6	7	Excellent efficacy for SWD.	Pyrethroids are highly effective against SWD especially Mustang Max but Taiwan has no alternate mode of action for rotation!
		Brigade 2 EC bifenthrin	US 1 Taiwan 1	3	12	5-7	5	7	May stimulate mite populations.	
		Asana XL esfenvalerate	US 1 Taiwan 1	7	12	5-7	4		7 day PHI makes Asana difficult to use. Don't apply Asana within 7 days of pollination.	
China	4A Neonicotinoid	Assail 30SG acetamiprid	US 1.6 China 2	1	12	1-3	5	7	Neonics are poor adulticides but they suppress oviposition and an application can simultaneously control aphids.	Most unlikely target market for US with current list of registered chemicals for SWD control. Highly unlikely a program with a single slow-acting adulticide will be successful in preventing infestation.

This is a list of insecticides registered on caneberries for SWD with US tolerances = or < those for Japan, Korea, Taiwan and China.

MRLs are subject to change. Always check the USDA FAS MRL database prior to use. (<http://www.mrlidatabase.com/>).

Residual activity is based on SWD % mortality after 24 hours on field-aged leaves brought to laboratory.

Proper resistance management requires rotating between mode-of-action classes as much as possible.

Questions and comments, contact: Beverly S. Gerdeman, bgerdeman@wsu.edu or Lynell K. Tanigoshi, tanigosh@wsu.edu

**2014 WASHINGTON RED RASPBERRY COMMISSION
RESEARCH PROPOSAL**

New Project Proposal

Proposed Duration: 2 years

Project Title: Quantifying Root Weevil Impacts and Post Treatment Root Recovery

PI: Pete W. Jacoby, Plant Ecologist

Unit: WSU Crop & Soil Sciences – Pullman

Telephone: 509-335-3495

Email: jacoby@wsu.edu

Co-PI: Lynell Tanigoshi, Entomologist

Unit: WSU Mount Vernon - NWREC

Telephone: 360-848-6152

Email: tanigosh@wsu.edu

Cooperator(s): Kevin Berendsen, Enfield Farms, Linden, WA

Year Initiated ____ **Current Year** 2014 **Terminating Year** 2015

Total Project Request: **Year 1:** \$ 13,918 **Year 2:** \$ 12,048

Other funding sources: Washington Commission for Pesticide Commission (pending)

Description: Root weevils continue to be a perennial problem for raspberry producers in northwestern Washington and in other production areas in the northern U.S. and southern Canada. The most damaging stage of the life cycle of this pest occurs beneath the soil surface as weevils develop from eggs into larvae prior to emerging as adults. We will introduce new technologies to enable early detection of root weevil larvae and to relate levels of root damage to overall plant productivity in a field research setting.

Justification and Background: Collectively, a group of 4 or more species of weevils in the genus *Otiorhynchus* comprises one of the most serious pests of raspberries, strawberries, and nursery stock in the Pacific Northwest. While much has been learned about the life cycles of these weevil species, early detection remains difficult and significant damage may occur in the early life cycle when larvae are feeding on roots of their host plants (Rosenstiel 1955; Tanigoshi & Bergen 2002). The larval stages of the root weevil can weaken newly established plantings, reducing production and impacting economic return for growers. Current management practices include foliar and drenching sprays of pesticides after adult weevils have emerged from the soil. While these treatments have been shown to reduce adult populations and reduce egg laying that could impact the next season's production, they have little or no impact on fall emerging larvae that will feed on roots until their adult emergence in the next spring.

This project will determine the feasibility of using a device known as a mini-rhizotron to examine root weevil development, their feeding behavior and impacts on root health. This root scanner may also allow estimates in efficacy of insecticide drenches in reducing overwintering weevil larval populations and root recovery. The mini-rhizotron is a modification from earlier forms of rhizotrons that were constructed by excavating a pit, placing a clear sheet of hard plastic against the soil surface, sheltering the pit from light, and periodically tracing the root growth of targeted plants on clear acetate sheets. The mini-rhizotron involves coring a 3-inch hole at an

angle of 30-45 degrees from vertical near or below a plant, and inserting a hard clear acrylic observation tube that is left in place for repeated observations of root growth. Miniaturization of electronic components has produced a small camera system that can be placed inside the observation tube and lowered incrementally to obtain digital 360 degree photographs at desired depths (Caldwell and Virginia 1989; Vamerli et al. 2012). Software allows these photographs to yield total root length data and to statistically analyze differences between periodic observations, depths and treatments (CID Bio-Sciences, Inc. www.cid-inc.com). We are unaware of this rhizotron technique being used to detect populations of soil-borne insects, but believe that, because of the size and coloration of root weevil larvae, this technique could provide detection and quantification of larval populations early in their development. We also believe this technique could help assess degree of pesticide efficacy, plant root damage, and rate of root recovery after pesticide drench applications to the soil surface.

Relationship to WRRC Research Priority(s): This project will promote a better understanding of soil ecology as an integral part of the rhizosphere and its effect on plant health and crop yield. It will advance a better understanding of plant responses to root weevil feeding injury and post-treatment root responses to larval control tactics.

Objectives: The overall objective of this project is to evaluate new technologies in monitoring and predicting severity of root weevil infestation in the larval stage. We will: (1) evaluate the use of mini-rhizotrons in estimating root weevil populations in the rooting zone of the soil; (2) monitor *in situ* impact of drench treatments on root weevil larvae; and (3) monitor rate of post-treatment root recovery of plants damaged by larval feeding. Additionally, we plan to: (4) link recovery of the above ground plant to rate of root recovery by employing a field portable infrared gas analyzer (ADC Bio-Scientific, Ltd. <http://www.adc.co.uk>) to determine photosynthesis rate between treated and non-treated plants in a field setting.

Procedures: Site locations will be established in collaboration with a private commercial grower near Linden, WA in raspberry fields with suspected populations of overwintering root weevil life stages. We will verify the presence or absence of pre-treatment root weevil larvae by obtaining equal size soil cores in 15 cm (6 inch) increments to a depth of 60 cm to quantify existing population levels. Some of the cores will receive a mini-rhizotron tube to obtain periodic digital photos to estimate root length and larvae densities from pre-treatment through post-treatment intervals. Randomized treatment plots, consisting of 5 adjacent plants each will receive various pesticide treatments. Core samples and rhizotron placement will be in the vicinity or beneath the central plant in each designated plot. The first treatments will consist of a basal drench spray(s) at pre-bloom (March-April) of the pyrethroids Asana and Brigade and systemic neonicotinoid Admire Pro for teneral adult weevils just below the soil surface around the base of the plant and maturing instar larvae feeding on feeder roots. Tank mixes of Admire with Asana and Brigade will also be evaluated at pre-bloom. Subsequent pre-emergence applications will be made if larval suppression has not been affected. November soil drenches will be applied to known infestations to evaluate insecticide efficacy for early instars that will remain in the soil until adult emergence in late May-June. Multiple untreated plants will be compared with larval infested plants to quantify root feeding injury with economic injury levels (i.e., larval root weevil feeding-days). Treatments will be replicated at least 3 times for all treatment dates.

Pre-treatment soil cores will be taken from the field locations to laboratory sites for separation of soil, plant roots, and larvae. Data will be separated into 15 cm (6 inch) sections to determine differences by soil depth. Measurement of photosynthesis rate will be taken on designated plants in each treatment plot to determine if high levels of root damage results in lowered levels of plant activity, as indicated by plant photosynthesis in the leaves. These measures will be made at intervals of 2-4 weeks during the growing season. One set of sensors with data logger (Decagon Devices, Inc. www.decagon.com) will be established in near proximity to the experimental plots to obtain a record of soil moisture and temperature. This data logger will have the capacity to be downloaded remotely, allowing periodic monitoring and data retrieval, conserving travel to site.

Anticipated Benefits and Information Transfer: An immediate output from this project will be the assessment of potential use of rhizotrons for use in estimating below-ground populations of visible insect larvae. A positive result could aid in future research investigations and to evaluate various treatment effects for earlier adoption by growers. Another immediate output will be a report to the WRRC on the benefits for accurately timed pesticide treatments for effecting reductions of root weevils and minimizing resulting damage to the roots of high value plantings such as red raspberries. An intermediate outcome will be presentations and publications reporting the results of this study in order to stimulate the use of instrumentation to advancement of precision pesticide-based management for growers and crop consultants by being able to monitor weevil development with improved accuracy. Another intermediate outcome would be to be the ability to relate levels of root damage to subsequent loss of plant and fruit productivity. A long-range impact of this research could be more effective timing and types of control measures and products to prevent high levels of plant damage from root weevil larvae.

References:

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Budget:	2014	2015	2016
Salaries	\$ 3,135	\$ 3,260	
Time-Slip	1,250	2,000	
Operations (goods & services)	4,650	1,000	
Travel	3,500	3,500	
Meetings	-0-	-0-	
Other	-0-	850	
Benefits	1,356	1,410	
<hr/>			
Totals	\$ 13,891	\$ 12,020	

Budget Justification:

Salary -1 month at 1 FTE Research Technician (Entomology) at Mount Vernon, \$3,135. Second year salary is estimated to increase by 4% (\$3,260)

Time Slip – WSU student labor will be employed at both Mount Vernon and Pullman to assist with installation of the field research and to reduce data from rhizotron digital photos and separate roots and weevil larvae from soil samples (no benefits required)

Operations (goods and services) – Materials required to establish this field research include the following:

	No.	Unit Price	\$
Mini-rhizotron access tubes (3 tubes x 4 trts =12)		@ \$150 ea.	= 1,800
Sensors (soil temp and moisture)	5	@ 275 ea.	= 1,375
Data logger	1	@ 975 ea.	= 975
Miscellaneous (expendable supplies)			<u>500</u>
Year 1 Total =			\$ 4,650

Travel – Jacoby – ca 5 trips (3-day trips-Pullman-Lynden-Pullman) = 3,500
 [Mileage each trip – ca 785 x \$0.51/mi = \$400]
 [Lodging and meals each trip – ca 3 days @ \$100/day = \$300]

Meetings – no funds from this project will be used for meetings, except on the request of the WRRC

Other – Publication expenses (estimated) \$ 850

Equipment – all major equipment, including rhizotron camera system, computers and software, as well as a hydraulic soil coring machine and photosynthesis porometer with infrared gas analyzer are available through Dr. Jacoby’s project on rhizosphere plant ecology – these will need to be transported to the study site, as they are being used in other research projects located in the central and southern Columbia basin

Benefits – Year 1 -1 month for Research Technician @ 43.25% = \$1,356
 Year 2 “ “ “ “ “ “ = 1,410

Pete W. Jacoby

Professor / Plant Ecologist

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Education

Ph.D. Range Ecology, University of Wyoming, 1971
M.S. Rangeland Remediation, University of Wyoming, 1968
B.S. Rangeland and Forestry Sciences, Texas A&M University 1966

Professional Experience

Specialist, Texas Agricultural Extension Service, Fort Stockton, TX 1970-73
Asst. Prof./ Extension Specialist, Univ. Arizona, Tucson, AZ 1974-76
Assoc. Prof./ Professor, Texas Agricultural Experiment Sta., Vernon, TX 1976-93
Director, Univ. Nebraska Res. & Ext. Center, North Platte, NE 1993-97
Assoc. Dean, WSU CAHNRS, Pullman, WA 1997-2013
Professor, WSU Crop & Soil Sciences Dept., Pullman, WA 2013-present

Research Emphasis

Ecophysiology of woody perennial plants with focus on root dynamics in the rhizosphere

Extension Emphasis

Weed ecology in high value irrigated crops

Teaching Emphasis

World Agricultural Systems
M.S. in Agriculture program

Selected Publications

- Ansley, R.J., T.W. Boutton, and **P.W. Jacoby**. 2007. *Mesquite root distribution and water use efficiency in response to long-term soil moisture manipulations*. Pp. 96-103. In: Sosebee, R.E.; Britton, C.M.; McArthur, E.D.; Kitchen, S.G., comp. 2007. *Proceedings: Shrubland dynamics – fire and water*; 2004 August 10-12; Lubbock, TX. Proceedings RMRS-P-47. Fort Collins, CO; U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 173 p.
- Ansley, R.J., W.E. Pinchak, W.R. Teague, B.A. Kramp, D.L. Jones, and **P.W. Jacoby**. 2004. Long-term grass yields following chemical control of honey mesquite. *J. Range Manage.* 57: 49-57.
- Cuomo, G.J., R.J. Ansley, **P.W. Jacoby** and R.E. Sosebee. 1992. Honey mesquite transpiration along a vertical site gradient. *J. Range Manage.* 45:334-338.
- Ansley, R.J., **P.W. Jacoby**, C.H. Meadors and B.K. Lawrence. 1992. Soil and leaf water relations of differentially moisture-stressed honey mesquite (*Prosopis glandulosa* Torr.) *J. Arid Environments.* 22:147-159.
- Ansley, R.J., **P.W. Jacoby** and R. Hicks. 1991. Leaf and whole plant transpiration in honey mesquite following severing of lateral roots. *J. Range Manage.* 44:577-583.
- Jacoby, P.W.** and R.J. Ansley. 1991. *Mesquite: classification, distribution, ecology and control*. Chapt. 36, pp. 366-378. In: L.F. James, J.O. Evans, M.H. Ralphs and R.D. Child (eds.) *Noxious Range Weeds*. Westview Press Inc., Boulder, CO. 466 p.
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- Ansley, R.J., **P.W. Jacoby** and B.K. Lawrence. 1989. *Influence of stress history on water use patterns on honey mesquite*. p. 75-82. In: A. Wallace, E.D. McArthur and M.R. Haferkamp (eds.). *Proc. Symp. on Shrub Ecophysiology and Biotechnology*. Logan, UT. U.S. Dept. Agric., For. Serv. Gen. Techn. Rep. INT-245. 183 p.
- Jacoby, P.W.**, R.J. Ansley and B.K. Lawrence. 1988. Design of rain shelters for studying water relations of rangeland shrubs. *J. Range Manage.* 41:83-85.
- Ansley, R.J., **P.W. Jacoby** and B.K. Lawrence. 1988. Root containerization for physiological studies of shrubs and trees on rangeland. *J. Range Manage.* 41:90-91.
- Heitschmidt, R.K., R.J. Ansley, S.L. Dowhower, **P.W. Jacoby** and D.L. Price. 1988. Some observations from the excavation of honey mesquite root systems. *J. Range Manage.* 41:227-231.

2014 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

New Project Proposal

Proposed Duration: (1 year)

Project Title: Determining the ability of Spotted-Wing Drosophila to overwinter in weedy blackberry.

PI: Tim Miller

Co-PI: Lynell Tanigoshi

Organization: Washington State University **Organization:** Washington State University

Title: Extension Weed Scientist

Title: Entomologist

Phone: (360) 848-6138

Phone: (360) 848-6152

Email: twmiller@wsu.edu

Email: tanigosh@wsu.edu

Address: 16650 State Route 536

Address: 16650 State Route 536

City/State/Zip: Mount Vernon, WA 98273 **City/State/Zip:** Mount Vernon, WA 98273

Cooperators: The cooperators will be identified during winter, 2013-14

Year Initiated 2014 **Current Year** 2014 **Terminating Year** 2014

Total Project Request: **Year 1** \$3,774 **Year 2** \$0 **Year 3** \$0

Other funding sources:

Agency Name: Washington Blueberry Commission

Amt. Requested: \$3,781

Notes: This preliminary project is expected to last for one year. Should results prove to be positive, it is likely that this research will be expanded to include a graduate student's research program.

Description: Himalaya and evergreen blackberry (*Rubus armeniacus* and *R. laciniatus*, respectively) are weedy introduced plants that are widespread in western Washington, Oregon, and British Columbia. Because both species produce large numbers of berries in late summer and fall, it is assumed that much of this fruit harbors populations of spotted-wing drosophila (*Drosophila suzukii*, SWD) larvae. It is currently unknown if SWD overwinters in immature stages of development (larvae or pupae), or as adults. Traps will be placed in weedy blackberry infestations near established raspberry fields to help determine if SWD utilize weedy blackberry during the winter, and if they do, whether they do so as overwintering adult or immature insects.

Justification and Background:

Himalaya and evergreen blackberry were introduced into the PNW in the late 1800's or early 1900's for fruit production as well as for breeding material to produce new blackberry cultivars. These species quickly escaped from cultivation and now cover thousands of acres in this region. Fruit is produced from late July through early October, with berries frequently hanging on the floricanes well into December.

Spotted-wing drosophila is a recent introduction into the Pacific Northwest. It is known

to parasitize raspberry, blackberry, and blueberry, although there currently is much to learn about its life cycle and biology in western Washington, Oregon, and British Columbia. For example, it is not currently known if SWD overwinters as larvae, pupae, or adults, and from what sites adults originate in early summer. It is also not known where the majority of the overwintering population resides in this region. Many have speculated that weedy blackberries offer an attractive food source for developing larvae after the majority of cultivated fruit crops such as red raspberry, blueberry, and blackberry have been harvested, and therefore offer refugia for SWD during the winter. If SWD populations overwinter in these weedy blackberry patches, perhaps the initial populations of this fly in the spring or early summer come from these infestations. Therefore it is of interest to know whether this insect makes use of weedy blackberries during the winter, either as adults among the canes or as larvae or pupae in the soil/duff underneath the canes.

Should SWD be found to populate these weedy blackberry patches, there is a question as to whether management of these sites during the winter will exert a negative effect on SWD winter survival. Modifying the weedy blackberry winter canopy through use of herbicides such as Crossbow (triclopyr ester + 2,4-D ester) applied in December or from mowing nontreated canes in the winter will change temperatures and exposure within these sites and may reduce the insect's survivability. An alternative SWD management option would be to treat these patches with insecticide to reduce the number of adults that come from these sites to begin the surge in insect population in spring or early summer.

Dr. Amy Dreves at Oregon State University is also investigating which habitats the majority of SWD overwinters. We are unaware of any weed management projects in raspberry occurring in Idaho or British Columbia, especially as it relates to SWD.

Relationship to WRRC Research Priority(s): #1 Priority, Management Options for Control of SWD and #3 Priority, Weed Management.

Objectives (2014): To determine (1) if SWD utilizes weedy blackberry patches as winter refugia for adult or immature insects, and (2) if chemical or mechanical control of weedy blackberry patches affects SWD use.

Procedures:

Plots will be established during winter 2013-14 in well-established Himalaya or evergreen blackberry patches. Weedy blackberry infestations will be identified that are within ¼ mile of red raspberry fields to improve the likelihood that such sites will harbor SWD. Three treatments will be applied to the blackberries (1) treatment with Crossbow at 1.5% in December, (2) mowing overwintering canes to the ground in December, (3) nontreated check. Plots will be at least 2500 ft² in size (50 ft by 50 ft). One trap of two types will be placed in each plot: (1) adult traps baited with yeast or apple cider vinegar or (2) emergence traps (inverted bucket with a collection jar at the top) to collect adults arising from overwintering pupae in the duff/soil. A total of six sites will be used, resulting in 18 traps of each type (3 blackberry treatments x 6 sites = 18 traps) for a total of 36 traps. Replications will be accomplished at different farms in Whatcom and Skagit counties, to broaden the area in which trapping occurs and to increase the likelihood that overwintering SWD will be encountered. Traps will be emptied once in both January and February (1 sample per trap x 2 months x 36 traps = 36 samples) and biweekly from

March through October (2 samples per trap x 8 months x 36 traps = 576 samples). The number of SWD occurring in each sample will be determined, with a subsample to be dissected to determine their reproductive ability at the time of sampling. The experimental design will be a completely random design with six replicates (sites). Means will be separated using Tukey's Honestly Different Significance test ($P < 0.05$).

Anticipated Benefits and Information Transfer:

Data from this experiment will be disseminated through extension bulletins and during grower meetings sponsored by extension faculty and the agricultural industry. If weedy blackberry is found to be a host for SWD, additional trials will subsequently be designed to determine the degree of threat from this source, probably as a project for a graduate student.

Budget:

	2014	2015	2016
Salaries¹	\$ 2,000	\$ 0	\$ 0
Time-Slip	\$ 0	\$ 0	\$ 0
Operations (goods & services)²	\$ 500	\$ 0	\$ 0
Travel³	\$ 500	\$ 0	\$ 0
Meetings	\$ 0	\$ 0	\$ 0
Other	\$ 0	\$ 0	\$ 0
Equipment	\$ 0	\$ 0	\$ 0
Benefits⁴	\$ 774	\$ 0	\$ 0
Total	\$ 3,774	\$ 0	\$ 0

Budget Details

¹Salary for A/P scientific assistant Carl Libbey (weed science, \$500) and research associate Beverly Gerdeman and research technician Hollis Spitler (entomology, \$1000 and \$500, respectively); \$2,000 total.

²Operations (goods and services) include insect traps, bait, and related office and field supplies.

³Travel is for plot establishment, maintenance, and sampling of traps, and for presentation of data at meetings (884.95 miles x 56.5 cents/mile = \$500).

⁴Benefits (39.25% for Libbey (\$196), 35.890% for Gerdeman (\$359), and 43.86% for Spitler (\$219)); \$781 total.

2014 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

New Project Proposal

Proposed Duration: (1 year)

Project Title: Testing herbicides for weed control in red raspberries.

PI: Tim Miller

Organization: Washington State University

Title: Extension Weed Scientist

Phone: (360) 848-6138

Email: twmiller@wsu.edu

Address: 16650 State Route 536

City/State/Zip: Mount Vernon, WA 98273

Cooperators: The anticipated cooperator is Darryl Ehlers, Lynden, WA

Year Initiated 2014 **Current Year** 2014 **Terminating Year** 2014

Total Project Request: **Year 1** \$4,807 **Year 2** \$0 **Year 3** \$0

Other funding sources:

Agency Name: Washington State Commission for Pesticide Registration

Amt. Requested: \$4,250

Notes: Herbicides are generally provided by the manufacturer. Partial funding by WRRRC for the 2013 project was augmented from British Columbia Raspberry Industry Development Council and may be again in 2014.

Description: Weeds continue to be problematic in red raspberry production. In order to better manage them, it would be beneficial to gain registrations for new herbicides. A thorough testing of new herbicides alone and in combination with existing products is necessary to achieve both improved weed control while maintaining crop safety. Products of interest are Stinger (clopyralid), Paramount (quinclorac), Treevix (saflufenacil), Zeus (sulfentrazone), and Alion (indaziflam). Sequential applications of these products will be tested for efficacy on various weed species and for crop safety in this project.

Justification and Background:

Perennial weed species generally become more important the longer raspberry blocks are left in production. These weeds often will outlive the raspberry crop and are also difficult to control in the break crop between raspberry plantings, so they generally remain a problem in the subsequent raspberry planting. Yet another difficulty with perennial weeds in raspberry is the physical interference to berry drop using machine harvesters, which may result in berry loss. They also impact harvest of hand-picked fruit, reducing the efficiency of hand harvest by making berries harder to find and pick.

Perennial weeds frequently become established the first couple of seasons on a new raspberry block, when raspberry plants are small and not as competitive. Often, these weeds are present in the field prior to transplanting baby raspberries. If not controlled when the infestation is relatively small, perennial weeds become increasingly difficult to kill, increasing herbicide and labor costs and becoming a major factor in reducing the longevity of raspberry plantings.

Trials with Stinger in healthy raspberries over the last few years have been encouraging. Importantly, florican injury and berry harvest have not been significantly impacted by these applications. Since postemergence (POST) treatments can be made when weeds are visible and thus to areas known to be infested with perennial weeds, cost of these treatments may be significantly lower than broadcast applications to the full block. Additionally, if good to

excellent weed control results from these applications, slight crop injury due to the herbicide is more acceptable if it occurs only on selected areas of the field. More reliable crop injury data resulting from applications of these products is needed to document that they are safe for use in raspberry if registrations are to result, however. Treevix (BASF) and Alion (Bayer) are two new herbicides that recently gained federal registrations in certain orchard crops. Initial data in raspberry is promising, with Treevix as a cane burning herbicide similar in activity to Goal (oxyfluorfen) and Alion providing residual control of germinating weed seeds.

Dr. Ed Peachey at Oregon State University is also conducting herbicide work in red raspberry, with both of us previously conducting preliminary crop safety (nonresidue) trials with IR-4 with Paramount. I am unaware of any weed management projects in raspberry occurring in Idaho or British Columbia.

Relationship to WRRRC Research Priority(s): #3 Priority, Weed Management

Objectives (2014): To test several non-labeled herbicides in various sequences or mixtures for control of several perennial weeds in established red raspberries.

Procedures:

Plots will be established in 2014 in established raspberries. Herbicide applications will be made for several combinations of herbicides at cane burning time in early spring (Treevix, Zeus, and Alion) and late spring (Stinger and Paramount). A typical application sequence could be Treevix + Alion in April followed by Stinger in late May. Most sequences/combinations of these herbicides will be included in this trial, as well as combinations and sequences with other registered residual herbicides such as Sinbar (terbacil), simazine, diuron, Callisto (mesotrione), Matrix (rimsulfuron), and Sandea (halosulfuron). Weed control will be evaluated, as will herbicide effects on raspberry yield, berry size, and primocane growth.

Anticipated Benefits and Information Transfer:

If positive, data from this experiment will be used to support new herbicide registrations in raspberries for these products. The data resulting from these studies will be disseminated through extension bulletins and during grower meetings sponsored by extension faculty and the agricultural industry.

Budget:

	2014	2015	2016
Salaries¹	\$ 1,500	\$ 0	\$ 0
Time-Slip	\$ 1,000	\$ 0	\$ 0
Operations (goods & services)²	\$ 500	\$ 0	\$ 0
Travel³	\$ 500	\$ 0	\$ 0
Meetings	\$ 0	\$ 0	\$ 0
Other	\$ 0	\$ 0	\$ 0
Equipment	\$ 0	\$ 0	\$ 0
Benefits³	\$ 1,307	\$ 0	\$ 0
Total	\$ 4,807	\$ 0	\$ 0

Budget Details

¹Salary for A/P scientific assistant Carl Libbey is completely funded by external grants.

²Operations (goods and services) include flags and related office and field supplies.

³Travel is for plot establishment, maintenance, and harvest, and for presentation of data at meetings (884.95 miles at 56.5 cents/mile = \$500)

⁴Benefits (39.25% for A/P scientific assistant, \$589; 71.8% for time-slip help, \$718; total \$1,307).

**2014 WASHINGTON RED RASPBERRY COMMISSION
RESEARCH PROPOSAL**

New Project Proposal

Proposed Duration: 2-years

Project Title: Integration of Factors to Improve Soil Health in Red Raspberry Production

Principal Investigator

Chris Benedict, WSU Whatcom County
Extension
Agriculture Extension Educator
1000 N. Forest St. Suite 201,
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Co-PI

Colleen Burrows, WSU Whatcom County
Extension,
Agriculture Special Projects Coordinator
1000 N. Forest St. Suite 201,
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cburrows@wsu.edu

Cooperators: Lisa Wasko DeVetter, WSU Small Fruit Horticulture Program Leader (Spring 2014)

Year Initiated 2012 **Current Year** 2014 **Terminating Year:** 2015

Total Project Request: \$14265 **Year 1** \$5787 **Year 2** \$8478 **Year 3** \$0

Other funding sources:

Description: Many red raspberry producers have witnessed a reduction in the harvestable productivity of a planting; this is suspected to be due to declines in soil health. Much of this can be attributable to increases in soil borne pathogens and reliance on cultural practices (such as cultivation and lack of introduction of organic matter) that can lead to soil health declines. To overcome these declines, producers are relying on increased inputs which have resulted in increased production costs. Past research has evaluated the causes of soil health decline and potential solutions to the issue in a reductionistic way; it has only evaluated a portion of the overall production system. This project will continue efforts that began in 2012 that includes use of ground covers (small grains and brassicas), alternative nutrient sources/rates (organically derived sources/spring soil test-driven), and efficient water delivery mechanisms (tensiometer driven) and their impacts on various soil health parameters (biological, chemical, and physical). Intrinsic in the design of these trials is a long-term viewpoint and an adaptive management strategy as basic knowledge is obtained.

Justification and Background: Raspberry growers have noticed a decrease in duration of productive plantings, costing significant amounts in income losses and replant costs. One suspected reason is the decline in soil health, a loss of soil organic matter and beneficial soil biological activity, and subsequent increase in soil-borne diseases such as root rot (*Phytophthora rubi*) and nematodes (*Pratylenchus penetrans*). A reliance on fumigation and post plant soil-applied pesticides has been used to attempt to overcome these issues, but is being compromised by changes in use-regulations.

While development of resistant varieties to these pests is the long-term goal, other methods have been tested to manage root rot and nematodes. Other cultural options, such as fallow periods, are not economically optional for many producers and management of this fallow period can lead to declines in other soil health parameters (Forge et al., 2000). The use of small grain ground covers to reduce *P. penetrans* populations has had mixed results; in some studies reductions have occurred (Forge et al., 2000), while in others populations were not significantly reduced (MacGuidwin and Layne, 1995).

Physical soil health issues such as reductions in the organic matter fraction can impact a number of other factors such as fertility, water availability, compaction, erosion, pest issues (Magdoff and van Es, 2000), and lower mineralization of nutrients (Forge and Kenney, 2011). Resulting is the requirement to increase inputs such as fertilizers and can lead to the need for additional tillage activities, thus further increasing soil organic matter losses. Introducing biomass to increase organic matter levels, such as through the use of ground covers, is a common practice in annual production systems.

Over-irrigation can cause leaching of valuable nitrogen fertilizer and can promote root susceptibility to *P. rubi*. Retention of nutrients within the root zone has shown to significantly increase through scheduled-based irrigation (Neilsen et. al 2011).

We will evaluate the use of a set of cultural practices in an integrated fashion so that the individual and combined impacts can be measured. This systems experiment utilizes promising results found in British Columbia (Forge 2012, Forge and Kempler 2009).

Relationship to WRRRC Research Priority(s): Because of the inter-disciplinary scope of this project it is related to many of the priorities including:

#1 priorities

- Understanding soil ecology and soil borne pathogens and their effects on plant health and crop yields.
- Soil fumigation techniques and alternatives to control soil pathogens, nematodes, and weeds

#2 priorities

- Nutrient/Irrigation management

Objectives:

1. Evaluate integrated measures to improve soil health including use of ground covers, brassica seed meals, alternative nutrient sources, and more efficient water delivery systems.

Procedures: This project is designed to be ongoing through the lifetime of a raspberry planting and will continue for a number of years utilizing similar methods described below, but with adaptive management as treatment results and additional tools become available.

1. Evaluate integrated measures to improve soil health including use of ground covers, alternative nutrient sources, and more efficient water delivery systems.

The trial was initiated in the spring of 2012 setup in a completely randomized split-split plot design consisting of three replications on two commercial farms in 'Meeker' raspberries. Variables include: 1.) Water Use (Tensiometer vs. Timed Irrigation)[Main-plot]; 2.) Ground Covers (No vs. Spring Planted vs. Postharvest planted vs. perennial ground cover [one site only])[split-plot]; and (Organically derived nutrient source vs. synthetic fertilizers [site 1]; spring soil sample driven vs. standard [site 2])[split-split plot]. Spring planted (SP) ground covers will be planted by late April and post-harvest (PH) ground covers will be planted after harvest operations. Ground cover biomass will be taken prior to harvest (Spring-planted only) and in October. A commercially available brassica seed meal (MustGrow™) is labeled for use in red raspberries will be included in micro-plots in the spring of 2014 and 2015. Soil samples will be taken from each split-split plot in late March and nutrient content analyzed to drive spring nutrient application rates by plot. As the plantings are entering into their 4th and 5th year, we will downsize the intensive soil biological sampling (nematode populations, *P. rubi*) that took place in 2012 and 2013 to reduce project costs in year 1

(2014), but will continue this sampling in 2015. Physical soil property assessments will include: compaction (penetrometer: pre- and post-harvest), bulk density (post-harvest), and infiltration (pre- and post-harvest). Soil temperature sensors (10 cm) are placed in each sub-plot and soil moisture meters (10 cm) are placed in each main-plot before irrigation begins along with flow meters. Dormant primocane assessment (diameter) will occur in late winter (Zebarth, Freyman and Kowalenko, 1993) and again in late fall. Chlorophyll content (SPAD 502Plus, Konica Minolta Sensing, Inc.) of leaves will be used to assess leaf nitrogen content in August (Privè et al., 1997).

Anticipated Benefits and Information Transfer: Resulting from this work is a better understanding of management practices that will lead to improved soil health. Results will be shared with producers at the annual Washington Small Fruit Conference in December in both years. Results will also be published in the Whatcom Ag Monthly Newsletter and on the WSU Whatcom County Extension website with photos and videos of the process for growers to get a better idea of what is involved throughout the year.

References:

- Forge, T.A. 2012. Alternative raspberry field renovation practices and the use of organic amendments: Implications for root diseases and nitrate leaching, 57th Annual North Willamette Horticulture Society Meeting, Canby, OR, USA, January 10-12, 2012, Berry Section.
- Forge, T.A. and E. Kenney. 2011. Soil health, organic amendments and alternative practices for renovation of raspberry fields. SAGES Workshop, Abbotsford, CA.
- Forge, T.A. and Kempler, C. 2009. "Organic mulches influence population densities of root-lesion nematodes, soil health indicators and root growth of red raspberry.", *Canadian Journal of Plant Pathology*, 31(2), pp. 241-249.
- Forge, T.A., R. E. Ingham, D. Kaufman, and J.N. Pinkerton. 2000. Population Growth of *Pratylenchus penetrans* on Winter Cover Crops Grown in the Pacific Northwest. *J. of Nematology*. 32: 42-51.
- MacGuidwin, A. E., and T. L. Layne. 1995. Response of nematode communities to sudangrass and sorghum-sudangrass hybrids grown as green manure crops. Supplement to the *Journal of Nematology* 27:609–616.
- Magdoff, F and H. van Es. 2000. Building soils for better crops: 2nd Ed.. Sustainable Agriculture Network. 230 pgs.
- Neilsen, D. S. Kuchta, T. Forge, B. Zebarth, C. Nichol, and M. Sweeney. 2011. Irrigation and Nitrogen Management. SAGES Workshop, Abbotsford, CA.
- Privè, J., J.A. Sullivan, and J.T.A. Proctor. 1997. Seasonal changes in net carbon dioxide exchange rates of Autumn Bliss, a primocane-fruiting red raspberry (*Rubus idaeus* L.). *Can. J. of Plant Sci.* 77: 427-431.
- Zebarth, B.J., S. Freyman, and C.G. Kowalenko. 1993. Effect of ground covers and tillage between raspberry rows on selected soil physical and chemical parameters and crop response. *Can. J. Soil Sci.* 73: 481-488.

Budget: Indirect or overhead costs are not allowed unless specifically authorized by the Board

	2014	2015	2016
Salaries ^{1/}	\$489	\$509	\$998
Time-Slip ^{2/}	\$2430	\$2160	\$4590
Operations (goods & services) ^{3/}	\$1210	\$4570	\$5780
Travel ^{4/}	\$848	\$848	\$1695
Meetings	\$0	\$0	\$0
Other	\$0	\$0	\$0
Equipment ^{5/}	\$400	\$0	\$400
Benefits ^{6/}	\$410	\$391	\$801
Total	\$5787	\$8478	\$14265

Budget Justification

- ^{1/} One WSU Whatcom County Extension Personnel at 0.01% FTE (\$998)
- ^{2/} One WSU Whatcom County Extension timeslip @ \$15/hr (yr1 170hr, yr2 168hr) (\$4,590)
- ^{3/} Soil Nutrient Analysis (\$15/sample-\$1260[yr1&yr2]); Soil Biological Assays [yr 2] (\$40/sample-\$3,360); Cover Crop Seed (\$880[yr1&yr2]); Sample Shipping (\$80[yr1&yr2]); Brassica Seed meal (\$200 [yr1&yr2]).
- ^{4/} Travel to and from research sites, 1500 miles at \$0.565/mile (\$1695)
- ^{5/} Includes irrigation fittings for tensiometer-driven irrigation lines (\$400)
- ^{6/} Employee benefits for Extension personnel (35.59%) and timeslip (9.7%)(801).

CURRENT & PENDING SUPPORT

Name: Chris Benedict

Instructions:

Who completes this template: Each project director/principal investigator (PD/PI) and other senior personnel that the Request for Applications (RFA) specifies

How this template is completed:

- Record information for active and pending projects, including this proposal.
- All current efforts to which PD/PI(s) and other senior personnel have committed a portion of their time must be listed, whether or not salary for the person involved is included in the budgets of the various projects.
- Provide analogous information for all proposed work which is being considered by, or which will be submitted in the near future to, other possible sponsors, including other USDA programs.
- For concurrent projects, the percent of time committed must not exceed 100%..

Note: Concurrent submission of a proposal to other organizations will not prejudice its review by CSREES.

NAME (List/PD #1 first)	SUPPORTING AGENCY AND AGENCY ACTIVE AWARD/PENDING PROPOSAL NUMBER	TOTAL \$ AMOUNT	EFFECTIVE AND EXPIRATION DATES	% OF TIME COMMITTED	TITLE OF PROJECT
	Active:				

Benedict, C. McMoran, D. Corbin, A.	WA State Dairy Products Commission	\$48,972	1/1/12-12/31/13	5%	Evaluation of Short-Season Silage Corn Varieties
Benedict, C. Burrows, C.	WA Red Raspberry Commission	\$5,921	1/1/13-12/31/13	2%	First Alert Scouting of SWD in Western WA.
Burrows, C. Benedict, C.	WA Red Raspberry Commission	\$7,500	1/1/13-12/31/13	5%	Integration of Factors to Improve Soil Health in Red Raspberry Production
Collins, D Corbin, A Benedict, C Cogger, C Bary, A	Utah State Univ/WSARE	\$196,624	10/1/10 – 9/30/14	5%	Selecting Management Practices & Cover Crops for Reducing Tillage, Enhancing Soil, Quality & Managing Weeds in Western WA Organic Vegetable Farms
Murphy, K Baik, B Benedict, C. Desta, K Dillon, M. Epstein, L. Goldberger, J. Machado, S. Maughan, J. Matanguihan, J. Petri, S. Reeve, J. Van Horn, M.	USDA-NIFA	\$1,236,000	1/1/13-12/31/16	5%	Developing adapted varieties and optimal management practices for quinoa in diverse environments
Collins, D. Ostrom, M. Benedict, C. Garcia-Pabon, J. Busboom, J. Flores, M. Heitsuman, M.	USDA BFRDP	\$749,999	8/1/12-7/31/15	10%	Cultivating New Generation and Immigrant Farmers in Washington State
Pending: Benedict, C. C. Burrows	WA Red Raspberry Commission	\$14,265		10%	Integration of Factors to Improve Soil Health in Red Raspberry (This proposal)

Washington Red Raspberry Commission Progress Report Format for 2013 Projects

Project No:

Title: Integration of Factors to Improve Soil Health in Red Raspberry Production

Personnel:

Principal Investigator

Chris Benedict, WSU Whatcom County
Extension
Agriculture Extension Educator
1000 N. Forest St. Suite 201,
Bellingham, WA, 98225.
(360) 676-6736
chrisbenedict@wsu.edu

Co-PI and/or cooperator(s)

Colleen Burrows, WSU Whatcom County
Extension,
Agriculture Special Projects Coordinator
1000 N. Forest St. Suite 201,
Bellingham, WA, 98225.
(360) 676-6736
cburrows@wsu.edu

Reporting Period: 1/1/13-12/31/13

Accomplishments:

This project has entered into its second year of treatment implementation. Plant health and growth was monitored through spring and fall primocane diameter measurements, monthly visual inspections, yield estimates (fruit weight over three collections periods, total weight, and average yield), pruning weights, and SPAD (chlorophyll) monitoring. Soil health measurements included chemical (spring nutrient), physical (bulk density, compaction, infiltration), and biological (*P. penetrans*, *P. rubi*, and *V. dahliae*). Tensiometer controlled irrigation lines and flow meters were re-installed in the spring. We've included a between-row application of composted dairy solids at one of the sites to determine if it can improve the imbalance of pathogenic to non-pathogenic microorganism (as has been show in other studies). Relationship analysis will determine what (i.e. management, soil, plant) factors are correlated with soil health indicators that may lead to improved management options.

Results:

To date soil nutrient, plant health/growth, estimated yield, water use, soil moisture, bulk density, compaction, infiltration, and SPAD readings have been compiled and analyzed. Soil biological assay results are still being finalized. Tensiometer controlled irrigation lines utilized 43% and 14% (29% average across sites) less water at each site, respectively while maintaining adequate soil moisture levels. Cover crops were well-established at both sites in both the spring and fall. Overwintering cover crop biomass ('Wheeler' rye) averaged 3.5 tons/A, a spring mix of SorghumXSudangrass + Oats (110 lbs/A) was planted at the other site, and a fall mix of white oats + 'Wheeler' Rye was planted at 110 lbs/A at both sites.

Publications:

We are in the process of developing a dedicated portion of WSU Whatcom County Extension website to the issues around soil health in red raspberries. We will include results from this project as they evolve. Results will be discussed at the Washington Small Fruit Conference in December. A project update will be published in the Whatcom Ag Monthly in January, highlighting results from 2013 work.

2014 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

New Project Proposal

Proposed Duration: (2 years)

Project Title: Mobile App for Pest Scouting and Record Keeping in Raspberry fields

PI: Chris Benedict

Organization: Washington State University

Title: Extension Educator

Phone: 360-676-6736 ext 21

Email: chrisbenedict@wsu.edu

Address: 1000 N. Forest St.

Address 2: Suite 201

City/State/Zip: Bellingham, WA 98225

Co-PI: Colleen Burrows

Organization: Washington State University

Title: Agriculture Special Projects

Phone: 360-676-6736 ext 22

Email: cburrows@wsu.edu

Address: 1000 N. Forest St.

Address 2: Suite 201

City/State/Zip: Bellingham, WA 98225

Cooperators:

Year Initiated 2014 **Current Year** 2014 **Terminating Year** 2016

Total Project Request: \$18767 **Year 1** \$11230 **Year 2** \$7537 **Year 3** \$0

Other funding sources:

Agency Name: Washington Blueberry Commission

Amt. Requested: \$18767

Notes:

Description:

Growers currently rely on paper notebooks and calendars for pest scouting record keeping; this data is difficult to refer back to or analyze and is not being used to its full potential for farm management. Through this project we will develop a mobile phone application to provide growers and consultants information on scouting methods, pest identification and biology, pest alerts, and the ability to input scouting information for future access. In the first year, a beta version will be developed and tested by selected grower and consultants. Input from those users will be incorporated into the application to develop the alpha version. Future versions will include weather information and pest degree day models. Pest alerts will be developed for those that have current models, so users can receive alerts at times when pest pressure is expected to be high. This tool will be available to all growers in 2015. It will allow growers to collect and analyze data and to share information between farm managers and consultants. The application will be developed as a foundation where additional components (such as cultural information) may be added in the future.

Justification and Background:

Most red raspberry growers currently keep scouting and other cultural management records using notebooks and/or calendars which make data analysis difficult. Many of these growers also use smart phones with capabilities of geolocation, taking photos, web access, and information entry and sharing. A few mobile phone or web applications currently exist for farm record keeping, but most are catering to large scale farms in other regions or require a costly subscription; none are catered to berry growers. Through this project, we will develop a field-friendly mobile application for smart phones and tablets that small fruit growers can use to record data and access information.

The web-based WSU Integrated Pest Management for Raspberries manual was updated in 2010 with input on scouting techniques and thresholds gleaned from growers and industry representatives. This manual contains valuable information on pests, but is not being fully utilized because of the format. By incorporating this information into a mobile phone application, users will have access to it in the field, when it is most needed. Additionally, collection and synthesis of pest population information enables small fruit growers to make more informed management decisions. With the addition of a web-accessible database of collected information (smartphone app uploads data), field managers and consultants can look for trends in decisions and identify efficiencies for future actions. We envision that the development of this application lays a foundation for future add-ons (i.e. WSU AgWeatherNet, Ask an Expert, cultural management tool, WSU's Irrigation Scheduler) that will provide increased access to information at the fingertips of growers.

This project will incorporate current pest management research being performed by researchers in Washington, Oregon, and British Columbia; consultation with researchers will be done to include new pest management strategies.

Relationship to WRRRC Research Priority(s):

This project relates to several pest management priorities by improving information access. Those priorities include:

- Management options for control of the Spotted Wing Drosophila
- Root Weevils
- Mite Management
- Weed management
- Foliar and Cane Diseases

Objectives:

- 2014: To develop and beta test a mobile phone application for growers and consultants to use for recording scouting information, receiving alerts, and receiving information about common pests.
- 2015: To incorporate user input into the application, and add weather and degree day models.

Procedures:

In the winter of 2014, a beta version of a mobile phone/tablet application will be developed. This application will allow growers to keep track of pest population data relating to different fields. It will be formatted as an easy-to use application with a customizable interface. An additional component will include a web-accessible user interface to manage account and field information. The following features will be included:

- Ability to map fields by drawing or uploading GPS data via a web-interface,
- Plant growth stage dependent scouting information will be pushed to users, pest identification tools and tips provided, and field scouting information recorded so that growers can be sure they are scouting for the correct pests at the correct times of year. Information will be incorporated from the on-line WSU Integrated Pest Management for Raspberries Manual, and
- Users will have login capability that they will be able to share (in a limited or full user capability) with other users if desired.

This beta version will be tested by 6-10 grower and consultant users during the 2014 season as well as by one WSU scout that will be monitoring select fields. Input given by users will be

incorporated into the application during and following the growing season.

For the 2015 growing season, additional features will be added:

- Integration of weather forecasts and past weather data relating to risk of increased pest and disease pressure, and
- Degree day models for pests that are currently available.

The application will be available for use by all growers in 2015; however, improvements may be made on a regular basis.

This application will be developed specifically for berry growers in Washington State, with pest information and alerts for these growers. Additional features (than those listed above) may be added if they are requested by test users.

This project will be coordinated by Chris Benedict and Colleen Burrows at WSU Whatcom County Extension. Content information will be provided by WSU and industry representatives. Researchers from WSU, OSU, USDA, and British Columbia agencies will be consulted for new or relevant scouting and pest management resources. The mobile phone/tablet software application will be developed by an external software development firm.

Anticipated Benefits and Information Transfer: (100 words maximum)

This mobile application will enable growers to better utilize existing information about pest identification, scouting techniques, and treatment thresholds. Users will enter scouting data directly into this application, rather than relying on notebooks and calendars, and can easily refer back to and compare information from the past or between various fields by utilizing a web-interface. This tool will be further developed to incorporate cultural practices such as irrigation and fertilizer input recommendations.

Information will be transferred to users by field days and breakfast meetings in 2014 and 2015, as well as at the Small Fruit Conferences.

Budget: Indirect or overhead costs are not allowed unless specifically authorized by the Board

	2014	2015	2016
Salaries ^{1/}	\$2445	\$2543	\$0
Time-Slip	\$0	\$0	\$0
Operations (goods & services)	\$325	\$0	\$0
Travel ^{2/}	\$339	\$339	\$0
Meetings	\$0	\$0	\$0
Other ^{3/}	\$7250	\$3750	\$0
Equipment ^{4/}	\$0	\$0	\$0
Benefits ^{5/}	\$871	\$905	\$0
Total	\$11230	\$7537	\$0

Budget Justification

^{1/}One WSU Whatcom County Extension Personnel at 0.10% FTE (Includes 4% raise in year 2) [Year 1 \$2445; Year 2 \$2543]: \$4988.

^{2/} Travel for scouting activities and meetings associated with this project (such as training sessions with beta users) 600 miles at \$0.565/mile [Year 1 \$339; Year 2 \$339]: \$678.

^{3/}Includes contract with software developer and necessary server space [Year 1 \$7250; Year 2 \$3750]: \$11,000.

^{4/}None requested.

^{5/} Employee benefits for Extension personnel (35.59%). [Year 1 \$871; Year 2 \$905]: \$1776.

CURRENT & PENDING SUPPORT

Name: Chris Benedict

Instructions:

Who completes this template: Each project director/principal investigator (PD/PI) and other senior personnel that the Request for Applications (RFA) specifies

How this template is completed:

- Record information for active and pending projects, including this proposal.
- All current efforts to which PD/PI(s) and other senior personnel have committed a portion of their time must be listed, whether or not salary for the person involved is included in the budgets of the various projects.
- Provide analogous information for all proposed work which is being considered by, or which will be submitted in the near future to, other possible sponsors, including other USDA programs.
- For concurrent projects, the percent of time committed must not exceed 100%..

Note: Concurrent submission of a proposal to other organizations will not prejudice its review by CSREES.

NAME (List/PD #1 first)	SUPPORTING AGENCY AND AGENCY ACTIVE AWARD/PENDING PROPOSAL NUMBER	TOTAL \$ AMOUNT	EFFECTIVE AND EXPIRATION DATES	% OF TIME COMMITTED	TITLE OF PROJECT
Benedict, C. McMoran, D. Corbin, A.	Active: WA State Dairy Products Commission	\$48,972	1/1/12-12/31/13	5%	Evaluation of Short-Season Silage Corn Varieties
Benedict, C. Burrows, C	WA Red Raspberry Commission	\$5,921	1/1/13-12/31/13	2%	First Alert Scouting of SWD in Western WA.
Burrows, C. Benedict, C.	WA Red Raspberry Commission	\$7,500	1/1/13-12/31/13	5%	Integration of Factors to Improve Soil Health in Red Raspberry Production
Collins, D Corbin, A Benedict, C Cogger, C Bary, A	Utah State Univ/WSARE	\$196,624	10/1/10 – 9/30/14	5%	Selecting Management Practices & Cover Crops for Reducing Tillage, Enhancing Soil, Quality & Managing Weeds in Western WA Organic Vegetable Farms
Murphy, K Baik, B Benedict, C. Desta, K Dillon, M. Epstein, L. Goldberger, J. Machado, S. Maughan, J. Matanguihan, J. Petri, S. Reeve, J. Van Horn, M.	USDA-NIFA	\$1,236,000	1/1/13-12/31/16	5%	Developing adapted varieties and optimal management practices for quinoa in diverse environments
Collins, D. Ostrom, M.	USDA BFRDP	\$749,999	8/1/12-7/31/15	10%	Cultivating New Generation and Immigrant Farmers in Washington State

Benedict, C. Garcia-Pabon, J. Busboom, J. Flores, M. Heitsuman, M.					
Pending: Benedict, C. C. Burrows	WA Red Raspberry Commission	\$18,768	1/1/14-12/31/15	5%	Mobile App for Pest Scouting and Record Keeping in Raspberry fields (This proposal)

Application Date: November 7, 2013

Name of Applicant Organization/Company:

USDA-ARS Horticultural Crops Research Laboratory
3420 NW Orchard Ave.
Corvallis, OR 97330

Principal Investigator:

Jerry Weiland
Research Plant Pathologist
(541) 738-4062
jerry.weiland@ars.usda.gov

Co-PI and/or cooperator:

Chris Benedict (Co-PI)
Regional Extension Specialist
Washington State University Extension
1000 N. Forest St., Suite 201
Bellingham, WA 98225
(360) 676-6736 ext. 21
chrisbenedict@wsu.edu

Title: Incidence and Detection of *Verticillium dahliae* in Red Raspberry Production Fields

Year Initiated 2013 Current Year 2013 Terminating Year 2014

Funding:

Total amount requested: \$13,582 (2014), year 2 for project started in 2013;

Other sources of funding: We will solicit funding from the Oregon Raspberry and Blackberry Commission for similar surveys.

Current funding already received for this project: \$10,000 from the Washington Red Raspberry Commission for the 2013 survey. Requesting additional funds to complete year 2 of survey in 2014.

Incidence and Detection of *Verticillium dahliae* in Red Raspberry Production Fields

Year Initiated 2013 Current Year 2013 Terminating Year 2014

Project Description and Expected Outcomes:

Verticillium dahliae was isolated from wilted and dying red raspberry plants found in several western Washington production fields in 2012. We propose to survey for the incidence of *V. dahliae* in production fields to determine if the pathogen is widespread in the red raspberry industry and to determine if the pathogen is consistently associated with fields containing wilting and dying plants. Our survey will utilize two common methods for *V. dahliae* detection, a culture-based method and a DNA-based method, to ensure robust detection and to compare methods for detection and quantification of the pathogen. We expect to determine the incidence of *V. dahliae* in red raspberry production fields and establish whether grower observations of cane wilting and death are associated with this pathogen. We also expect to determine which method of pathogen detection is more effective at detection and quantification of *V. dahliae* in field soils and plant samples.

A fifth objective was added this year in view of preliminary results from 2013. We additionally propose to determine the association of other potential pathogens, including nematodes, *Phytophthora rubi*, and true fungi (*Rhizoctonia*, *Alternaria*, and *Fusarium* species, for example) with diseased plants of red raspberry collected during the survey. This will ensure that we obtain a more complete census of the microbes associated with diseased red raspberry plants in Washington State in addition to determining the prevalence of *V. dahliae*. This research directly addresses a #1 priority of the Washington Red Raspberry commission, understanding soilborne pathogens and their effects on plant health.

Justification and Background:

Verticillium dahliae is a common soilborne pathogen in many agricultural soils and has recently been suspected of infecting and killing red raspberry plants in the state of Washington. Historically, relatively little research has addressed the role of *V. dahliae* in red raspberry production. An early report indicated that *V. dahliae* killed canes and reduced yields of *Rubus idaeus* cultivars in England (Harris, 1925). The pathogen was also described from Ontario, Canada, where 5 - 20% of the plants in red raspberry fields were infected (Berkeley and Jackson, 1926). Symptoms of the disease have been described as yellowing and wilting leaves followed by cane death. Occasionally, a blue discoloration was observed on canes, but this was not found consistently in all infected plants. Often, the plants were not completely killed and new shoots were able to grow from noninfected roots. But, in other cases, entire hills of red raspberry were killed. The disease usually has a patchy distribution and severity may differ from year to year; fields of Cuthbert that had severe *Verticillium* wilt one year only had a trace of disease in the following two years (Berkeley and Jackson, 1926).

Red raspberry cultivars are often considered as resistant to *Verticillium* (Fiola and Swartz, 1994). However, *V. dahliae* was recently found in plant and soil samples collected from declining red raspberry fields in western Washington in 2012 by two commercial analytical laboratories and one research laboratory (Weiland, unpublished data). In addition, two years of greenhouse

inoculations of Meeker red raspberry with *V. dahliae* have shown that this cultivar can be infected and killed by the pathogen (Figure 1).

Although the pathogen has been detected from naturally-infected plants and it is known to cause disease, additional information is needed in order to establish whether *V. dahliae* is a threat to red raspberry production. First, the incidence of *V. dahliae* in production fields as well as the amount of inoculum associated with disease remains unknown. A survey is needed in order to determine: a) whether *V. dahliae* is widely present in the red raspberry industry; b) if its presence is associated with the pockets of disease observed in fields in 2012; and c) to determine if particular inoculum levels are associated with disease. Second, results from each of the three laboratories were somewhat contradictory in terms of the presence and quantity of *V. dahliae* in plant and soil samples. It is unclear whether these differences were due to the variability inherent in sampling (e.g., some samples had *V. dahliae* and some samples did not) or whether the differences were due to the method of detection. Two methods were used by the laboratories for the detection of *V. dahliae*: A culture-based method and a DNA-based method. The culture-based method relies on identification of living cultures of *V. dahliae* from soil and plant samples whereas the DNA-based method is based on the selective detection of *V. dahliae* DNA. Both methods have advantages and disadvantages, but a comparison should be made to determine which method is the most sensitive at detection and which method is the best at quantification of pathogen inoculum levels.

Relationship to WRRRC Research priority(s):

This proposed research directly addresses a #1 priority of the WRRRC, understanding soilborne pathogens and their effects on plant health.

Objectives:

1. Determine the incidence of *V. dahliae* in red raspberry production fields;
2. Determine if the presence of *V. dahliae* in soil and plant samples is associated with disease symptoms of wilting and cane death;
3. Determine which inoculum levels are associated with disease symptoms;
4. Compare culture-based and DNA-based methods for effectiveness of *V. dahliae* detection and quantification.
5. To determine the association of other potential pathogens, including nematodes, *Phytophthora rubi*, and true fungi (e.g., *Rhizoctonia*, *Fusarium*, and *Alternaria* species) with disease symptoms of wilting and cane death.

Procedures:

Over a two-year period we propose to evaluate the incidence and quantity of *V. dahliae* in red raspberry production fields using two available methods for detection and quantification. In the first year of the survey, 2013, we sampled 18 diseased sites and 5 nondiseased sites in Washington. For 2014, we propose to identify 25 additional diseased sites and 10 nondiseased sites in red raspberry production fields to sample for the presence of *V. dahliae*. Sampling will begin in late June or as soon as symptoms associated with Verticillium wilt are discovered. Diseased sites are defined as areas within the production field where symptoms of wilting, cane dieback, sparse canopy, stunted growth, and/or blue stem discoloration are observed. Within each diseased site, three to five symptomatic plants 20-30 feet from each other will be sampled

for *V. dahliae* infection by collecting 2-3 symptomatic canes/plant. Soil cores (12”) will also be collected immediately adjacent to each symptomatic plant (within hill) and then bulked for each disease site. For nondiseased sites, canes from five healthy plants and adjacent soil cores will be collected as described above. Nondiseased sites are defined as apparently healthy areas within the production field where no Verticillium wilt symptoms are observed and plants have normal coloration and robust growth.

Two methods will be used to detect and quantify the pathogen from soil and plant samples. Each plant and soil sample will be divided into two samples with one half of each sample to be tested by the culture-based method and the other half of each sample to be tested by the DNA-based method. The culture-based method has been widely used to detect and quantify *V. dahliae* by research and diagnostic labs for decades (Butterfield and DeVay, 1977), whereas the DNA-based methods are more recent (Bilodeau et al., 2012, Duressa et al., 2012, Markakis et al., 2009). For the culture-based method, plant samples are surface-disinfested with a 10% bleach solution, cut into segments, and then plated into petri dishes containing a semi-selective medium from which diagnostic *V. dahliae* cultures will grow if the pathogen is present. Symptomatic plant samples will also be plated on other media to determine if other potential pathogens such as *Phytophthora rubi*, *Rhizoctonia* species, *Fusarium* species, and *Alternaria* species are also present. Soil samples will be dried for 2 weeks to eliminate competing contaminant fungi and then spread on the same semi-selective medium used above. The presence of nematodes will also be surveyed from all plant and soil samples. For the DNA-based method, *V. dahliae* DNA will be extracted and amplified from plants and soil samples using established protocols (Bilodeau et al., 2012, Duressa et al., 2012, Markakis et al., 2009).

Pathogen incidence for each site (diseased and nondiseased) will be determined by the presence or absence of *V. dahliae* as detected by each method. Quantification of *V. dahliae* from plant samples will be based on the number of plants from which the pathogen was detected by each method. Quantification of the pathogen from soil will be based on the number of *V. dahliae* cultures that grow from each soil sample or the amount of *V. dahliae* DNA amplified by the DNA-based method. Data will be compiled and results from both diseased and nondiseased sites will be compared to determine the incidence of *V. dahliae* in red raspberry production fields (Objective 1). Results from plant and soil samples at diseased sites will be used to determine if the presence of *V. dahliae* in the soil is associated with disease (Objective 2) and to determine which inoculum levels are associated with disease symptoms (Objective 3). Finally, results from each method will be compared to determine which method is best at detecting and at quantifying *V. dahliae* from plant and soil samples (Objective 4). Incidence data will be analyzed using the chi-square test for independence and quantity data will be analyzed using ANOVA. Finally, the number of other soilborne and potential pathogens (*Phytophthora rubi*, nematodes, *Rhizoctonia*, *Fusarium*, and *Alternaria* species) will be quantified to determine if their presence is correlated with disease symptoms in the field (Objective 5).

Timeline:

Activity	2013	2014
Plant and soil sample collection	June-October	June-October
Plant and soil sample processing	June-December	June-December

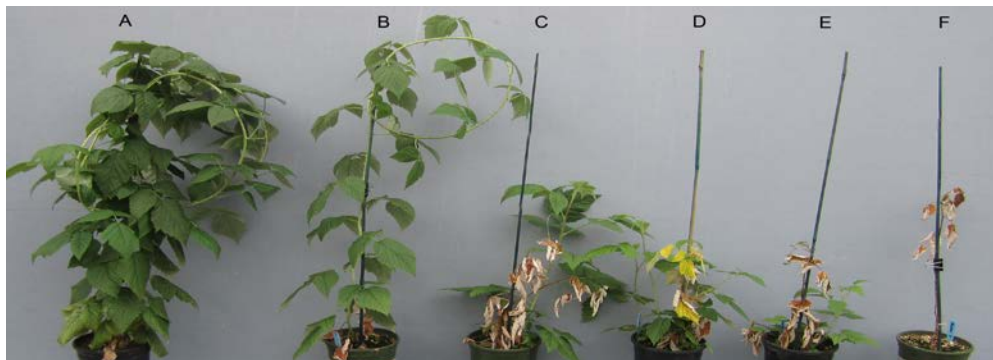
Anticipated Benefits and Information Transfer:

This research will identify if *V. dahliae* is widely prevalent and causing disease in Washington red raspberry production fields. It will also identify inoculum levels that are associated with disease and identify appropriate methods for *V. dahliae* detection and quantification. Results will lay the foundation for future research including disease screen assays for resistance among red raspberry cultivars and the evaluation of production (e.g. fertilization, irrigation, etc.) and disease management practices (fumigant rates, chemistries, etc.) for their effects on inoculum levels and disease development. Findings will also help growers interpret results from diagnostic laboratories. Our research results will be presented to red raspberry growers at meetings (Small Fruit Conference, Lynden) and communicated to the Washington Red Raspberry Commission, Peerbolt Crop Management, and Whatcom County Extension for inclusion in their newsletters.

References:

- Berkeley, G. H. and Jackson, A. B. 1926. Verticillium wilt of the red raspberry. *Scientific Agriculture* 6: 261-270.
- Bilodeau, G. J., Koike, S. T., Uribe, P., and Martin, F. N. 2012. Development of an assay for rapid detection and quantification of *Verticillium dahliae* in soil. *Phytopathology* 102: 331-343.
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- Duressa, D., Rauscher, G., Koike, S., Mou, B., Hayes, R. J., Subbarao, K. V., and Klosterman, S. J. 2012. A real-time PCR assay for detection and quantification of *Verticillium dahliae* in spinach seed. *Phytopathology* 102: 443-451.
- Fiola, J. A. and Swartz, H. J. 1994. Inheritance of tolerance to *Verticillium albo-atrum* in raspberry. *HortScience* 29: 1071-1073.
- Harris, R. V. 1925. The blue stripe wilt of the raspberry. *The Journal of Pomology and Horticultural Science* 4: 221-229.
- Markakis, E. A., Tjamos, S. E., Antoniou, P. P., Paplomatas, E. J., and Tjamos, E. C. 2009. Symptom development, pathogen isolation and real-time qpcr quantification as factors for evaluating the resistance of olive cultivars to *Verticillium* pathotypes. *European Journal of Plant Pathology* 124: 603-611.

Figure 1. Increasing severity of verticillium wilt symptoms (from left to right) on inoculated plants of ‘Meeker’ red raspberry in a 2012 greenhouse disease assay. A) healthy, noninoculated control plant. B) less vigorous growth of inoculated plant. C-E) stunted growth, wilting, cane death, and leaf yellowing on inoculated plants. F) entire plant died as a result of inoculation. Picture was taken two months after inoculation with *V. dahliae*.



Budget:

	2013	2014
Salaries^{1/}: Lab Technician	\$1630	\$1630
Salaries^{2/}: Sample collection	\$1175	\$1175
Shipping^{3/}: Samples	\$160	\$160
Culture-based sample processing	\$1000	\$1000
DNA-based sample processing	\$6000	\$6000
Misc sampling supplies^{4/}	\$200	\$200
Travel^{5/}	\$2430	\$2430
Meetings	\$200	\$200
Benefits^{6/}	\$787	\$787
Total	\$13,582	\$13,582

Budget Details

^{1/} One USDA Laboratory Technician at 0.05% FTE.

^{2/} One WSU Whatcom County Extension Personnel at 0.03% FTE.

^{3/} Shipping samples from Bellingham, WA to Corvallis, OR for processing.

^{4/} Sampling bags (500 @ \$0.40 = \$200).

^{5/} Travel to and from research sites, 3000 miles at \$0.51/mile (\$1530) for WSU Extension Personnel. Weiland will travel to Washington three times a year to assist in surveying, sample collection, and to present research findings at the Small Fruit Conference (\$900).

^{6/} Benefits USDA laboratory technician (16.6% = \$271) and WSU Extension Personnel (43.9% = \$516)

Other Sources of Funding: Dr. Weiland's base CRIS funds cover the salary of the PI as well as his technician (~\$42,500/year for 25% of their time). Additional base CRIS funds (~\$2000) were also used to cover expenses to isolate other potential soilborne pathogens from symptomatic cane tissue in 2013. Dr. Inga Zasada, USDA-ARS additionally contributed \$2500 in base CRIS funds to assess the 2013 soil and plant samples for the presence of plant parasitic nematodes. We expect to contribute the same amount of funds in 2014.

2013 Progress Report

for

Incidence and Detection of *Verticillium dahliae* in Red Raspberry Production Fields

Jerry Weiland, USDA-ARS
Chris Benedict, WSU Extension

Our research proposed to survey 25 diseased sites and 5 healthy sites in red raspberry production fields for the presence of *Verticillium dahliae*, a soilborne pathogen known to infect red raspberry plants. The purpose of the survey was to determine if the pathogen is widespread in the red raspberry industry and to determine if the pathogen is associated with wilting and dying plants. Our specific objectives were to:

1. Determine the incidence of *V. dahliae* in red raspberry production fields;
2. Determine if the presence of *V. dahliae* in soil and plant samples is associated with disease symptoms of wilting and cane death;
3. Determine which inoculum levels are associated with disease symptoms;
4. Compare culture-based and DNA-based methods for effectiveness of *V. dahliae* detection and quantification.

Beginning in June 2013, we began collecting soil and plant (cane) samples from diseased and healthy sites in red raspberry production fields. We finished sampling in September 2013. In all, 20 diseased sites and 3 healthy sites were sampled. Most sites were fairly dry and in locations where *Phytophthora rubi* was not expected to be a problem. A smaller number of sites were sampled than proposed due to the reduced budget. Two to five plant and soil samples were collected at each site. For the diseased sites, we tried to find plants that were actively wilting late in the growing season with canes that were just beginning to die. Most of these canes were at least 6 feet tall with dark reddish-black lesions spreading up the cane from the roots. This was opposed to the large number of shorter canes (less than 3 feet tall) that had died earlier in the season at the wetter locations. We did not collect many canes from these latter sites because they had been dead for more than a week, which makes it extremely difficult to isolate pathogens. We also assumed these shorter, dead canes were primarily affected by *Phytophthora rubi* earlier in the growing season and not by *V. dahliae* which tends to cause symptoms in late summer. After collecting, each plant and soil sample was divided into two samples, with one half of each sample to be tested by the culture-based method (isolation of *V. dahliae* on semi-selective media) and the other half of each sample to be tested by the DNA-based method (quantitative PCR).

To date, we have completed testing all plant samples with the culture-based method. Attempts to isolate *V. dahliae* from the soil samples are in progress, but were delayed due to the federal government furlough. Both plant and soil samples to be tested by the DNA-based method are currently in storage at -80°C while we refine our quantitative PCR protocol to ensure that it is working correctly. Therefore, all preliminary results (below) are from plant samples tested by the culture-based method only.

Of the 78 plants collected from 20 diseased sites, *Verticillium dahliae* was only isolated from one plant at 1 site. As mentioned above, results have not been confirmed with the DNA-based method, which may be much more sensitive to the presence of *V. dahliae* than the culture-based method. In addition, other fungi

may have outcompeted/overgrown *V. dahliae* in infected stems, making it less likely to be isolated on the semi-selective medium. Therefore, our priority this winter is to finish optimizing the DNA-based method in order to double check our results from the culture-based method. Results from both the culture-based and DNA-based methods on the soil samples will provide further evidence for the presence or absence of *V. dahliae* once they are completed. These latter assays may provide a better measure of the prevalence of *V. dahliae* in production fields, particularly if *V. dahliae* infection is restricted to the root region, which would make it less likely that we would have detected the pathogen in the cane samples that we collected during the survey. Finally, *V. dahliae* was also not isolated from any of the plants obtained from the three healthy sites.

Because of the low isolation rate of *V. dahliae* from red raspberry plants, we also decided to attempt to isolate other potential pathogens from the diseased plants we collected during the survey. Stem tissue was plated on a semi-selective medium for *Phytophthora* species (PARP) and on a general medium that allows the isolation of fungi. We attempted to isolate *Phytophthora* from plants at all 20 diseased sites. However, the decision to isolate other pathogenic fungi was made later during the survey. Therefore, we only have results for “other pathogenic fungi” from 11 sites. Finally, Dr. Inga Zasada also extracted nematodes from soil and root samples that were collected from the same 11 sites mentioned above. Results from the nematode analyses are in progress.

Of the 78 plants collected from the 20 diseased sites, *Phytophthora* (probably *P. rubi*) was isolated from 23 plants (29% of the plant samples) from 12 of the 20 (60%) diseased sites. *Fusarium* and *Alternaria* species were also consistently isolated from the lesions extending up from the root system of symptomatic plants. Of the 43 plants that we collected from 11 diseased sites, we isolated *Fusarium* species from the cane lesions at 10 sites and *Alternaria* species from cane lesions at 9 sites. Work is in progress to quantify the number of diseased plants from which we were able to isolate these fungi. Conversely, we rarely isolated these two fungi from 10 healthy plants from three sites, so they do appear to be associated with disease. Both *Alternaria* and *Fusarium* have been implicated as pathogens in other crops, but have not been extensively tested in red raspberry. Siva Sabaratnam at Agriculture and Agri-Food Canada did isolate *Fusarium* species in his survey of fungi from diseased red raspberry roots and found that they caused symptoms of root rot (unpublished). Because of the prevalence of *Phytophthora*, *Fusarium*, and *Alternaria* at many diseased sites, we added a fifth objective to the 2014 grant proposal to also assess the presence of plant parasitic nematodes, *Phytophthora*, and other potentially pathogenic fungi from diseased plants in red raspberry fields. We therefore are proposing to determine whether there is a disease complex present and to identify the key pathogens involved.

2014 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

New Project Proposal

Proposed Duration: 2 years

Project Title: Characterizing the soil ecology of red raspberry produced under different production regimes

PI: Inga Zasada

Organization: USDA-ARS Horticultural Crops Research Unit

Title: Research Plant Pathologist

Phone: (541)738-4051

Email: inga.zasada@ars.usda.gov

Address: 3420 NW Orchard Ave.

City/State/Zip: Corvallis, OR 97330

Co-PI: Christopher Dunlap

Organization: USDA-ARS Crop Bioprotection Research Unit

Title: Research Scientist

Phone: (309)681-6339

Email: christopher.dunlap@ars.usda.gov

Address: 1815 N. University St.

City/State/Zip: Peoria, IL 61604

Cooperator: Chris Benedict, WSU, Bellingham WA

Year Initiated 2014 **Current Year** 2014 **Terminating Year** 2015

Other sources of funding: Dr. Zasada's base CRIS funds cover the salary of the PI as well as her technician (~\$42,500/year for 25% of their time). Dr Dunlap's base CRIS funds cover the cost of the technician that performs the NGS library preparation and running the sequencing equipment. In addition, Dr Dunlap's lab will provide the DNA barcodes required for multiplexing samples, which saves \$100 a sample (\$16,000) relative to life technologies retail kit. No support is requested to offset \$120,000 in annual equipment maintenance agreements of the sequencing facility.

Description:

Understanding soil ecology and effects on plant health and productivity has long been a #1 research priority of the Washington Red Raspberry Commission. A unique opportunity has presented itself to enable the first look at raspberry soil ecology. Dr. Dunlap is the technical coordinator of the next-generation sequencing (NGS) facility at National Center for Agricultural Utilization Research in Peoria, IL. He recently contacted Dr. Inga Zasada offering his expertise in soil microbial ecology to address this issue for the raspberry industry. Dr Dunlap's leadership in this area has lead to a capital investment of over \$1 million in the facility and has greatly increased the technical capabilities of the center. Dr Dunlap has lead NGS projects on beneficial bacterial genomics and has international collaborations on microbial ecology (Brazil) and fungal

transcriptomics (Australia). We also now have two long-term raspberry experiments in Whatcom County established by Mr. Chris Benedict. These experiments provide an ideal venue in which to explore the impact of different raspberry production regimes (compost, nutrient management, cover crops) on soil ecology and raspberry productivity. This team of scientists will work together and begin to understand raspberry soil ecology and effects on plant health and productivity.

Justification and Background:

Soil health is driven by the individual and combined contributions of the physical, chemical and biological processes occurring in soil. From an agricultural perspective the services that are provided by the soil to sustain plant productivity include such things as nutrient availability, pest suppression, and increased water holding capacity. In perennial production systems there has been increasing interest in understanding soil health, and specifically soil microbiology and the role it may play in replant disorders (Mazzola, 1998; Yang et al., 2012). Based upon conversations with Washington raspberry growers there is a feeling that the raspberry production system may be experiencing similar replant issues as have been observed in other perennial production systems. Beginning to explore the ecology of microorganisms in the soil may provide insights into raspberry production systems that support unique microbial communities that encourage long-term productivity and economic viability. It may also be possible to identify microbial agents of disease that are not currently known or understood in raspberry.

Mazzola and Manici (2012) highlighted the dynamic ways by which a soil microbial community can function within a perennial cropping system. A microbial community can cause disease. Molecular methods have been used to identify differences in microbial communities between replant and nonreplant impacted orchards (Rumberg et al., 2007; Yang et al., 2012). Three different fungi, one oomycete, and a plant-parasitic nematode were found to contribute to apple replant disorder (Mazzola and Manici, 2012). Clearly different strategies will be required to manage such a diverse array of organisms. Molecular methods have also been used to identify microorganisms that provide valuable ecosystem services, such as disease suppression. Disease suppressive soils are exceptional ecosystems in which crop plants suffer less from specific soil-borne pathogens than expected owing to the activities of other soil microorganisms. Mendes et al. (2011), (Dr Dunlap's Brazilian collaborator) recently reported upon fungal root pathogen attack where plants can exploit the soil microbial consortia to prevent infection.

The rapid decrease in the cost of high-throughput DNA sequencing has opened new opportunities to study microbial ecology. **Figure 1** shows the cost of sequencing is 10,000x cheaper than six years ago. This technological revolution has permitted population metagenomics to

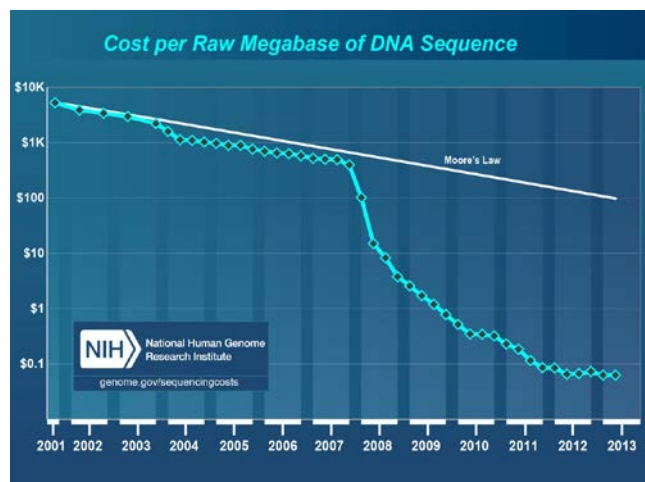


Figure 1. Cost per raw megabase of DNA sequence.

become a more routine experiment. Population metagenomics is an emerging molecular biology field to study environmental microbial populations by sequencing DNA extracted from the environment to quantify the species present. We will use this technology to explore soil and root microbial ecology in raspberry production systems.

Relationship to WRRRC Research priority(s):

This proposed research directly addresses a #1 priority of the WRRRC, understanding soil ecology and soil borne pathogens and their effects on plant health and crop yields.

Objectives:

1. Characterize microbial ecology (fungal, bacterial and nematode) under different raspberry production regimes, and;
2. Evaluate plant productivity (yield and disease) and relate that to microbial ecology.

Procedures:

We propose to evaluate microbial communities in two long-term on-farm trials established in Whatcom County by Mr. Chris Benedict. These trials have been established to compare different raspberry production regimes including irrigation, fertilizer, compost applications, and cover crops. For the purpose of this research we will compare the microbial communities in the roots and soil surrounding roots of raspberries produced conventionally (i.e. with standard fertilizer program) to those produced with a modified fertilizer program (either organic or based upon soil test data). Within the established experimental design samples will be collected from five replications of each treatment.

Soil and root samples will be collected at both sites in the spring and fall of 2014 and 2015. At each collection date, samples will be collected from plants in the center of plots. Cores, 4 x 4 x 4 inches, will be collected from two plants directly in the root zone (about 6 inches from the base of the plant). The samples containing roots and soil will be combined, placed in a plastic bag and transported to the laboratory. Once in the lab the samples will be partitioned to allow for the following analyses:

1. Soil and root microbial ecology – A subsample of soil and fine roots will be shipped overnight to Dr. Dunlap. A flow chart of sample handling is provided is **Appendix A**. After DNA extraction, PCR will be performed to generate 400 bp amplicons using standard primers for the 16S gene (bacterial) or ITS gene (fungal) for the samples. The resultant amplicons will be barcoded and prepared for NGS sequencing. The samples will be sequenced in our facility using Ion Torrent sequencing technology. The sequencing reads will be analyzed using MG-RAST metagenomics analysis server. The ratio of microbial species present will be determined and statistically analyzed.
2. Soil nematode ecology – The soil nematode community will be extracted from a 250 g subsample of soil by decant sieving Baermann funnel methods. The total number of extracted nematodes will be determined and nematodes will be identified to genus. Using this data and having knowledge of the ecological services provided by different types of soil nematodes, various ecological indices will be calculated do discern differences in nematode community assemblages under different raspberry production regimes.

Plant productivity will be evaluated by measuring the number and height of primocanes in July of both years. Dormant primocane assessment (diameter) will occur in later winter.

Timeline:

Activity	2014	2015
Sample soil and roots	April, Sept	April, Sept
Plant vigor assessment	July	Feb and July
Data processing and analysis	All year	All year

Anticipated Benefits and Information Transfer:

This research will provide the first information on soil ecology in red raspberry production systems. Our research will establish a baseline upon which to build. Plant genotype is believed to be the driving force in rhizosphere ecology; this baseline provides a method to compare raspberry cultivars. Another long term goal would be to understand the potential of disease suppressive soils in red raspberry production systems. Our research results will be presented to red raspberry growers at meetings (Berry Workshop, Lynden). Results will also be communicated to the Washington Red Raspberry Commission and to Peerbolt Crop Management for inclusion in their newsletters.

References:

Mazzola, M., Manici, L.M. Apple replant disease: role of microbial ecology in cause and control (2012). *Annual Review of Phytopathology* 50, pp. 45-65

Mendes, R., Kruijt, M., De Bruijn, I., Dekkers, E., Van Der Voort, M., Schneider, J.H.M., Piceno, Y.M., DeSantis, T.Z., Andersen, G.L., Bakker, P.A.H.M., Raaijmakers, J.M. Deciphering the rhizosphere microbiome for disease-suppressive bacteria (2011) *Science*, 332 (6033), pp. 1097-1100.

Rumberg, A. Merwin, I.A., Thies, J.E. Microbial community development in the rhizosphere of apple trees at a replant disease site (2007). *Soil Biology and Biochemistry* 39, pp. 1645-1654

Yang, J., Ruegger, P.M., McKenry, M.V., Becker, J.O., Borneman, J. Correlations between root-associated microorganisms and peach replant disease symptoms in a California soil (2012). *Plos One*, 7 (10), pp. 1-10.

Budget:

	2013	2014
Salaries	\$	\$
Administrative Professional	\$	\$
Time-Slip^{1/}	\$5,000	\$5,000
Operations (goods & services)^{2/}	\$12,400	\$12,400
Travel^{3/}	\$525	\$525
Other	\$0	\$0
Equipment	\$0	\$0
Benefits	\$0	\$0
Total	\$17,925	\$17,925

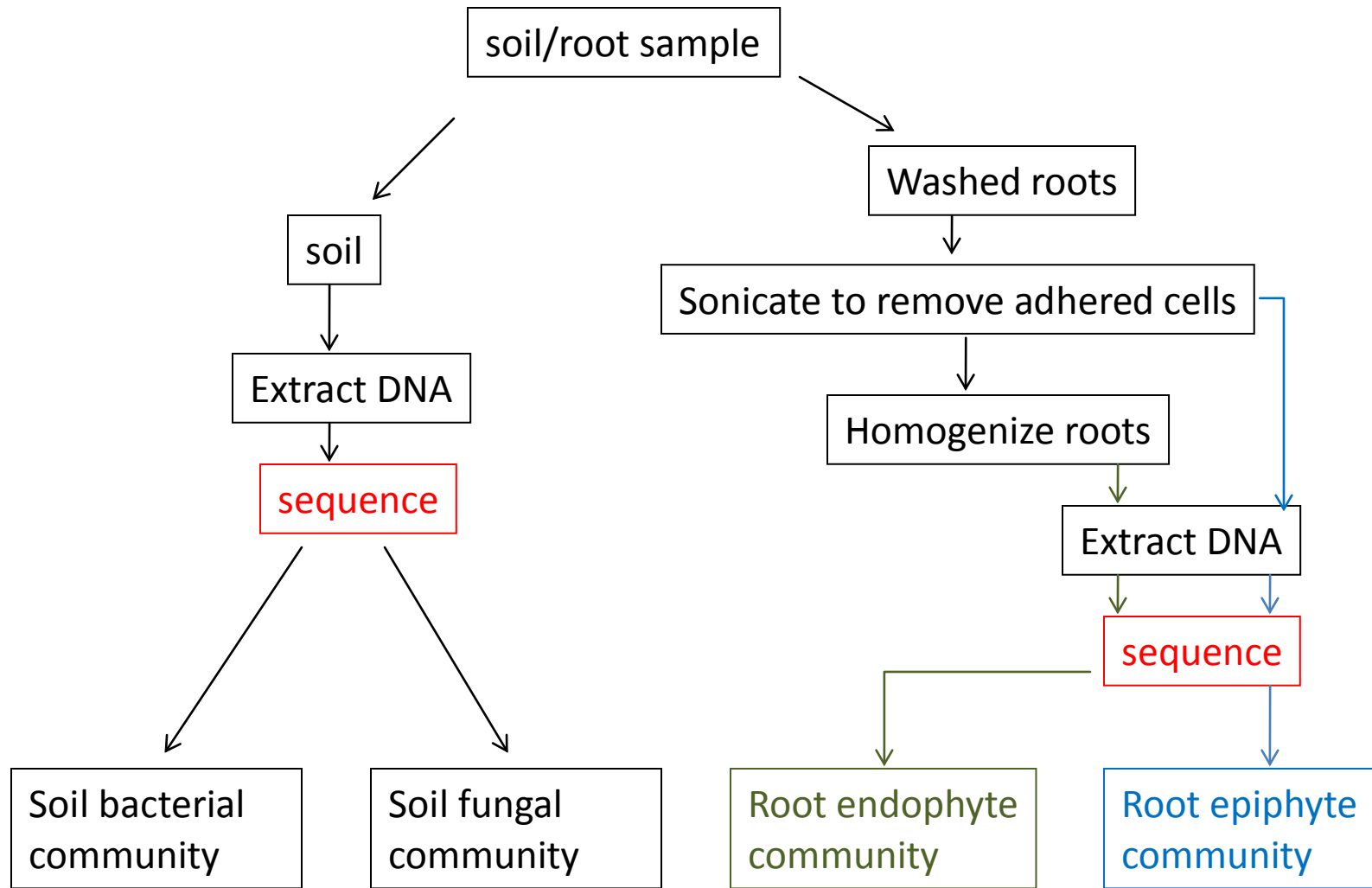
Budget Details

^{1/}One part time student will be hired in each year to support Dr. Dunlap's research efforts

^{2/}Soil and root samples will be processed for nematodes; a total of 40 samples/year at \$30/sample (\$2,400/year). Microbial community analysis for 40 samples, processed for four communities = 160 sequencing samples at \$62.5 sample (\$10,000/yr).

^{3/}Zasada will travel to Washington three times to help Benedict with research and to present research findings to the commission and at the WSU Small Fruits Workshop (\$175/trip).

APPENDIX A: Metagenomics sample handling



CURRENT & PENDING SUPPORT

Name: Christopher Dunlap

Instructions:

Who completes this template: Each project director/principal investigator (PD/PI) and other senior personnel that the Request for Applications (RFA) specifies

How this template is completed:

- Record information for active and pending projects, including this proposal.
- All current efforts to which PD/PI(s) and other senior personnel have committed a portion of their time must be listed, whether or not salary for the person involved is included in the budgets of the various projects.
- Provide analogous information for all proposed work which is being considered by, or which will be submitted in the near future to, other possible sponsors, including other USDA programs.
- For concurrent projects, the percent of time committed must not exceed 100%..

Note: Concurrent submission of a proposal to other organizations will not prejudice its review by CSREES.

NAME (List/PD #1 first)	SUPPORTING AGENCY AND AGENCY ACTIVE AWARD/PENDING PROPOSAL NUMBER	TOTAL \$ AMOUNT	EFFECTIVE AND EXPIRATION DATES	% OF TIME COMMITTED	TITLE OF PROJECT
David Shapiro- Ilan, Ted Cottrell, Dan Horton, Russ Mizell, Christopher Dunlap	Active: Sustainable Agriculture Research and Education Grant, SARE #: LS11- 241	\$9790	3/31/2012- 3/31/2014	2%	Enhancing Natural Enemy Systems: Biocontrol Implementation for Peachtree Borers
	Pending:				

Current and Pending Support – Inga Zasada

NAME (List/PD #1 first)	SUPPORTING AGENCY AND AGENCY ACTIVE AWARD/PENDING PROPOSAL NUMBER	TOTAL \$ AMOUNT	EFFECTIVE AND EXPIRATION DATES	% OF TIME COMMITTED	TITLE OF PROJECT
	Active:				
Tzanetakis et al.	USDA-NIFA-SCRI	1,463,234	9/2009 – 8/2014	5%	Management of virus complexes in Rubus
Walters, Moore, Zasada, Grunwald, Peerbolt, Bolda	USDA-NIFA-RAMP	831,869	9/2010 - 8/2014	10%	A new way of managing soil borne diseases of raspberry in western states: development of decision making tools and sustainable management systems
Moyer & Zasada	Washington State Grape & Wine Research	12,301	6/2013 - 5/2014	5%	Essential nematode data to enable decision-making by Washington grape growers
Zasada & Walters	Washington Red Raspberry Commission	9,000	4/2013 – 3/2014	5%	Fine-tuning vdate applications in red raspberry for <i>Pratylenchus penetrans</i> control
Ingham & Zasada	Oregon Potato Commission	10,000	5/2013 – 4/2014	5%	Pathogenicity of <i>Globodera ellingtonae</i> to potato
Navarre et al.	USDA-ARS	76,712	6/2013-5/2014	5%	Developing methods to eradicate PCN
	Pending:				
Dardarand et al.	USDA-APHIS	367,663	2013	10%	<i>Globodera</i> eradication research

2014 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

Continuing Project Proposal

Proposed Duration: 2 years

Project Title: Fine-tuning Vydate applications in red raspberry for *Pratylenchus penetrans* control

PI: Inga Zasada

Organization: USDA-ARS Horticultural Crops Research Laboratory

Title: Research Plant Pathologist

Phone: (541)738-4051

Email: inga.zasada@ars.usda.gov

Address: 3420 NW Orchard Ave.

City/State/Zip: Corvallis, OR 97330

Co-PI: Thomas Walters

Organization: Walters Ag Research

Title: Consultant

Phone: (360)420-2776

Email: waltersagresearch@frontier.com

Address: 2117 Meadows Lane

City/State/Zip: Anacortes, WA 98211

Cooperator: Norm McKinley, Dupont

Year Initiated 2013 **Current Year** 2014 **Terminating Year** 2014

Other sources of funding: During the last 5 years approximately half of my research program has been focused on nematode issues in Washington red raspberry and I have consistently addressed two of the five #1 priorities of the industry. This research endeavor has required extensive funding, most of which has come from USDA-ARS CRIS base funds, USDA-NIFA competitive grants, and the Washington Pesticide Commission. Over the past 5 years approximately \$1.3 million has been obtained in competitive grants to support our (Walters, Grunwald, Peerbolt, Moore, Zasada) work on soil borne pathogens and nematodes in red raspberry. In addition, over the past 5 years half of my salary plus my technicians has been leveraged to achieve this research for a total of approximately \$400,000; this does not include any of the infrastructure costs such as laboratory and greenhouse space. In addition we have found industry collaborators with Dupont funding the 2012 Vydate trail described in the Justification and Background section of this proposal (\$5,840).

Agency Name: None

Amt. Requested/Awarded: None

Notes: None

Description:

Recently, Vydate (oxamyl) was registered for use on non-bearing raspberry for the control of plant-parasitic nematodes in Washington, specifically the root lesion nematode, *Pratylenchus penetrans*. This means that Vydate can be applied to red raspberry at least 12 months prior to a harvest (EPA SLN No. WA-120005). This is an important addition to the raspberry growers' nematode management toolbox, and being able to maximize the efficacy of this product should provide growers with another way to control nematodes. *Pratylenchus penetrans* attacks the roots of raspberry and slowly causes feeder roots to die, limiting the capacity of the plant to uptake water and nutrients. If *P. penetrans* is not managed it can result in the reduced lifespan of a raspberry planting. We propose to conduct on-farm field trials to determine the best time in the spring after planting to apply Vydate to maximize nematode control. This research directly addresses a #1 priority of the Washington Red Raspberry commission, soil fumigant techniques and alternatives.

Justification and Background:

Root lesion nematodes, *Pratylenchus penetrans*, are migratory endoparasites, migrating between the soil and roots. This nematode is prominent in western Washington soils and has been shown to reduce raspberry vigor and yield (Gigot et al., accepted; Zasada and Walters, unpublished data). Currently, *P. penetrans* is managed in red raspberry by pre-plant fumigation with Telone C-35; there are few reliable post-plant nematicides registered for use in raspberry. We previously evaluated 13 post-plant nematicides for ability to suppress *P. penetrans* in raspberry (Zasada et al., 2010). Of the tested products only Vydate (oxamyl) and fosthiazate suppressed this nematode on raspberry plants. It is unlikely that fosthiazate will be labeled for us on red raspberry. However, Vydate was recently labeled for us on non-bearing raspberry for the control of nematodes in Washington (EPA SLN NO. WA-120005). This is a different type of use compared to that in Canada where Vydate is labeled for use on bearing plants; therefore it is essential that we identify the optimal way to apply Vydate to maximize nematode control.

In 2012, we conducted three on-farm trials with the goal of evaluating the phytotoxicity of Vydate to raspberry. Vydate was applied to either newly planted tissue cultured or dormant cane plants twice in May or June. Phytotoxicity was evaluated and soil samples were collected before treatment and soil and root samples collected in the fall for quantification of *P. penetrans* populations. Initially, there were very few nematodes detected in these fields with average populations of 0, 3, and 1 *P. penetrans*/100 g soil in Fields 1 (Wakefield), 2 (Chemainus), and 3 (Meeker), respectively. In the fall *P. penetrans* population densities in roots were significantly lower (indicated by * in **Figure 1**) in plots treated with vydate compared to the nontreated control. The same trend was

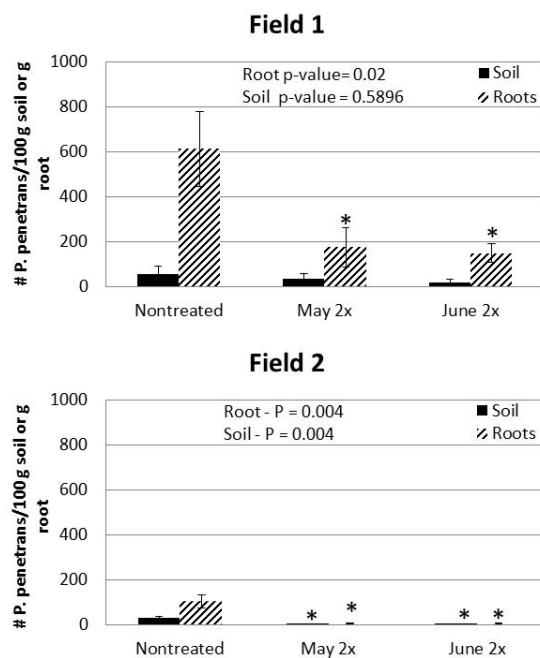


Figure 1. Effect of vydate applications in May and June on the suppression of *P. penetrans* in non-bearing red raspberry.

observed in Field 3 (data not shown), however population levels were low. Vydate was not phytotoxic to raspberry in these trials.

These results are significant because they demonstrate that a non-bearing application of Vydate has the potential to be an important nematode management tool for Washington raspberry growers. Currently the only application timing recommendation for Vydate is that soil temperature be at least 45 °F at a depth of 8 inches. The Vydate technical bulletin states “For perennial crops, the period of root growth is critical. Applications should be timed to coincide with periods of root flush or growth”. Nematode control by Vydate in relationship to root growth in northern Washington needs to be considered to maximize the efficacy of this product.

Relationship to WRRRC Research priority(s):

This proposed research directly addresses a #1 priority of the WRRRC, soil fumigant techniques and alternatives.

Objectives:

1. Optimize the use of Vydate for nematode suppression in non-bearing raspberry, and;
2. Continue to monitor nematode population dynamics in raspberry plants treated with Vydate in 2012 (**completed, see progress report for results**).

Procedures:

We propose to evaluate Vydate for the control of *P. penetrans* in non-bearing raspberry. All trials will be conducted in grower fields. During year 2 of this research we will monitor *P. penetrans* population densities in three field trials established in 2013. In 2013 the following treatments were established in a randomized block design with four replications in each field (total of 48 plots):

1. **Nontreated control**
2. **April Vydate 2x** – Vydate will be applied at 2 pints/acre twice in April; applications will be separated by 3 weeks. Due to reliable rainfall in April this is a very practical time of year since the application will not need to be watered in if applied prior to a rain. This treatment was not evaluated in 2012 trials.
3. **May Vydate 2x** – Same 2x rate as for April. Soil temperatures will be warmer in May and nematodes may be more active and root growth more abundant making this a better time to apply Vydate.
4. **June Vydate 2x** – Same 2x rate as for April. Soil temperatures will be even warmer than in May, however, rainfall will not be as reliable as in April/May making it necessary to follow the application with 0.25 or 0.50 acre inch of water.

Soil and root samples will be collected again in the spring and fall of 2014 from plots. Roots will be separated from soil and nematodes will be extracted from roots and soil and quantified. Plant vigor (cane height and number of canes) will be evaluated in July 2014.

Timeline:

Activity	2013 (completed)	2014
Sample 2012 trials for nematodes	April, Sept	

Sample 2013 trials for nematodes	April, Sept	April, Sept
Apply 2013 Vydate applications	April, May, June	
Evaluate 2012 trials for phytotoxicity	July	
Evaluate 2013 trials for phytotoxicity	July	July

Anticipated Benefits and Information Transfer:

This research will provide raspberry growers with information to maximize the efficacy of non-bearing applications of Vydate for *P. penetrans* control. Our research will also provide information about potential phytotoxic side effects of Vydate to newly planted raspberry. Two applications of Vydate at 2 quarts/acre will cost a grower approximately \$150/acre (chemical plus labor); our research will ensure that this is money well spent. Our research results will be presented to red raspberry growers at meetings (Berry Workshop, Lynden) and the annual WSU-NWREC field day. Results will also be communicated to the Washington Red Raspberry Commission and to Peerbolt Crop Management for inclusion in their newsletters.

References:

Gigot, J., Walters, T.W., and Zasada, I.A. Impact and Occurrence of *Phytophthora rubi* and *Pratylenchus penetrans* in commercial red raspberry (*Rubus idaeus*) fields in northwestern Washington. International Journal of Fruit Science (submitted).

Zasada, I.A., Walters, T.W., and Pinkerton, J.N. 2010. Post-plant nematicides for the control of root lesion nematode in red raspberry. HortTechnology 20:856-862.

Budget:

	2013	2014
Salaries	\$0	\$0
Administrative Professional^{1/}	\$2,040	\$2,450
Time-Slip	\$780	\$0
Operations (goods & services)^{2/}	\$5,760	\$2,880
Travel^{3/}	\$1,000	\$553
Other	\$0	\$0
Equipment	\$0	\$0
Benefits	\$867	\$0
Total	\$10,447	\$5,883
	(received \$9000)	

Budget Details

^{1/}Tom Walters will be responsible for the day-to-day management of this research.

^{2/}A total of 92 soil/root samples will be collected in 2014. Cost of processing a single sample is \$30.

^{3/}Walters 3 trips, Anacortes – Lynden to collect samples (\$68/trip). Zasada will travel to Washington twice to help Walters with research and to present research findings to the commission and at the WSU Small Fruits Workshop (\$175/trip).

Washington Red Raspberry Commission Progress Report

Title: Fine-tuning Vydate applications in red raspberry for *Pratylenchus penetrans* control

Personnel:

Inga Zasada, USDA-ARS HCRU and Thomas Walters, Walters Ag Research

Reporting Period: 2013

Accomplishments:

Identified that late spring (May or June) applications of Vydate to non-bearing raspberry are the most effective at reducing population densities of *P. penetrans* in raspberry roots.

Results:

2012 Vydate Trials. Three trials were established in newly planted raspberry fields in 2012. Field 1 was planted to the variety Wakefield, field 2 to Chemainus and field 3 to Meeker. At each location, 12 plots were established. To each of four plots, Vydate was applied at a rate of 2 pints/acre twice in May and twice in June. The remaining four plots at each location served as a nontreated control. *Pratylenchus penetrans* population data in soil and roots was determined in spring and fall of 2012 and 2013. In addition, plant vigor was assessed in July of both years to determine if Vydate is phytotoxic to young raspberry plants. In Fall 2012, regardless of application timing, Vydate reduced *P. penetrans* population densities in roots in all fields (**Figure 1**). The length of protection appeared to be driven by the *P. penetrans* pressure present in a field. In field 1, which had the greatest nematode pressure, suppression was only observed 5-6 months after Vydate application. In field 2, which had moderate nematode pressure, protection of plants from *P. penetrans* was observed up to 10-11 months post application. In the field that had the lowest nematode pressure suppression of *P. penetrans* persisted 1.5 years post application. Vydate was never found to be phytotoxic to the tested raspberry varieties.

2013 Vydate Trials. Three trials were established in 2013 in newly planted fields of Wakefield (fields 4 and 5) and Meeker (field 6). These trials included the same treatments as the 2012 trials: two May applications, two June applications, and a nontreated control. An additional treatment of April Vydate applications was included; at all dates Vydate was applied similar to that described above. The first April application was timed to occur when soil temperatures were above 45 °F, followed by a second application three weeks later (the recommendation on the label). Nematode population dynamics and plant vigor was determined as in 2012. The soil and root *P. penetrans* population data is presented in **Figure 2**. In field 4 significantly better *P. penetrans* suppression was achieved with a June application of Vydate compared to the other treatments. In the other two field where *P. penetrans* populations were much lower, there was a nonsignificant trend for greater nematode suppression with May and June Vydate applications compared to April and nontreated. Similar to 2012, no phytotoxicity was observed.

Conclusion:

Vydate should be applied to non-bearing raspberry in May or June to achieve maximum *P. penetrans* suppression.

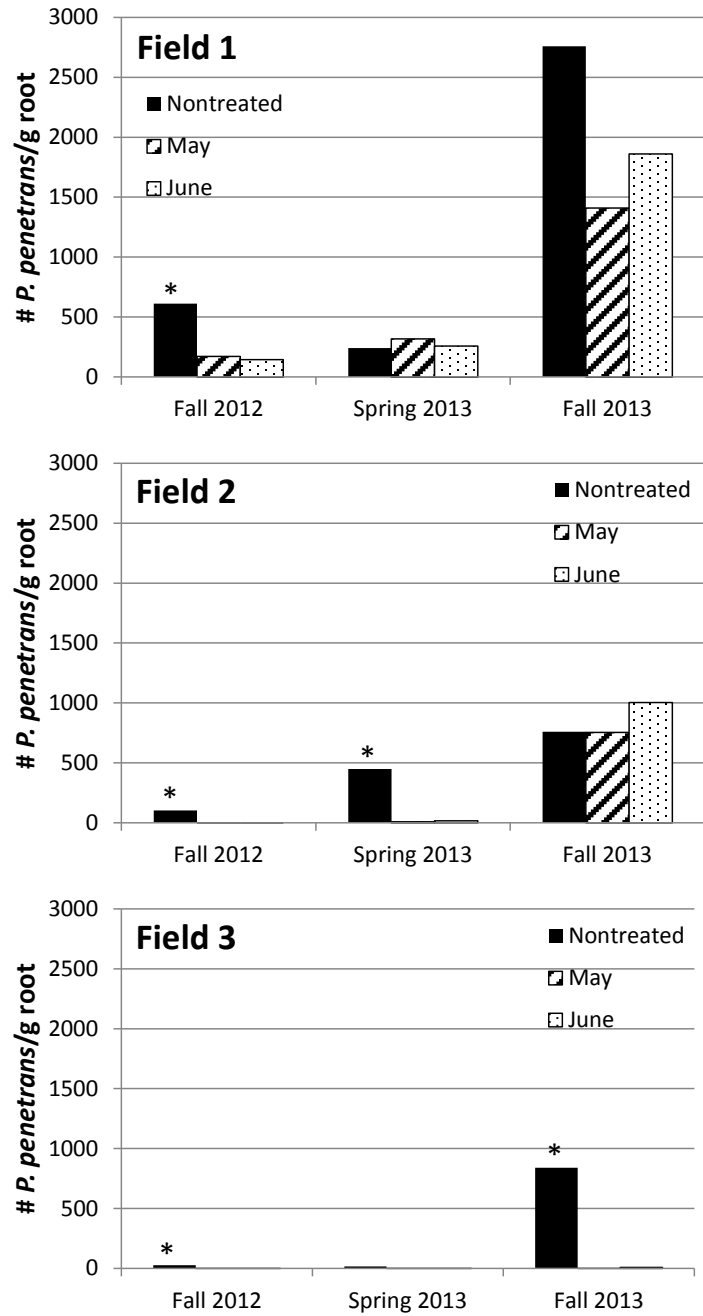


Figure 1. Effect of application timing of vydate for the suppression of *Pratylenchus penetrans* in red raspberry. Bars with a * are significantly different from other treatments within a sampling time ($P < 0.05$).

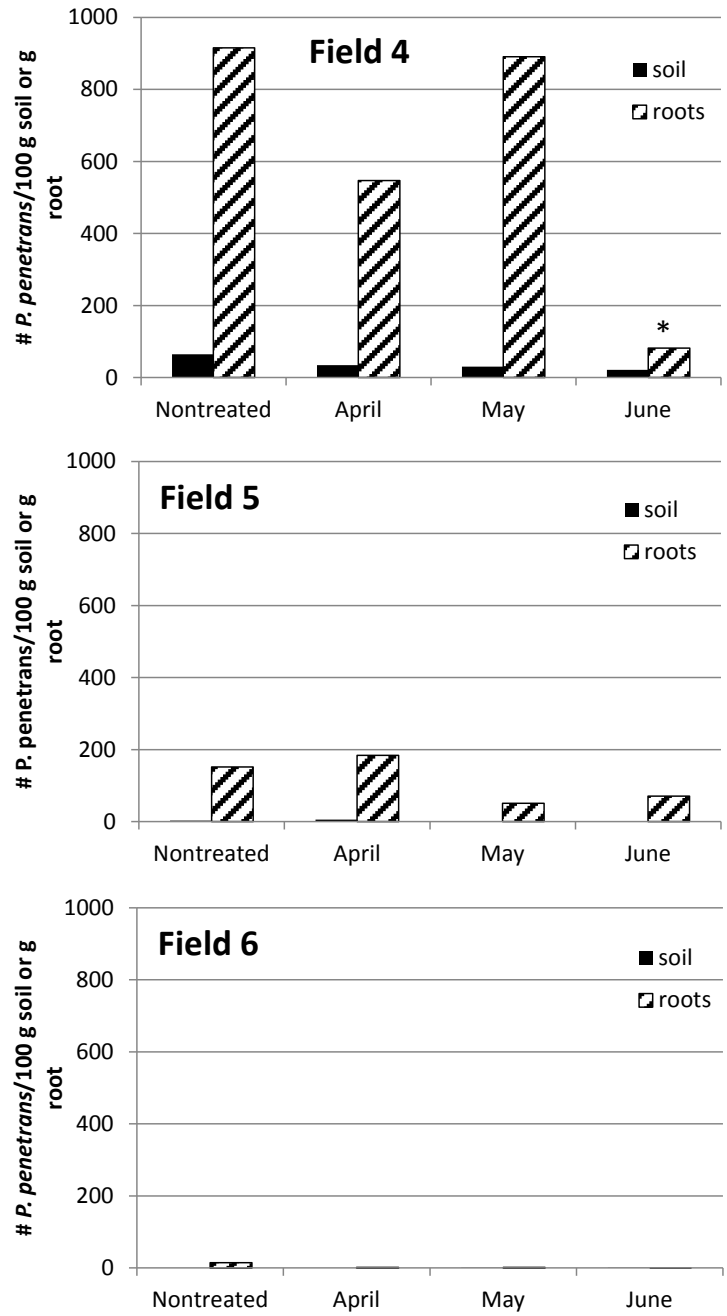


Figure 2. Effect of application timing of vydate for the suppression of *Pratylenchus penetrans* in non-bearing red raspberry. Bars with a * are significantly different from other sampling dates within a sample type (roots or soil) ($P < 0.05$).

2014 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

New Project Proposal

Proposed Duration: 3 years

Project Title: Effectors as a new strategy to control plant-parasitic nematodes

PI: Axel Elling

Organization: Department of Plant Pathology, Washington State University

Title: Assistant Professor

Phone: (509) 335-3742

Email: elling@wsu.edu

Address: P.O. Box 646430

City/State/Zip: Pullman, WA 99164

Year Initiated 2014 **Current Year** 2014 **Terminating Year** 2017

Total Project Request: **Year 1** \$13,872 **Year 2** \$14,427 **Year 3** \$15,005

Other funding sources:

Agency Name: Northwest Potato Research Consortium (NPRC), WA Grain Commission (WGC)

Amt. Requested/Awarded: Three-year proposals have been submitted to both NPRC (\$200,000) and WGC (\$90,000). Both funding agencies provided funds for year 1 (see Current & Pending Support) and are currently considering funding requests for year 2.

Notes: WSU is including this information on other resources 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.

Description:

Plant-parasitic nematodes have evolved highly sophisticated interactions with their hosts that are based on secretions from the nematode's salivary glands. These secretions (*i.e.*, effectors) are released into the plant during the infection process and enable the nematode to overcome plant defense responses (Mitchum et al. 2013). The **objective** of this project is to study how nematode effectors manipulate plant defense mechanisms and to identify weak links in the plant-nematode interaction that can be developed into new control strategies. A specific **outcome** of this project will be a list of nematode and plant genes that play key roles in the infection process and that make attractive new targets for control strategies. Additionally, we will gain a better understanding of how nematodes infect plants and how the infection process can be interrupted.

Justification and Background:

The root-lesion nematode *Pratylenchus penetrans* is a widespread and economically important pest in Washington's raspberry production areas (McElroy 1992). Current nematode control strategies strongly depend on pre-planting applications of synthetic nematicides, but recent decisions by the EPA make it likely that fewer nematicides will be available going forward. Non-

fumigant nematicides are always under regulatory threat because they are generally older, acutely toxic products. Alternative control strategies are very limited: rotations are not a feasible option in perennial crops, and resistant cultivars, which could be one of the most economic and sustainable control tactics, are not available. Therefore, the urgent development of new control methods is necessary to prevent significant yield losses due to *P. penetrans* infestations in the future. Current strategies to develop alternative control tactics against *P. penetrans* in raspberries are aimed at testing new synthetic nematicides and cover crops/green manure. The research proposed here will complement ongoing efforts to control plant-parasitic nematodes in raspberries as part of an integrated pest management program.

Plant-parasitic nematodes release secretions produced in their salivary glands into host plants during infection. These secretions degrade plant cell walls, thereby enabling nematode invasion and interfere with the normal functions of host plant physiology. Plant-parasitic nematodes depend on these secretions for their survival and their ability to establish themselves as parasites, which makes these secretions central to their infection strategy and an attractive target for new control methods. Recent studies have shown that disabling the nematode genes (effector genes) that produce these secretions can result in dramatically increased resistance of the host plant (Huang et al. 2006, Elling et al. unpublished). This can be achieved through biotechnology, traditional breeding or new chemicals. In a recently expired project that was funded by this commission, we identified putative effector genes in *P. penetrans*. As a direct extension of these efforts, we are now well positioned to study the roles effectors play in the infection process.

We propose to exploit a weak link in the infection strategy of plant-parasitic nematodes, namely their dependency on effectors to develop new control strategies against nematodes. In order to use nematode effectors as new control targets, their function and the plant genes they interact with need to be characterized, which is the goal of this project. No similar research is conducted in Washington, Oregon or Idaho.

Relationship to WRRRC Research Priorities:

This project will improve our understanding about plant-parasitic nematodes and can lead to the development of new control strategies. This project directly addresses two #1 WRRRC priorities: *i*) understanding soil ecology and soil borne pathogens and *ii*) alternatives to control soil pathogens and nematodes.

Objectives:

- 1. Identify plant proteins that interact with nematode effectors.***
- 2. Study the role of nematode effectors during the infection process.***

We anticipate that the entire project will take 3 years to complete. In year 1, we will focus on the first objective. Once plant proteins that are targeted by the nematode are identified, we will study the role of the nematode effector and plant protein in this interaction, which we expect to take up the majority of years 2 and 3.

Procedures:

- 1. Identify plant proteins that interact with nematode effectors.***

Nematode effectors disable plant defenses and interfere with physiological processes of plant cells. Effectors achieve this by binding to host plant proteins and altering their functions. To date, it is largely unknown with which plant proteins nematode effectors interact. It is important

to determine to which host plant proteins nematode effectors bind because this will allow us to disrupt these interactions, which will lead to increased nematode resistance. For this project we will study root-knot nematode effectors. Previous research showed that root-lesion and root-knot nematodes are very closely related and that many of their effectors are the same. Root-knot nematodes offer the advantage of a more rapid reproduction cycle, which significantly facilitates and speeds up the proposed research. Findings will be transferred to root-lesion nematodes at the conclusion of this project.

A widely used method to study protein-protein interactions is the yeast two-hybrid assay, in which a known protein binds to an unknown protein, which can then be studied. We have previously identified nematode effectors through a WRRC-funded project, which now allows us to determine which unknown plant proteins bind to known nematode effectors. Once we have identified nematode and plant proteins that interact with each other in yeast, we will confirm that this interaction also occurs in plants. Once we know what plant proteins are manipulated by the nematode, we can develop strategies to protect them from nematode attack in future projects and in collaboration with breeders.

2. Study the role of nematode effectors during the infection process.

During the infection process, nematodes secrete a mix of effectors into the plant. The quantities of the secreted effectors are minute and their effects are very difficult to study if we were to imitate the nematode on a 1:1 scale. Therefore, we will break down this complex interplay into a more manageable system. Once we know to which plant proteins nematode effectors bind, we will study single pairs of interacting nematode effectors and plant proteins. Specifically, we will drastically increase the quantity of the effector (and associated plant protein) in question. This will enable us to magnify the effect of a single effector and understand its function during the infection process by analyzing changes in the appearance of the plant and/or the nematode's ability to infect, thereby leading to new control tactics.

Anticipated Benefits and Information Transfer:

This research will **benefit** growers and the raspberry industry because it will aid in developing new cultivars that are resistant against nematodes and identify new control targets in the nematode. This will benefit growers because it will minimize the need for costly, time-consuming and potentially toxic application of currently available chemicals to control nematodes.

Information transfer will be based on presentations at the WA Small Fruit Conference, pesticide recertification days and various grower and crop consultant meetings. Regular updates will be sent to the commission. In addition, we will publish results in peer-reviewed journals and trade magazines.

References:

Huang, G., Allen, R., Davis, E.L., Baum, T.J., and Hussey, R.S. 2006. Engineering broad root-knot resistance in transgenic plants by RNAi silencing of a conserved and essential root-knot nematode parasitism gene. *Proc. Natl. Acad. Sci. USA* 103:14302-14306.

McElroy, F.D. 1992. A plant health care program for brambles in the Pacific Northwest. *Journal of Nematology* 24:457-462.

Mitchum, M.G., Hussey, R.S., Baum, T.J., Wang, X., Elling, A.A., Wubben, M. and Davis, E.L.

2013. Nematode effector proteins: an emerging paradigm of parasitism. *New Phytologist* 199:879-894.

Budget:

	2014	2015	2016
Salaries	\$0	\$0	\$0
Time-Slip^{1/}	\$6,720	\$6,989	\$7,269
Operations (goods & services)	\$6,000	\$6,240	\$6,490
Travel^{2/}	\$300	\$320	\$341
Meetings	\$200	\$200	\$200
Other	\$0	\$0	\$0
Equipment	\$0	\$0	\$0
Benefits^{3/}	\$652	\$678	\$705
Total	\$13,872	\$14,427	\$15,005

Budget Justification

^{1/}Undergraduate student at close to minimum wage (\$10.00/hour for 12 hrs/week for 56 weeks).

^{2/}Travel to Small Fruit Conference in Lynden and other grower meetings. In addition, a modest contribution of \$200/year is requested to attend national scientific meetings, which directly benefit this project because they provide the best way to receive critical feedback and set up new collaborations with researchers in the field.

^{3/}Regular benefits for temporary employees at 9.70% as mandated by WSU.

Washington Red Raspberry Commission Progress Report Format for 2013 Projects

Project No: 3061-4744

Title: New strategies to control root-lesion nematodes

Personnel: Dr. Axel Elling, Department of Plant Pathology, Washington State University, Pullman, WA 99164, Phone (509) 335-3742, Email: elling@wsu.edu

Reporting Period: Final report

Accomplishments:

The root-lesion nematode *Pratylenchus penetrans* is a significant pest in raspberries in the Pacific Northwest. Its control is largely limited to the application of pesticides, which are costly and potentially harmful to the environment and applicator. Increasing EPA-mandated restrictions necessitate new nematode control strategies. However, the development of novel control tactics is hampered by a lack of knowledge about genes that the nematode requires for infection and survival. Prior to this study, the available genetic information about *P. penetrans* was limited to a relatively small survey (Mitreva, Elling et al. 2004, *Mol. Genet. Genomics* 272:138-148). **With the completion of this project, we have for the first time an overview of almost all *P. penetrans* genes and are able to predict which genes are in all likelihood involved in plant-nematode interactions.** This is an important advance because it not only significantly increases our understanding about root-lesion nematode infection strategies but also sets the stage for the development of new nematode control tactics that have the potential to reduce the use of pesticides and could translate into substantial cost savings for producers.

Results:

Plant-parasitic nematodes produce effector proteins in their salivary glands from where they are secreted into plant cells during the infection cycle. Effector proteins are critically important for the nematode's ability to establish itself as a parasite and to survive and reproduce in its host plant. This project has provided the first complete analysis of almost all *P. penetrans* genes. We used high-throughput DNA sequencing techniques to create a catalog of *P. penetrans* genes. Using bioinformatic analyses, we compared *P. penetrans* genes to those of other plant-parasitic nematodes to identify genes that are only found in *P. penetrans* and others that are found in many different plant-parasitic nematode species. This is important because it allows us to target genes that are of fundamental importance to nematode infection and that could lead to a broad-band nematode resistance. We are specifically interested in nematode effectors and found about 350 *P. penetrans* genes that represent effector candidates. We are currently trying to narrow down this list further. Interestingly, we discovered that a significant number of *P. penetrans* effector candidates are similar to plant genes, which suggests that they are involved in changing physiological pathways in plants. This is an important finding because it provides an excellent starting point for future resistance breeding projects that would aim at counteracting the changes the nematode is trying to accomplish within host plant cells. In summary, this project has provided a list of *P. penetrans* effector gene candidates whose role in the infection process can now be

characterized. Once their roles are understood, it will be possible to disrupt their functions and to increase nematode resistance in plants.

Publications:

Peer-reviewed journal articles

Mitchum, M.G., Hussey, R.S., Baum, T.J., Wang, X., Elling, A.A. and Davis, E.L. 2013. Nematode effector proteins: An emerging paradigm of parasitism. *New Phytologist* 199:879-894.

Kandel, S.L., Smiley, R.S., Garland-Campbell, K., Elling, A.A., Abatzoglou, J., Huggins, D., Rupp, R. and Paulitz, T.C. 2013. Relationship between climatic factors and distribution of *Pratylenchus* spp. in the dryland wheat production areas of eastern Washington. *Plant Disease* 97:1448-1456.

Elling, A.A. and Jones, J.T. 2014. Functional characterization of nematode effectors in plants. In: *Plant-pathogen interactions: Methods and protocols*, second edition, eds. Birch, P., Jones, J. and Bos, J.I.B., *Methods in Molecular Biology* (in press).

Elling, A.A., Rosa, B. and Mitreva, M. 201-. Comparative analysis of the *Pratylenchus penetrans* transcriptome. (Manuscript in preparation)

Abstracts/Posters

Rosa, B., Elling, A.A. and Mitreva, M. 2013. Comparative, functional genomic analysis of plant-parasitic nematodes. (Presentation at Genome Meeting in St. Louis, MO)

Popular articles

Kantor, S. 2014. Discovering an unseen threat to agriculture. (Article featuring Dr. Elling's work, currently under consideration for publication in *WSU Magazine* and *Discover*).

Paulitz, T.C., Elling, A.A., Garland-Campbell, K. and Smiley, R.S. 2012. Rooting out the problem: unseen nematodes take a visible bite out of profits. *Wheat Life* 55:66-69.

Other outputs

Shutting the door on nematodes: the role of effectors. (Presentation at Small Fruit Conference, Lynden, WA, December 6-7, 2012)

Lessons from a plant-parasitic nematode: genomes and host-parasite relationships. (presentation in Department of Biology, University of Idaho, Moscow, ID, May 1, 2012)

Things to know about nematodes. (Presentation at Annual Basin Producers Pesticide Recertification Day, Moses Lake, WA, January 21, 2011)

New control strategy against root-lesion nematodes. (Presentation to Washington Red Raspberry Commission, Lynden, WA, December 6, 2010)

Sequence data for *Pratylenchus penetrans* (All sequence data will be uploaded into public databases)

**2014 WASHINGTON RED RASPBERRY COMMISSION
RESEARCH PROPOSAL**

New Project Proposal

Proposed Duration: 3 years

Project Title: Biology and control of *Botrytis* fruit rot of red raspberry

PI: Tobin L. Peever

Organization: Department of Plant Pathology, Washington State University

Title: Associate Professor

Phone: 509-335-3754

Email: tpeever@wsu.edu

Address: P.O. Box 646430

City/State/Zip: Pullman, WA 99164-6430

Year Initiated 2014 **Current Year** 2014 **Terminating Year** 2016

Total Project Request: Year 1 **\$21996** Year 2 **\$22596** Year 3 **\$23220**

Other funding sources: None

Description

The objective of this project is to improve management of *Botrytis* fruit rot of raspberry. Despite intensive fungicide application programs aimed at control of this disease in the US PNW, it is estimated that fruit losses and downgrades in fruit quality exceed 25% of the harvestable fruit due to incomplete disease control. Additionally, fungicides used for control are losing effectiveness due to the development of resistance, further limiting management options. Applications of fungicides in the PNW are currently timed on a calendar basis rather than according to inoculum availability and infection risk largely because the life cycle of the pathogen and inoculum sources are poorly understood. Specific **outcomes** of this project will include a detailed study of the disease cycle of *Botrytis cinerea* infecting raspberry and identification of sources of inoculum of the pathogen. Simultaneously, we will conduct a survey for resistance to fungicides currently used to *Botrytis* fruit rot control. This will provide important baseline data necessary for the design of control strategies to minimize the development of fungicide resistance and improve disease control.

Justification and Background

Current control strategies for *Botrytis* fruit rot of raspberry in WA involve up to 7 fungicide sprays at an average cost of \$75 per application per acre. Despite this intensive fungicide application schedule, fruit losses average 20-25% suggesting that much improvement in disease control is possible. The intensity of fungicide spray programs currently used to control *Botrytis* fruit rot in raspberry, coupled with the appearance of fungicide resistant isolates, suggests that improved control strategies are needed. Reducing the number of fungicide applications while maintaining or improving disease control will improve profitability for

producers and reduce selection pressure for resistance. Improved Botrytis fruit rot control will depend on a much greater understanding of the life cycle of the pathogen and a better understanding of the current levels of resistance to fungicides used for Botrytis fruit rot control and control of other fungal pathogens in PNW raspberries.

Botrytis fruit rot results from infections of mature raspberry fruit by *B. cinerea* but the initial source of the pathogen is thought to be latent or quiescent infections of floral parts (Dashwood & Fox 1988, Jarvis 1962). The fungus infects open flowers but remains latent before moving into developing fruit. The majority of fruit infections of both strawberry and raspberry appear to result from latent infections (Bristow et al. 1986, Jarvis 1962). Open flowers are rapidly colonized by *B. cinerea* and necrotic stamens and styles are an important source of inoculum for fruit rot (Dashwood & Fox 1988, McNicol et al. 1985). A fungicide timing study in raspberry showed that restriction of fungicide applications to the bloom period resulted in more Botrytis fruit rot compared to sprays applied throughout season (Ellis et al 2008). This result may indicate that the infection window for raspberry is longer than that of strawberry and not necessarily restricted to the flowering stage. The effect of machine harvesting on Botrytis fruit rot of raspberry has not been investigated but it has been suggested that successive machine harvestings may wound developing fruit allowing airborne Botrytis to invade fruit from the exterior or allowing Botrytis spores on the surface of ripening fruit a chance to penetrate the fruit (McNicol et al. 1990, Williamson & McNicol 1986).

Botrytis cinerea also has the ability to infect raspberry leaves and canes (Xu et al 2009, Williamson et al. 2007) and this phase of the disease is known as “cane Botrytis” (Williamson et al. 2007). Xu et al (2009) found that older canes were more susceptible to infection than younger canes and that most cane infection resulted from direct infection of canes rather from leaves through petioles as suggested by Williamson et al. (2007). Cane blight can be highly destructive to floricanes and may reduce lateral shoot formation (Williamson & Hargreaves 1981). Infected canes also provide a potential source of inoculum for the fruit rot phase of the disease (Williamson et al. 2007). Another potential source of inoculum for fruit rot epidemics are pruned floricanes from the previous year that are left within rows and eventually incorporated into the soil. Based on the biology of *B. cinerea* and other similar fungi, it is highly likely that sclerotia and mycelium of *B. cinerea* could survive in these senescent canes on the soil surface and provide a source of inoculum for Botrytis fruit rot epidemics.

Five classes of fungicides are currently registered for control of *Botrytis cinerea* on small fruit worldwide (Williamson et al. 2007) and in the US PNW (Coyne et al. 2012). Most classes of modern fungicides are at high risk for resistance development due to their specific modes of action. Intensive spray programs are expected to select strongly for resistance. *B. cinerea* sampled from German strawberry fields were resistant to up to 6 different fungicides (Leroch et al. 2013) and 15 to 80% of *B. cinerea* isolates sampled from raspberry, blueberry, currants and strawberry fields in northern Germany were each resistant to one of five different fungicides and 18% were resistant to all five fungicides (Weber 2011). These results indicate that fungicide resistance is a severe problem in small fruit production and that measures are required to reduce selection pressure in order to extend the useful lifespan of these chemicals. Currently there is no published data on levels of resistance to fungicides used to control Botrytis fruit rot and other

fungus diseases of raspberry in the US PNW but anecdotal reports suggest reductions in efficacy of certain products.

Relationship to WRRC Research Priorities

This research project addresses one of the #1 priorities of the WRRC namely “*Fruit rot including pre harvest, post harvest, and/or shelf life*”.

Objectives

- 1) Identify and quantify sources of inoculum for *Botrytis* fruit rot epidemics
- 2) Study dynamics of fruit infection by *Botrytis cinerea* during growing season particularly in relation to machine harvesting
- 3) Determine baseline resistance levels to fungicides commonly used for control of *Botrytis* fruit rot

Procedures

1) Identify and quantify sources of inoculum for *Botrytis* fruit rot epidemics

Raspberry tissues will be sampled for *Botrytis cinerea* at several times throughout the year in order to identify potential sources of inoculum. Both necrotic and live tissues will be sampled and samples will include senescent leaves and stems on soil beneath canopy, leaves on primocanes and floricanes, primocanes and floricane stems, pruned floricanes between rows, flowers and fruits. Samples will include both symptomatic and asymptomatic tissues. Tissues will be removed from raspberry plants in 4-6 commercial production fields approximately 6 times per year including dormant periods and active growth periods. Samples will be surface disinfested and placed in moist chambers to induce sporulation (Boyd-Wilson et al. 2013) and also plated on agar media containing antibiotics (Dashwood & Fox 1988) to isolate the pathogen. Colonies emerging from tissues will be identified using morphological and molecular methods. Numbers of samples scored as positive for *B. cinerea* for both the sporulation and isolation assays will be recorded for each tissue type at each sampling period in order to identify and quantify inoculum sources. Isolates will be placed in long-term storage for further study. The dynamics of *B. cinerea* infection during the harvest season will focus on sampling ripe fruit during harvest and assaying for the pathogen. Remaining ripe fruit left behind during successive harvests will be assayed for infection. Fruit will be sectioned and plated on agar medium amended with antibiotics (Dashwood & Fox 1988) and the percentage of fruit infected with *B. cinerea* isolated recorded.

2) Resistance of *Botrytis cinerea* to commonly used fungicides

Isolates obtained and stored in pure culture under Objective 1 will be screened for resistance to currently registered and commonly used fungicides in raspberry production in the US-PNW. Conidial germination, germ tube elongation and mycelial growth assays on

discriminatory concentrations of technical grade fungicides will be employed as is commonly used to assay fungicide resistance in *B. cinerea* (Leroch et al 2013, Weber 2011). Levels of resistance and frequencies of isolates in each resistance category will be recorded as well as frequencies of cross-resistance to multiple fungicides.

Anticipated Benefits and Information Transfer

This research will address critical gaps in our knowledge of the disease cycle of *Botrytis cinerea* causing Botrytis fruit rot of raspberry in the US-PNW and provide important baseline data on the status of fungicide resistance in raspberry fields. The identification and quantification of sources of inoculum may result in modified cultural practices and will allow more precise and effective timing of fungicide applications allowing producers to move away from calendar-based spray schedules in favor of more biologically-based application schedules. The specific effects of fungicide spray timing will be the focus of future studies and will build on knowledge of the life cycle of the pathogen learned from this research. Improved timing will allow reductions in overall fungicide use, reduced selection for fungicide resistance and reduced fungicide residues in fruit. Knowledge of baseline levels of resistance to each fungicide as well as cross-resistance to multiple fungicides will allow the design of spray schedules that minimize resistance risk and extend the lifespan of currently registered fungicides while maintaining effective disease control.

References

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- Coyne, D., G. Menzies, C. Burrows and C. MacConnell. 2012. Integrated pest management for raspberries. WSU Whatcom County Extension.
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- Williamson, B. and R.J. McNicol. 1986. Pathways of infection of flowers and fruits of red raspberry by *Botrytis cinerea*. *Acta Horticulturae* 183: 137-141.
- Williamson, B., B. Tudzynski, P. Tudzynski and J.A.L. van Kan. 2007. *Botrytis cinerea*: the cause of grey mould disease. *Molecular Plant Pathology* 8: 561-580.
- Xu, X.M., E. Wedgewood, A.M. Berrie and T. O'Neill. 2009. Infection of raspberry leaves by *Botrytis cinerea* in relation to leaf and cane age. *Communications in Agricultural and Applied Biological Sciences* 74: 761-770.

Budget:

	2014	2015	2016
Salaries – post-doctoral 0.25 FTE ¹	11250	11700	12168
Time-slip	0	0	0
Operations (Goods & services)	4500	4500	4500
Travel ²	2500	2500	2500
Meetings	0	0	0
Other	0	0	0
Equipment	0	0	0
Benefits ³	3746	3896	4052
Total	\$21996	\$22596	\$23220

Budget Justification:

¹ 0.5 FTE post-doctoral salary paid by Washington State University and 0.25 FTE paid by WA Blueberry Commission. Salary for 1 PhD-level graduate student provided by Washington State Commission on Pesticide Registration does not appear in this budget.

² Five trips to field sites per year at three days duration per trip. Cost shared with WA Blueberry Commission

³ Benefits rate for post-doc = 33.3%

Current & Pending Support

Instructions:					
1. Record information for active and pending projects. 2. All current research to which principal investigator(s) and other senior personnel have committed a portion of their time must be listed whether or not salary for the person(s) involved is included in the budgets of the various projects. 3. Provide analogous information for all proposed research which is being considered by, or which will be submitted in the near future to, other possible sponsors.					
Name (List PI #1 first)	Supporting Agency and Project #	Total \$ Amount	Effective and Expiration Dates	% of Time Committed	Title of Project
	Current:				
	Pending:				
Peever	WA Blueberry	22496	04/01/14 to 03/30/15	20	Biology and Control of Mummy Berry and Botrytis fruit rot of blueberry
Peever	WSCPR	22496	04/01/14 to 03/30/15	20	Biology and Control of Mummy Berry and Botrytis fruit rot of blueberry
Peever	WSCPR	21996	04/01/14 to 03/30/15	20	Biology and Control of Botrytis fruit rot of red raspberry

2014 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

New Project Proposal

Proposed Duration: (3 years)

Project Title: Evaluating soil fumigation alternatives in Washington raspberry fields.

PI: Thomas Walters

Organization: Walters Ag Research

Title: Owner

Phone: 360-420-2776

Email: waltersagresearch@frontier.com

Address: 2117 Meadows Lane

Address 2:

City/State/Zip: Anacortes, WA 98221

Co-PI: Inga Zasada

Organization: USDA-ARS HCRL

Title: Plant Pathologist

Phone: 541-738-4051

Email: inga.zasada@ars.usda.gov

Address: 3420 NW Orchard Ave

Address 2:

City/State/Zip: Corvallis, OR 97330

Cooperators: Randy Honcoop, Rob Dhaliwal, Jon Maberry, possibly others

Year Initiated 2014

Current Year 2014

Terminating Year 2016

Total Project Request:

Year 1 \$ 8,095

Year 2 \$ 8,795

Year 3 \$ 8,805

Other funding sources: None, but this project builds upon a previous USDA-RAMP grant to Walters, Zasada and others.

Description: We will help growers interested in alternative fumigation practices establish trials of these on their farms, we will help them evaluate the strengths and weaknesses of these practices, and we will present results of these trials to the raspberry industry. These practices may include, but are not limited to: bed fumigation, use of metam products with a rotary spader and incorporation of mustard cover crops or seed meals.

Our objective is to identify soil fumigation practices that are effective, economical and manageable for growers. Washington growers must comply with buffer zones, fumigant management plans and other features of new labels as a result of EPA's reregistration of metam and cloropicrin. The process is underway for Telone, as well.

The outcome we seek is a wider understanding, shared among industry members, of the merits, costs and limitations of alternative fumigation practices. We envision that growers will not limit their options to the current standard of broadcast, nontarped deep shank injection with Telone C-17 or C-35 for every field to be replanted. Instead, they will be able to knowledgeably choose from several options, appropriate to the needs of their particular field and operation.

Justification and Background: (400 words maximum)

We will address the issue of a lack of fumigation options for raspberry growers in Washington State. In spite of enormous changes in fumigant labels and their requirements, fumigation practices have changed very little so far. Growers break up fields into smaller sections for fumigation to reduce buffer zone size, but continue to use the same products, rates and application methods.

The practices used are mostly successful, but not entirely. Nontarped deep shank fumigation does not allow much fumigant to accumulate in the upper part of the soil profile. This makes it possible to sow a small grain cover crop in the soil at the same time it is fumigated. We've documented root lesion nematode survival in fields fumigated this way: for example, our progress report on fine-tuning Vydate applications describes work in six fields. In four of these, we found substantial populations of *P. penetrans* one year after fumigation. Broadcast deep-shanked applications are also sometimes weak in weed control, for the same reason.

We conducted trials of bed fumigation with Whatcom and Skagit berry growers. In these trials, which are still being evaluated, we have been surprised to see that tarped bed fumigation has held up to nematode and disease pressure at least as well as nontarped broadcast fumigation. The costs at time were comparable to nontarped broadcast fumigation, about \$1000/Acre. Some of the participating growers liked this approach, and are considering using it in their own fields in the future. Others were put off by the lack of a bed shaper appropriate to their use. More widespread trials will help growers determine whether this practice may be useful to their own fields and production systems.

Some growers are interested in Metam application incorporated with a rotary spader and power roller. A specialty equipment manufacturer, Imants, makes an applicator for this purpose. Rotary spader application of metam has been highly successful in strawberry nurseries in Spain. It is now considered a preferred method of soil fumigation for that crop (Garcia-Sinovas et al, 2008), although it was not successful before the use of the rotary spader to incorporate the Metam.

Non-chemical alternatives to soil fumigation, such as Brassica cover crops, incorporation of mustard seed meals and Anarobic soil disinfestation, have been explored by numerous researchers and growers, but have not yet found widespread use by raspberry growers. These practices may be adequate for fields without large disease or nematode populations.

Relationship to WRRRC Research Priority(s): This project directly addresses a #1 priority of the WRRRC, "Soil fumigation techniques and alternatives to control pathogens, nematodes and weeds".

Objectives:

Our overall objective is to identify soil fumigation practices that are effective, economical and manageable for growers. This year, we will:

- Help growers establish fumigation trials in their fields, by researching potential new practices, planning trial layouts and sampling timelines, and coordinating with custom fumigators.
- Host a field day, where growers will be able to learn about the practices being tried, the reasons for trying them, and their costs.

Procedures: (400 words maximum)

- Anticipated length of project
- What will be done and when

In the spring and summer of 2014, we will contact growers to find those replanting acreage in 2014 and interested in trying new practices. Walters will collect field histories from these growers, will obtain soil type analyses, and will research all options of interest to the grower. Zasada will provide nematode analysis for the fields of interest. We have already begun discussions with three growers (Maberry, Honcoop, Dhaliwal), and hope to include others (up to 10).

Each participating grower will receive a written evaluation of the field, with projected disease, weed and nematode problems based upon the information collected. They will also receive research on the strengths, weaknesses and costs of the practices they are considering. Once growers decide which, if any, of the alternatives to try, we will work with them to design the trial to facilitate a comparison with standard practices.

Walters will organize a field day early in September 2014. At this event, he will present alternatives to be trialed by growers this season. Researchers, custom fumigators and product representatives will also be invited to share their work and their thoughts, as has been done at this field day for the past two years.

Walters will work with participating growers to implement these practices and collect samples prior to fumigation, and following treatment. Disease pressure and weed populations will be assessed before the crop is pulled out, Nematode populations will be assessed prior to fumigation. Expenses associated with fumigation practices will be recorded.

In year 2, weed pressure will be recorded several times March through June. Soil nematode populations will be assessed at planting, soil and root populations will be assessed in the fall. Disease and plant vigor assessments will take place at least twice during the growing season at each location. Similar nematode and disease assessments will be made in years 2 and 3; field days will be conducted in these years, as well.

Anticipated Benefits and Information Transfer:

Producers will benefit from this work by learning from their peers' experiments with fumigation and alternative practices. They will learn what kind of fields practices have been tried in, how much they cost, and how they succeeded and failed.

This work will be transferred to users in the usual venue of the small fruit meeting in Lynden. We will also continue to hold a fumigation field day in Lynden during the first part of September. The field day is generally well-attended, and affords a good opportunity for exchange of ideas, as well as conveying research results.

References:

Garcia-Sinovas D., Garcia-Mendez E., Andrade M.A., Becerril, M., DeCal A., Melgarejo P., Salto T., Martinez-Beringola M.L., Redondo C., Martinez-Treceno A., Medina J.J., Soria C., Lopez-Aranda J.M. 2008. Strawberry nurseries in Spain: Alternatives to MB, 2007 results. 2008 conference on Methyl Bromide Alternatives, accessed at mbao.org.

Budget: Indirect or overhead costs are not allowed unless specifically authorized by the Board

	2014	2015	2016
Salaries ^{1/}	\$ 5,000	\$5,000	\$5,000
Time-Slip	\$	\$	\$
Operations (goods & services) ^{2/}	\$ 1,800	\$ 2,500	\$ 2,500
Travel ^{3/}	\$ 1,195	\$1,195	\$1,195
Meetings ^{4/}	\$ 100	\$100	\$110
Other	\$	\$	\$
Equipment	\$	\$	\$
Benefits	\$	\$	\$
Total	\$ 8,095	\$ 8,795	\$ 8,805

Budget Justification

^{1/}Walters, 0.05 FTE. , estimated 5 days for preliminary visits to growers, 5 days research/data tabulation, 2 days for field day preparation and hosting, 6 days assisting growers with practices and collecting samples. This amount includes what I must set aside for my own benefits (retirement and health insurance).

^{2/} Soil samples to commercial lab: 10 samples @\$35 each in 2014. Nematode samples to Zasada: 20 soil and 20 root samples in 2014 @\$30 each, amounts in 2015 and 2016 dependent upon nature of trials established. \$250 for sample shipping in 2014

^{3/} Walters 15 trips Anacortes to Lynden, each 120 miles @\$0.565/mile. Zasada, one trip Corvallis-Lynden for field day, \$175.

^{4/}Supplies and refreshments for field day

**2014 WASHINGTON RED RASPBERRY COMMISSION
RESEARCH PROPOSAL**

New Project Proposal

Proposed Duration: 1 year

Project Title: Evaluating fungicide efficacy for Botrytis control

PI: Thomas Walters

Organization: Walters Ag Research

Title: Owner

Phone: 360-420-2776

Email: waltersagresearch@frontier.com

Address: 2117 Meadows Lane

Address 2:

City/State/Zip: Anacortes, WA 98221

Co-PI:

Organization:

Title:

Phone:

Email:

Address:

Address 2:

City/State/Zip:

Cooperators: Don Wallace, Tobin Peever, Alan Schreiber (?)

Year Initiated 2014 **Current Year** 2014 **Terminating Year** _____

Total Project Request: **Year 1** \$ 9,624 **Year 2** \$ **Year 3** \$

Other funding sources: None

Description:

This project will extend the 2013 project initiated by Schreiber, and largely executed by Wallace and Walters. Wallace supervised the fungicide applications in that project and Walters conducted the harvest and storage evaluations.

We will conduct replicated fungicide efficacy tests in a Whatcom county growers' field to assess the efficacy of selected fungicides, and to assess the efficacy of fungicide programs alternating fungicides from various FRAC groups. Our selection of products to include will be driven by those with demonstrated efficacy in 2013, as well as those already labeled or with potential for labeling on raspberry. These trials will also be used to provide Dr. Tobin Peevers, WSU plant pathologist, with infected plant material treated with known fungicide programs.

Justification and Background:

Fruit rots, largely caused by *Botrytis cineria*, account for significant harvest losses in Washington raspberry fields. Some infected fruit fails to release from the plant, while others become a harvest contaminant, increasing harvest costs and decreasing the value of the harvested crop.

Raspberry growers have a number of labeled fungicides to choose from, but the most commonly used ones fall within a few chemical families: Abound, Cabrio and Quadris fall within FRAC category 11, Elevate is in category 17. Pristine contains one active ingredient in FRAC category 7, and another in category 11, and Switch contains one active ingredient each from categories 9 and 12. Because compounds in the same FRAC category are chemically similar, growers should not make consecutive applications of compounds in the same category, to avoid development of fungicide resistance. The Resistance potential of Abound, Cabrio and Quadris and Elevate is high, and that of Pristine and Switch is Medium (Adaskaveg et al., 2013).

Raspberry growers are routinely using a small number of chemical classes to control botrytis, and most of these have a high risk of resistance development. It's therefore essential for them to use these products in ways which will be both effective and minimize the risk of resistance development.

This project also relates to the small fruit pathology program initiated by Dr. Tobin Peever at WSU. We will be able to provide Dr. Peever with Botrytis-infected field material from the same field, but treated with a full range of different fungicides, to further his work on determining baseline resistance levels to fungicides commonly used for fruit rot management.

Relationship to WRRRC Research Priority(s): This project address a WRRRC #1 priority, "Fruit rot including pre harvest, post harvest, and/or shelf life".

Objectives:

In this year, we will validate the efficacy of fungicides identified in 2013. We will also document effective fungicide programs to prevent fungicide resistance development.

Procedures:

Jan-March, 2014. We will consult with Schreiber to document the most promising products and programs from the 2013 trial. We will also consult with Whatcom Farmers' Co-op to assure that programs to be tested are realistic and representative of fungicide programs used by growers. We will arrange for a grower to host the trial (preferably Rob Dhaliwal, who hosted the trial in 2013).

April-June, 2014. In April, we will assemble all needed equipment and products for the trials. We will use the same WSU sprayer as was used for the 2013 trials (a Rears over-the-row plot sprayer); since this trial will be a direct benefit to Dr. Tobin Peever's program, he has indicated that we should be able to work this out with WSU. We have budgeted for tractor rental, but in the event that a tractor can be arranged through WSU as well, we will return that portion of the funds to WRRRC. We will lay out randomized, replicated plots for an efficacy trial and a separate program trial, as was done in 2013. We will use the colored marking system we developed in 2013 for this. Beginning at approximately 10% bloom, we will make three applications at 7-10 day intervals in the efficacy trial and six applications at similar intervals in the program trial. Disease development will be evaluated 7-14 days after the last application in the efficacy trial, and again 7-14 days after the last application in the program trial.

July 2014. Plots will be individually harvested using the grower's harvester, as we did in 2013. We will be present to collect yield data at all or nearly all of the harvests (in 2013 we successfully obtained plot data from 8 out of 9 harvests). We will collect samples for storage evaluations, based on the procedure we used in 2013. Samples of harvested fruit will be placed in labeled clamshells and stored on ice for transport. The following morning, they will be transferred to ambient (68-74 F) temperatures and stored in the dark for 4-5 days. The percentage of fruits with visible signs of mold will be evaluated.

August-December 2014. Data will be analyzed, reviewed with Peever, shared with cooperators and product representatives, and presented at the Small Fruit Workshop

Anticipated Benefits and Information Transfer:

Raspberry growers will see examples of fungicide rotations that worked well for Botrytis control, and minimize the risk of resistance development. They will also benefit from the efficacy trial, which will help identify those products which are currently most effective against Botrytis. They will also benefit from the efficacy data this trial will generate to help generate product

registrations for raspberry. These results will be transferred to users at the Small fruit workshop, and in a summary sheet, summarizing the 2013 and 2014 results.

References:

Adaskaveg J., Gubler G., Michailides T. 2013. Fungicides, Bacteriacides and Biologicals for Deciduous Tree Fruit, Nut, Strawberry and Vine Crops 2013. Accessed at (<http://www.ipm.ucdavis.edu/PDF/PMG/fungicideefficacytiming.pdf>) 11/7/2013.

Budget: Indirect or overhead costs are not allowed unless specifically authorized by the Board

	2014	2015	2016
Salaries^{1/}	\$ 6,500	\$	\$
Time-Slip	\$	\$	\$
Operations (goods & services), tractor rental, supplies	\$ 1,400	\$	\$
Travel^{2/}	\$ 1,224	\$	\$
Meetings	\$	\$	\$
Other, grower compensation	\$ 500	\$	\$
Equipment^{3/}	\$	\$	\$
Benefits^{4/}	\$	\$	\$
Total	\$ 9,624	\$	\$

Budget Justification

^{1/}Walters, 0.045 FTE, \$4500; Wallace 0.02 FTE \$2000

^{2/}Estimated 18 trips to site: one for setup, seven for product applications, ten for harvests.

^{3/}Justify equipment funding requests. Indicate what you plan to buy, how the equipment will be used, and how the purchase will benefit the growers. Include attempt to work cooperatively with others on equipment use and purchase.

^{4/}Included here are tuition, medical aid, and health insurance for Graduate Research Assistants, as well as regular benefits for salaries and time-slip employees.

Project Proposal to WRRC**Proposed Duration:** 2014-2015**Project Title:** Management of Fungicide Resistant Botrytis in Red Raspberry**PI:** Alan Schreiber**Organization:** Agriculture Development Group, Inc.**Title:** Researcher**Phone:** 509 266 4348 (office), 509 59 4537 (cell)**Email:** aschreib@centurytel.net**Address:** 2621 Ringold Road, Eltopia, WA 99330**Cooperators:** Dr. Tobin Peever-WSU, Tom Walters-Walters Ag Research**Year Initiated:** 2013**Current Year:** 2014**Terminating Year:** 2015**Total Project Request:** Year 1 \$10,000 Year 2 \$10,000 Year 3 \$10,000

Other Funding Sources: I plan to submit a parallel proposal to the Washington State Commission on Pesticide Registration for \$15,000. I expect that registrants will be involved in this project and will contribute funds to this effort.

Description: Resistance has been documented to three of five active ingredients used for control of botrytis in 2012. The project proposes to screen currently used products, other products that are registered but not used and products not registered for raspberry for control of botrytis. This project will be a basic efficacy trial that is based on the 2013 trial, but with some improvements based on what was learned during the course of this trial.

Justification and Background: This project will generate data on which fungicidal products are effective for control of botrytis and which products are not. Dr. Peever will take the lead on berry pathology work and at this time does not plan to take the lead on efficacy trials in raspberries. Based on a conversation on November 8th, 2013, he does not plan to conduct any efficacy trials in raspberries. I am submitting this proposal at the request of the WRRC to ensure that the necessary information is generated for the raspberry industry of Washington. It is our expectation as in berry pathology expertise is developed in Washington there will be less of a need for out of state assistance. We plan to coordinate our work with Oregon State University and University of California, as we did in 2013.

Botrytis cinerea, is a fungus that causes blossom blight, preharvest rot, postharvest rot, and cane infections. On raspberry, it overwinters as sclerotia on canes and mycelia in dead leaves and mummified fruit. Sclerotia produce conidia in spring. A moist, humid environment is ideal for pathogen sporulation and spread. All flower parts except sepals are very susceptible. Initial infections of flowers are latent such that the fungus is dormant until fruit ripens. Fruit rot may be more prevalent in wet weather, in fields under overhead set irrigation systems, or where fruit

ripens in the field for mechanical harvest. Conidia can infect mature or senescent leaves, resulting in primocane infections through petioles.

This is the most treated for disease of berries in Washington with growers applying three to six applications per season starting with a prebloom application and continuing through to harvest. (Raspberry growers who are applying only three or four applications are probably incurring significant economic losses from the disease.) There is no threshold for this disease. If you find it, think you have it or are at risk of having it, you have to start a treatment program. The PNW Small Fruit Research Center ranks it as a number one priority for research for blueberry and raspberry. Raspberry and blueberry have the same disease, are planted adjacent to each other and have the same fungicides used for control of the pest. Raspberry has fruit that is susceptible earlier than blueberry and has heavier selection pressure. It is likely that spores that survive raspberry fungicide programs infect blueberry fields that mature later and then are subjected to another fungicide program in the same season.

Despite aggressive treatment programs, growers incur annual losses to the pest. Botrytis is well known for developing resistance to fungicides. Growers, crop advisors, researchers and extension representatives are concerned that resistance may be developing. The PNW Disease Management Handbook states this about Botrytis on raspberry “Fungal strains can become tolerant to a fungicide when it is used exclusively in a spray schedule. To reduce the possibility of tolerance, alternate or tank-mix fungicides that have different modes of action. Strains resistant to 5 different modes of action have been reported from Germany.” Based on complaints of poor control in 2011, Dr. Christopher Clemens, Technical Service Representative, Syngenta Crop Protection, worked with berry crop advisors and collected infected raspberry fruit from northwest Washington and submitted them to UC-Davis plant pathologist, Dr. Jim Adaskavig to challenge the diseased fruits for tolerance to fungicides. Samples were collected from nine fields and ten isolates from each of the nine fields were screened.

Pristine (boscalid+pyraclostrobin), Captan, Elevate (fenhexamid) and Switch (cyprodinil+fludioxonil) are the four products used for Botrytis control. Fifteen percent of isolates from five fields were found to be resistant to boscalid. Pyraclostrobin does not have efficacy against botrytis to start and is not a factor in this conversation. Every isolate from every field came back resistant to fenhexamid. Twelve percent of isolates from four fields were resistant to cyprodinil. No isolates were resistant to fludioxonil. These results were a source of great concern to the industry. What made these results more worrisome for the industry is the fact that there are almost no new products that have potential for registration on raspberries that have a mode of action different from existing products. Additional concerns for the industry are limitations placed on growers due to MRL restrictions. Due to MRL issues, some products have limited use (e.g. berries going to Canada cannot have any Captan residues: Canada is the number one export market for Whatcom County raspberries.)

Growers try using all four products (some can only use three products due to MRLs) during a season for resistance management and due to label restrictions such as number of application restrictions, REI and PHIs. The loss of even one product (which is being proposed for fenhexamid) could mean a significant problem; the loss of two products would cause a crisis in the industry.

Relationship to WRRC Research Priority: This directly addresses the fruit rot priority.

Objectives: Our objective is to generate botrytis efficacy data for as many products as is possible for red raspberry. A secondary objective is to use this data and information provided by Dr. Peever to develop better Botrytis control recommendations for raspberry.

Procedures: It is my experience (AAS) that it generally requires 3 years to get an adequate assessment of what products work for a particular pest. We plan to conduct efficacy trials in 2014 and 2015. The testing techniques would be similar to that of 2013 with some improvements. Although testing details have not been finalized, we would like to use the same or similar site as in 2013. Two trials were conducted in 2013; a 24 treatment trial with 6 applications and included yield data and evaluations and an 18 treatment trial with 3 applications with no yield data were successfully completed. The logic for two trials are that for unregistered products unfamiliar to raspberry could be easily screened and would not require crop destruct and a fuller trial that is season long would be done with registered products or products that did not require a crop destruct. Accordingly, we propose to conduct two trials in 2014, one that would a quick screen for unregistered products and a second trial that would be season long. A commercial style applicator would be used. Each treatment would be replicated four times.

Applications would start prebloom and would continue into harvest. The start and end data and number of applications depend on environmental and weather conditions and disease pressure. Botrytis samples from the trial plots will be provided to Dr. Peever to determine the degree of resistance to various fungicides. Dr. Tom Walters would be involved in applying fungicides and collecting of efficacy and yield data.

We would expect significant additional funding would be provided by chemical companies wishing to have their products tested.

Anticipated Benefits and Information Transfer: We would provide a written report to the WRRC, would make a presentation at the Small Fruit Conference and would work closely with WSU extension, crop advisors and members of the raspberry industry to make sure the outcome of the research was well known through the grower community.

Budget:	2014	2015
Salaries	6,500	6,500
Operations		
Travel	675	675
Other	1,200	1,200
Benefits	1,625	1,625

Budget Notes: Salaries and benefit are to support an ag research employed by Alan Schreiber. Travel is to partially offset travel to the site. Other is funds that are expected to be used to pay for a subcontractor to collected data or make applications.

WSCPR funds would be used fund the effort to make applications and collect data.

Chemical company funds would be used to support the grower/crop destruct, support Schreiber's time, travel and operational costs (buy product that is not donated, etc)

In 2013 approximately 90% of funds associated with this project were provided to subcontractors such as WSU, Tom Walters, and Tom Peerbolt.

Related Information

We feel that based on the 2013 results that the likelihood of successful results in 2014 and 2015 are very high.

Results from 2013: Unfortunately, we are still waiting on results from the raspberry, blueberry and strawberry survey results from University of California Riverside. The efficacy data are complete and a final report is nearing completion. The mid season evaluations indicated that most products, even Elevate, provided significant control of botrytis. Some experimental products clearly did not. Some biofungicides were largely ineffective. Unfortunately disease pressure collapsed during harvest so it was hard to distinguish much difference between products based on the yield data.

We have some areas for improvement in 2014, including increasing sample size of berries in the mid season evaluation, and changing our evaluation methods at harvest particularly if the disease is not present at a high level.

2014 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

New Project Proposal

Proposed Duration: Three years

Project Title: Mechanizing red raspberry pruning and cane tying

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Email: manoj.karkee@wsu.edu

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Address 2:

City/State/Zip: Prosser, WA 99350

Co-PI: Julie Tarara

Organization: USDA-ARS

Title: Research Horticulturist

Phone: 509-786-9392

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Address 2:

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Cooperators: Qin Zhang, WSU-CPAAS; Tom Walters, Walters Ag Research

Year Initiated 2014 **Current Year** 2014 **Terminating Year** 2016

Total Project Request: Year 1 \$72,007 Year 2 \$63,731 Year 3 \$64,188

Other funding sources: For Year 1, \$62,007 was requested as a sub-contract to WSU from the funding that WRRC and WSU scientists recently received through the WA Specialty Crop Block Grant program. An additional \$10,000 is requested from WRRC to complement engineering research activities under this grant. In-kind supports of \$2,500 from WRRC and \$24,225 from WSU have been provided to the project. In addition, Maberry Packing and Enfield Farms have respectively offered \$18,167 and \$15,833 of in-kind support to this project. Similar funding and matching support is available for Year 2 and 3 of this project. Additional funding will be sought through the SCRI program when it is introduced again in 2014.

Description:

Cane management in red raspberry production is highly labor intensive. Labor availability is uncertain at best and labor cost is increasing. Currently, Washington growers estimate the pruning and tying cost in red-raspberry production to be from \$500 to \$800 per acre. In addition, labor is at risk for chronic and acute injury. Mechanization has the potential to substantially reduce labor use from cane management. In this project, we plan to develop a systematic approach for cane management through horticultural modifications and engineering solutions. A horticultural system for physically separating one-year and two-year old canes will be evaluated for its feasibility of allowing mechanized pruning of two-year old canes while maintaining the desired level of yield. In addition, techniques will be developed to bundle one-year old canes together and tie them to the trellis wires. We expect that the successful completion of the proposed approach will lead to a practical cane management system. In the long term, commercial adoption of the system will improve economic sustainability of WA red raspberry production. The system will also have potential to be adapted to other WA specialty crops such as black raspberry and blackberry.

Justification and Background:

Red raspberry is a premium crop for WA, which produces more than 85% of total US production of frozen red raspberries. This is a bi-annual crop where two-year old canes (floricanes) must be pruned out selectively every year without damaging one-year old canes (primocanes) (Fig. 1).

Following pruning, a number of primocanes must be bundled and trained to trellis wires. This operation is highly labor intensive, costing about \$500 - \$800 per acre per year. Because labor availability is increasingly uncertain and labor costs are continually increasing (Fennimore and Doohan, 2008), an automated or mechanized solution

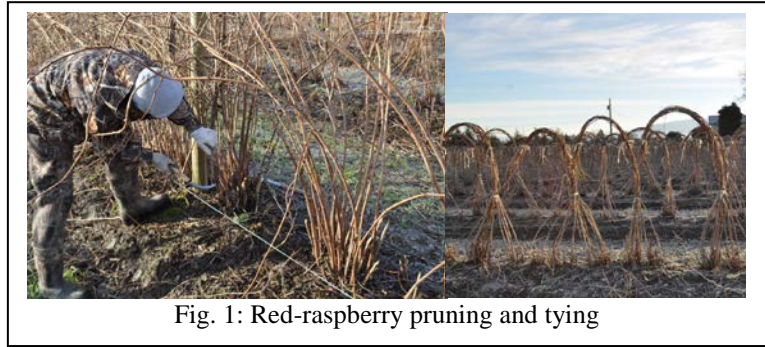


Fig. 1: Red-raspberry pruning and tying

for pruning and training is a critically important need for the WA red raspberry industry. With immigration from Mexico to the USA expected to be net negative within the next five years (Pew Research Center, 2012) and Congressional reform of immigration law uncertain, it is expected that labor may soon become a critical constraint on red raspberry production. Therefore, it is crucial that we begin now to develop mechanization technologies so that the technology is ready for industry adoption before its competitiveness and sustainability may be compromised. During this project we will systematically evaluate horticultural modifications to

“Labor shortage, quality of available labor, need for labor training and cost associated with all of this are the biggest issues we are [currently] facing in raspberry production.” – A WA Grower, Feb, 2012.

raspberry bush training and engineering solutions to cane training and pruning. Our goal is to develop viable, practical techniques of performing training and pruning that reduce labor from its current requirements and consequently reduce the cost of production while minimizing crop loss.

This project will impact all red raspberry growers in WA who use the floricanes production system - the entire industry relies on manual labor to prune and tie canes. This combined operation represents about 35% of the total variable costs of production (MacConnell and Kansiger, 2007). The project will generate industry-applicable horticultural and engineering techniques to improve labor productivity and reduce labor demand. Success in this project will dramatically reduce labor demand and costs, amounting to as much as \$500 per acre per year for combined pruning and cane tying. These savings will lead to millions of dollars of economic benefit to WA red raspberry industry, which will substantially improve the competitiveness and long-term sustainability of the industry.

Relationship to WRRC Research Priority(s): This project directly addresses priority #2: “Labor saving cultural practices including mechanical pruning and tying techniques.” By evaluating current major varieties with a range of plant architectures, it also contributes to the commission’s #1 priority of cultivar development.

Objectives to be accomplished in 2014:

The primary goal of the proposed work is to minimize labor demand in red raspberry pruning through horticultural modifications and mechanization, or automation solutions. To achieve the overall goal, we will particularly focus on the following objectives over the three year duration of this project:

1. Establish at Washington State University's Center for Precision Agricultural and Automation Systems (WSU-CPAAS) a block of red raspberries that will include three commercial cultivars;
2. Evaluate row and training configurations to physically separate floricanes from primocanes;
3. Develop and evaluate mechanization technologies for cane management, which will include

- a. Bundling and tying mechanisms for the primocanes that will bear the following year's crop, and
- b. Sensing systems for floricanes identification and a floricanes pruning mechanism

Particularly in the Year 2014, progress will be made in the following research activities.

1. Preparation and planting red raspberry plot
2. Horticultural management of red raspberry planting
3. Design cane bundling and tying machine
4. Develop prototype machine
5. Evaluate and improve in the lab
6. Outreach activities

Procedures:

Objective #1&2 - Horticultural Studies (Lead –Tarara): All cultural practices will be according to commercial standards. Two trellis wire configurations will be established at standard row spacing. The first configuration is that of the commercial standard planting. The second configuration physically separates floricanes from primocanes during the growing season: floricanes are constrained between two central catch wires spaced 18" apart; primocanes are trained to the outside of the floricanes. This arrangement requires the addition of two central wires to the trellis. The following horticultural attributes will be measured: number of canes per plant; cane length at harvest; number of canes damaged by the bundler (evaluated via necrosis); number of fruiting laterals per sample cane; yield; and weight of dormant-pruned spent floricanes. Three cultivars will be included: Meeker, Wakefield, and Chemainus.

Objective #3 - Engineering Approaches (Lead – Karkee): We will focus on designing and developing bundling and tying technologies in year 1 and year 2. The design will include a pair of augers to move primocanes to a vertical position. Canes will then be bundled together and anchored with a tying mechanism. PI Karkee has been working on an automated knot twining machine for hop production (He et al., 2012). A knot tying mechanism for bundled canes will be developed based on the technology developed in the hop string tying work (Fig. 2).

A second graduate student will be hired to develop and evaluate a method to identify and locate floricanes for cutting or pruning. A preliminary study was performed in 2011 for identifying red raspberry floricanes for pruning and the results were promising (Fig. 3). In this work, a color

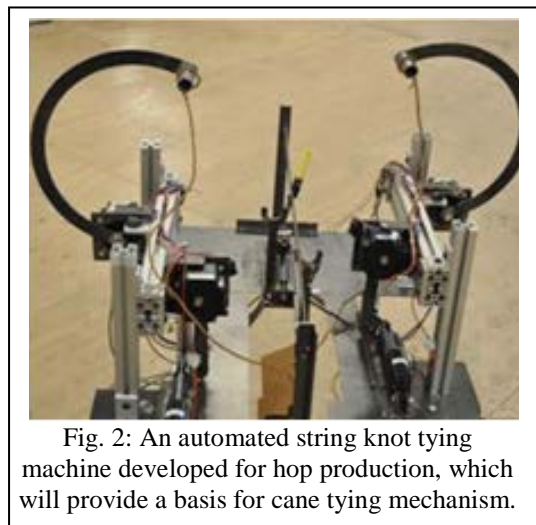


Fig. 2: An automated string knot tying machine developed for hop production, which will provide a basis for cane tying mechanism.

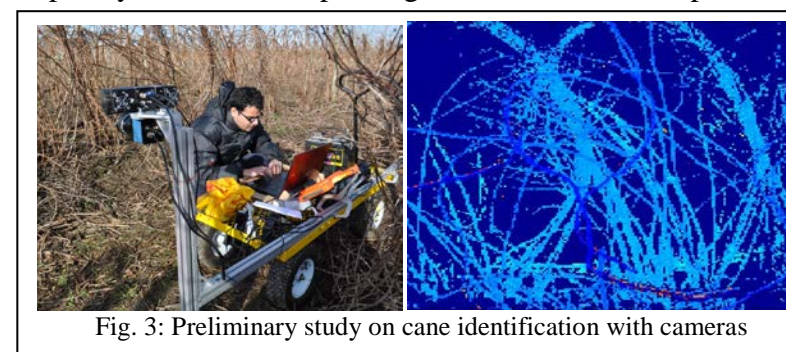


Fig. 3: Preliminary study on cane identification with cameras

vision camera and a laser scanner will be used to identify and locate floricanes for pruning. An over-the-row tunnel system (already developed by PI Karkee and available to this project) will be used to reduce the effect of variable lighting conditions during imaging.

We will investigate the use of food-grade red paint as well as red string-tying to provide the additional information for the image processing system. Because the canes will be physically

separated through horticultural modification, one colored tie or a few colored canes will be sufficient to locate the region of floricanes to be pruned.

Anticipated Benefits and Information Transfer:

This project will evaluate new training systems with the potential to facilitate development of mechanized approaches to both training and pruning, which will ultimately reduce the estimated \$500-\$800 per acre cost of these practices. Working connections among growers, horticulturists, and engineers will be fostered by this well-defined project. Following this, we expect smooth and effective cooperation among parties on future mechanization projects. Results will be transferred to users at the planned workshops and at annual berry meetings, including the Washington Small Fruit Conference. The direct participation of growers in this project will also facilitate transfer to growers through peer-to-peer connections.

References:

Fennimore, S. A., and D. J. Doohan, 2008. The Challenges of Specialty Crop Weed Control, *Future Directions. Weed Technology*, 22: 364-372.

He, L., Q. Zhang, X. Du, R. Luo, and M. Karkee. 2012. A Twining Robot for High-Trellis String Tying in Hops Production. *Transactions of the ASABE*, 55(5): 1667-1673.

MacConnell, C., and M. Kangiser. 2007. Washington Machine Harvested Red Raspberry Cost of Production Study for Field Re-establishment. Washington State University, Whatcom County Extension.

Pew Research Center. 2012. Net Migration from Mexico Falls to Zero—and Perhaps Less. Available at: http://www.pewhispanic.org/files/2012/04/Mexican-migrants-report_final.pdf; assessed on: accessed 6 Nov, 2013.

Budget: *Indirect or overhead costs are not allowed* unless specifically authorized by the Board

	2013	2014	2015
Salaries	\$23,993	\$23,993	\$23,993
Time-Slip	\$1,080	\$3,160	\$3,608
Operations (goods & services)	\$22,383	\$9,482	\$8,038
Travel	\$3,048	\$3,048	\$3,048
Other/Miscellaneous	\$1,370	\$1,370	\$1,369
Benefits	\$20,133	\$22,678	\$24,132
Total	\$72,007	\$63,731	\$64,188

Budget Justification

Salaries (Sub-Total: \$71,979) – All values are in accordance with Washington State University's mandated rates for salaries, wages, and salary inflation. A salary of \$14,294 per annum (Year 1 rate), is requested to hire a graduate student (under Dr. Karkee’s supervision) to work on the day-to-day research activities in developing algorithms and prototype machines and conducting field tests in red raspberry bundling and tying. Partial support for another graduate student will be provided at \$5,473 per year. This graduate student will be responsible for carrying out sensing and pruning mechanism development tasks. In addition, a salary of \$4,226 (5% of his full time appointment) per year (Year 1 rate) is requested for Co-PI Karkee, who will

direct the design, planning, and implementation of engineering research activities. Dr. Karkee will provide an additional 11.25% or more of his time to this project as matching support.

Wages (Sub-Total: \$7,848) – Wages are required for installation and maintenance of, and data collection in the field plot at the hourly rates of \$12.00 for field labor. Total estimated wages are \$1,080 for Yr1, \$3,160 for Yr2, and \$3,608 for Yr3.

Supplies (Sub-Total: \$39,903) – Engineering materials and supplies are estimated at \$34,503 for the duration of the project. The budget will cover the cost of materials and supplies for horticultural studies as well as engineering prototype development. The bulk of horticultural goods and services are required for year 1--field establishment and maintenance (\$13,483). These include contract fumigation service, a drip irrigation system, trellis materials, plants, standard fertilizers and agri-chemicals, and land use charges. Also included are WSU-Prosser's crop maintenance service fees for land preparation, planting, installing the irrigation system, applying irrigation, and applying pesticides and herbicides as needed. In years 2 and 3 (\$4,982 and \$5,038), materials and services include WSU-Prosser's crop maintenance service fees, necessary chemicals and fertilizers, trellis and irrigation repairs, land charges, and field supplies for collecting horticultural data (including picking bins, field notebooks, and measuring tapes and counters). For the materials and supplies (including aluminum sections, iron sections, nuts, and bolts) for engineering prototype development, a budget of \$3,500, \$4,500 and \$3,000 is requested respectively for year 1, year 2 and year 3. In-house fabrication and technician support is available to PI Karkee to build research prototypes, which has reduced the requested budgeted for prototype development. Finally, irrigation supplies of \$5,400 will be incurred in Year 1 only to equip the drip irrigation system with the required sand filter.

Travel (Sub-Total: \$9,144) – Each year, one graduate student will travel to Lynden, WA to conduct field experiments in collaboration with grower collaborators. At the standard per diem rate, this travel will cost \$988 each year (mileage – 574 miles @ \$0.57 = \$327; lodging – 5 night @ \$77 = \$385; and meals – 6 days @ \$46 = \$276). Two-day long trips to the data collection site are also included for PI Karkee and Co-PI Tarara each year to supervise the field experiment as well as attend the project annual meeting, which will cost \$619 each person-trip (Mileage = \$327, Lodging = \$154, and Meals = 138). In addition, travel funds are requested for Students, PI Karkee, and Co-PI Tarara to attend professional conferences during three years of this project. Total travel cost will be \$3,048 per year.

Other (Sub-Total: \$4,109): A miscellaneous budget of \$1,370 per year is requested to cover unexpected minor expenses during the project duration.

Benefits (Sub-Total: \$66,943) – All values are in accordance with Washington State University's mandated rates for benefits and benefit inflation according to staff classification: \$13,582 per annum for one graduate student (Year 1 rate); \$4,527 per annum for partial support to a second graduate student (Year 1), \$1,251 for PI Karkee (Year 1 rate); and \$8.64/hour for field labor. Total estimated benefits costs are \$20,133 for Yr1, \$22,678 for Yr2, and \$24,132 for Yr3.

Current & Pending Support-Karkee

NAME (List/PD #1 first)	SUPPORTING AGENCY AND AGENCY ACTIVE AWARD/PENDI NG PROPOSAL NUMBER	TOTAL \$ AMOUNT	EFFECTIVE AND EXPIRATION DATES	% OF TIME COMMIT TED	TITLE OF PROJECT
ACTIVE					
Whiting (PD); Zhang; Allard; Almenar; Dhingra; Grant; Harte; Karkee ; Long; Oraguzie; Pierce; Ross; Seavert	USDA-NIFA- SCRI	\$5,000,000	09/09 to 08/14	10%	A Total Systems Approach to Developing a Sustainable, Stem-free Sweet Cherry Production, Processing, and Marketing System
Grieshop (PD); Brunner; Agnell; Perry; Nye; Zhang; Karkee; Hoheisel; etc.	USDA-NIFA- SCRI	\$2,472,895	09/11 to 08/14	12%	Development and optimization of solid-set canopy delivery systems for resource-efficient, ecologically sustainable apple and cherry production
Karkee (PD); Zhang; Lewis	WTFRC	\$67,506	06/11 to 05/14	2%	3D Machine Vision for Improved Apple Crop Load Estimation
Karkee (PD); Zhang; Lewis; De Kleine	WTFRC	\$53,395	06/12 to 05/14	2%	Design and Development of Apple Harvesting Techniques
Hashimoto (PD), Chiang; Cooper; Eggeman; Karkee; Vaugh; Yanagida; Zhang; etc.	USDA-NIFA- BRDI	\$6,000,000	04/12 to 03/16	15%	Conversion of High-Yield Tropical Biomass into Sustainable Biofuels
Karkee (PI), Tarara	WA Red Raspberry Commission	\$15,000	03/13 to 02/16	2%	Establishing Red Raspberry Trellising Plot
Karkee (PI) ; Lewis ; Mo; Zhang	National Robotics Initiative - NSF and USDA- NIFA	\$548,735	10/13 to 09/16	15%	Human machine collaboration for automated harvesting of tree fruit
PENDING					
Bierlink (PD), Karkee (WSU- PD); Tarara	WSDA Special Crop Block Grant/WRRC	\$199,926	10/13 to 09/16	8%	Mechanizing Red Raspberry Pruning and Tying
Karkee; Leachman, Taylor, Zhang	WSU CAHNRS	\$50,000	01/14 to 06/15	10%	Autonomous Unmanned Aerial Systems (UASs) for Mitigating Bird Damage in Fruit and Berry Crops: Proof of Concept

Current & Pending Support-Tarara

Instructions:					
1. Record information for active and pending projects. 2. All current research to which principal investigator(s) and other senior personnel have committed a portion of their time must be listed whether or not salary for the person(s) involved is included in the budgets of the various projects. 3. Provide analogous information for all proposed research which is being considered by, or which will be submitted in the near future to, other possible sponsors.					
Name (List PI #1 first)	Supporting Agency and Project #	Total \$ Amount	Effective and Expiration Dates	% of Time Committed	Title of Project
Hirst et al.	<u>Current:</u> NIFA-SCRI	\$3,027,747	9/1/12 to 8/31/16	20%	Automation of dormant pruning of specialty crops
Bryla and Tarara	Oregon Blueberry Commission and	\$6,880	7/1/13 to 6/30/14	5%	Practices to Reduce Heat-Related Fruit Damage in Highbush Blueberry
	Washington Blueberry Commission	\$8,670	7/1/13 to 6/30/14		
Bierlink (PD), Karkee (WSU-PD); Tarara	<u>Pending:</u> WSDA Special Crop Block Grant/WRRC	\$199,926	10/13 to 09/16	15%	Mechanizing Red Raspberry Pruning and Tying

**2014 WASHINGTON RED RASPBERRY COMMISSION
RESEARCH PROPOSAL**

New Project Proposal Proposed Duration: 2 years

Project Title: Exploring opportunities for developing value-added dried products from raspberries.

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Address 2:
City/State/Zip: Pullman, WA, 99164

Co-PI: Giuliana Noratto
Organization: WSU
Title: Assistant Professor
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Email: giuliana.noratto@wsu.edu
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Address 2:
City/State/Zip: Pullman, WA, 99164

Cooperators: Enfield Farms, Lynden, WA

Year Initiated Current Year 2014 Terminating Year 2015

Total Project Request: Year 1 \$18,365/-; Year 2 \$7,306/-

Other funding sources: *None*

Notes: The data obtained from this project will be used to solicit larger funding from the USDA-FSMIP program. They fund projects to evaluate the expansion of the markets for the US produce and raspberries fit into this program.

Description: (less than 200 words) describing objectives and specific outcomes

Raspberries carry important antioxidants such as anthocyanins, ellagitannins, and other phenolic acids with proven health benefits. It is well known that the raspberries have a very short shelf life after harvest and are very fragile. These have been the major hurdles for both marketing and processing. A significant percentage of the harvested raspberries are frozen. The remaining portion goes into making processed products, such as juice, jams, and purees.

One of the strongest desires of the raspberry producers is to increase the markets for their produce both nationally and internationally. The requirement of cold storages is the biggest hurdle. To be able to reach the international markets in Asia, South America, Africa, and parts of Europe, there is a huge need to develop value added shelf stable products. Some of the simplest forms are dried fruit and dried fruit powders. Although it is important to ensure that the value added products retain their quality and health properties.

We propose to evaluate the dehydration and powdering characteristics of red raspberry to develop more shelf stable products. The dehydration studies will be done by hot air drying and

freeze drying methods. Impact of the dehydration on the bioactive compounds and nutritional properties will also be studied.

The major outcome of this work will be development of validation data for viability of dehydration of raspberries. Data on the dehydration characteristics will be obtained from the work.

Justification and Background: (400 words maximum)

The most commonly processed raspberry products include; Individually Quick Frozen (IQF). Small percent of the raspberries are processed into purees, juices and jams. A very small percentage of raspberries are subjected to dehydration. There is no published information readily available for the processors in WA to quickly start a processing facility to make dehydrated raspberry powders. There is a need to develop this information for the growers of WA.

Since majority of the harvested raspberry crop is processed as frozen product, exploring broader national and many international markets becomes difficult. IQF raspberries markets have been explored to a great extent but internationally there are still markets that need to be explored. Major hurdles to get into international markets are the lack of cold storage facilities and distribution systems in many of those markets. Even if the international markets had the cold storage facilities and distribution systems, the costs of shipping the frozen products will make the products less affordable. Because of these barriers, we have the challenge to develop an alternative process to develop a value added product with extended shelf life.

In this project we propose to develop basic data about the drying characteristics of red raspberries. Along with the drying characteristics we also intend to determine stability of health promoting compounds such as phenolics as the effects of drying. This will help the processors to understand the potential of expanding the markets for the WA grown raspberries. The outcomes of this project will help the raspberry growers and processors in the region tremendously, as this will help ultimately help to increase the markets for the raspberries.

Relationship to WRRRC Research Priority(s):

This proposal is being submitted based on the needs that were assessed in discussions with many processors and farmers in the Lynden are. We believe this project will lead to opportunities to increase the markets of the Washington grown raspberries.

Objectives:

Our objectives for this proposal, is to thoroughly understand the drying characteristics of the red raspberries by using air drying method and compare it with freeze drying process. The impacts of the drying processes on the bioactive compounds and nutritional properties of the raspberries will also be studied.

There is no well published literature for the practical aspects of making dehydrated products from raspberries. It is critical to the raspberry industry to have this information available, so they can explore these opportunities.

Specific objectives:

1. Evaluate existing dehydration technologies to process the raspberries into value added shelf stable powders
2. Determine the effects of dehydration methods on the bioactive components.

Procedures: (400 words maximum)

We anticipate the total length of the project to be 2 years. During the first year time will be spent on the drying experiments. During this year, the major goal will be develop optimum drying conditions for the drying of raspberries. Final drying experiments and the extension work will be conducted during the 2nd year of the project. Analysis of the dried products for the bioactive compounds will be studied during the first year and the 2nd year.

All the drying work will be led by Dr. Girish Ganjyal. Dr. Ganjyal will use his pilot scale processing facilities at the School of Food Science and will work with selected processing facilities for part of the processing work. The budget will be used to support one technician and one part time student helper for conducting the drying experiments. The work on analysis of the bioactive components will be led by Dr. Giuliana Noratto. Dr. Noratto will use her laboratories at the School of Food Science for all of this work.

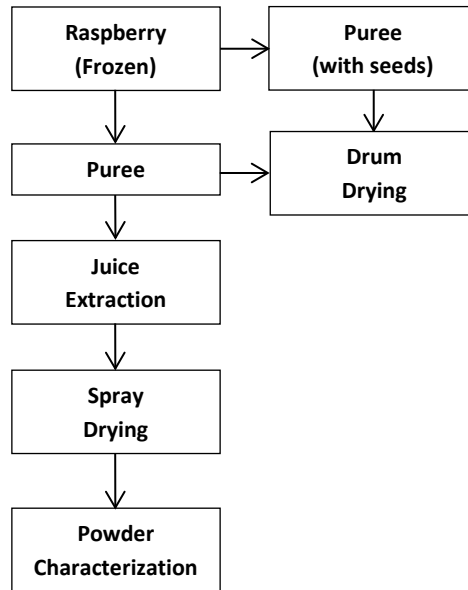
The flowchart #1 in next page, show the brief outline of the proposed work.

Anticipated Benefits and Information Transfer: (100 words maximum)

The major benefits of the work will be the establishment of the standard drying characteristics for the red raspberries. The outcomes of the work will help the processors to determine if the dehydration of the raspberries will be a viable option to increase their markets or not. The information will be very useful, if any of the processors move with the idea of expanding their business with raspberry powders.

A serious effort will be undertaken to transfer the results to the stakeholders. The extension work will be led by Dr. Girish Ganjyal. The team will work together with the growers and processors to disseminate the information as the key milestones of the project are reached and in depth at the end of the project. The team will use various methods, including short conferences, bulletins, research and extension publications for disseminating the information to the stakeholders. This work will be completed by December 2015.

Flow Chart 1: Outline of the processing research



References:

Agricultural Marketing Resource

Center. http://www.agmrc.org/commodities_products/fruits/raspberries/

Anekella, K., & Orsat, V. (2013). Optimization of microencapsulation of probiotics in raspberry juice by spray drying. *LWT - Food Science and Technology*, 50(1), 17-24.

AOAC (2006). Official Analytical Chemist. Official methods of analysis. 18th ed.: Gaithersburg, Md.: AOAC Intl. 2000 p

Bakkalbasi, E., Menten, O., & Artik, N. (2009). Food Ellagitannins - Occurrence, Effects of Processing and Storage. *Critical Reviews in Food Science and Nutrition*, 49(3), 283-298.

Beekwilder, J., Jonker, H., Meesters, P., Hall, R. D., van der Meer, I. M., & Ric de Vos, C. H. (2005). Antioxidants in raspberry: on-line analysis links antioxidant activity to a diversity of individual metabolites. *J Agric Food Chem*, 53(9), 3313-3320.

Bhandari, B. R., Senoussi, A., Dumoulin, E. D., & Lebert, A. (1993). Spray drying of concentrated fruit juices. *Drying Technology*, 11(5), 1081-1092

Budget: Indirect or overhead costs are not allowed unless specifically authorized by the Board

	2014	2015	2016
Salaries ^{1/}	\$5,840	\$3,037	\$
Time-Slip	\$4,000	\$	\$
Operations (goods & services)	\$5,000	\$1,500	\$
Travel ^{2/}	\$1,000	\$1,500	\$
Meetings	\$	\$	\$
Other	\$	\$	\$
Equipment ^{3/}	\$	\$	\$
Benefits ^{4/}	\$2,524	\$1,269	\$
Total	\$18,365	\$7,306	\$

Budget Justification

^{1/}Specify type of position and FTE.

^{2/}Provide brief justification for travel requested. All travel must directly benefit project. Travel for professional development should come from other sources. If you request travel to meetings, state how it benefits project.

^{3/}Justify equipment funding requests. Indicate what you plan to buy, how the equipment will be used, and how the purchase will benefit the growers. Include attempt to work cooperatively with others on equipment use and purchase.

^{4/}Included here are tuition, medical aid, and health insurance for Graduate Research Assistants, as well as regular benefits for salaries and time-slip employees.

Budget requested is to cover the labor costs for a technician and a time-slip for conducting the dehydration experiments and the analysis of the samples. Part of the amount is also requested to cover the supplies costs for both the dehydration studies and the analysis of the bioactive compounds. A small portion of the amount is requested for travel related to the project activities.

The salaries requested are to cover part of the technician salary. The amount of \$8281/- (\$5,840 salary + \$2441 benefits) for the first year is requested to cover the salary of the technician for a period of 10 weeks. For the 2nd year we anticipate the cost will be around \$4306/- (\$3036 in salary and \$1269 in benefits) only. Technician will conduct the drying experiments in the first year and partly in the 2nd year. The technician will also do part of the analysis work. We are also requesting the amount of \$4000/- in time slip salaries and \$84/- in benefits for the first year to cover the costs of one time-slip. The time-slip will help with the dehydration experiments and analysis of the samples during the first year.

We are requesting \$5000/- for the first year and \$1500/- for the second year to cover all the supplies costs for the dehydration and samples analysis work. Travel amounts are also requested for the amounts of \$1500/- for the first year and \$1000/- for the second year. Travel will be related to the project work and not to attend any workshops or conferences. We will use the travel monies to visit the processing facilities in Lynden area and to disseminate the information from the research to the stakeholders.

CURRENT & PENDING SUPPORT

Name: **Girish Ganjyal**

NAME (List/PD #1 first)	SUPPORTING AGENCY AND AGENCY ACTIVE AWARD/PENDING PROPOSAL NUMBER	TOTAL \$ AMOUNT	EFFECTIVE AND EXPIRATION DATES	% OF TIME COMMITTED	TITLE OF PROJECT
G. Ganjyal	Active: New Faculty Seed Grant Competition – Washington State University	\$21,000	06/01/2013 to 06/30/2014	2%	Enhancing value of the Washington State’s processed fruit and vegetable by-products through innovative processing techniques
A. Fatland, G Ganjyal, A. Holzer, J. Leachman, C. Love, V. Yadama, et. al.	U.S. Economic Development Administration	\$136,000 (WSU - \$80,000)	01/01/2014 to 12/31/2014	2%	Investing in Manufacturing Communities Partnership Planning Grant
G. Ganjyal	Lamb Weston (ConAgra Foods., Inc.), Richland, WA	\$26,923	11/15/2013 to 04/15/2014	1%	Understanding the Changes in Physico- Chemical and Structural Characteristics of Potato Strips during Frying and Freezing Unit Operations in French Fry Processing
Pending:					
G. Ganjyal, S. Sablani, Y. Wang	Washington Cherry Commission	\$50,000	05/01/2014- 05/01/2016	3%	Strategies to reduce postharvest cracking and splitting of cherries by processing and packaging techniques
R. Visvanathan, G. Ganjyal, C. Morris, K. Ambrose., D. Maier, S. Hiregoudar	Indo-US Science and Technology Forum (IUSSTF)	\$145,760 (WSU - \$31,460)	01/01/2014 to 12/31/2015	2%	Indo-US Joint Centre on “Value added processing of millets”
G. Ganjyal L. Lei K. Englund	Emerging Research Issues for Washington Agriculture 2014 Internal Competitive Grant Program	\$78,593	01/01/2014- 12/31/2016	5%	Development of Biobased Materials from Low Value Byproducts of Agricultural Processing of Washington State
J. Tang, M. Zhu, S. Sablani, G. Ganjyal, D. Shah	Emerging Research Issues for Washington Agriculture 2014 Internal Competitive Grant Program	\$79,189	01/01/2014- 12/31/2016	1%	Understanding of Food and Microbiological Properties at Elevated Temperatures to Improve Low- moisture Food Safety

S. Sablani, S. Chen, M. Bule, G. Ganjyal	Emerging Research Issues for Washington Agriculture 2014 Internal Competitive Grant Program	\$79,932	01/01/2014-12/31/2016	1%	Smart Sustainable Functional Foods (S2F2) Development Using Bioactive Compounds for Improving Human Health
K. Murphy, G. Ganjyal, J. Matanguihan, G. Noratto	Emerging Research Issues for Washington Agriculture 2014 Internal Competitive Grant Program	\$77,500	01/01/2014-12/31/2016	3%	Putting Barley back on our Plates: Development and extension of nutritious whole grain hullless barley
S. Sablani, K. Killinger, G. Ganjyal	Washington Raspberry Commission	\$31,455	01/01/2014-12/31/2015	1%	Ultraviolet light (UV-C) treatment for improving safety of red raspberries
G. Ganjyal, G. Noratto	Washington Raspberry Commission	\$25,671	01/01/2014-12/31/2015	1%	Exploring opportunities for developing value-added dried products from raspberries.
G. Ganjyal, G. Noratto	Washington Raspberry Commission	\$25,570	01/01/2014-12/31/2015	1%	Developing edible coatings for raspberries for their use in muffins.

Giuliana Noratto

CURRENT AND PENDING SUPPORT

Instructions:

1. Record information for active and pending projects, including this proposal.
2. All current efforts to which project director(s) and other senior personnel have committed a portion of their time must be listed, whether or not salary for the person involved is included in the budgets of the various projects.
3. Provide analogous information for all proposed work which is being considered by, or which will be submitted in the near future to, other possible sponsors.

NAME (List/PD #1 first)	SUPPORTING AGENCY AND AGENCY ACTIVE AWARD/PENDING PROPOSAL NUMBER	TOTAL \$ AMOUNT	EFFECTIVE AND EXPIRATION DATES	% OF TIME COMMITTED	TITLE OF PROJECT
Giuliana Noratto	Active: Washington Grain Commission. 13C-3057-4668	\$60,000	07/01/2013-06/30/2015	15%	Wheat as a source of bioactive compounds that can protect from obesity
	Pending: National Processed Raspberry Council Washington Tree Fruit Research Commission, NW Cherry Review	\$123,558 \$45,000		15% 10%	Protective effects of raspberries against diabetes and associated complications through modulation of gut microbiota Positioning cherries among “Superfruits” for intestinal health

	Emerging Research Issues Internal Competitive Grant	\$80,000		15%	Protective effects of apples against obesity and associated complications through modulation of gut microbiota
	Emerging Research Issues Internal Competitive Grant	\$76,000		5%	Putting Barley back on our Plates: Development and extension of nutritious whole grain hulless barley

**2014 WASHINGTON RED RASPBERRY COMMISSION
RESEARCH PROPOSAL**

New Project Proposal Proposed Duration: 2 years

Project Title: Developing edible coatings for raspberries for their use in muffins.

PI: Girish Ganjyal

Organization: WSU

Title: Assistant Professor

Phone: 509-335-5613

Email: girish.ganjyal@wsu.edu

Address: School of Food Science

Address 2: FSHN 228

City/State/Zip: Pullman, WA, 99164

Co-PI: Shyam Sablani

Organization: WSU

Title: Assistant Professor

Phone: 509-335-7746

Email: ssablani@wsu.edu

Address: Biological Systems Engineering

Address 2: L J Smith Hall # 209

City/State/Zip: Pullman, WA, 99164

Cooperators: TIC Gums, Tate & Lyle Co.

Year Initiated Current Year 2014 Terminating Year 2015

Total Project Request: Year 1 \$12,661/-; Year 2 \$12,909/-

Other funding sources: *None*

Notes:

Description: (less than 200 words) describing objectives and specific outcomes

The Washington red raspberry industry has major challenges to remain competitive including expansion of markets for fresh and processed raspberries. Fruit and fruit pieces are widely used as ingredients in many food formulations such as muffins, pastry and confections. Maintaining the characteristic form, color, flavor and texture are important elements in consumer acceptance of raspberries in a number of these applications.

The textural properties and integrity of raspberry is presents a challenge as the fruit is delicate and breaks down and/ or bleeds pigment (water soluble anthocyanin migration) into the crumb of baked products. Thus the problem is two-fold: maintaining the integrity of the berry and retaining the anthocyanin inside the berry. Given the potential health benefits of raspberry phytochemicals including anthocyanins, it is desirable to devise processes that reduce losses of these beneficial compounds while allowing ready and convenient incorporation of berries into attractive food products.

Thus the objective of this project is to develop solutions for inclusion of raspberries in muffins. We hope to develop solutions involving the coating of the raspberries while they are frozen and then holding the integrity of the muffins during after the baking process. The outcomes will be the formulations for the right coatings solutions.

Justification and Background: (400 words maximum)

The most commonly processed raspberry products include; Individually Quick Frozen (IQF). Small percent of the raspberries are processed into purees, juices and jams. Raspberry growers and processors are always looking to increase the markets without cannibalizing the existing markets. One of the potential markets include, the baked products such as muffins. Other small fruits such as blueberries are used in muffins very efficiently.

Raspberries are very fragile and thus have the tendency to break apart and make the muffins soggy. There is a need to address this issue. If the raspberries can be coated with an appropriate edible coating, they could be effectively used in baked products.

Addressing this issue will help open new markets for the raspberry fruit. Increasing the markets for the Washington red raspberries will help increase the value of the fruit and bring more returns to the farmers and the processors.

Relationship to WRRRC Research Priority(s):

Objectives:

The overall objective of this proposal is to develop solutions that will allow the incorporation of whole raspberries into pastry products (muffins) while maintaining the integrity of the fruit and reducing bleeding of pigment into the crumb.

Specific objectives of the project include:

1. Develop a coating solution for frozen raspberry fruit that can be stable in muffin system.
 - This objective will be addressed in the first year of funding.
2. Attempt to develop an edible coating for partially dried raspberry fruit that can be stable in muffin system.
 - This objective will be addressed in the second year of funding.

Procedures: (400 words maximum)

Raspberry fruit procured from industry will be coated with edible films after various pretreatments.

We will take the following two approaches to determine the best coating for the raspberries:

1. Apply a mixture of hydrocolloid and dextrin's that will coat the fruit in their frozen state. The mixture would gel while baking and help hold the fruit together.

- First we will screen various hydrocolloids that can form a solutions in the cold temperatures.
 - Then we will screen various starches with different dextrin levels that can help with the coating of the fruit
 - The optimum mixture of the hydrocolloid and dextrin will be determined based on various experimental trials.
2. In the second approach, we will attempt to develop an edible coating for partially dried fruit.
- Initially we will treat whole fruit with a range of low methoxy pectin and calcium solutions to firm and stabilize the raspberry tissue.
 - Subsequently the fruit will be air dried at low temperature to moisture content in the 50-60% range.
 - The product will be coated with composite polymer dispersions and the surface allowed, to dry, yielding an edible coating.
 - A wide range of materials will be tested including surfactants and polysaccharide gums such as chitosan, sodium caroxymethyl cellulose and lipophilic materials to result in a barrier to anthocyanin diffusion.
 - The resulting edible film coated raspberries will be frozen and stored at minus 20°C.

To test integrity of the fruit, an anthocyanin leakage test will be done in which weighed treated berries will be submerged on a screen suspended in a beaker containing an aqueous buffer (at room temperature) and the buffer stirred by a magnetic mixer. Aliquots of the liquid will be removed periodically and anthocyanin content measured by visible spectroscopy at 520nm. Fruit will be further tested by placing a weighed amount of frozen berries into a closed container lined with paper towels and allowed to thaw for 4 hours, the berries weighed and drip loss calculated.

Muffins will be made as described by Rosales Soto (2008) using treated berries with berry incorporation at the end of the batter development step. After baking at 350°F in a conventional oven, muffins will be cut horizontally into 0.8cm slices and digital photos taken followed by image analysis to evaluate bleeding. Portions of the muffins will be frozen, freeze-dried and extracts evaluated for anthocyanin, total phenolics and antioxidant activity as described by Rosales Soto (2008).

References:

Rosales Soto, Maria U. 2008. Phenolics, anthocyanins and antioxidant activity in red raspberry muffins, M.S. Thesis. School of Food Science, Washington State University, Pullman, WA.

Budget: Indirect or overhead costs are not allowed unless specifically authorized by the Board

	2014	2015	2016
Salaries ^{1/}	\$4,380	\$4,555	\$
Time-Slip	\$2,400	\$2,400	\$
Operations (goods & services)	\$3,000	\$3,000	\$
Travel ^{2/}	\$1,000	\$1,000	\$
Meetings	\$	\$	\$
Other	\$	\$	\$
Equipment ^{3/}	\$	\$	\$
Benefits ^{4/}	\$1,881	\$1,954	\$
Total	\$12,661	\$12,909	\$

Budget Justification

^{1/}Specify type of position and FTE.

^{2/}Provide brief justification for travel requested. All travel must directly benefit project. Travel for professional development should come from other sources. If you request travel to meetings, state how it benefits project.

^{3/}Justify equipment funding requests. Indicate what you plan to buy, how the equipment will be used, and how the purchase will benefit the growers. Include attempt to work cooperatively with others on equipment use and purchase.

^{4/}Included here are tuition, medical aid, and health insurance for Graduate Research Assistants, as well as regular benefits for salaries and time-slip employees.

Budget requested is to cover the labor costs for a technician and a time-slip for conducting the experiments and the analysis of the samples. Part of the amount is also requested to cover the supplies costs for both the coating development studies and the analysis of the samples. A small portion of the amount is requested for travel related to the project activities.

The salaries requested are to cover part of the technician salary. The amount of \$6211/- (\$4380 in wages and \$1811 in benefits) for the first year is requested to cover the salary of the technician for a period of 4 weeks. For the 2nd year we anticipate the cost will be around the same with a total of \$6509 out of which \$1954 will be for benefits. Technician will conduct the experiments related to the coatings development in the 1st year and in the 2nd year. The technician will also do part of the analysis work. We are also requesting the amount of \$3000/- during the 1st year and the 2nd year to cover the costs of one time-slip employee, with benefits of \$50/year for the benefits. The time-slip employee will help with the experiments and analysis of the samples.

We are requesting \$3000/- for both the 1st and 2nd year to cover all the supplies costs for the experiments related to the coating development and samples analysis work. Travel amounts are also requested for the amounts of \$1000/- for both years. Travel will be related to the project work and not to attend any workshops or conferences. We will use the travel monies to visit the processing facilities in Lynden area and to disseminate the information from the research to the stakeholders.

2014 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

New Project Proposal

Proposed Duration: (1, 2 or 3 years): 2 Years

Project Title: Processed fruit quality of newly released raspberry selections

PI: Carolyn Ross

Organization: WSU School of Food Science

Title: Associate Professor

Phone: 509-335-2438

Email: cfross@wsu.edu

Address: FSHN 122

Address 2:

City/State/Zip: Pullman, WA 99164-6376

Cooperators: Patrick Moore, Horticulture and Landscape Architecture, Washington State University, Puyallup WA. Catherine Daniels, Dept. of Entomology, Washington State University, Puyallup WA.

Year Initiated 2014 **Current Year** 2014 **Terminating Year** 2016

Total Project Request: **Year 1** \$11,010 **Year 2** \$11,010

Other funding sources: None

Description:

The objectives of this study are to evaluate the processed quality of raspberry selections recently released by the WSU Small Fruit Breeding Program. The Meeker raspberry cultivar will be included as a control. Each selection will be processed into individually quick frozen (IQF), puree and juice. Various parameters for each product following processing will be evaluated, including color, total anthocyanins, acidity, proteins, sugars, Vitamin C, folic acid and pH. Following these measurements, end-use products [IQF (smoothie), puree (Greek yogurt) and juice (stand-alone juice)] will be made from each processed raspberry selection. Chemical measurements will be made again to determine any effects of processing.

Consumer liking for each product will be determined at WSU-Pullman and at the Small Fruit Conference in Lynden, WA. The outputs for this proposal include the generation of extension bulletins, presentations at field days, industry seminars and the generation of a peer-reviewed publication. The outcome of this research, and communicating its results, is that growers will be able to select raspberry cultivars specific to new or existing markets, based on the cultivars sensory and chemical profile.

Justification and Background:

The majority of raspberries produced in Washington State are consumed in processed form. These include individually quick frozen (IQF), puree (block frozen) and juice. Major processing cultivars include Meeker, Wakefield and Chemainus, and while these are popular, the WSU Small Fruit Breeding Program continues to introduce new cultivars to meet consumer and grower needs. Fruit composition data for these raspberry selections, as well as plant growth conditions and hardiness is known.

However, what we don't know is the chemical qualities of the raspberry products once they have been submitted to processing methods used on Washington fruit. It is known that different raspberry selections vary in their antioxidant profile; what we don't know is if processing differentially affects these antioxidant profiles (Boyles and Wrolstad, 1993). Thus, to better understand the characteristics of new selections and how this fruit best be used, information on chemical makeup (sensory properties and antioxidant content) of these selections in their three processed forms is important.

The overall goal of this project is to assess the processed fruit quality of new raspberry selections introduced by the WSU Small Fruit Breeding Program. The objective is to process five raspberry selections in three forms--IQF, puree and juice--and then produce food products from each of these three forms: IQF included in fruit smoothies, puree included in Greek yogurt and juice prepared as a stand-alone product. We also plan on determining the stability of the antioxidants as the product moves through processing. In addition, formal sensory evaluation testing will be performed to determine whether consumers fall in groups regarding preference.

In 2013, a Specialty Crops Block Grant was funded by Oregon Department of Agriculture. The objectives of this training grant are to encourage the use of caneberries in new product and menu development, and it's primarily directed at food manufacturing and food service professionals. This proposal differs from grants funded by the ORBC in that the nutritional and chemical parameters of the processed raspberry products will be determined, thus giving specific information useful to niche marketing.

Relationship to WRRC Research Priority(s): With this proposal, we will address Priority 1 by evaluating the newly released raspberry selections for their processed fruit quality.

Objectives:

The overall goal of this project is to assess the processed fruit quality of new raspberry selections introduced by the WSU Small Fruit Breeding Program. Specific objectives include:

- 1) Process five raspberry selections in three forms: IQF, puree and juice,
- 2) Evaluate each of these processed products for their nutritional and chemical characteristics,
- 3) Produce end-use products from each of these three forms – IQF included in fruit smoothies, puree included in Greek Yogurt and juice prepared as a stand-alone product,
- 4) Evaluate the chemical and nutritional characteristics of each end-use product, as well as consumer acceptance of each product.

To achieve this goal of evaluating different raspberry selections, we will employ a series of experiments over two harvest years (2014 and 2015).

Procedures:

Objective 1: Raspberries will be harvested in Puyallup at commercial ripeness in July 2014. These raspberries will be randomly assigned to 3 treatments: IQF, puree or juice. Juice and puree will be prepared as described by Boyles and Wrolstad (1993) and Macerías et al. (2007), and in consultation with the industry so as to produce a product similar to that seen in the industry. These products will be prepared in the WSU Pilot Plant Facilities (Pullman WA).

Objective 2: The puree, juice and IQF berries will be evaluated for chemical and nutritional parameters, including color, total anthocyanins, acidity, proteins, sugars, Vitamin C, folic acid and pH (Hager et al., 2008; Macerías et al., 2007; Boyles and Wrolstad, 1993). The IQF berries will also be assessed for

percent crumble.

Objective 3: End-use products will be prepared. The IQF berries will be incorporated into a smoothie, and the puree will be incorporated into a Greek yogurt – these products were selected based on their popularity. Greek yogurt will be prepared in the WSU Creamery facilities. We will explore using raspberries in baked products given previous research that suggests using coated raspberries to maintain the antioxidant content of the raspberries (Rosales, 2008).

Objective 4: The chemical and nutritional characteristics of each end-use product (smoothie, yogurt and juice) will be evaluated as described in Objective 2. In addition, consumer acceptance of each product will be examined in the WSU Sensory Evaluation Facility and at the Small Fruit Conference in Lynden (December 2014). Demographic data about each consumer will be collected to determine if consumers fall into groups, that is, whether different parts of the population like different products. Such grouping was previously demonstrated in our lab for fresh and frozen raspberries (Villamor et al., 2013).

We anticipate completing the first year of the project by December 2014. Kenny McMahon and Allison Baker, two graduate students already employed in the Ross lab, will start in Spring 2014 to develop all methodologies. Once the raspberries are received in July, the graduate students will complete processing (Objective 1) and the necessary chemical measurements (Objective 2). End-use products will be prepared and optimized, with Objective 3 completed in September 2014. Objective 4 will be completed by December 2014. In early 2015, the final report will be prepared, along with extension materials and a peer-reviewed manuscript. During 2015, data will be shared with industry via seminars and field day presentations.

Anticipated Benefits and Information Transfer:

Knowledge will be transferred to users via extension bulletins, field days, and industry seminars. For this outreach, we will coordinate with the WSU Small Fruits Breeding Program and appropriate commissions. We will submit a manuscript to the Journal of Food Science.

As a benefit to the study, producers will gain valuable information on the processing quality of different raspberry selections – this will assist them in selecting a cultivar appropriate for their anticipated market. Breeders will learn which raspberry attributes are most desired by consumers. Answering these questions has importance for Washington State and the Pacific Northwest given the region's large raspberry production.

References:

- Boyles, M.J. and Wrolstad, R.E. (1993). Anthocyanin composition of red raspberry juice: Influences of cultivar, processing and environmental factors. *Journal of Food Science*. 58: 1135-1141.
- Hager, A., Howard, L.R., Prior, R.L. and Brownmiller, C. 2008. Processing and storage effects on monomeric anthocyanins, percent polymeric color and antioxidant capacity of processed black raspberry products. *Journal of Food Science*. 73: H134-140.
- Macerias, R., Alvarez, E. and Cancela, M.A. 2007. Rheological properties of fruit purees: Effect of cooking. *Journal of Food Engineering*. 80: 7630769.
- Rosales-Soto, M.U. 2008. Phenolics, anthocyanins and antioxidant activity in red raspberry muffins. Washington State University: Thesis.
- Villamor, R. R., Daniels, C.H., Moore, P.P. and Ross, C.F. 2013. Preference mapping of frozen and fresh raspberries. *Journal of Food Science*. 78: S911-S919.

2014 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

Budget: Indirect or overhead costs are not allowed unless specifically authorized by the Board

	2014	2015
Salaries	\$	\$
Time-Slip ^{1/}	\$9610	\$9610
Operations (goods & services) ^{2/}	\$1400	\$1400
Travel	\$	\$
Meetings	\$	\$
Other	\$	\$
Equipment	\$	\$
Benefits ^{3/}	\$	\$
Total	\$11,010	\$11,010

Budget Justification

^{1/}Funds are requested to provide summer salary for two graduate students. Funding includes \$1610/month x 2 students x 3 months = \$9610

^{2/}We request funds for the chemicals, glassware and lab consumables (ie. pipette tips, cuvettes) required for the chemical assays and texture measurements. These funds also include the sensory evaluation-related costs, including panelist incentives and other consumables required to conduct the sensory evaluation panels.

WASHINGTON STATE UNIVERSITY: Assurance Statement(s)

Investigator: Carolyn Ross	Phone: 509-335-2438
OGRD #: pending	E-mail: cfross@wsu.edu
Project Title: Processed fruit quality of newly released raspberry selections =	
Sponsor: CAHNRS - ARC	WSU Account #:

Biosafety and Chemical Safety

* includes livestock pathogens and toxins

Project does not involve recombinant DNA/Vectors/Plasmids; Infectious/Select Agents; Carcinogens, Mutagens, or Teratogens.

Project involves:

Recombinant DNA/Vectors/Plasmids

Carcinogens, Mutagens, or Teratogens

Infectious/Select Agents*

Reserved

MUA #: (if known)

Care and Use of Animals

Project does not involve vertebrate animals. Project involves vertebrate animals.

ASAF #: (if known)

Protection of Human Subjects

(includes the use of human tissue or bodily fluids)

Project does not involve human subjects.

Project does involve human subjects.

IRB #: (if known)

Approval is pending

Use of Radioactive Materials or Radiation Equipment

Project does not involve the use of radioactive materials or radiation equipment.

Project does involve the use of radioactive materials or radiation equipment

Conflict of Interest

Conflict of Interest definitions and questions (www.ogrd.wsu.edu/coi_dq/)

Project does not involve a Conflict of Interest

Project does involve a Conflict of Interest

Carolyn F. Ross
Sensory Scientist/Associate Professor
School of Food Science, Washington State University

EDUCATION:

- 2003 Certificate in University Teaching, Department of Teaching Resources and Continuing Education, University of Waterloo, Waterloo, ON.
- 2001 Doctorate of Philosophy in Food Science/Environmental Toxicology, Department of Food Science and Human Nutrition. Michigan State University, East Lansing, MI.
- 1997 Master of Science in Food Science, Department of Food Science, University of Guelph, Guelph, ON.
- 1995 Bachelor of Human Ecology in Foods and Nutrition, Department of Human Ecology, University of Manitoba, Winnipeg, MB.

PROFESSIONAL EXPERIENCE:

- 2010-present Associate Professor, WSU, School of Food Science, Pullman, WA.
- 2011 Visiting Researcher, Dept. of Wine, Food and Molecular Biosciences, Lincoln University, Lincoln New Zealand.
- 2004-2010 Assistant Professor, Washington State University, Department of Food Science and Human Nutrition, Pullman, WA.
- 2004 Post-doctoral Fellow, Cool Climate Oenology and Viticulture Institute, Brock University, St. Catharines, ON.
- 2003 Director of Research, CC Technology, Laramie, WY.
- 2003 Post-Doctoral Fellow, Dept. Chemistry, Laboratory Manager and Industrially Focused Analytical Research Laboratory, University of Waterloo, Waterloo, ON.
- 2002 Post-doctoral Fellow, Georgia Tech Research Institute, Food Processing Technology Division, Atlanta, GA.

TEACHING EXPERIENCE:

Current major advisor to 2 M.Sc. students and 4 Ph.D. students. Former major advisor to 14 graduate students (2 PhD and 12 MS).

Courses taught: Sensory Evaluation of Food and Wine Lecture (FS 422 3 cr annual); FS522 annual 3 cr), Sensory Evaluation of Food and Wine Laboratory (422 2 cr annual) , Introduction to Vines and Wines (FS 113, 3 cr annual), Graduate Oral Seminar (FS 518, 1 cr).

EXTENSION ACTIVITIES:

Teach the Sensory Evaluation Module in the WSU Viticulture and Enology Certificate Program annually x 120 hours of commitment. On-line instruction (lectures, exams and homework) and hands-on practicum. Extension presentations including wine sensory evaluation short courses and webinars in WA State, Idaho, Oregon and New Zealand.

AWARDS AND HONORS:

Finalist, Early Career Excellence Awards, College of Agriculture, Human and Natural Resource Science, Washington State University, Pullman WA (2012, 2013).

Provost's Leadership Development Program, Washington State University, Pullman, WA (2011-2012).

Certificate of Honor, Outstanding Mentor, Women & Leadership Alliance Forum, Washington State University (2006, 2007)

PROFESSIONAL SERVICE:

- IFT – Sensory and Consumer Science Division. Chair of SCSD White Paper Program. 2011 –present.
- IFT – Sensory and Consumer Science Division. Member of Scientific Review Committee. 2009 –present.
- IFT-Lewis & Clark Section. Executive Committee (Secretary). 2012 –present.
- Past Secretary 2005-2007.

Sensometric Society. 2005-present
American Chemical Society. 2004-present
American Society for Enology and Viticulture (ASEV). 2004-present.
ASEV Enology Technical Program Committee. 2011 – present.
Institute of Food Technologists (IFT) - National. 1996-present
IFT-Sensory Evaluation Division. Executive Committee (Member-at-Large). 2008 – 2010.
IFT – Sensory Evaluation Division. Chair of the Scientific Program Committee. 2009 –2011.
IFT-Lewis & Clark Section. 2004-present

SPECIAL TRAINING:

Provost's Leadership Development Program, WSU, Pullman, WA. 2011-2012

Certificate in Sensory Statistics, Hal MacFie Training Services, UK. 2004.

Certificate in University Teaching, Department of Teaching Resources and Continuing Education
University of Waterloo, Waterloo, ON. 2002.

Federal Institute of Food and Drug Law Summer Internship Program, Food and Drug Law Institute, Washington, DC. 1999.

PUBLICATIONS AND PRESENTATIONS:

Refereed journal articles (57, 51 since appointment at WSU 2004)
h-index=14 (as determined from Google Scholar)

Book chapters (5)

Extension/outreach publications and materials: Contributions to WSU Viticulture and Enology Extension News, 2006 – present.

Abstracts and papers presented at technical meetings: 61

PI Name(s): Carolyn Ross				YR1	YR2	YR3	YR4	YR5	YR6	TOTAL
Agency Name: WA Raspberry Commission				01/01/14 12/31/14	01/01/15 12/31/15	01/01/16 12/31/16				1/1/2014 12/31/2016
00 - SALARIES										
	Pay Rate	# Mos.	% FTE							
PI:	0.00	0.00	0.00%	Salary						
				Benefits						
PI:	0.00	0.00	0.00%	Salary						
				Benefits						
Co-PI:	0.00	0.00	0.00%	Salary						
				Benefits						
Co-PI:	0.00	0.00	0.00%	Salary						
				Benefits						
Co-PI:	0.00	0.00	0.00%	Salary						
				Benefits						
Post Doc/Research Assoc.:	0.00	0.00	0.00%	Salary						
				Benefits						
Classified Staff:	0.00	0.00	0.00%	Salary						
				Benefits						
PhD Student ~ Step #				Salary						
				QTR						
				Health						
				1.5%						
Master Student ~ Step # 26 one semester				Salary						
				QTR						
				Health						
				1.5%						
01 - WAGES										
	\$ Per Hr.	Hrs/Wks	# Wks.							
Student:	\$0.00	0	0	Wages						
				Benefits						
Student (enrolled part-time):	\$0.00	0	0	Wages						
				Benefits						
*Non-Student Temporary	\$18.25	20	24	Wages	8,760	8,760				17,520
				Benefits	850	850				1,700
MA student InSummer				Wages						
**Non-Student Temporary	\$0.00	0	0	Wages						
				Benefits						
***Non-Student Temporary	\$0.00	0	0	Wages						
				Benefits						
				Total Salary						
				Total Wages	8,760	8,760				17,520
				Total Salary & Wages	8,760	8,760				17,520
07 - BENEFITS										
				Total Benefits	850	850				1,700
				Total Salaries/Wages/Benefits	9,610	9,610				19,220
02 - PERSONAL SERVICES CONTRACTS (Consultants)										
				Total Personal Services Contracts						
03 - GOODS/SERVICES										
				Goods and services	1,400	1,400				2,800
				Total Goods/Services	1,400	1,400				2,800
04 - TRAVEL										
				Domestic						
				Foreign						
				Total Travel						
05 - COMPUTER SERVICES										
				Total Computer Services						
06 - CAPITAL EQUIPMENT (>\$5,000)										
				Total Capital Equipment						
08 - STIPENDS/SUBSIDIES/PARTICIPANT SUPPORT COSTS										
				Total Stipends/Subsidies/Participant Support Costs						
14 - RESTRICTED: incl. SUBAWARDS/SUBCONTRACTS										
	University One	YR-1	YR-2	YR-3	YR-4	YR-5				
	SBCT DC									
	SBCT IC (7%)									
	University Two	YR-1	YR-2	YR-3	YR-4	YR-5				
	SBCT DC									
	SBCT IC (7%)									
				Total Subcontractor DC						
				Total Subcontractor IC						
				Total Subcontracts/Restricted						
16 - NON-CAPITALIZED EQUIPMENT (e.g. Laptops w/agency approval) http://web1.wa.gov/Forms/116116122PM10_Purchasing/1010_Liquidize_Vocets_and_Subcontract/Item#88										
				Total Non-Capitalized Equipment ("Small & Attractive" Items)						
TOTAL DIRECT COSTS					11,010	11,010				22,020
EXCLUSIONS										
				QTR						
				Equipment (Over 5k)						
				Subcontracts, amt excluding first 25K (per subcontract), HAND ENTER.						
				Other (Off-Site Rental & Stipends, Etc)						
				Total Exclusions						
MTDC BASE				Base	11,010	11,010				22,020
13 - FACILITIES & ADMINISTRATIVE COSTS (F&A, IDCs, OVERHEAD)				F&A Rate:						
TOTAL COSTS					11,010	11,010				22,020
F&A Base Type:				MTDC	TD	TC	SWB	Other		

Approved Ben Weller 10/25/2013

New Project Proposal: Yes
Proposed Duration: One year

Project Title: Innovative Packaging Technologies To Enhance The Safety And The Quality Of Fresh Raspberry

PI: Thomas J Gianfagna

Organization: Department of Plant Biology and Pathology, Rutgers University, New Brunswick, NJ 08901,

Title: Professor

Phone: (848) 932- 6369

Email: gianfagna@aesop.rutgers.edu

Co-PI: Kit L Yam

Organization: Department of Food Science, Rutgers University, New Brunswick, NJ 08901

Title: Professor

Phone: (848) 932- 5467

Email: yam@aesop.rutgers.edu

Cooperators: Von Thun's Farms, South Brunswick, NJ

Year Initiated 2014 **Current Year** 2014 **Terminating Year** 2014

Total Project Request: \$\$6,777

Other funding sources: Yes

Agency Name: American Floral Endowment

Amt. Requested: \$9,943

Notes: Currently, we are studying the effects of these types of packaging systems for extending the freshness and shelf life of cut flowers with a grant from the American Floral Endowment.

Description: The major goal of this project is to develop innovative packaging systems for fresh raspberry by effectively integrating the existing and emerging technologies available to enhance the quality and safety of fresh raspberry and extend its shelf-life. Currently, we are studying the effects of these types of packaging systems for extending the freshness and shelf life of cut flowers with a grant from the American Floral Endowment (AFE). Funds from WRRC will be leveraged with the AFE grant by our interdisciplinary team to complete the following objective:

Develop a science-based approach to extend the shelf life of fresh produce, using packaging as a delivery means, based on the following strategies:

- a. minimize microbial safety hazards using controlled release of natural antimicrobials.
- b. extend the shelf life of raspberry using MAP bags.

Justification and Background: This project integrates several existing or emerging technologies (natural antimicrobials, modified atmosphere packaging, and controlled release packaging) to develop new packaging systems for enhancing food quality and safety of fresh raspberries.

The major postharvest pathogen of strawberry is *Botrytis cinerea*, the causal agent of gray mold disease. Essential oils (EO) have anti-microbial activity against *Botrytis cinerea* (1, 2, 3, 4), and are regarded as safe (GRAS). The EO's are incorporated into controlled release packaging (CRP) that release active compounds at differentiable rates suitable for short-term or intermediate-term inhibition of microorganisms in fresh foods. For our delivery system, we encapsulated EO's into cyclodextrin (CD) and wrapped these capsules into a sachet, which can be placed inside the

packaging. The motivation for developing CRP is to prolong stable shelf life without overloading foods with additives. CRP can continuously replenish active compounds from packaging, which is necessary for achieving long term stability of foods.

While there are no other projects being conducted in British Columbia, Oregon, and Idaho that deal directly with postharvest shelf life extension of red raspberry, there are projects that focus on improving raspberry cultivars to have greater appeal and fruit quality, and for preharvest fungicide treatment that provides postharvest control of Botrytis. Innovative packaging would have synergy with both of these projects.

Experiments were done to evaluate the antimicrobial effectiveness of 3.5% thyme oil (*Thymus vulgaris*) (TO) incorporated into CD on strawberries stored for 8 d, and blueberries stored for 30 d at -1 °C with 94% humidity, with or without MAP (VFA bag), or TO:CD sachets. We found that fruit that was stored using MAP (VFA bag) and TO:CD sachets had significantly less decay and less weight loss other treatments as shown in Table 1 and Table 2.

Table 1. Effects of TO:CD sachets on strawberry fruit quality after 7 d storage at -1 °C^a

Treatment	Disease Incidence (%)	Change in wt after 7 d (g)	Firmness (N/cm ²)	TSS (° Brix)
- TO:CD sachet + VFA bag	36.4a	-1.81b	6.75b	7.22b
- TO:CD sachet - VFA bag	22.4b	-9.99a	7.94b	8.03a
+ TO:CD sachet - VFA bag	13.9c	-8.21a	7.94b	7.13b
+ TO:CD sachet+ VFA bag	11.3c	-5.88b	9.39a	8.03a

^a Means in the same column with the same letter are not significantly different ($P \leq 0.05$).

Table 2. Effects of TO:CD sachets on blueberry fruit quality after 30 d storage at -1 °C and 3 d at 15 °C^a

Treatment	Disease Incidence (%)	Change in wt after 7 d (g)	Firmness (N/cm ²)	TSS (° Brix)
- TO:CD sachet + VFA bag	42a	-10.2b	13.5a	12.6b
- TO:CD sachet - VFA bag	40a	-27.6a	12.2b	14.3a
+ TO:CD sachet - VFA bag	42a	-25.6a	12.3ab	12.7b
+ TO:CD sachet+ VFA bag	28b	-12.8c	13.4ab	12.3b

^a Means in the same column with the same letter are not significantly different ($P \leq 0.05$).

Relationship to WRRC Research Priority: #1 fruit rot including pre harvest, post harvest, and shelf life.

Objectives

The following objectives will be addressed this funding year:

- Encapsulation of TO into cyclodextrin
- Production of TO:CD Tyvek sachets
- Fruit Storage Experiment using sachets and MAP

Procedures

Essential Oil Sachets, and MAP

Controlled release Tyvek™ sachets of 3.5% thyme oil, encapsulated by β-cyclodextrin will be made (5). MAP bags will be donated by Orchard View Farms, The Dalles, OR.

Plant materials

Raspberry fruit will be obtained from local commercial sources and transported to the laboratory at Rutgers University, New Brunswick, NJ. The fruit will be sorted so as to select only well formed, rot free, unblemished fruit.

Treatments

4 cm x 4cm Tyvek™ sachets will be filled with 0.5 g TO:CD prepared from a ratio of 14:86 TO:CD and heat sealed.. Alternatively, 4 cm x 4cm Tyvek™ sachets will be filled with different amounts of CD without TO (control) and heat sealed. Two sachets will be adhered to the bottom of a clamshell package raspberries will be added. The raspberries will be pre-cooled to 5 °C prior to packing. The clamshells will be enclosed in MAP bags and placed in a 0 °C cold storage room for 5 days at 94% humidity. Each treatment will have 12 replicates. Fruit quality parameters such as weight loss, firmness, total soluble solids (TSS) (6), and fungal decay will be measured after storage.

Statistical Analysis

Data for the physical, chemical and microbiological parameters will be subjected to analysis of variance (ANOVA) and to test significant differences between means with $P < 0.05$. The proof of concept will be considered successfully achieved if the treatments are statistically significant compared to the controls.

Timetable (one year)

Objective	Tasks	Begin	Complete
Minimize gray mold	Measure fungal decay	2/1/2014	11/7/2014
Extend shelf life	Measure fruit quality parameters	2/1/2014	11/7/2014
Final Report			11/7/2014

Anticipated Benefits and Information Transfer: The raspberry industry will benefit from fruit that has less decay and less weight loss, which will extend shelf life. The results will be transferred to the users as a commercially available sachet.

References:

- 1) Antunes, M.D.C.; Cavaco, A.M. (2010) The use of essential oils for postharvest decay control. A review. *Flavour Fragr. J.* 25: 351-366.
- 2) Serrano, M.; Martínez-Romero, D.; Guillén, F.; Valverde, J.M.; Zapata, P.J.; Castillo, S.; Valero, D. (2008) The addition of essential oils to MAP as a tool to maintain the overall quality of fruits. *Food Sci. Tech.* 19: 464-471.

- 3) Tzortzakis, N.G. (2006) Maintaining postharvest quality of fresh produce with volatile compounds. *Innov. Food Sci. Emerg.* 8: 111-116.
- 4) Reddy, B.M.V.; Angers, P.; Gosselin, A.; Arul, J. (1998) Characterization and use of essential oil from *Thymus vulgaris* against *Botrytis cinerea* and *Rhizopus stolonifer* in strawberry fruits. *Phytochem.* 47: 1515-1520.
- 5) Del Toro-Sánchez, C.L., Ayala-Zavala, J.F., Machi, L., Villegas-Ochoa, M.A., Alvarez-Parrilla, E., González-Aquilar, G.A. Controlled release of antifungal volatiles of thyme essential oil from β -cyclodextrin capsules. (2010) *J. Incl. Phenom. Macrocycl. Chem.* 67:431-441.
- 6) Almenar, E.; Catala, R.; Hernandez-Muñoz, P.; Gavara, R. (2009) Optimization of an active package for wild strawberries based on the release of 2-nonanone. *Food Sci Tech.* 42: 587-593.

Budget (2014)

Item	AFE	WRRC	Combined Total
Salary (part time technician)	\$5,440 (\$40/h for 136 h)	\$5,440 (\$40/h for 136 h)	\$10,880
Operations (supplies)	\$4,095 (chemicals, greenhouse rental)	\$929 (cold room rental, raspberries, clamshells)	\$5,024
Fringe Benefits	\$408	\$408	\$816
Total	\$9,943	\$6,777	\$16,720

Budget Justification: The funds from WRRC will be leveraged with funds from AFE to fund a part-time technician at the rate of \$40.00 an hour plus fringe. The FTE is 0.33 for the WRRC project. The fringe benefit rate for a part-time or time-slip employee is 7.5%.

**2014 WASHINGTON RED RASPBERRY COMMISSION
RESEARCH PROPOSAL**

New Project Proposal

Proposed Duration: 2 years

Project Title: Ultraviolet light (UV-C) treatment for improving safety of red raspberries

PI: Shyam Sablani Organization: WSU Title: Associate Professor Phone: 509-335-7746 Email: ssablani@wsu.edu Address: Biological Systems Engr Address 2: PO Box 646120 City/State/Zip: Pullman, WA 99164	Co-PI Karen Killinger WSU Associate Professor 509-335-2970 karen_killinger@wsu.edu FSHN 229, Sch of Food Sci PO Box 646376 Pullman, WA 99164	Co-PI Girish Ganjyal WSU Assistant Professor 509-335-5613 girish.ganjyal@wsu.edu FSHN 228, Sch of Food Sci PO Box 646376 Pullman, WA 99164
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Cooperators: Enfield Farms

Year Initiated 2014

Current Year 2014

Terminating Year 2015

Total Project Request: **Year 1** \$ 13,695 **Year 2** \$17,760 **Year 3** \$0/-

Other funding sources: *None*

Description:

Most of the raspberries grown in the state of Washington are Individual Quick Frozen (IQF). Due to the delicate nature of the fruit, they are minimally treated with a chlorine spray wash before freezing. This chlorine spray used in the current process is not adequate. The raspberries cannot be washed thoroughly with chlorine water like apples or other fruits. Fragility, shape and surface characteristics of the raspberries limit the usage of thorough washing of the fruit. There have been incidences when bacterial pathogens such *Salmonella* and *Listeria monocytogenes* are isolated in fresh and frozen berries. The FDA Food Safety Modernization Act requires growers and packers of fresh produce, and processors to identify and adopt effective preventive controls to reduce microbial risk in fresh produce and ensure the safety of processed products. The *specific objective* is to investigate the efficiency of ultraviolet light-C in inactivating at least two foodborne pathogens (*e.g. E. coli, Listeria* or *Salmonella*) on frozen raspberries and to investigate its effects on berry quality. Improved safety of frozen fruit will provide economic incentives for berry growers and processors. This research addresses priority #3 Product and Production Certification Systems - food safety & security, standards, traceability.

Justification and Background:

Of all the small fruits, raspberries have the shortest shelf life. They become very soft in a matter of hours after harvest if not stored in cold temperatures, and mold grows easily on raspberries kept on consumer's counters at room temperature. The lack of effective postharvest control of gray mold is the most important single factor limiting the sale of raspberry fruit for distant markets (Ellis et al., 2008). Although hot water treatments, biological control, and chemical applications have shown to reduce postharvest rots of fruits, each has limitations that can affect commercial applicability. Due to the delicate nature of raspberry fruit, hot water treatment is not a viable option to control fruit rot. Biological methods have not lived up to their early promise, and only a very limited number of biofungicides are available on the market. Moreover, the intensive use of chemicals has resulted in the buildup of chemically resistant fungi. Therefore, the emphasis in postharvest fruit protection has shifted from chemical to physical methods.

Washington State is one of the leading producers of raspberry fruit. Due to short postharvest life most of the raspberries grown in the state of Washington are individual Quick Frozen. The raspberries are minimally treated with minimal chlorine spray wash before freezing. Recent outbreaks of *E. coli* O157:H7, *Salmonella* and *Listeria monocytogenes* in fresh fruits, fruit juices and frozen berries have demonstrated that microbial risks exist for these products (Bower et al, 2003). Moreover, the U.S. Food and Drug Administration (FDA) Food Safety Modernization Act requires growers and packers of fresh produce, and processors to identify and adopt effective preventive controls to reduce microbial risk in fresh produce and ensure the safety of processed products.

The use of ultraviolet light (UV) light is well established for water treatment, air disinfection and surface decontamination. Despite the efficacy of UV-C light to disinfect smooth surfaces, there are relatively few applications of this technology in the food processing industry. Since complex interactions may occur between microorganisms and surface materials, such as shielding effects from incident UV, efficacy of UV light depends on surface structure or topography (Syamaladevi et al., 2013 and 2014). There is a need to develop effective interventions that can help reduce the pathogen load on fruit surface. There is not much reported peer reviewed literature on the effectiveness of UV-C light in frozen fresh products. Because of the various unanswered questions this research is proposed.

Relationship to WRRRC Research Priority(s):

#3 priorities

- Product and Production Certification Systems - food safety & security, standards, traceability

Objectives:

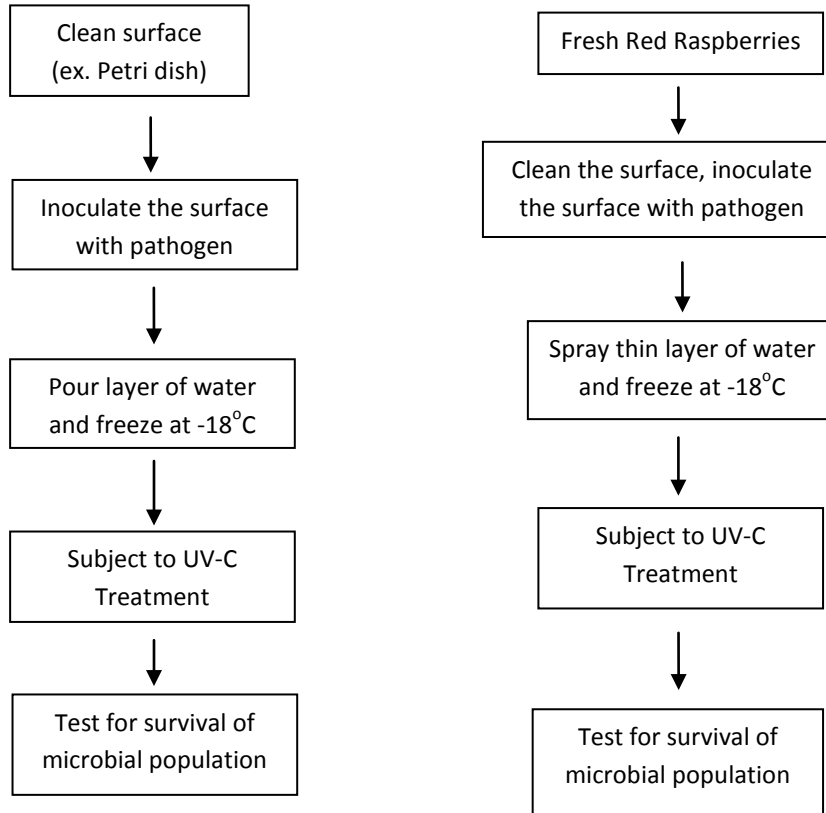
The *specific objectives* are:

1. to investigate the efficiency of ultraviolet light-C in inactivating at least two foodborne pathogen (*e.g. E. coli, Listeria* or *Salmonella*) on frozen surfaces and on frozen red raspberries (**year 1**),
2. to continue UV-C inactivation study on other pathogens and investigate the influence of UV-C treatment on physical and chemical quality of berries during frozen storage (**year 2**).

Procedures:

Anticipated length of project is two years.

We propose to address the two questions on the laboratory scale at WSU laboratories, with process shown in the flow-charts below.



Materials: Washington-grown red raspberries will be obtained from local farmers and grocery stores. *E. coli* O157:H7, *Salmonella* or *Listeria monocytogenes* will be cultured and purified in the WSU Food Microbiology Laboratory.

UV-C treatment of bacteria on fruit surfaces: The UV-C irradiation device (Reyco Systems, Inc., Meridian, ID), consisting of four Steril-Aire™ 16SE food-grade, shatter-resistant, sleeved UVC Emitters™ mounted in bulkhead fittings will be used for bacteria inactivation study. The UV-C dose will be controlled by altering the distance between UV lamps and the fruit surfaces, as well as and time of exposure. Initially, the efficacy of UV-C will be tested on bacteria grown in the petri dish covered by ice layer. Later similar experiments will be conducted with bacteria on frozen fruit surfaces. Pathogens will be inoculated on the fruit surface before and after freezing of raspberries (flow chart shown above). The UV-C illumination duration will be 5 to 180 seconds, and illumination dosages will be 0.28 to 10 kJ/m². We will follow a standard protocol of dilutions, plating, and incubation recommended by Schenk et al. (2008). The overall preparation of the samples for UV-C treatment will be conducted inside a Class II security cabinet to avoid post-contamination.

Kinetics of microbial inactivation: To quantify the UV-C dose required to achieve the desired reduction in the microbial load, the experimental data of microbial inactivation will be analyzed using kinetic equations.

Physical and chemical analysis: Fruit quality will be determined before and after UV-C treatments. To determine whether treatment with UV-C light has negative effects on the visual appearance of fruit surface, instrumental color analysis will be performed. The Hunter L (lightness), a (redness-greenness), and b (yellowness-blueness) values will be measured with a Chroma Meter CR-300 (Konica Minolta Co. Ltd., Ramsey, NJ), which will be calibrated with the standard white tile prior to use. Fruit firmness will be determined using a texture profile analyzer (Stable Micro Systems, Limited, NY). Total anthocyanin will be extracted with 1% HCl:methanol, measured spectrophotometrically, and quantified using the appropriate molar extinction coefficient. Total phenolic content will be determined according to the Folin-Ciocalteu procedure, and will be expressed as gallic acid equivalents (GAE) in milligrams per kilogram of fresh weight (Singleton et al., 1999). Ellagic acid in raspberries will be determined with HPLC, as described by Crozier et al. (1997). Analysis of total antioxidant activity will be conducted using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) method (Gordon, 2001).

Anticipated Benefits and Information Transfer: (100 words maximum)

The research project will enhance scientific understanding of UV-C light inactivation of selected microorganisms on frozen raspberries. New research methodology developed from this project can be applied to other human pathogenic bacteria. The information acquired will provide a viable, alternate physical method to sanitize raspberries, increasing the microbial safety frozen berries. This improved microbial safety of raspberries will provide economic incentives to Washington red raspberry industry. The findings will be communicated to fruit growers and processors in the NW region. We also plan to present results at regional meetings such as the Northwest Food Processors Association.

References:

- Bower, C. K., Stan, S., Daeschel, M. and Zhao, Y. (2003). Oregon State University., Extension Service.
- Crozier, A., Lean, E. J. L., McDonald, M. S., Black, C. (1997). Quantitative analysis of the flavonoid content of commercial tomatoes, onions, lettuce, and celery. *Journal of Agricultural and Food Chemistry* 45, 590-595.
- Ellis, M. A., Madden, L. V., Wright, S. R., Madden, L. V., and Wilson, L. L. 2008. Plant Health Progress doi:10.1094/PHP-2008-1015-01-RS.
- Gordon, M. H. (2001) In *Antioxidants in food: practical applications*. J. Pokorny, N. Yanishlieva, M. Gordon (eds.). CRC Press, Boca Raton, 2001: 71-84.
- Schenk, M, Guerrero, S, and Alzamora, S. M. (2008). Food Bioprocess Technol. 1:384 -392
- Singleton V. L., Orthofer R., Lamuela-Raventos R. M. (1999). Analysis of total phenols and other oxidation substrate and antioxidants by means of Folin-Ciocalteu reagent. *Methods in Enzymology* 299: 152-179.
- Syamaladevi, R. M., Lupien, S., Bhunia, K., Sablani, S. S., Dugan, F., Rasco, B., Killinger, K., Dhingra, A., and Ross, C. 2014. *Postharvest Biology and Technology* 87: 27-32
- Syamaladevi, R. M., Xiaonan Lu, Sablani, S. S., Insan, S. K., Adhikari, A., Killinger, K., Rasco, B., Dhingra, A., Bandyopadhyay, A. and Annapure, U. S. 2013. *Food and Bioprocess Technology* 6: 2959-2973

Budget: Indirect or overhead costs are not allowed unless specifically authorized by the Board

	2014	2015	2016
Salaries^{1/}	\$ 5,324	\$ 7,087	\$
Time-Slip	\$1,500	\$ 1,560	\$
Operations (goods & services)	\$4,500	\$ 5,500	\$
Travel^{2/}	\$	\$ 500	\$
Meetings	\$	\$	\$
Other	\$	\$	\$
Equipment^{3/}	\$	\$	\$
Benefits^{4/}	\$ 2,371	\$ 3,113	\$
Total	\$ 13,695	\$ 17,760	\$

Budget Justification -

Budget requested is to cover the partial support of a research technician II (12.5% year one and 16% year two) and a time-slip for conducting the microbial and quality evaluation experiments, respectively. Goods and Services/Supplies is also requested to cover the costs for the UV instrument, microbial media and physical and chemical quality evaluation. Travel in the amount of \$500 is requested during the second year. Travel will be related to project work and not to attend any workshops or conferences. We will use the travel funds to visit the processing facilities in Lynden area and to disseminate information from the research to the stakeholders.

Project Timeline –

Year 1: Feasibility of testing transparency of UVC light to pass through ice and inactivate pathogens.

Year 2: Inactivation kinetics of at least two pathogens, physical and chemical quality of fruit after UVC treatment and during storage, and feasibility study of integrating UV-C system on processing and packing lines.

CURRENT & PENDING SUPPORT

Name: **Shyam S. Sablani**

Instructions: Who completes this template: Each project director/principal investigator (PD/PI) and other senior personnel that the Request for Applications (RFA) specifies **How this template is completed:**

- Record information for active and pending projects, including this proposal.
- All current efforts to which PD/PI(s) and other senior personnel have committed a portion of their time must be listed, whether or not salary for the person involved is included in the budgets of the various projects.
- Provide analogous information for all proposed work which is being considered by, or which will be submitted in the near future to, other possible sponsors, including other USDA programs.
- For concurrent projects, the percent of time committed must not exceed 100%.

NAME (List/PD #1 first)	SUPPORTING AGENCY AND AGENCY ACTIVE AWARD/PENDING PROPOSAL NUMBER	TOTAL \$ AMOUNT	EFFECTIVE AND EXPIRATION DATES	% OF TIME COMMI TTED	TITLE OF PROJECT
Barbosa- Canovas, Bermudez- Aguirre, Sablani,	Active: Defense Logistics Agency, Department of Defense	\$767,481	March, 2010 to February, 2014	5%	Improving Thermal Processing of Foods Sealed in Military-Ration Polymeric Trays
Tang, Rasco, Sablani, Davidson, D'Souza, Huang, Dunne, Yang, Wright, Gray	USDA-NIFA-AFRI	\$3,999,998	March, 2011 to March, 2014	9%	Control of Food-borne Bacterial and Viral Pathogens using Microwave Technology

Tang, Barbosa-Canovas, Sablani, Davis	USDA-NNF	\$238,500	March 2012 to March 2016	5%	Educating Food Engineers to Develop High-Performance Integrated Processing and Packaging Technologies that Enhance
Sablani, Killinger	BIOAg program	\$51,445	April 2012-June 2014	5%	Sustainable Sanitation Technique for Postharvest Quality and Safety of Organic Fruits
Ganjyal, Sablani, Wang	<u>Pending</u> Washington Cherry Commission	\$50,000	May 2014 – April 2016	5%	Strategies to Reduce Postharvest Cracking and Splitting of Cherries by Processing and Packaging Techniques
Sablani, Chen, Bule, Ganjyal	ERI: ARC/WSU	\$79,932	January 2014 to December 2015	5%	Smart Sustainable Functional Foods using Bioactive Compounds for Improving Human Health
J. Tang, M. Zhu, S. Sablani, G. Ganjyal, D. Shah	ERI: ARC/WSU	\$79,189	January 2014 to December 2015	2%	Understanding of Food and Microbiological Properties at Elevated Temperatures to Improve Low-moisture Food Safety
Sablani, Killinger, Ganjyal	Washington Red Raspberry Commission	\$31,455	January 2014 to December 2015	2%	Ultraviolet light (UV-C) treatment for improving safety of red raspberries

CURRENT & PENDING SUPPORT

Name: **Karen Killinger**

Instructions:

Who completes this template: Each project director/principal investigator (PD/PI) and other senior personnel that the Request for Applications (RFA) specifies

How this template is completed:

- Record information for active and pending projects, including this proposal.
- All current efforts to which PD/PI(s) and other senior personnel have committed a portion of their time must be listed, whether or not salary for the person involved is included in the budgets of the various projects.
- Provide analogous information for all proposed work which is being considered by, or which will be submitted in the near future to, other possible sponsors, including other USDA programs.
- For concurrent projects, the percent of time committed must not exceed 100%.

NAME (List/PD #1 first)	SUPPORTING AGENCY AND AGENCY ACTIVE AWARD/PENDING PROPOSAL NUMBER	TOTAL \$ AMOUNT	EFFECTIVE AND EXPIRATION DATES	% OF TIME COMMIT TED	TITLE OF PROJECT
	<u>Active</u>				
Killinger, Bary, Cogger, Dougherty	Washington State Specialty Crop Block Grant	\$103,246	2011-2014	10%	Advancing food safety GAPs among Washington specialty crops growers
Fan, Gurtler and Killinger	Center for Produce Safety	\$168,000	2012-2013	5%	Sanitization of soft fruits with non-aqueous antimicrobials

Killinger, Hanrhan, Doughert, Fellman	Washington Tree Fruit Research Commission	\$72,313	2012-2013	10%	Enhancing apple packing programs while ensuring fruit quality
Killinger, Adhikari	Center for Produce Safety	\$171,313	2013-2015	10%	Assessment of sanitization techniques for tree fruit storage bins
Killinger, Cogger, Bary	Washington State Specialty Crop Block Grant	\$232,423	2012-2015	10%	Evaluation of irrigation practices for specialty crop risk reduction
Killinger, Coles, Morrissey	Washington State Specialty Crop Block Grant	\$152,129	2012-2014	10%	Strengthening food safety management systems for specialty crop production, packing and processing in the Pacific Northwest
Killinger, Meschke	Washington Tree Fruit Research Commission	\$120,000	2012-2014	10%	Evaluation of irrigation practices for specialty crop risk reduction
Sablani, Killinger	BIOAg program	\$51,445	2012-2014	5%	Sustainable Sanitation Technique for Postharvest Quality and Safety of Organic Fruits
Sablani, Killinger, Ganjyal	<u>Pending</u> Washington Red Raspberry Commission	\$31,455	2014 to 2015	5%	Ultraviolet light (UV-C) treatment for improving safety of red raspberries

CURRENT & PENDING SUPPORT

Name: Girish Ganjyal

Instructions:

Who completes this template: Each project director/principal investigator (PD/PI) and other senior personnel that the Request for Applications (RFA) specifies

How this template is completed:

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- Provide analogous information for all proposed work which is being considered by, or which will be submitted in the near future to, other possible sponsors, including other USDA programs.
- For concurrent projects, the percent of time committed must not exceed 100%..

Note: Concurrent submission of a proposal to other organizations will not prejudice its review by CSREES.

NAME (List/PD #1 first)	SUPPORTING AGENCY AND AGENCY ACTIVE AWARD/PENDING PROPOSAL NUMBER	TOTAL \$ AMOUNT	EFFECTIVE AND EXPIRATION DATES	% OF TIME COMMITTED	TITLE OF PROJECT
<u>G. Ganjyal</u>	Active: New Faculty Seed Grant Competition – Washington State University	\$21,000	06/01/2013 to 06/30/2014	3%	<i>Enhancing value of the Washington State’s processed fruit and vegetable by-products through innovative processing techniques</i>
A. Fatland, <u>G Ganjyal</u> , C. Love, J. Leachman, A. Holzer, V. Yadama, et. al.	U.S. Economic Development Administration	\$163,000	01/01/2014 to 12/31/2014	2%	<i>Investing in Manufacturing Communities Partnership Planning Grant</i>
<u>G. Ganjyal, S. Sablani, Y. Wang</u>	Pending: Washington Cherry Commission	\$50,000	05/01/2014- 05/01/2016	5%	<i>Strategies to reduce postharvest cracking and splitting of cherries by processing and packaging techniques</i>

R. Visvanathan, <u>G. Ganjyal</u> , C. Morris, K. Ambrose., D. Maier, S. Hiregoudar	Indo-US Science and Technology Forum (IUSSTF)	\$145,760 (WSU - \$31,460)	01/01/2014 to 12/31/2015	2%	<i>Indo-US Joint Centre on “Value added processing of millets”</i>
<u>Ganjyal, G</u>	Lamb Weston (ConAgra Foods., Inc.), Richland, WA	\$32,000	11/15/2013 to 01/15/2014	2%	<i>Understanding the Changes in Physico-Chemical and Structural Characteristics of Potato Strips during Frying and Freezing Unit Operations in French Fry Processing</i>
<u>G. Ganjyal</u> L. Lei K. Englund	Emerging Research Issues (ERI) for Washington Agriculture 2014 Internal Competitive Grant Program	\$78,593	01/01/2014-12/31/2016	5%	<i>Development of Biobased Materials from Low Value Byproducts of Agricultural Processing of Washington State</i>
J. Tang, M. Zhu, S. Sablani, <u>G. Ganjyal</u> , D. Shah	ERI for Washington Agriculture 2014 Internal Competitive Grant Program	\$79,189	01/01/2014-12/31/2016	1%	<i>Understanding of Food and Microbiological Properties at Elevated Temperatures to Improve Low-moisture Food Safety</i>
S. Sablani, S. Chen, M. Bule, <u>G. Ganjyal</u>	ERI for Washington Agriculture 2014 Internal Competitive Grant Program	\$79,932	01/01/2014-12/31/2016	1%	<i>Smart Sustainable Functional Foods (S2F2) Development Using Bioactive Compounds for Improving Human Health</i>

K. Murphy, <u>G. Ganjyal</u> , J. Matanguihan, G. Noratto	ERI for Washington Agriculture 2014 Internal Competitive Grant Program	\$77,500	01/01/2014-12/31/2016	3%	<i>Putting Barley back on our Plates: Development and extension of nutritious whole grain hullless barley</i>
Sablani, Killinger, ganjyal	Washington Red Raspberry Commission	\$31,455	01/01/2014-12/31/2015	1%	<i>Ultraviolet light (UV-C) treatment for improving safety of red raspberries</i>

Washington Red Raspberry Commission Progress Report for 2013 Projects

Project No: 14C-4799-1440

Title: Connecting Raspberry Breeding to Crop Markets (amount: \$1,000)

Personnel: Catherine H. Daniels and Patrick Moore

Reporting Period: CY2013

Accomplishments/Results:

- The WRRRC funds are being held (unspent) until the USDA-SCRI program releases an RFA (they did not release one in 2013 as we had hoped)
- We expect the USDA-SCRI RFA to be released in early 2014
- When that RFA is released, we will submit our proposal “Integrating Consumer Preferences with Molecular Mapping and Traditional Breeding Efforts within the US Red Raspberry Industry” to USDA
- The WRRRC monies will be used as part of the industry match for the USDA-SCRI proposal we have under development. If funded by USDA, we will use the WRRRC monies to support Dr. Moore’s breeding program.

Publications:

- None yet.

Washington Red Raspberry Commission Progress Report for 2013 Project

Project No:

Title: First Alert Scouting of SWD in Western WA

Personnel: Colleen Burrows, Chris Benedict, and Katherine East, WSU Whatcom County Extension

Reporting Period: January to November 2013

Accomplishments:

A variety of trap designs and baits were tested at 4 farms in Whatcom County. Traps were placed in fields on June 3, 2013 and were monitored weekly for SWD males and females trapped until the end of raspberry harvest (late July or early August). Trap and bait combinations were chosen in collaboration with the project at Oregon State University with Dr. Amy Dreves.

Traps tested were:

- Standard, 10-hole clear cup
- Clear cup with mesh side
- Red cup with mesh side
- Contech commercial trap
- Captiva commercial trap

Baits tested were:

- Apple cider vinegar
- Bread yeast and sugar combination
- Monterey Insect Bait (corn steep liquor)
- Suzukii Trap (from Spain)
- Biolure bait
- Torula Yeast Bait

Results:

The most attractive baits early in the season and later in the season were the bread yeast and sugar combination and the Suzukii Trap bait (from Spain). Growers would attract more SWD to these baits and potentially get a better idea of fly numbers in the field. The bread yeast bait is not as convenient as the standard apple cider vinegar, but it is less expensive. The Suzukii Trap bait is not yet available in the United States, but may be more convenient as it does not need to be mixed prior to use.

The most attractive trap model was the clear cup with a mesh panel on the side. These traps allow a greater surface area for the fly to enter the cup, perhaps resulting in a higher catch. Unfortunately, they are time consuming to build, but can be re-used year to year.

Publications:

A final report of this work, including results and pictures of traps will be posted on the website: <http://whatcom.wsu.edu/ipm/swd/index.html>

Project No: 13C-3755-6040

Title: Evaluation of resistance to *Verticillium dahliae* and *Phytophthora rubi* in red raspberry

PI: Patrick Moore, Horticulture, Washington State University, Puyallup Research and Extension Center, 2606 W Pioneer Ave., Puyallup, WA 98371; moorepp@wsu.edu, 253-445-4525.

Co-PIs: Jerry Weiland, Plant Pathology, USDA-ARS, 3420 NW Orchard Avenue, Corvallis, OR 97330; jerry.weiland@ars.usda.gov, 541-738-4062.

Wendy Hoashi-Erhardt, Horticulture, Puyallup Research and Extension Center, 2606 W Pioneer Ave., Puyallup, WA 98371; wkhe@wsu.edu, 253-445-4641.

Reporting period: 2013

Accomplishments: Thirty-six raspberry genotypes representing important cultivars and breeding material were subjected to inoculation with microsclerotia of *Verticillium dahliae* in a greenhouse study commenced in May 2013 (Test A). A second identical study was initiated a month later in June 2013 (Test B).

Verticillium dahliae (originally isolated from red raspberry cultivar ‘**Saanich**’) inoculum was produced using a method adapted from López-Escudero et al. (2007). The number of microsclerotia/gram of inoculum was determined and mixed into potting media at rates suitable to obtain a density of 100 microsclerotia/gram before planting with the raspberry genotypes. Potting media is prepared from collected field soil that was sieved and mixed with equal parts sand, then autoclaved.

Raspberry plants with 4-6 fully expanded leaves and three months out of tissue culture were planted and evaluated in potting media with five replicates and two treatments, 1) a non-inoculated control and 2) inoculated with *V. dahliae*. Plants were maintained in the greenhouse with ambient seasonal daylengths.

Cane heights were measured 12 Sept 2013 and analyzed (Table 1). There was a significant effect of cultivar for both tests ($p=0.0001$ on both). There was a significant effect of verticillium inoculation for both tests ($p=0.0001$ and $p=0.0207$, respectively), but there was no significant interaction of cultivar and *Verticillium* inoculation, indicating that a difference in response to *Verticillium* among the cultivars was not apparent at the time of measurement. *Verticillium* inoculation resulted in smaller cane heights for both tests, with the earlier test showing a greater disparity in height between non-inoculated and inoculated plants. ‘Munger’, a black raspberry known for its susceptibility to *Verticillium*, showed a major difference between non-inoculated and inoculated plant height, more dramatically so in the first and older test.

Results: These are preliminary results only, on experiments only 3 to 4 months after initial exposure to the pathogen. Observations of verticillium disease development in black raspberry and red raspberry indicate that plant symptoms becomes more apparent after a period of dormancy and spring growth. To capture this disease progression, plants will be maintained over the winter, through dormancy, and into the spring and summer of 2014 for further monitoring of response to *Verticillium*.

No funds are requested for 2014.

Table 1. Mean cane heights in cm of 36 red raspberry cultivars inoculated with microsclerotia of *Verticillium dahliae* on two planting dates in the greenhouse.

		Non-	Verticillium-			Non-	Verticillium-
Clone		inoculated	inoculated	Clone		inoculated	inoculated
4-month				3-Month			
study	Algonquin	28	34	study	Algonquin	32	30
	Autumn Bliss	48	47		Autumn Bliss	43	41
	Cascade Bounty	50	44		Cascade Bounty	40	34
	Cascade Delight	35	22		Cascade Delight	30	29
	Chemainus	58	46		Chemainus	45	39
	Cherokee	40	26		Cherokee	31	25
	Chief	9	10		Chief	30	21
	Chilliwack	57	54		Chilliwack	47	58
	Citadel	33	20		Citadel	29	24
	Dinkum	64	44		Dinkum	52	45
	Glen Prosen	34	27		Glen Prosen	34	35
	Haida	28	20		Haida	32	27
	Heritage	56	36		Heritage	43	41
	Killarney	28	21		Killarney	33	29
	Latham	58	41		Latham	.	.
	Lewis	72	46		Lewis	58	55
	Malahat	28	22		Malahat	28	29
	Mandarin	18	20		Mandarin	10	10
	Meeker	120	92		Meeker	62	68
	Munger	128	92		Munger	109	101
	Newburgh	53	57		Newburgh	48	41
	Nootka	55	41		Nootka	43	40
	Nova	20	9		Nova	30	36
	Puyallup	30	22		Puyallup	32	31
	Qualicum	27	27		Qualicum	33	36
	Reveille	26	20		Reveille	29	31
	Saanich	43	52		Saanich	48	41
	Skeena	49	54		Skeena	40	34
	Success	57	31		Success	4	4
	Summit	54	56		Summit	50	52
	Sumner	27	22		Sumner	30	31
	Tahoma	28	28		Tahoma	28	29
	Tulameen	48	51		Tulameen	48	41
	Wakefield	32	26		Wakefield	33	32
	Washington	40	52		Washington	46	42
	Willamette	71	76		Willamette	56	50