



2017 Research Proposals

and

2016 Research Reports

WRRC Board of Directors - with term expiration date, December 1, 20__

<u>Year</u>	<u>Seat</u>		
18	1	Glenn Sakuma Burlington	
19	2	Randy Honcoop Lynden	<u>Advisory Members</u> Steve Midboe – Lynden – Agronomy
17	3	Jessy Ghuman Everson	Joan Yoder – Everson – Food Safety/Treasurer
17	4	Jon Cotton Battle Ground	
15	5	<i>Open</i>	WRRC Office Henry Bierlink, Executive Director <i>henry@red-raspberry.org</i>
19	6	Jonathan Maberry, President Lynden	Stacey Beier, Office Manager 1796 Front Street, Lynden, WA 98264 (360) 354-8767
WSDA	7	Joel Kangiser Olympia	

Research Priorities 2017

#1 priorities

- Develop cultivars that are summer bearing, high yielding, winter hardy, machine-harvestable, disease resistant, virus resistant and have superior processed fruit quality
- Maximum Residue Limits (MRL) – residue decline curves, harmonization
- Fruit rot including pre harvest, post harvest, and/or shelf life.
- Labor saving practices – ex. Pruning, AY, public/private technology partnerships
- Management options for control of the Spotted Wing Drosophila

#2 priorities

- Soil fumigation techniques and alternatives to control soil pathogens, nematodes, and weeds.
- Understanding soil ecology and soil borne pathogens and their effects on plant health and crop yields.
- Root weevils
- Nutrient Management – Revise OSU specs, Consider: timing, varieties, appl. techniques
- Irrigation management – application techniques including pulsing

#3 priorities

- Mite management
- Viruses/crumbly fruit, pollination
- Weed management – especially horsetail
- Foliar & Cane diseases – i.e. spur blight, yellow rust, cane blight, powdery mildew, etc.
- Cane Management including suppression
- Pest Management as it affects Pollinators
- Effect on BRIX by fungicide and fertility programs
- Season extension: improve viability of fresh marketing

2017 WRRC RESEARCH PROPOSALS 2016 REPORTS

PAGE	PROJECT TITLE	RESEARCHER (S)	REQUEST	Draft #1	Other \$	Source	Approved
PLANT BREEDING							
4	Developing the Genomic Infrastructure for Breeding Black Raspberries - Final Report	Finn					
9	Cooperative raspberry cultivar development	Finn	\$11,861				
23	Red Raspberry Breeding	Moore	\$78,000		\$66,500	NWCSF	
31	Red Raspberry Cultivar Development	Dossett	\$12,000				
39	Regional on-farm Trials of Advanced Raspberry Selections	Peerbolt	\$11,200		\$11,500	ORBC	
ENTOMOLOGY							
45	Non-toxic RNAi based bioinsecticide to Control SWD	Choi	\$10,000		\$52,500	ARF	
51	Survey for Egg Parasitoids of Brown Marmorated Stink Bug, <i>Halyomorpha halys</i>	Gerdeman	\$0		\$8,083	WSCPR	
55	WSU NWREC Raspberry Field Plot for Invasive Pests and Foliar/fruit Disease Mgt Trial	Gerdeman/DeVetter	\$8,958		\$8,958	NARF	
57	Insecticide Degradation for Red Raspberry in the PNW - Final Report	Gerdeman/Joe DF					
WEEDS							
63	Weed Control in Red Raspberries - Final Report	Miller					
66	Determining whether raspberry plants should be caneburned	Miller	\$3,221		\$3,000	RIDC	
PHYSIOLOGY							
74	Application of Biodegradable Mulches in Tissue Culture Red Raspberry	DeVetter	\$10,457		\$17,312	WSCPR	
78	Impact of Nitrogen on Nematode Parasitism of Red Raspberry	DeVetter/Zasada	\$10,182				
81	Comparison of Alternate- and Every-Year Production in Summer-Bearing Red Raspberries	DeVetter	\$6,635				
86	Raspberry trellising demonstration plot for development of automation technologies	Karkee/Tarara	\$9,832				
PATHOLOGY/VIROLOGY							
93	Management of Fungicide Resistance in Botrytis in WA Berries	Schreiber	\$12,000		\$22,500	WSCPR	
106	Fungicide Decline Curves for meeting MRLs for Raspberry - Final Report	Schreiber					
118	Managing SWD in Red Raspberry with Reduced Insecticide Residues	Schreiber	\$15,000				
122	Biology of <i>Botrytis</i> causing fruit rot of red raspberry and fungicide resistance	Peever	\$20,610		\$26,442	WSCPR	
144	Laboratory Equipment for Small Fruit Pathology at NWREC	Peever/Harteveld	\$0		\$42,250	WBC	
146	Development of novel disease management methods for fruit rots of raspberry	Stockwell	\$5,919				
152	Evaluation of Raspberry Bushy Dwarf Virus strains	Moore/Martin	\$4,386				
156	Regional Survey for a Resistance-Breaking Strain of Raspberry bushy dwarf virus	Lanning	\$15,700				
SOILS							
161	Evaluating soil fumigation alternatives - Final Report	Walters/Zasada					
164	Fungicide Sensitivity of <i>Phytophthora rubi</i> - Final Report	Weiland					
165	Vapam cap, crop termination, and bed fumigation treatments to improve soil fumigation	Walters/Zasada	\$13,407				
Total Production Research			\$259,368	\$0	\$259,045		\$0
	Research Related	WRRC expenses	\$5,250	\$5,250			
	Small Fruit Center fee		\$2,500	\$2,500			
TOTAL			\$267,118	\$7,750			\$0

2017 Research Budget

\$227,750

applied

PROJECT TITLE	RESEARCHER (S)	REQUIRED ANNUAL MATCH	CASH	IN-KIND	SERVICES
Potassium Phosphanate MRL	U C Davis	\$5,000		\$5,000	admin, Crop Protection committee
Managing Washington Berry Diseases	Peever	\$15,000	\$15,000		
Caneberry Pesticide Degradation	Gerdeman/De Francisco	\$6,000		\$6,000	SFU sponsorship, SFC support
Improved Irrigation Practices	Bryla	\$10,000		\$10,000	on-farm plots, communications

PLANT BREEDING

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: 2016 (Final)

Accomplishments:

Major project goals: The overall goal of this proposal was to develop and make available genomic tools for the improvement of black raspberry and apply these tools for crop improvement using wild germplasm. These resources will significantly aid in the integration of novel traits from wild germplasm into elite cultivars and are necessary tools for molecular breeding of black raspberries and related species (e.g., red raspberry, blackberry) and to address the needs of the industry for improved cultivars. Objectives were presented as solutions to address problems in production and breeding that were identified by the industry and the USDA-ARS Small Fruits Crop Germplasm Committee. Conversations with black raspberry growers and processors over the last decade revealed disease and short planting longevity as their top production concerns. The USDA-ARS Northwest Center for Small Fruits Research (an academic/commercial industry partnership) and the Oregon Raspberry and Blackberry Commission have identified cultivar improvement as a number one research priority for the commercial raspberry industry.

Obj. 1: Transcriptome sequencing and high-throughput genomic sequencing: We completed the sequencing and assembly of the genome of a black raspberry individual using the facilities at Oregon State University, Corvallis, OR and The Donald Danforth Center, St. Louis, MO. We also obtained transcriptome sequence information from a variety of plant tissue types to better understand the genes that are expressed in each tissue. We used the expressed gene data to identify gene locations on our genome sequence. The genome of black raspberry (*Rubus occidentalis*) accepted and published in The Plant Journal on line May 12, 2016.

Obj. 2: Developing molecular markers from EST and genomic sequences: We mined the genome sequence for Simple Sequence Repeat (SSR) markers not previously available in black raspberry. We completed the high-throughput sequencing of our two mapping populations and a third population to identify specific differences within a single population and among the three populations. We used this information to develop targeted SSR and genome-wide SNP molecular markers and have placed these markers on a genetic linkage map. We are in the process of constructing a linkage map for our second mapping population of 192 progeny.

Obj. 3: Studying genotype by environment interaction on specific traits of interest in crosses involving diverse wild black raspberry germplasm: Interest in black raspberry production has expanded far beyond upstate New York and the Ohio River Valley where production was once concentrated; however, the industry today is reliant on cultivars developed for this region. The extent to which they are adapted to other production regions is not well understood. Studying the performance of seedling populations segregating for adaptation and other important traits in four production regions, Oregon, New York, Ohio, and North Carolina will provide valuable information on relative performance for these traits and effectiveness of selection for them in very different locations with strong small fruits industries and an interest in

improved black raspberry cultivars. We successfully completed three years of data collection to conduct this analysis. Preliminary results using a subset of data indicate that an individual's performance is influenced by the environment in some cases. This analysis will be completed this year.

Obj. 4: Using molecular markers for mapping specific traits of interest in crosses involving diverse wild black raspberry germplasm: We constructed a genetic linkage map for one mapping population. We are in the process of developing the linkage map for the second mapping population. Genetic linkage maps provide a framework of how the chromosomes of black raspberry are assembled and which regions are inherited together and will be used for identifying the regions of the genome involved in the expression of traits of interest. Next we plan to map loci involved in disease and insect resistance, vigor, phenology, fruit chemistry properties, and quality traits across locations as well as specific to each production region. The resulting linkage maps and QTL association will be used for the development of marker-based tests for important traits.

Obj. 5: Evaluate transferability of SSR markers developed in black raspberry to red raspberry: The completion of the first genetic linkage map for black raspberry will provide us with the means to address this objective as we are prioritizing evaluating transferability of markers mapped in black raspberry to red raspberry to allow comparative mapping in both crops. To date, 37 SSR markers are polymorphic in both species and 14 of these markers are located on the linkage map for ORUS 4305 with 1 to 4 markers per linkage group. These and other markers are useful as anchor markers for comparing maps between red and black raspberry and other Rose Family crops.

Obj. 6: Better understanding of consumer preferences and factors promoting black raspberry market expansion: We managed a replicated planting of advanced black raspberry selections for use in sensory evaluation. Fruit harvested from all fruiting plants commenced on 28 June and continued to 10 July. Fresh fruit harvested from these plots were submitted to an 11-member trained sensory panel for quantitative descriptive analysis (QDA) of appearance, aroma, flavor and texture characteristics. QDA panelists were exposed to 3 or 4 entries per test; each genotype was evaluated twice. There were significant differences among genotype means for many fruit characteristics. Oregon-grown fruit of the same selections and standards were machine-harvested and processed into puree by the Oregon State University (OSU) Department of Food Science and Technology. Purees were randomly assigned to two groups of four purees. Groups were subjected to consumer preference analysis at the OSU Sensory Science Laboratory on August 6th and 7th and on September 17th and 18th using 109-member and 115-member consumer panels, respectively. Purees will be analyzed by the QDA panel in mid-Oct. 2014. We are also exploring messaging techniques to improve black raspberry market share. Survey instruments and protocols to ascertain purchasing incentives of larger buyers (processors, retail grocery chains, etc.) have also been developed. We have also explored several analytical techniques for extracting and evaluating flavor compounds present in these fruit and have developed an analytical library of over 30 flavor compound standards. This information will provide us with consumer acceptance targets when selecting germplasm for breeding. Additional work is on-going at the Ohio State University.

A survey instrument entitled "Opportunities and Challenges Facing Black Raspberry Producers" was developed to delineate current production and marketing strategies and to outline important grower needs/concerns for future expansion of acreage. This instrument was presented at the 2014 Oregon Raspberry and Blackberry Commission (ORBC) Annual Growers Meeting, December 17 at the Wellspring Conference Center, in Woodburn, Oregon and at the North American Raspberry & Blackberry Association (NARBA) Conference, February 24-27, 2015 in Fayetteville, Arkansas. An on-line version was made available and was promoted by ORBC, NARBA and the Ohio Produce Growers & Marketers Association (OPGMA). The survey

revealed that most black raspberry producers (68%) farm less than 100 acres and 32% of respondents reported to grow less than 5 acres of this crop and realized gross receipts of less than \$50,000 annually. They tend to grow a mixture of berry crops. They sell through farm stands, farmers markets, pick your own, wholesale and retail. Responding growers indicated that production costs, product perishability and shipping constraints, disease and insect problems, consumer unfamiliarity with the product (often confused with blackberries), and the lack of cultivar diversity to be major impediments to industry growth. Varietal characteristics most highly desired by producers included excellent pest resistance and fruit quality characteristics, thornlessness, season extension capacity and the primocane fruiting habit.

Obj. 7: Delivering research results and training in molecular breeding to the industry, breeders, and students through a multifaceted outreach and extension program: Over the course of the project we have presented our research at more than 20 different conferences and field days and have nine peer-reviewed publications. We hired and trained high school students and trained volunteers in North Carolina and Oregon in field and molecular components of the project. Research was also highlighted on several social media sites. We conducted training in germplasm assessment and characterization, molecular breeding, and applied use of molecular tools in breeding at the 2015 American Society for Horticultural Science Annual Conference in New Orleans, LA.

Publications:

- Dossett M, Bassil NV, Lewers KS, Finn CE. **2012**. Genetic diversity in wild and cultivated black raspberry (*Rubus occidentalis* L.) evaluated by simple sequence repeat markers. *Gen. Res. Crop Evol.* 59:1849-1865.
- Lee J, Dossett M, and Finn CE. **2012**. *Rubus* fruit phenolic research: the good, the bad, and the confusing. *Food Chem.* 130:785-796.
- Lee J, Dossett M, and Finn CE. **2013**. Anthocyanin fingerprinting of true bokbunja (*Rubus coreanus* Miq.) fruit. *J. Funct. Foods* 5:1985-1990.
- Paudel L, Wyzgoski FJ, Scheerens JC, Chanon AM, Reese RN, Smiljanic D, Wesdemiotis C, Blakeslee JJ, Riedl KM, Rinaldi PL. **2013**. Non-anthocyanin secondary metabolites of black raspberry (*Rubus occidentalis* L.) fruits: Identification by HPLC-DAD, NMR, HPLC-ESI-MS and ESI-MS/MS analyses. *J. Agric. Food Chem.* 61:12032-12043.
- Lee J, Dossett M, and Finn CE. **2014**. Mistaken identity: clarification of *Rubus coreanus* Miquel (bokbunja). *Molecules- special Anthocyanin issue* 19:10524-10533.
- Lee J. **2014**. Marketplace analysis demonstrates quality control standards needed for black raspberry dietary supplements. *Plant Foods Hum. Nutr.* 69:161-167
- Paudel L, Wyzgoski FJ, Giusti MM, Johnson JL, Rinaldi PL, Scheerens JC, Chanon AM, Bomser JA, Miller AR, Hardy JK, Reese RN. **2014**. NMR-based metabolomic investigation of bioactivity of chemical constituents in black raspberry (*Rubus occidentalis* L.) fruit extracts. *J. Agric. Food Chem.* 62:1989-1998.
- Dossett M, Bushakra JM, Gilmore BS, Koch CA, Kempler C, Finn CE, Bassil NV. **2015**. Development and transferability of black and red raspberry microsatellite markers from short-read sequences. *J. Amer. Soc. Hort. Sci.* 140:243-252.
- Bushakra JM, Bryant DW, Dossett M, Vining KJ, VanBuren R, Gilmore BS, Lee J, Mockler TC, Finn CE, Bassil NV. **2015**. A genetic linkage map of black raspberry (*Rubus occidentalis*) and the mapping of *Ag4* conferring resistance to the aphid *Amphorophora agathonica*. *Theor. Appl. Genet.* 128:1631-1646.
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- Lee, J. **2015**. Analysis of bokbunja products show they contain *Rubus occidentalis* L. fruit. *J. Funct. Foods.* 12:144-149.

- Lee, J. **2015**. Sorbitol, *Rubus* fruit, and misconception. *Food Chem.* 166:616-622.
- VanBuren R, Bryant D, Bushakra JM, Vining KJ, Edger PP, Rowley ER, Priest HD, Michael TP, Lyons E, Filichkin SA, Dossett M, Finn CE, Bassil NV, Mockler TC. **2016**. The genome of black raspberry (*Rubus occidentalis*). *The Plant Journal* (online)
- Lee J. **2016**. Further research on the biological activities and the safety of raspberry ketone are needed. *NFS Journal* 2:15-18

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- Dossett M, Bassil NV, Finn CE. **2012**. Fingerprinting of black raspberry cultivars shows discrepancies in identification. *Acta Hortic.* 946:49-53.
- Dossett M, Lee J, and Finn CE. **2012**. Anthocyanin content of wild black raspberry germplasm. *Acta Hortic.* 946:43-47.
- Bassil N, Gilmore B, Hummer K, Weber C, Dossett M, Agunga R, Rhodes E, Mockler T, Scheerens JC, Filichkin S, Lewers K, Peterson M, Finn CE, Graham J, Lee J, Fernández-Fernández F, Fernandez G, Yun SJ, and Perkins-Veazie P. **2014**. Genetic and Developing Genomic Resources in Black Raspberry. *Acta Hortic.* 1048:19-24.
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- Lee J, Dossett M, Finn CE. **2014**. Chemotaxonomy of black raspberry: deception in the marketplace? *Polyphenols Comm.* 2014:347-348.
- Bassil N, Gilmore B, Hummer K, Dossett M, Mockler T, Filichkin S, Peterson M, Finn C, Lee J, Fernandez G, Perkins-Veazie P, Weber C, Agunga R, Rhodes E, Scheerens JC, Lewers K, Graham J, Fernández-Fernández F, Yun SJ. **2014**. Genetic and Developing Genomic Resources in Black Raspberry. *Acta Hortic.* 1048:19-23.
- Bradish CM, Fernandez GE, Bushakra JM, Perkins-Veazie P, Dossett M, Bassil NV, Finn CE. **2016**. Evaluation of vigor and winter hardiness of black raspberry breeding populations (*Rubus occidentalis*) grown in the southeastern US. *Acta Hortic.* 1133:129-134.
- Bushakra JM, Bassil NV, Weiland JE, Finn CE, Vining KJ, Filichkin S, Dossett M, Bryant DW, Mockler TC. **2016**. Comparative RNA-seq for the investigation of tolerance to *Verticillium* wilt in black raspberry. *Acta Hortic.* 1133:103-114.
- Perkins-Veazie P, Ma G, Fernandez GE, Bradish CM, Bushakra JM, Bassil NV, Weber CA, Scheerens JC, Robbins L, Finn CE, Dossett M. **2016**. Black raspberry fruit composition over two years from seedling populations grown at four US geographic locations. *Acta Hortic.* 1133:335-338.

Abstracts

- Bushakra J, Bassil N, Dossett M, Gilmore B, Mockler T, Bryant D, Filichkin S, Weiland J, Peterson M, Bradish C, Fernandez G, Lewers K, Graham J, Finn C. **2013**. Black raspberry genomic resource development. *International Plant & Animal Genome XXI*.
- Bushakra JM, Bryant D, Mockler T, Finn CE, Dossett M, Peterson M, Gilmore B, Bassil NV. **2013**. Black raspberry genotyping by sequencing. *American Society of Horticulture Science Meeting. 2013 American Society for Horticultural Science Annual Conference.*
- Bradish CM, Fernandez GE, Bushakra JM, Perkins-Veazie P, Dossett M, Bassil NV, Finn CE. **2014**. North Carolina's role in a nationwide effort to improve black raspberry. *HortScience Annual Meeting Supplement 49:S56-S57 (abstract).*
- Bryant D, Bushakra JM, Dossett M, Vining K, Filichkin S, Weiland J, Lee J, Finn CE, Bassil N, Mockler T. **2014**. Building the genomic infrastructure in black raspberry. *HortScience Annual Meeting Supplement 49:S233 (abstract).*
- Bushakra JM, Bradish CM, Weber CA, Scheerens JC, Dossett M, Peterson M, Fernandez G, Lee J, Bassil N, Finn CE. **2014**. Toward understanding genotype x environment interactions in black raspberry. *HortScience Annual Meeting Supplement 49:S298 (abstract).*

Perkins-Veazie P, Fernandez G, Bradish CM, Ma G, Scheerens JC, Weber CA, Finn CE, Bassil N, Bushakra JM. **2014**. Black raspberry fruit composition from seedling populations planted at multiple locations. HortScience Annual Meeting Supplement 49:S248 (abstract).

Bushakra J, Dossett M, Lee JC, Lee J, Bassil NV, Finn CE. **2015**. Molecular evaluation of aphid-resistant black raspberry germplasm for improved durability in black and red raspberry [abstract]. American Society of Horticulture Science Meeting. 2015 American Society for Horticultural Science Annual Conference.

Bushakra J, Bassil NV, Bryant D, Mockler T, Dossett M, Gilmore BS, Peterson ME, Bradish C, Fernandez G, Lee J, Finn CE. **2015**. Black raspberry genomic and genetic resource development to enable cultivar improvement [abstract]. Plant and Animal Genome XXIII Conference.

Bushakra J, Bryant D, Dossett M, Vining K, VanBuren R, Gilmore BS, Filichkin S, Weiland JE, Peterson ME, Bradish CM, Fernandez G, Lewers KS, Graham J, Lee J, Mockler T, Bassil NV, Finn CE. **2015**. Developing black raspberry genetic and genomic resources [abstract]. International *Rubus* and *Ribes* Symposium

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: 2016

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 WSU and BC advanced selections is available and can aid in making decisions on the commercial suitability of their materials. ‘Lewis’, and multiple florican selections, are in grower and machine harvest trials in Washington. ORUS 4373-1 was identified in Puyallup as having good root rot tolerance and ORUS 4607-2, and ORUS 4465-3 have looked promising in machine harvest trial and all three are being propagated for more extensive grower trials. ‘Kokanee’ primocane fruiting raspberry was released and several primocane selections are being propagated for grower trials. We made 36 selections this year. We have 65 florican and 47 primocane selections from our crosses in trial, in addition to 24 WSU and BC selections (Table RY1).

Results: Forty-seven crosses were made in spring 2016 and a new seedling field (~2000 seedlings) was established. We made 15 florican and 21 primocane selections that have cultivar potential. 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 2016 or will be harvested in 2017. Presented in Tables RY2-RY9 are the results from 2016. Promising selections have been sent to growers in Lynden to evaluate machine harvestability; 1st year results of this evaluation can be seen in Table RY4. ORUS 4373-1, ORUS 4607-2, and ORUS 4465-3 and five primocane fruiting selections are being cleaned up and propagated for grower trial. ‘Kokanee’, a primocane fruiter, was released; it is a late season high quality raspberry suited for fresh market sales. Multiple selections were identified as having excellent root rot resistance in Puyallup.

While indirectly related to red raspberry, our efforts in black raspberry have identified verticillium wilt and aphid resistance (that should translate into virus resistance for the aphid transmitted viruses). Genetic markers are being developed for sources of aphid resistance.

Publications:

- VanBuren, R., D. Bryant, J.M. Bushakra, K.J. Vining, P.P. Edger, E.R. Rowley, H.D. Priest, T.P. Michael, E. Lyons, S.A. Filichkin, M. Dossett, C.E. Finn, N.V. Bassil and T.C. Mockler. 2016. The genome of black raspberry (*Rubus occidentalis*). The Plant J. 87:533-680 (and cover).
- Bushakra, J.M., C.M. Bradish, C.A. Weber, M. Dossett, G. Fernandez, J. Weiland, M. Peterson, J.C. Scheerens, L. Robbins, S. Serçe, C.E. Finn, and N.V. Bassil. 2016. Toward understanding genotype × environment interactions in black raspberry (*Rubus occidentalis* L.). Acta Hort. 1117:25-30.
- Bradish, C.M., G.E. Fernandez, J.M. Bushakra, P. Perkins-Veazie, M. Dossett, N.V. Bassil, C.E. Finn. 2016. Evaluations of sustained vigor and winter hardiness of black raspberry (*Rubus occidentalis*) grown in the Southeastern U.S. Acta Hort 1133:129-134.
- Bushakra, J.M., N.V. Bassil, J.E. Weiland, C.E. Finn, K.J. Vining, S. Filichkin, M. Dossett, D.W. Bryant, and T.C. Mockler. 2016. Comparative RNA-seq for the investigation of tolerance to verticillium wilt in black raspberry. Acta Hort. 1133:103-114.
- Perkins-Veazie, P., G. Ma, G.E. Fernandez, C.M. Bradish, J.M. Bushakra, N.V. Bassil, C.A. Weber, J.C. Scheerens, L. Robbins, C.E. Finn, and M. Dossett. 2016 Black raspberry fruit composition over two years from seedling populations grown at four US geographic locations. Acta Hort. 1133: 335-338.

Appendices

Table RY1. Red raspberry genotypes in trial at OSU-NWREC in 2016; 151 genotypes evaluated

<i>Floricanne Fruiteurs</i>				<i>Primocane Fruiteurs</i>	
ORUS 1040-1	ORUS 4462-1	ORUS 4607-1	BC 1-16-38	ORUS 1167-2	ORUS 4495-1
ORUS 3230-1	ORUS 4462-2	ORUS 4607-2	BC 93-6-30	ORUS 3842-1	ORUS 4564-2
ORUS 3528-1	ORUS 4463-1	ORUS 4608-1	BC 97-30-20	ORUS 4086-1	ORUS 4566-1
ORUS 3568-1	ORUS 4465-1	ORUS 4608-2		ORUS 4086-2	ORUS 4566-2
ORUS 3702-3	ORUS 4465-2	ORUS 4611-1		ORUS 4090-2	ORUS 4595-1
ORUS 3713-1	ORUS 4465-3	ORUS 4613-1	WSU 1914	ORUS 4097-3	ORUS 4599-2
ORUS 3718-1	ORUS 4471-1	ORUS 4616-1	WSU 1956	ORUS 4155-3	ORUS 4599-3
ORUS 3722-1	ORUS 4473-2	ORUS 4639-1	WSU 1980	ORUS 4289-3	ORUS 4617-1
ORUS 3767-3	ORUS 4473-3	ORUS 4641-1	WSU 1985	ORUS 4289-4	ORUS 4619-1
ORUS 3958-1	ORUS 4476-1	ORUS 4641-2	WSU 1996	ORUS 4291-1	ORUS 4619-2
ORUS 3959-1	ORUS 4476-2		WSU 2001	ORUS 4384-1	ORUS 4619-3
ORUS 3959-2	ORUS 4482-2		WSU 2010	ORUS 4386-1	ORUS 4621-2
ORUS 3959-3	ORUS 4482-3	Cascade Bounty	WSU 2011	ORUS 4388-2	ORUS 4622-2
ORUS 4179-1	ORUS 4482-4	Cascade Delight	WSU 2029	ORUS 4388-3	ORUS 4622-3
ORUS 4371-1	ORUS 4483-1	Cascade Gold	WSU 2068	ORUS 4412-5	ORUS 4716-1
ORUS 4371-2	ORUS 4598-1	Chemainus	WSU 2075	ORUS 4469-1	ORUS 4716-2
ORUS 4371-3	ORUS 4600-1	Lewis	WSU 2122	ORUS 4486-1	ORUS 4719-1
ORUS 4371-4	ORUS 4600-2	Meeker	WSU 2130	ORUS 4486-2	ORUS 4723-1
ORUS 4371-5	ORUS 4600-3	Rudi	WSU 2133	ORUS 4487-1	ORUS 4725-1
ORUS 4373-1	ORUS 4600-3	Saanich	WSU 2166	ORUS 4487-2	
ORUS 4375-1	ORUS 4600-4	TulaMagic (Frutafri)	WSU 2188	ORUS 4487-3	Anne
ORUS 4380-1	ORUS 4600-5	Tulameen	WSU 2200	ORUS 4487-4	BP-1
ORUS 4380-2	ORUS 4601-1	Wakefield	WSU 2205	ORUS 4493-1	Kokanee
ORUS 4380-3	ORUS 4603-1			ORUS 4494-1	Heritage
ORUS 4380-4	ORUS 4603-2			ORUS 4494-2	TulaMagic (Frutafri)
ORUS 4461-1	ORUS 4606-2			ORUS 4494-3	Vintage

Table RY2. Mean yield and berry size in 2015-16 for floricanne fruiting raspberry genotypes at OSU-NWREC planted in 2013.

Genotype	Berry size (g) 2015-16 ^z	Yield (tons·a ⁻¹)		
		2015	2016	2015-16
2015	2.8 a			2.84 a
2016	3.6 b			2.65 a
<i>Replicated</i>				
Meeker	2.7 b	2.01 a	3.29 a	2.65 a
ORUS 4371-4	3.6 a	2.81 a	2.41 a	2.61 a
ORUS 4373-1	3.3 a	2.03 a	2.24 a	2.13 a
<i>Nonreplicated</i>				
ORUS 4465-1	3.6	2.13	3.60	2.86
WSU 1914	3.1	2.87	2.37	2.62
WSU 2010	2.2	2.90	1.77	2.33
ORUS 4371-3	2.8	2.14	2.04	2.09

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

Table RY3. Mean yield and berry size in 2016 for floricanes fruiting red raspberry genotypes in replicated and observation trials at OSU-NWREC planted in 2014.

Genotype	Berry size (g) ^z	Yield (tons·a ⁻¹)
<i>Replicated</i>		
WSU 1980	5.2 ab	4.89 a
Lewis	4.8 ab	4.83 a
WSU 2166	5.3 a	4.11 ab
ORUS 4462-2	4.6 bc	4.09 ab
ORUS 4482-3	4.8 ab	3.74 ab
Meeker	3.3 e	3.71 ab
ORUS 4465-3	4.0 cd	3.56 ab
ORUS 3713-1	3.6 de	3.21 ab
WSU 2188	5.1 ab	3.15 ab
WSU 2122	3.7 de	2.75 b
<i>Nonreplicated</i>		
WSU 1956	4.3	4.82
ORUS 4473-3	3.6	4.28
ORUS 4465-2	3.7	3.83
WSU 1985	4.0	3.78
WSU 2068	3.7	3.78
ORUS 3767-3	3.0	3.60
ORUS 4463-1	4.4	3.34
WSU 2001	3.9	3.32
WSU 2075	2.8	3.24
WSU 2200	2.6	2.78
WSU 2010	2.5	2.76
ORUS 3959-3	5.4	2.74
WSU 2133	2.6	2.45
WSU 2205	3.0	2.23
ORUS 4473-2	3.4	2.21
ORUS 4462-1	3.3	2.17
WSU 2130	2.9	2.11

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

Table RY4. Performance of ORUS selections in machine harvest trials in Lynden, Washington. Planted in 2015.

Grower	Selection	Yield	Yield as %	Brix	Firm	Harvest		Comments
		(pounds/plot)	of Meeker			First	Last	
Enfield	Wake@field	84.3	135	12.6	43.9	15-Jun	3-Aug	-
Enfield	ORUS 4607-2	75.3	121	13.0	26.6	7-Jun	19-Jul	Firm enough. Often noted for lots of moldy berries & lots of green berries/stems showing up on belt
Enfield	Meeker	62.3	100	11.3	30.4	7-Jun	26-Jul	-
Enfield	ORUS 4603-1	56.9	91	10.6	31.3	7-Jun	19-Jul	Often noted for lots of moldy berries and lots of green berries/stems showing up on the belt
Enfield	ORUS 4600-3	53.0	85	11.6	36.6	11-Jun	19-Jul	Often noted for lots of moldy berries and lots of green berries/stems showing up on the belt
Enfield	ORUS 4600-1	41.7	67	11.1	40.6	11-Jun	19-Jul	Few comments about moldy berries
Enfield	ORUS 4462-1	39.5	64	10.8	46.4	11-Jun	19-Jul	Lots of comments about berries crumbling on the belt
Enfield	Cascade Harvest	31.8	62	10.7	29.8	7-Jun	26-Jul	-
Maberry	Cascade Harvest	85.7	153	-	-	13-Jun	25-Jul	-
Maberry	Willamette	58.5	104	-	-	13-Jun	25-Jul	-
Maberry	Meeker	56.2	100	-	-	13-Jun	25-Jul	-
Maberry	ORUS 4465-3	53.9	96	-	-	13-Jun	25-Jul	Nice frt, good color *
Maberry	ORUS 4283-1	49.5	88	-	-	13-Jun	25-Jul	Gs. Dark mod Soft *
Maberry	ORUS 3722-1	48.4	86	-	-	13-Jun	25-Jul	-
Maberry	ORUS 3713-1	39.8	71	-	-	13-Jun	25-Jul	Lrg, few frt
Maberry	ORUS 3767-3	21.9	39	-	-	13-Jun	25-Jul	Broken, mold, greens
Maberry	ORUS 3234-1	11.5	20	-	-	13-Jun	25-Jul	-

Table RY5. Mean yield and berry size in 2014-2016 for primocane fruiting raspberry genotypes at OSU-NWREC planted in 2013.

	Berry size (g) 2014-16	Yield (tons·acre ⁻¹)			
		2014	2015	2016	2014-16
<i>Replicated</i>					
2014	2.2 b				1.70 a
2015	1.7 c				1.10 b
2016	2.8 a				1.42 ab
ORUS 4487-1	2.2 b	2.88 a	1.50 a	1.40 a	1.93 a
Heritage	1.9 c	2.03 b	1.24 ab	1.75 a	1.68 a
Vintage	2.5 a	1.13 c	0.75 c	1.07 a	1.11 b
ORUS 4090-2	2.4 a	0.77 d	0.90 bc	1.09 a	0.92 b
<i>Non replicated</i>					
ORUS 4086-1	2.3		0.26	3.84	2.05
ORUS 4486-1	2.0	3.01	1.05	1.58	1.88
ORUS 4388-2	2.7	1.67	1.11	2.70	1.83
TulaMagic	2.8	1.49	0.27	3.04	1.60
ORUS 4086-2	2.3	1.66	0.22	1.45	1.11

Table RY6. Mean yield and berry size in 2015-16 for primocane fruiting red raspberry genotypes at OSU-NWREC planted in 2014.

	Berry size (g) 2015-16	Yield (tons·a ⁻¹)		
		2015	2016	2015-16
<i>Nonreplicated</i>				
ORUS 4493-1	3.0	2.06	7.61	4.84
Heritage	2.0	1.62	3.72	2.67
ORUS 4599-3	4.7	0.19	3.66	1.93
Vintage	2.9	1.04	2.55	1.79
Kokanee (ORUS 4090-1)	3.5	0.68	2.73	1.71
ORUS 4487-4	3.0	1.17	2.20	1.69
ORUS 4090-2	3.2	1.80	1.39	1.60

Table RY7. Mean yield and berry size in 2016 for primocane fruiting red raspberry genotypes at OSU-NWREC planted in 2015.

Genotype	Berry size (g)	Yield (tons·a ⁻¹)
<i>Replicated</i>		
Kokanee	3.0 a	3.16 a
Heritage	2.1 b	1.96 b
Vintage	3.3 a	1.77 b
<i>Non replicated</i>		
ORUS 4719-1	4.4	4.66
ORUS 4622-2	3.8	3.93
ORUS 4716-1	3.4	3.09
ORUS 4725-1	3.9	2.79
ORUS 4291-1	3.0	1.96
BP1 (=Amira)	4.3	1.32

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

Table RY8. Ripening season for floricanne fruiting red raspberry genotypes at OSU-NWREC. Planted in 2013 or 2014 and harvested 2015 and/or 2016.

Genotype	Year planted	Harvest season			No. years in mean	Rep/ Obsv
		5%	50%	95%		
ORUS 3767-3	2014	31-May	14-Jun	28-Jun	1	Obsv.
ORUS 4465-2	2014	31-May	14-Jun	28-Jun	1	Obsv.
WSU 2075	2014	31-May	14-Jun	28-Jun	1	Obsv.
WSU 2200	2014	31-May	14-Jun	28-Jun	1	Obsv.
WSU 2205	2014	31-May	14-Jun	28-Jun	1	Obsv.
ORUS 4473-2	2014	7-Jun	14-Jun	28-Jun	1	Obsv.
WSU 2068	2014	7-Jun	14-Jun	28-Jun	1	Obsv.
WSU 2130	2014	7-Jun	14-Jun	28-Jun	1	Obsv.
WSU 2166	2014	7-Jun	14-Jun	28-Jun	1	Rep
ORUS 4462-1	2014	7-Jun	14-Jun	5-Jul	1	Obsv.
WSU 2133	2014	7-Jun	14-Jun	5-Jul	1	Obsv.
ORUS 3722-1	2013	4-Jun	18-Jun	25-Jun	2	Rep
ORUS 3702-3	2013	4-Jun	18-Jun	29-Jun	2	Rep
WSU 2010	2013	4-Jun	18-Jun	29-Jun	2	Obsv.
ORUS 4465-1	2013	8-Jun	18-Jun	25-Jun	2	Obsv.
ORUS 4371-3	2013	8-Jun	18-Jun	29-Jun	2	Obsv.
WSU 1914	2013	11-Jun	18-Jun	2-Jul	2	Obsv.
WSU 2010	2014	7-Jun	21-Jun	28-Jun	1	Obsv.
Meeker	2014	7-Jun	21-Jun	5-Jul	1	Rep
ORUS 3713-1	2014	7-Jun	21-Jun	5-Jul	1	Rep
ORUS 4465-3	2014	7-Jun	21-Jun	5-Jul	1	Rep
ORUS 4473-3	2014	7-Jun	21-Jun	5-Jul	1	Obsv.
WSU 1985	2014	7-Jun	21-Jun	5-Jul	1	Obsv.
WSU 2122	2014	7-Jun	21-Jun	12-Jul	1	Rep
ORUS 3959-3	2014	14-Jun	21-Jun	5-Jul	1	Obsv.
WSU 2188	2014	14-Jun	21-Jun	5-Jul	1	Rep
Meeker	2013	8-Jun	22-Jun	2-Jul	2	Rep
ORUS 4371-4	2013	8-Jun	22-Jun	10-Jul	2	Rep
ORUS 4380-3	2013	15-Jun	22-Jun	29-Jun	2	Obsv.
ORUS 4462-2	2014	14-Jun	24-Jun	12-Jul	1	Rep
ORUS 4373-1	2013	8-Jun	25-Jun	10-Jul	2	Rep
ORUS 4463-1	2014	12-Jun	28-Jun	12-Jul	1	Obsv.
Lewis	2014	14-Jun	28-Jun	12-Jul	1	Rep
ORUS 4482-3	2014	14-Jun	28-Jun	12-Jul	1	Rep
WSU 1956	2014	14-Jun	28-Jun	19-Jul	1	Obsv.
WSU 1980	2014	14-Jun	28-Jun	19-Jul	1	Rep
WSU 2029	2013	2-Jul	13-Jul	24-Jul	2	Obsv.

Table RY9. Ripening season for primocane fruiting red raspberry genotypes at OSU-NWREC. Planted in 2012, 2013, or 2014 and harvested 2013-16.

Genotype	Year planted	Harvest season			No. years in mean	Rep/ Obsv
		5%	50%	95%		
ORUS 4493-1	2014	20-Jul	27-Jul	24-Aug	2	Obsv.
ORUS 4291-1	2015	26-Jul	2-Aug	16-Aug	1	Obsv.
BP-1 (=Amira)	2015	26-Jul	9-Aug	16-Aug	1	Obsv.
ORUS 4725-1	2015	26-Jul	9-Aug	16-Aug	1	Obsv.
ORUS 4719-1	2015	2-Aug	9-Aug	30-Aug	1	Obsv.
ORUS 4599-3	2014	6-Aug	13-Aug	20-Aug	2	Obsv.
ORUS 4090-2	2013	1-Aug	15-Aug	5-Sep	3	Rep
ORUS 4086-2	2013	3-Aug	15-Aug	10-Sep	3	Rep
ORUS 4622-2	2015	2-Aug	16-Aug	30-Aug	1	Obsv.
Vintage	2015	2-Aug	16-Aug	30-Aug	1	Rep
ORUS 4716-1	2015	9-Aug	16-Aug	30-Aug	1	Obsv.
ORUS 4486-1	2013	8-Aug	20-Aug	5-Sep	3	Obsv.
Kokanee	2015	9-Aug	23-Aug	13-Sep	1	Rep
Heritage	2015	16-Aug	23-Aug	30-Aug	1	Rep
Vintage	2014	3-Aug	24-Aug	14-Sep	2	Rep
Heritage	2013	10-Aug	24-Aug	7-Sep	3	Rep
ORUS 4388-2	2013	13-Aug	27-Aug	10-Sep	3	Obsv.
Heritage	2014	10-Aug	27-Aug	14-Sep	2	Rep
ORUS 4086-1	2013	13-Aug	27-Aug	7-Sep	2	Obsv.
TulaMagic (Frutafri)	2013	22-Aug	5-Sep	12-Sep	3	Obsv.
Kokanee	2014	20-Aug	7-Sep	21-Sep	2	Obsv.
ORUS 4487-4	2014	24-Aug	10-Sep	21-Sep	2	Obsv.

Project Title: Cooperative raspberry cultivar development program

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Year Initiated __2013__ **Current Year** 2017-2018__ **Terminating Year** _Continuing__

Total Project Request: Ongoing.

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.

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' or 'Wake@field' 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 floricanes 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/crumblly fruit, pollination
- Foliar & Cane Diseases – i.e. spur blight, yellow rust, cane blight, powdery mildew, 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 floricanes 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. Selections are included in the formal

WRRC machine harvest and in separate grower trials in Lynden. 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 were 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 three sources of resistance and have developed molecular markers for these that are being verified with the goal of being able 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: \$ 5,000

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 (1 student GS-2-5, 4 months)	\$8,361
Operations (goods & services)	1,000
Travel ¹	1,500
Other: “Land use charge” (\$3,500/acre)	1,000
Total	\$11,861

¹To visit Puyallup, Lynden, and/or grower trials, field days and small fruit conferences 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	7/2016-6/2017	1	Funds towards industry matching on SCRI grant "Developing the Genomic Infrastructure for Breeding Improved Black Raspberries"
Strik, BC, and Finn, C.E.	Oregon Blueberry Commission	\$18,920	7/2016-6/2017	2	Cooperative Breeding Program-Blueberries
Finn, C.E.	Oregon Blueberry Commission	\$11,488	7/2016-6/2017	4	Developing PNW Cultivars That May Resist Blueberry Shock Virus
Finn, C.E.	Oregon Raspberry and Blackberry Commission	\$1,000	7/2016-6/2017	1	Funds towards industry matching on SCRI grant "Developing the Genomic Infrastructure for Breeding Improved Black Raspberries"
Strik, B.C. and C.E. Finn	Oregon Raspberry and Blackberry Commission	\$44,500	7/2016-6/2017	4	Production System/Physiology Research and Cooperative Breeding Program-Raspberries and Blackberries
C.E. Finn	Oregon Strawberry Commission	\$8,342	7/2016-6/2017	2	Breeding day-neutral strawberries in Corvallis, OR
Strik, B.C. and C.E. Finn	Oregon Strawberry Commission	\$16,500	7/2016-6/2017	4	Cooperative Breeding Program - Strawberries
Bassil, N.V., J.M. Bushakra, C.E. Finn, and M. Dossett	OSU ARF	12,500	2/2016-1/2018	1	Assessment of aphid resistance in black raspberry and development of trait-associated molecular markers for breeding improvement

Iezzoni, A., C. Peace, K. Gasic, J. Luby, C. Finn, J. Norelli, D. Main and 27 others (including P. Moore)	USDA Specialty Crop Research Initiative	\$10 million total; \$1.8 million annual; \$15K to USDA Breeding	10/2014-9/2019	5	RosBREED: Combining Disease Resistance With Horticultural Quality In New Rosaceous Cultivars
Finn, C.E.	Washington Blueberry Commission	\$15,108	7/2016-6/2017	4	Developing commercial blueberry cultivars adapted to the Pacific Northwest with an emphasis on tolerance of Blueberry shock virus (BIShV)
Finn, C.E.	Washington Red Raspberry Commission	\$4,000	7/2016-6/2017	2	Cooperative raspberry cultivar development program.
Finn, C.E.	Washington Red Raspberry Commission	\$1,000	7/2016-6/2017	1	Funds towards industry matching on SCRI grant "Developing the Genomic Infrastructure for Breeding Improved Black Raspberries"
Finn, C.E.	Washington Strawberry Commission	\$3,500	7/2016-6/2017	2	USDA-ARS Cooperative Strawberry Breeding Program
Pending:					

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: 2016

Accomplishments:

Grower Trials. Four selections were planted in grower trials in 2014. All four selections were productive, machine harvested well, with good fruit quality. One selection appears very promising, machine harvests well, productive, firm, producing fruit at the same time as ‘Willamette’. This selection will be evaluated in 2017 and may be considered for release.

Crosses/seedlings/selections. In 2016, there was a strong emphasis to improve root rot resistance by crossing selections that had good machine harvestability with cultivars and selections that are highly root rot resistant. Fifty-one of the sixty crosses for cultivar development had at least one parent that was root rot resistant. The seedling field planted in 2014 was in an area with a high level of root rot. Thirty selections were made, with six selections from the cross of WSU 1914 and Cascade Harvest. WSU 1914 has Boyne (highly root rot resistant) as a parent.

Machine Harvesting Trials. A new machine harvesting trial was planted in Lynden with 77 WSU selections, 6 ORUS selections, 5 BC selections and ‘Cascade Harvest’, ‘Meeker’ and ‘Willamette’ for reference. This planting will be harvested in 2018 and 2019.

The machine harvesting planting established in 2013 was evaluated in 2015-16 seasons. Harvest data for each harvest in the 2013 planting were collected in 2015, but only subjectively evaluated in 2016. The 20 plots with the highest yields in 2015 are given in Figure 1 WSU 2069 had the highest yield and WSU 2068 and WSU 1962 were in the top seven for yield. The 2014 planting was evaluated subjectively on five dates in 2016 and harvest data collected for each harvest.). Forty-three WSU selections and standard cultivars were machine harvested along with the BC and ORUS selections in this planting. Cascade Harvest had the second highest yield and WSU 2188 and WSU 2166 were in the top 10 for yield (Figure 2).

Selection Trial Puyallup. Cascade Harvest had the highest yield in both the 2013 selection trial at Puyallup and the 2014 planting (Tables 1 and 2). Other high yielding selections in the 2013 planting were WSU 2075, WSU 2069, WSU 2068 and WSU 1914. WSU 2188 had a good yield in the 2014 planting. WSU 2166 had very low yield in the first year of harvest as a result of producing few canes. This fall there is a normal amount of canes, hopefully the 2017 yield will be higher.

Publications

Machine Harvesting Field Day Lynden, WA June 23, 2016

Lanning, K.K., P.P. Moore, K.E. Keller, R.R. Martin. 2016. First report of a resistance-breaking strain of Raspberry bushy dwarf virus in red raspberry (*Rubus idaeus*) in North America. Plant Disease 100:868.

Figure 1

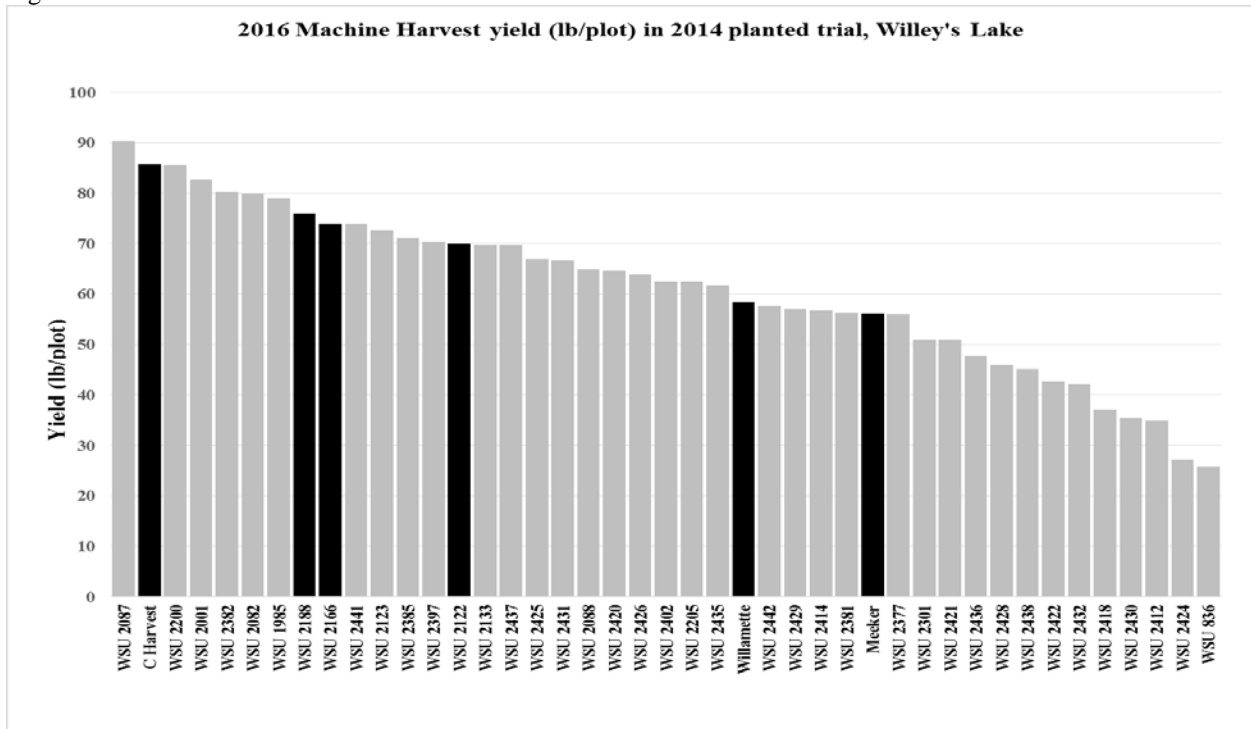


Figure 2

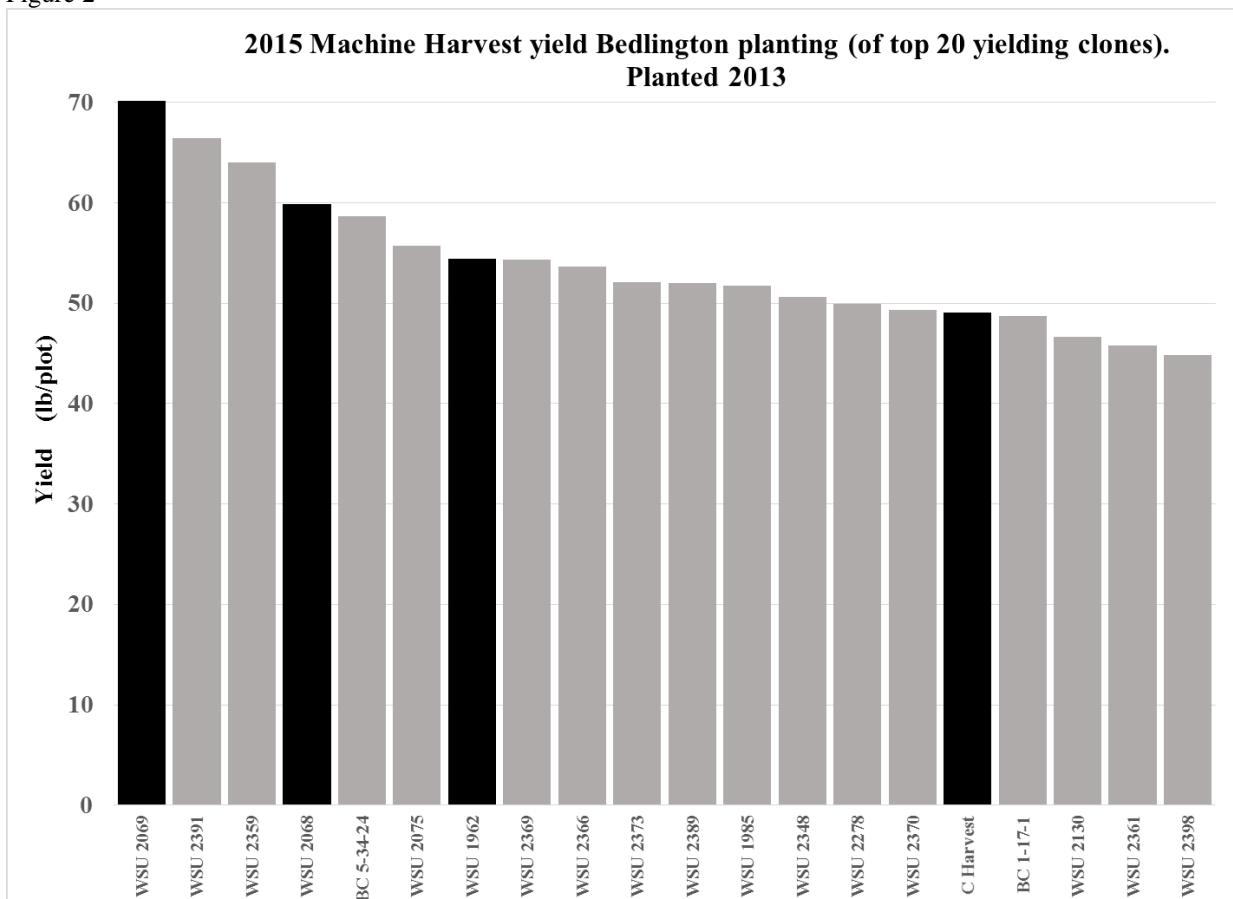


Table 1. 2015 and 2016 harvest of 2013 planted raspberries, Puyallup, WA

	Yield (t/a)			Fruit weight (g)		Culls (%)		Fruit firmness (g)		Midpoint of harvest	
	2016	2015	Total	2016	2015	2016	2015	2016	2015	2016	2015
C Harvest	10.3 a-c	7.8 a	18.1 a	4.09 ab	3.21 a	6.31 b	11.2 a	80 ab	192 ab	6/23 bc	6/28 cd
WSU 2075	12.2 a	5.7 a	17.9 a	3.02 b	2.02 ef	2.53 b	2.7 d	68 a-d	127 f	6/21 cd	6/22 e
WSU 2069	11.7 ab	5.8 a	17.4 a	4.00 ab	2.86 a-c	5.52 b	5.5 b-d	85 ab	163 b-d	6/18 ef	6/25 dc
WSU 2068	11.3 ab	6.1 a	17.4 a	5.41 a	2.94 ab	5.29 b	6.2 b-d	89 a	140 d-f	6/18 ef	6/23 e
Meeker	10.8 a-c	5.8 a	16.6 a	3.40 b	2.38 c-e	13.57 a	9.6 ab	81 ab	157 c-e	6/27 a	7/1 a-c
WSU 1914	10.0 a-d	5.4 a	15.3 a	3.81 ab	3.14 a	6.43 b	4.6 cd	67 a-d	141 d-f	6/28 a	6/30 bc
Willamette	9.7 a-d	4.7 ab	14.4 a	3.38 b	2.34 d-f	4.17 b	4.0 cd	67 a-d	131 ef	6/17 f	6/22 e
WSU 2010	9.7 a-d	4.7 ab	14.4 a	2.97 b	1.85 f	2.68 b	4.9 cd	64 b-d	137 d-f	6/20 de	6/24 dc
WSU 1985	8.4 a-e	5.1 ab	13.5 ab	3.91 ab	2.39 c-e	4.89 b	7.2 a-c	80 a-c	214 a	6/26 ab	7/3 ab
WSU 1962	7.9 b-e	5.0 ab	13.0 ab	3.35 b	2.37 c-e	5.84 b	7.3 a-c	57 cd	173 bc	6/27 a	7/5 a
WSU 2022	5.9 de	5.7 a	11.7 ab	3.48 b	2.97 ab	3.58 b	7.9 a-c	69 a-d	159 c-e	6/20 de	6/25 dc
WSU 1958	6.8 c-e	4.8 ab	11.6 ab	2.76 b	2.12 d-f	6.92 b	7.2 a-c	56 d	96 g	6/18 ef	6/23 e
WSU 1908	5.5 e	2.3 b	7.8 b	3.01 b	2.59 b-d	4.01 b	4.9 cd	49 d	82 g	6/19 d-f	6/23 e
Average	9.3	5.3	14.5	3.6	2.55	5.5	6.4	70	147	6/22	6/26

Table 2. 2016 harvest of 2014 planted raspberries, Puyallup, WA

	Yield (t/a)	Yield g/cane	Fruit weight (g)	Culls (%)	Fruit firmness (g)	Midpoint harvest
C Harvest	11.0 a	533 a	4.16 ab	9.79 a-c	90 a-c	6/23 d-f
WSU 2188	8.1 ab	493 ab	4.41 a	7.85 bc	102 a	6/27 a-d
Willamette	7.8 a-c	349 c	3.06 d-f	7.21 bc	74 b-e	6/19 fg
WSU 2001	7.8 a-c	537 a	3.86 a-c	14.33 a	87 a-d	6/30 a
WSU 2122	6.9 a-c	384 bc	3.64 bc	11.95 ab	88 a-d	6/26 b-e
Meeker	6.8 a-c	337 c	3.10 de	11.44 ab	74 c-e	6/28 ab
WSU 2200	6.8 a-c	336 c	2.49 f	6.40 bc	59 e	6/22 ef
WSU 1985	6.7 a-c	388 bc	3.43 b-d	9.16 a-c	64 de	6/27 a-c
WSU 2205	6.2 bc	322 c	3.16 de	4.25 c	74 b-e	6/17 g
WSU 0836	5.4 bc	315 c	2.92 ef	12.42 ab	63 de	6/17 g
WSU 2133	4.4 bc	438 a-c	2.93 ef	6.28 bc	60 e	6/23 c-f
WSU 2082	4.0 bc	393 bc	4.27 a	9.41 a-c	100 ab	6/23 c-f
WSU 2166	3.7 c	409 a-c	4.30 a	4.37 c	101 a	6/19 fg
Average	6.6	403	3.5	8.8	79.9	6/23

PROJECT: 13C-3755-5641

TITLE: Red Raspberry Breeding, Genetics and Clone Evaluation

CURRENT YEAR: 2017

PI: Patrick P. Moore, Professor
253-445-4525

moorepp@wsu.edu

WSU Puyallup Research and Extension Center
2606 W Pioneer
Puyallup, WA 98372

Co-PI: Wendy Hoashi-Erhardt, Scientific Assistant
253-445-4641

wkhe@wsu.edu

Year initiated 1987 Current year 2017 Proposed Duration: continuing

Project Request: \$78,000 for 2017-2018

Other funding sources:

USDA/ARS Northwest Center for Small Fruits Research

Amount Awarded \$32,419 for 2016-2017 for both raspberry and strawberry breeding

USDA/ARS Northwest Center for Small Fruits Research

Amount Awarded \$34,144 for 2016-2017 for “Enhanced Tools for Improving Root Rot Resistance in Red Raspberry”

ORBC

Amount Awarded \$4,500 for 2016-2017 “Development of New Raspberry Cultivars for the Pacific Northwest”

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, root rot tolerance and raspberry bushy dwarf virus (RBDV) resistance 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, tolerant to root rot and RBDV resistant while maintaining fruit quality. Replacement cultivars for 'Willamette' and 'Meeker' and new cultivars that extend the season are needed. With over 90% 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 2016 will be sown in 2016-2017. The goal will be to plant 108 plants for each cross.
3. Selections will be made among the seedlings planted in 2015. 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 2014 and 2015 will be harvested in 2017. 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 2014 and 2015 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 land use fees and \$5,000 for equipment use fees

Budget	2017-18
00 Salaries	\$20,102
Scientific Assistant (0.15 FTE)	
Ag Res Tech 2 (0.15 FTE)	
Ag Res Tech 1 (0.15 FTE)	
01 Timeslip Labor	12,000
03 Service and Supplies	31,868¹
Machine Harvest Trials	13,000
Land use fees	3,000
Equipment use fees	5,000
Supplies	10,868
04 Travel	4,000²
07 Benefits	10,030
SA, ART2, ART1	8,786
Timeslip	1,244
Total	\$78,000

¹ Includes: Field, greenhouse, and laboratory supplies; \$3,000 for WSU farm service fees, \$5,000 for WSU equipment use 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 2014 – Maberry Packing	\$3,000
Machine harvesting trial established in 2015 – Honcoop Farms	\$3,000

Maintenance of test plantings

Machine harvesting trial established in 2016 - Honcoop Farms	\$3,000
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Establishment and maintenance of new test planting

Machine harvesting trial to be established in 2017

Will work with the WRRRC to identify a suitable grower for the 2017 machine harvesting trial	\$4,000
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²Travel 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	% of time committed	Title of Project
Moore, P.P. and Hoashi- Erhardt	Northwest Center for Small Fruit Research	\$32,362	2016-17	5%	Small Fruit Breeding in the Pacific Northwest
Moore, P.P. and Hoashi- Erhardt	Northwest Center for Small Fruit Research	\$34,078	2016-17	5%	Enhanced Tools for Improving Root Rot Resistance in Red Raspberry
Moore, P.P. and Hoashi- Erhardt	Washington Red Raspberry Commission	\$63,000	2016-17	10%	Red Raspberry Breeding, Genetics and Clone Evaluation
Moore, P.P. and Hoashi- Erhardt	Washington Strawberry Commission	\$19,000	2016-17	5%	Genetic Improvement of Strawberry
Moore, P.P., K.K. Lanning and R.R. Martin	Washington Red Raspberry Commission	\$8,229	2015-16	1%	Evaluation of Raspberry Bushy Dwarf Virus strains
Moore, P.P. and Hoashi- Erhardt	Oregon Raspberry and Blackberry Commission	\$4,500	2016-17	2%	Genetic Improvement of Raspberry
Moore, P.P. and Hoashi- Erhardt	Oregon Strawberry Commission	\$4,500	2016-17	2%	Genetic Improvement of Strawberry
Moore, P.P. and Hoashi- Erhardt	Washington State Department of Agriculture	\$32,109	2014-17	2%	Fresh Market Strawberry Pre- Breeding for Repeat Flowering and Powdery Mildew Resistance

Pending Support

Name (List PI #1 first)	Supporting Agency and Project #	Total \$ Amount	Effective and Expiration Dates	% of time committed	Title of Project
B Strik, L.W. DeVetter, C. Finn, D. Bryla, Y. Zhao and G Fernandez	USDA SCRI	\$6- 7,000,000	2017-21	10%	Preparing berry crops for climate change through breeding and modification of horticultural systems
N. Grunwald, I Zasada, M. Bolda, P. Moore, S. Galinato, L.W. DeVetter, D. Klemer, T Walters	USDA SCRI	\$3,500,000	2017-21	10%	Novel tools and translational approaches for managing replant disease threatening the raspberry industry.

Washington Red Raspberry Commission Progress Report Format for 2016 Projects

Project No:

Title: Red Raspberry Cultivar Development

Personnel: Michael Dossett, BC Blueberry Council
C/O Agriculture and Agri-Food Canada
Agassiz Research and Development Centre,
PO Box 1000, 6947 #7 Hwy.
Agassiz, BC, Canada, V0M 1A0
Michael.Dossett@agr.gc.ca Tel: 604-796-6084

Reporting Period: 2016

Accomplishments:

- In 2016, more than 350 raspberry selections were evaluated in replicated and unreplicated machine-harvest plots. About 230 of these were given their first evaluation from the machine harvester, while the remainder were in their second year (though last year most of these were quite poor, in part because of the weather).
- A new machine-harvest trial plot was established at the Clearbrook substation, which includes about 70 plots.
- 38 new selections were made.
- More than 4800 new seedlings were established from 2015 crosses. The combination of planting seedlings as dormant plugs on plastic-covered raised beds means that most of these seedlings will get a first look in 2017 to select for easy release and fruit quality.
- New sources of resistance to RBDV were identified in the program, including germplasm that appears to be slow to become naturally infected – a trait which could be particularly valuable if strains capable of breaking the resistance gene *Bu* become prevalent in the region.

Results:

This last year was a significant year for our program in that we evaluated the largest number of machine-harvested trial plots and selections that we've ever managed at the Clearbrook site. The vast majority of these were selections evaluated for the first time, while most of the remainder were the first year that we got a really good look. From these, there are just over a dozen selections that are being propagated for more trials, and about 15 that we will be keeping a close eye on in the coming year. Nearly all of these have a parent that has strong root rot tolerance. Additional selections of interest had high yield and/or machine picked well, but are being reserved for use as parents because they are not expected to carry a sufficient level of root rot tolerance based on their pedigree. There were relatively few selections evaluated this year (~2700) because of uncertainty in funding for labor in 2013 when the program first transitioned to industry control. That said, 38 new selections were made. Most of these are from either wild germplasm, or backcrosses to species material for particular traits we are interested (e.g. backcrosses from salmonberry hybrids, for root rot tolerance and earliness). There were a few primocane-fruiting selections made that may interest fresh-market growers. These came from

~800 seedlings that were sent from the breeding program in Simcoe, Ontario after they lost all of their funding a few years ago. Because things were pushed back a few years ago, approximately 10,000 seedlings are in the que for evaluations in the coming season, including a trial specifically designed to look at heritability of traits that are associated with ability to machine-harvest.

We have a project evaluating a population that segregates for two different aphid resistance genes, gene *Bu* for RBDV resistance, and root rot resistance. This past year, we have bulked up plants to screen for root rot resistance and begun testing for resistance in the greenhouse. The same plants are in the field in Puyallup so that we'll be able to compare field and greenhouse results. In addition, we have grafted this population and ~90% of our breeding germplasm for RBDV to identify resistance. This has allowed us to identify individuals carrying gene *Bu* and test genetic markers for resistance (the best one so far is predictive ~98% of the time, but is only useable in ~60% of our germplasm, we are working to optimize this to make it more specific and widely useable). In addition, this work has uncovered individuals which do not carry gene *Bu* (i.e. we can graft the virus into the plant) but which have never tested positive for natural infection in the field, sometimes following 15+ years of exposure in multiple replicated trial plots in which 'Tulameen' and 'Chemainus' typically start becoming infected after 2-3 seasons. This trait will probably be our best option, moving forward, for controlling RBDV and associated crumbly fruit problems given that it appears strains of the virus capable of overcoming the resistance gene *Bu* are now established in Washington State.

Preliminary observations from selections of interest in machine-harvested plots (please contact me if you would like more information on any of these or other selections in our program):

BC 10-5-10 and BC 10-5-26: These two are siblings that both have firm fruit that appears to harvest quite well. One parent is 'Cherokee' so these two might have inherited some root rot tolerance. BC 10-5-10 is a bit earlier and a little smaller, while an early read on BC 10-5-26 is that it appears to have somewhat better yield. Both of these are being propagated for further trial. Yields BC 10-5-10 were a bit lower than Meeker in an unreplicated grower test plot in Whatcom County in 2016, while those of BC 10-5-26 were slightly higher than Meeker in the same planting.

BC 9-8-81: This is a 2nd backcross from wild *Rubus strigosus* collected from the Adirondacks in the mid 1980s. It machine harvests exceptionally well and has potential for extremely good root rot tolerance. It is slightly later than Meeker and may or may not have enough yield.

BC 7-32-29: This selection produced fruit that picked very well. The fruit were not quite as firm as we'd like to see, but are still probably firm enough. It was among the highest yielding selections in our 2012-planted trial. 'Cascade Bounty' is a parent so it has potential for good root rot tolerance. At the same time, color may be a bit borderline (a quality that 'Cascade Bounty' tends to give to its progeny). The combination of yield, harvestability, fruit quality and potential for root rot tolerance make it worth looking at further.

BC 9-26-18: Another progeny of 'Cascade Bounty'. This is another one that picks quite well and is probably "firm enough" though not as firm as we would like to see. It is getting propagated for a better look.

BC 7-20-30: A first backcross from *Rubus niveus*, this selection is susceptible to *Raspberry bushy dwarf virus* (RBDV) but might have inherited good root tolerance. It picks well and is quite impressive for being ¼ species material.

BC 9-10-104, BC 9-22-10, and BC 9-22-11: are all 2nd backcrosses from wild *Rubus strigosus* collected in the Adirondacks (parent is a sib of the parent of 9-8-81), and as such may carry good root rot tolerance. All three are slightly later than 'Meeker' and look like they harvest reasonably well in their first season of machine-picking.

BC 9-11-3: A bit later than Meeker, but with potential for root rot tolerance and nice looking dark fruit.

BC 9-15-69: This selection was consistently noted for its flavor in evaluations in 2016. The color looks very good and it may have RBDV resistance as well as root rot tolerance (too soon to tell, but based on parents, both are a possibility).

BC 10-9-5: Another progeny of Cascade Bounty that machine picks very nicely. Color is better on this one than some of the others. Beautiful conic fruit is probably “firm enough”

WSU 2166 and **WSU 2188** both looked outstanding in our replicated plots. WSU 2166 starts a week or so earlier than WSU 2188 and isn't quite as firm, but looks to probably be firm enough and harvests exceptionally well. WSU 2188 has very firm berries that harvest extremely well, but may be more susceptible to RBDV (3 out of 4 plots infected after first season).

Publications:

There are 2 or 3 additional publications from work performed in 2016 that are currently in preparation.

Peer-reviewed publications:

Dossett, M. and C. Kempler. 2016. Breeding raspberries for aphid resistance in British Columbia: progress and challenges. *Acta Horticulturae*. 1133:115-119.

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, WRRC, RIDC, LMHIA	\$801,266	April 1, 2013 – March 31, 2018	50%	Current funding comes from AAFC’s Growing Forward 2 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. Red Raspberry Breeding for the Pacific Northwest
	AAFC, BCBC, WBC, 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

2017 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

Continuing Project Proposal

Proposed Duration: (3 years)

Project Title: Red Raspberry Cultivar Development

PI: Michael Dossett

Organization: BC Blueberry Council

Title: Research Scientist

Phone: 604-796-6084

Email: Michael.Dossett@agr.gc.ca

Address: C/O Pacific Agri-Food Research Centre

Address 2: 6947 Hwy #7, PO Box 1000

City/State/Zip: Agassiz, BC V0M 1A0

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 2016 **Current Year** 2017 **Terminating Year** 2018

Total Project Request: **Year 1** \$12,000 **Year 2** \$12,000 **Year 3** \$12,000

Other funding sources: Funding for the raspberry portion of the breeding program also comes from the BC Raspberry Industry Development Council. A total of \$43,315 in industry cash for raspberries is needed as matching funds for the large federal grant from which our program operates. This money is leveraged 1:3 to pay for the breeder, student labor, machine harvest of trial plots and all supplies needed for operating the program.

Description: This project is to support the continued effort to breed raspberry cultivars adapted to the PNW. 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 germplasm 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:

The red raspberry industry is facing challenges with diseases, increased production costs and competition 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. Historically, one of the difficulties we have encountered is that our material with a high degree of root rot tolerance has not been machine-harvestable and has been a bit soft. The 2016 season will be our first year of evaluating yield and multi-plant plots of selections that were made from running the machine harvester over seedling plots, including many that are expected to have a moderate or high degree of root rot tolerance and have good firmness.

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 committed to providing office and lab space in support of the continuation of this program, as well as limited greenhouse and 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 efforts 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. As a result, we work in a rotating system where each year we are making new crosses, selecting from previous selections and discarding selections which don't make the grade during testing.

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. We are currently 3 years into a 5 year funding program called Growing Forward 2. 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, and we are currently in the process of validating two potential markers for RBDV resistance in this population as well as their transferability to our overall germplasm.

Anticipated Benefits and Information Transfer:

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

	2016	2017	2018
Salaries ^{1/}	\$	\$	\$

Time-Slip	\$10,000	\$10,000	\$10,000
Operations (goods & services)	\$2,000	\$2,000	\$2,000
Travel^{2/}	\$	\$	\$
Meetings	\$	\$	\$
Other	\$	\$	\$
Equipment^{3/}	\$	\$	\$
Benefits^{4/}	\$	\$	\$
Total	\$12,000	\$12,000	\$12,000

The costs we are asking WRRRC to support represent approximately 1/4 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 (this has run around \$8,000/year over the last three years). Hiring students for the summer period costs approximately \$10,000/student. With the leveraged support, the budget we are proposing to WRRRC 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.

Washington Red Raspberry Commission Progress Report 2016

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: 2016

Accomplishments:

Infrastructure developments to date

- Completed development of the infrastructure to support a functioning, ongoing network of regional on-farm grower trials for evaluating raspberry advanced selections linking participating growers, propagators, breeders, and other industry and commission participants.
- Expanded grower cooperator network to include sites with heavier soils and wider regional distribution.
- Completed practical yearly timeline for trial activities.
- Improved draft overall budget for determining annual costs for an ongoing program.
- Improved protocols for coordinating a joint on-farm trial program with British Columbia and Oregon caneberry growers.

Areas still in need of work

- Developing protocols for consist evaluation of trials and site visits.
- Determining more accurate annual fixed costs (labor, office, travel expenses, etc.) for an ongoing program.
- Acquiring a source of stable funding to maintain an ongoing, long-term program.
- Improving and stabilizing information dissemination.

Information Dissemination Methods

- Cultivar/selection factsheet handouts being produced annually.
- Ongoing inclusion of information in the Small Fruit Update newsletter.
- Posting on the Northwest Berry Foundation Website.
- Email and phone interaction with growers and processors.
- Meeting presentations.

Cultivars/Selections Included in Trails to date

Raspberries:

- | | |
|-------------------------|-----------------------------------------------------|
| • Rudi | • WSU 2166 |
| • Cascade Harvest | • WSU 2180 |
| • WSU 1912 | Selections now planned for inclusion in 2017 |
| • WSU 1948 | • WSU 1914 |
| • Lewis | • WSU 2010 |
| • Squamish (BC 92-9-15) | • WSU 2069 |
| • WSU 1980 | • WSU 2166 |
| • WSU 2122 | Selections now planned for inclusion in 2018 |
| | • WSU 2069 |

2017 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

Project Proposal

Proposed Duration: (1year)

Project Title: Coordinated Regional on-farm Trials of Advanced Raspberry Selections—Sixth Year

PI:

Tom Peerbolt

Organization: Northwest Berry Foundation

Title: Executive Director

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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** 2016 **Terminating Year** 2017

Total Project Request: \$11,200

Other funding sources:

Agency Name: Oregon Raspberry and Blackberry Commission

Amt. Requested/Awarded: \$11,500

Notes: This is a similar project that will allow us to test caneberries in Oregon.

Description: Develop and maintain a network of regional on-farm grower trials for evaluating raspberry advanced selections issuing from the USDA-ARS/OSU breeding program in Corvallis, the WSU breeding program in Puyallup and the British Columbia raspberry breeding program combining public and private resources in ways that would accelerate the commercialization of our genetic resources. Over the first five years of this project the grower/cooperator network has been developed; trials have been established; the infrastructure has been created and implemented for collecting, recording, and disseminating trial information. Correcting some of the logistical problems from past years, we will in 2017 implement an improved advanced planning system to ensure cooperating growers of getting the plant material at the optimal time and in the right amounts. We've also implemented a system for advanced planning for selecting advanced selections for future trials 2-4 years in advance.

Justification and Background:

The northwest raspberry breeding programs have been a cornerstone of the industry's success. Its 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 its 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:

- Maintain and improve the established network of regional on-farm grower trials for evaluating raspberry advanced selections issuing from the USDA-ARS/OSU breeding program in Corvallis, the WSU breeding program in Puyallup and the British Columbia raspberry breeding program.
- Evaluate trials established over the past five years on farms located in a variety of regional growing conditions.:
 - 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.
- Establish new trials in 2017 of 3-4 WSU advanced selections.
- Develop list of draft selections to be included in onfarm trials in future years.

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 2016.

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.

2017 procedures

- Establish new 2017 plantings following procedures similar to those used in previous years.
- Evaluations will be made of previous year plantings concentrating on fruit quality and yields.
- Plantings over four years old will have reached the end of their evaluation period within this program. They can be removed after this year's harvest. However, if determined useful some could be left in for longer term observations.
- Evaluations will be made in the fall to determination whether to continue for another year's data of previous plantings. Some could be removed earlier than the planned maximum four-year trial period if the information needed to determine their value to the industry has been collected.
- 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 include the system-wide efficiencies achieved by replacing the ad hoc grower trial system by one that is coordinated and supervised.
- The results will be transferred to users by the Northwest Berry Foundation which 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:

	2016
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.

ENTOMOLOGY

2017 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

New Project Proposal

Proposed Duration: (3 years)

Project Title: Non-toxic RNAi-based biopesticide to control spotted wing drosophila

PI: Man-Yeon Choi

Title: Research Entomologist

Phone: 541-738-4026; **Email:** mychoi@ars.usda.gov

City/State/Zip: Corvallis/OR/97330

Cooperators: Dr. Jana Lee, Research Entomologist; Robert R. Martin, Research Pathologist (Virology), USDA-ARS

Year Initiated 2017

Current Year 2017

Terminating Year 2019

Total Project Request: Year 1 \$10,000 Year 2 \$10,000 Year 3 \$10,000

Other funding sources:

Agency Name: Commissions of Oregon Blueberry, Washington Blueberry, and Oregon Red & Raspberry

Amt. Requested/Awarded: Funded \$52,500 from OBC, ORBC and Agricultural Research Foundation for 2015 and 2016.

Notes: Will request OBC, WBC, and ORBC (\$10,000 each).

Project Description: Spotted wing drosophila (SWD) is a destructive Dipteran pest that attacks a wide range of ripening fruits including almost all small and stone fruits. Since the first arriving in U.S. mainland 2008 the infestation of SWD is rapidly expanding across the North American and Europe. The estimated economic impact from crop yield loss, drop in market value, and higher management cost is hundreds of millions of dollars in the U.S. alone, and increasing every year. Current control methods depend on chemical insecticides carrying many negative effects. Therefore, novel approaches such as non-toxic insecticides or biologically-based environmentally friendly alternatives are requested by growers.

RNA interference (RNAi) for insect control represents a new direction and promising tool for insect pest management. One of the key advantages of RNAi technology is its high degree of species-specificity for the target pests; this is a unique point compared other conventional insecticides. To develop RNAi application there are several major challenges that must be overcome. Our previous RNAi studies on SWD that addressed these technical problems, and it now can bridge a gap to develop a novel RNAi-based SWD control option. This technology enables us to develop biologically-based control alternatives for SWD to protect the small fruit industries.

Justification and Background:

SWD is a serious invasive pest from Asia that is now in the United States, Canada, Mexico, South

America, and Europe. The severe damage caused by this destructive pest affects ripening small fruits, and the infestation area is rapidly spreading through North America as well as Europe. Growers are facing economic losses by increased spending on management costs, the loss of production and market values, and rejection of exports if unacceptable levels of insecticide residues and damage are found. Current control of SWD relies heavily on chemical insecticides which have negative impacts on agricultural ecosystems affecting non-target insects, pollinators, and human health. In addition, there is an inevitable risk that SWD populations in the field will develop insecticide resistance with the continuous use of chemical controls. Therefore, the heavy reliance on chemical insecticides should be replaced or at least complemented with biologically-based environmentally friendly alternatives.

During the past decade the availability of insect genomics and computational biology has further enabled the implementation of RNAi technology to target economically important insect pests. It has shown striking results in various insect groups, suggesting that it will be a promising tool for the next generation of pest management. Recently, intensive studies of the RNAi application for insect pest management in academia and commercial entities has enabled a breakthrough by having the first RNAi product as a commercial bio-pesticide in the field soon. To date, a variety of RNAi targets are being screened and evaluated for specific impacts applicable to pest management of agricultural crops or insect vector-borne diseases.

To successfully develop RNAi applications, a critical initial step is screening for appropriate RNAi target genes because degrees of gene silencing impacts vary from RNAi target genes and insects. The challenge with gene selection is to select suitable insect-specific target genes that provide fast-acting mortality or suppression and long-term population suppression without affecting other non-target organisms. Therefore, it is important to screen multiple and key RNAi candidates to improve the chance for identifying an effective RNAi target. To find the most effective RNAi target(s), our project proposal will be focused on the screening of RNAi targets in SWD.

Relationship to Commission Research Priorities: Prevention and management options for spotted wing *Drosophila* control which is related in WRRC's research priorities #1.

Objectives: The goal of this research objectives is the development of a novel environmentally-friendly control that is non-toxic insecticide and non-genetically modified strategy to control SWD as well as other potential pests. RNAi approach to pest management consider three major challenges: 1) selection and identification of suitable target genes, 2) cost effective RNAi material production, and 3) development of a suitable delivery method into target pest. A large scale production of RNAi in vitro using kits is too expensive, and not a practical approach for growers (#2). Therefore, there is required a mass production system to synthesis dsRNA through a microbial-based process provides more practical application. To solve this problem, we have established a mass production system using a microbial-induced dsRNA production to increase the feasibility of RNAi application for SWD control. To control SWD the strategy of our RNAi approach is non-planted incorporated delivery method such as spray and/or bait-station application (#3).

In the present proposal, therefore we focus on the screening and identification of suitable RNAi target(s) from SWD (#1). A feasible approach for RNAi target gene screening is to search previous targets or systems observed already from same or similar insect groups. Therefore, our approach for RNAi target gene screening is based on our current RNAi research and previous RNAi results. We recently started the screening of RNAi candidates from SWD, and currently evaluate their impacts on the fly. In this proposal we continue to screen more target genes from

SWD, and evaluate and identify suitable RNAi targets. In order to achieve this goal the following specific objectives need to be accomplished in this project:

1. Cloning and identify potential RNAi target genes from SWD (Yr. 1)
2. Construct, design and biosynthesis dsRNAs for target genes (Yrs. 1 & 2)
3. Screen for efficacy using bioassay to measure RNAi impacts on SWD (Yrs. 2& 3)
 - 3-1. Inject dsRNA into adult flies and monitor RNAi impacts (Yrs. 2& 3)
 - 3-2. Feed dsRNA to larvae and adults, and evaluate RNAi impacts (Yr. 3)

Procedures

PI has expertise on insect RNAi and published research results in several peer-reviewed papers and the USDA-ARS news (USDA-ARS, 2014) that demonstrated the selection of RNAi targets, construct dsRNA, micro-injection and bioassay in insect pests. In addition, those research results have been submitted for patent applications and awarded an RNAi patent to develop RNAi control method, and are being developed for practical use. Therefore, PI is well-positioned to conduct all experimental procedures, and supervise technical assistants or graduate students for this project.

1. Identify potential RNAi target genes: We will employ a BLAST search with the published SWD genome to identify homologous genes in SWD. Using routine molecular biology techniques and software, specific primers and/or degenerate primer set will be designed to amplify target genes. Once confirmed the sequence DNA fragments will served as the template for dsRNA synthesis. With PI's molecular biology knowledge and experience this approach is expected to be straightforward without possible pitfalls.

2. Evaluate RNAi impact(s) on SWD: DsRNAs of each target SWD gene will be dissolved in RNase free water and injected into pupal or adult stages of SWD using a nanoliter injector. PI has experience with micro-injecting dsRNA into small insects such as ants. After injection SWD will be monitored for negative impacts including mortality, longevity, fecundity and other parameters. Dr. Lee's lab has developed a system to monitor longevity and fecundity of flies. Dr. Martin's lab has experience and tools to investigate the silencing of RNAi-targeted genes. Once we identify best RNAi target genes, feeding assays will be conducted if incorporated into a bait and kill approach.

3. Screening RNAi targets of SWD: For adult feeding assays, various dsRNA concentrations determined from the injection experiment will be mixed in a dry bread yeast. The mixed yeast with dsRNA material will be sprayed on the surface of the artificial diet in a petri-dish to allow adult flies to feed in the cage. After feeding, flies will be monitored for phenotypic changes, and verified for gene silencing as described above.

List of Accomplishments

1st year: Identify partial and/or full sequences for more target genes, and obtain actual DNA data.

2nd year: Design templates for dsRNA synthesis, synthesis dsRNAs for all target genes and evaluate each dsRNA amount and purity.

3rd year: Determine negative phenotype and/or genotype impacts on SWD, obtain narrowed down SWD RNAi targets for further evaluation.

Anticipated Benefits and Information Transfer: At the completion of these studies we expect to have identified potential RNAi target(s) that can be used to develop a biologically-based insecticide as a chemical insecticide alternative to control SWD and other pests of small fruits. We

also expect to identify specific physiological impacts from RNAi treatments on SWD. Thus, outcomes are not only expected to address specific questions in RNAi research for SWD control, but also to have fundamental impacts for the application of RNAi for biological pest control.

References selected

Choi, M.Y., Vander Meer, R.K., Coy, M., Scharf, M.E., 2012. Phenotypic impacts of PBAN RNA interference in an ant, *Solenopsis invicta*, and a moth, *Helicoverpa zea*. J Insect Physiol 58, 1159-1165.

Huvenne, H., Smagghe, G., 2010. Mechanisms of dsRNA uptake in insects and potential of RNAi for pest control: a review. J Insect Physiol 56, 227-235.

Lee, J.C., Bruck, D.J., Dreves, A.J., Ioriatti, C., Vogt, H., Baufeld, P., 2011b. In Focus: Spotted wing drosophila, *Drosophila suzukii*, across perspectives. Pest management science 67, 1349-1351.

Vander Meer, R.K., Choi, M.Y., 2013. Formicidae (ant) control using double-stranded RNA constructs. The United States of America, as represented by the Secretary of Agriculture, Washington, DC (US), USA.

Vander Meer, R.K., Choi, M.Y., 2015. Control of insect pests through RNAi of pheromone biosynthesis activating neuropeptide receptor, in: The United States of America, a.r.b.t.S.o.A., Washington, DC (US) (Ed.).

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Budget

This project is being submitted to OBC, WBC, ORBC and WRRC (\$10,000 each) for FY17-18. USDA-ARS base funds in Dr. Choi’s programs will be used to fund additional technical support and supplies for the project.

	2017	2018	2019
Salaries ^{1/}	\$23,000	\$23,000	\$14,000
Time-Slip	\$0	\$0	\$0
Supplies & Services	\$13,000	\$13,000	\$4,000
Travel ^{2/}	\$2,000	\$2,000	\$1,000
Meetings	\$0	\$0	\$0
Other	\$0	\$0	\$0
Equipment ^{3/}	\$0	\$0	\$0
Benefits ^{4/}	\$2,000	\$2,000	\$1,000
Total	\$40,000	\$40,000	\$20,000

Budget Justification

- ^{1/}Postdoctoral associate (0.5FTE)
- ^{2/}Travel to commission and grower meetings
- ^{4/}Benefit (50%)

Total Budget for Project 2017 \$40,000

Funding Breakdown
WBC, OBC, WRRC and ORBC (\$10,000 each)

Washington Red & Raspberry Commission Budget Request \$10,000

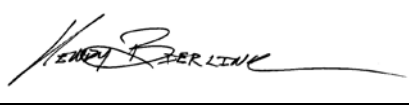
Current & Pending Support

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
Choi & Lee	Current: Northwest Center for Small Fruits Research	\$104,514	05/01/2014- 12/31/2016	10	Development of Bacteria Expressing an inducer of RNAi as a Biologically-Based Non-transgenic insecticide to control spotted wing drosophila
Choi & B. Martin	OBC & ORBC	\$20,000 \$20,000	10/01/2015- 09/30/2017	10	Development of biologically-based RNAi Insecticide to control spotted wing drosophila
Choi & R. Martin	Oregon Department of Agriculture	\$19,641	01/01/2016- 12/31/2016	5	Development of RNAi-based pesticide to control slugs in nurseries
Choi & Lee	Oregon Department of Agriculture	\$22,141	01/01/2016- 12/31/2016	5	Development of RNAi-based insecticide to control brown marmorated stink bug in nurseries
Choi	Agricultural Research Foundation	\$12,500	02/01/2016- 1/31/2017	5	Screening of RNAi targets to develop a novel RNAi-based control method for spotted wing drosophila
Choi, R. Martin & Ahn	Agricultural Research Foundation	\$12,495	02/01/2016- 1/31/2017	3	Development of RNAi-based biopesticide to control slugs on agricultural crops

Choi & B. Martin, Lee	Pending: OBC, WBC, ORBC	\$30,000	01/01/2017-12/31/2017	10	Development of biologically-based RNAi Insecticide to control spotted wing drosophila
Choi, Lee & Ahn	USDA-NIFA	\$313,248	10/01/2016-9/30/2019	15	Development of a non-nutritive sugar-based RNAi cocktail to control spotted wing drosophila
Choi, R. Martin & Ahn	Oregon Department of Agriculture	\$14,700	01/01/2017-12/31/2017	3	Genomic sequencing of gray garden slug: A molecular foundation for slug research
Choi, Lee, Martin & Ahn	Washington Tree Fruit Research Commission	\$43,880	01/01/2017-12/31/2017	5	Non-toxic RNAi-based biopesticide to control spotted wing drosophila

Washington State Commission on Pesticide Registration Number NEW

PROJECT # _____ (To be filled in by WSCPR)

1) Project Title: Survey for Egg Parasitoids of Brown Marmorated Stink Bug, <i>Halyomorpha halys</i> in Skagit and Whatcom Counties in Western Washington	
2) Applicant (user group) Name and Address: Washington Red Raspberry Commission 1796 Front Street Lynden, WA 98264	3) Project Contact Name, Phone and Email: Henry Bierlink 360-354-8767 henry@red-raspberry.org
3) Details of Project: Crop/Site <u>Raspberry/</u> Chemical <u>NA</u> (if specific to a particular chemical(s)) Pest Management Issue <u>BMSB egg parasitoid survey</u> Pest <u>brown marmorated stink bug, <i>Halyomorpha halys</i></u>	4) Research Lead: Name, Institution and Email Beverly Gerdeman WSU Mount Vernon NWREC bgerdeman@wsu.edu
4) Project Category: Check all that describe the focus of your project. Registration ___% Non Registration <u>100%</u> ___ Efficacy Trial ___ Integrated Pest Management ___ GLP ___ Phytotoxicity Study ___ Pesticide Resistance Study ___ Residue Study ___ Other _____ <u>X non-GLP</u>	
5) Project Duration Start Date : <u>June 2017</u> End Date : <u>September 2016</u>	
7) Total Project Cost \$ <u>20,363</u> WSCPR Request \$ <u>8,083</u> Co-funding \$ <u>\$12,280</u>	
8) Project Summary: Briefly (in 150 words or less) describe the pest control situation your project will address, its impact on the crop, and how WSCPR support will resolve the problem within a 5-year time frame. The brown marmorated stink bug, <i>Halyomorpha halys</i> , is a direct pest and harvest contaminant of blueberry and red raspberry. In 2015, the key egg parasitoid exhibiting 60-90% parasitization rate, <i>Trissolcus japonicus</i> , was identified in southern Washington State. Northwest Washington, the epicenter for small fruit production in the PNW has not been included in any of the parasitoid surveys. Presence of the parasitoid would increase probability that parasitoid releases could establish. Parasitoid introductions early in the invasion process can prevent outbreaks. The detection of a single BMSB in Skagit County in 2016 and insipient numbers discovered in BC, indicate populations in the PNW are at levels most susceptible to biocontrol mass releases. Due diligence prior to release of exotic natural enemies requires a pre-release survey. Areas not supportive of pre-release surveys would be low priority for releases. We propose to survey Skagit and Whatcom Counties for the presence of <i>T. japonicus</i> .	
9) Signatures <i>I certify to the best of my knowledge that the information in this application is true and correct.</i> <u>Henry Bierlink</u> Printed Name of Applicant <u>Executive Director, WRRC</u> Title of Applicant  <u>November 10, 2016</u> Date Signed	
10) Send original application to: Washington State Commission on Pesticide Registration; 2621 Ringold Road, Eltopia, WA 99330	

Description of Problem

Small fruit production (canberries and blueberries) in Western Washington is estimated at \$21.5 million. The brown marmorated stink bug, *Halyomorpha halys*, BMSB, is a serious, direct pest of small fruit with few natural predators in North America, allowing it to quickly spread throughout the United States. BMSB was detected in Skagit County in 2016 and also in British Columbia. In October 2016, Drs. Betsy Beers and Michael Bush reported a jump in BMSB numbers in Wenatchee and Yakima from a 35 yearly total for 2015 to over 200 in 5 days in 2016 (<http://cahnrs.wsu.edu/news-release/2016/10/18/stink-bugs-invade-more-counties-more-homes-in-washington-state/>). The sudden jump in numbers has entomologists on edge for the coming year. BMSB will feed and reproduce on blueberries, raspberries and blackberries. Buds and fruit of both wild and cultivated *Rubus* spp are prone to BMSB attack and infestations can result in off-flavors. On blueberries it will feed on all stages of fruit development causing sunken discolored areas. In addition, BMSB is a machine harvest contaminant threatening domestic trade and international exports.

Egg parasitoids are the key natural enemies that have helped keep numbers in check, in its country of origin, China but native North American egg parasitoids thus far, exhibit low levels of parasitism. The main Asian parasitoid, *Trissolcus japonicus*, exhibiting 60-90% rate of BMSB parasitization, was identified in Southern Washington State, in 2015 by Betsy Beers, Professor of Entomology with the WSU Tree Fruit Research and Extension Center, TFREC. This discovery is significant and could be a *game-changer* for the small fruit industry but the parasitoid's distribution in the state remains unknown. Surveys have been conducted by Betsy Beers and Nik Wiman, (Assistant Professor with OSU NWREC, Corvallis, OR), but Betsy is concentrating on the main fruit-growing region in eastern WA and Nik is concentrating in Oregon. Northwest Washington, the epicenter for small fruit production in the Pacific Northwest, has not been included in any funded survey research. Knowledge of the distribution of the egg parasitoid, *T. japonicus* in Washington State may help to determine its natural range and suitability of mass releases. BMSB populations in the PNW are at levels most susceptible to biological control mass releases. Due diligence prior to release of exotic natural enemies requires a pre-release survey. Areas not supportive of pre-release surveys would be low priority for releases. We propose to survey Skagit and Whatcom Counties for the presence of *T. japonicus*, to provide growers with maximum opportunities for future releases.

Funding Categories

Category B - Protection of the Environment

I. Protection of wildlife

II. Protection of natural resources

III. Control of non-native, invasive pests

The BMSB egg parasitoid survey will determine if the wasp is present in Skagit and/or Whatcom Counties. Occurrence of the wasp in these counties would likely prevent damaging populations of BMSB, eliminating the need for excessive use of insecticides. Timing of the survey however is particularly critical now as BMSB is just entering Skagit and Whatcom Counties and populations are low. Reducing insecticide use will reduce pesticide runoff, in turn protecting the vulnerable salmon populations, already suffering from lack of food resulting from unusual warmth in the Pacific Ocean. Reducing pesticide use will also conserve native and commercial pollinators.

Category C - III. Development of an integrated pest management tactic

Significance to Local or Regional Economy

Integrated pest management includes biological control along with cultural and chemical controls.

Detecting the egg parasitoid, *Trissolcus japonicus* in Northwest Washington would indicate the suitability of the area for mass releases and demonstrate the region's commitment to include biological control as an important component along with cultural and chemical methods in their BMSB control program.

Project Description

Rearing BMSB for egg masses

WSU NWREC will establish a colony of BMSB to provide fresh sentinel egg masses for the survey based on Medal et al (2012) and Tatman et al (2013).

Fifteen reproductive pairs of BMSB will be placed into fifteen rearing containers 30 cm x 23 cm x 10 cm along with 50 ml glass containers covered with cotton to provide moisture. The bottom of the containers is lined with Kimwipes® and paper towels are inserted into each box along with each reproductive pair of BMSB to provide protection. The containers will be stacked on shelves and exposed to 16-h photoperiod (16:8 h L:D) at 26°C ±2 and 50-55% RH. BMSB adults will be provided a variety of vegetables and nuts to promote egg development. Egg masses will be collected daily and stored at 10-12°C to prevent further development. Prior to deployment, non-viable egg masses will be affixed to cards using double-sided sticky tape.

Deployment and Collection

Skagit County Extension personnel will be responsible for the Skagit survey and Whatcom County Extension personnel will be responsible for their county survey. Each county will set out the sentinel egg masses on Mondays from June through September in 12 locations, to detect presence of BMSB parasitoids, both native and the exotic, *Trissolcus japonicus* by clipping egg masses in a natural position on the underside of leaves. Wooded locations near fruiting raspberry fields will be targeted for monitoring. At each survey site, vegetation will be inspected for presence of BMSB using a beating tray. Sticky cards with an attached *H. halys* lure (Alpha Scents, Inc, HalHal) will be set and checked weekly. Lures will be changed after 6 weeks. Egg masses will be recovered on Thursdays after 3 days in the field to prevent losses from predation and weathering. Parasitoids found on the egg masses in the field will also be collected using an aspirator and placed into separate vials for shipment and identification. Egg masses will be returned to WSU NWREC to rear out the parasitoid wasps.

Parasitoid Rearing and Shipment

Parasitized egg masses will be held for emergence in crispers stacked on shelves and exposed to 16-h photoperiod (16:8 h L:D) at 26°C ±2 and 50-55% RH. Recovered wasps will be shipped to Josh Milnes, at WSU TFREC in Wenatchee, WA. Betsy Beers, entomologist WSU TFREC and Nik Wiman, entomologist at OSU NWREC in Corvallis will provide their expertise if needed. Significant findings will be presented at the Washington Small Fruit Conference in 2017 in Lynden, WA and will be available on the WSU NWREC website

<http://mtvernon.wsu.edu/ENTOMOLOGY/main/index.html>.

References:

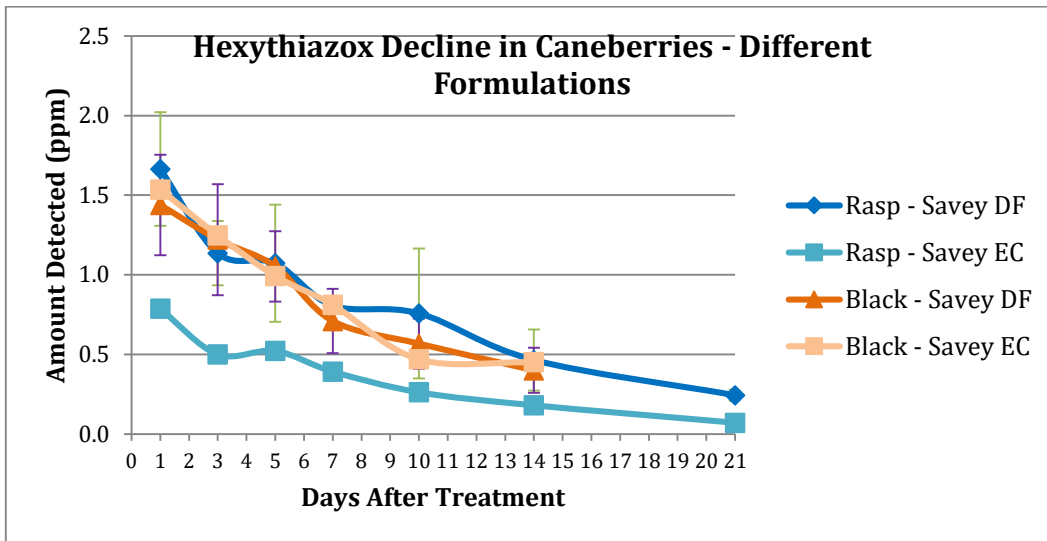


Fig. 8. Hexythiazox decline in caneberries with two different

Malathion is an organophosphate and like all Ops, residues fall precipitously after application and by 3-5 DAT are not as effective for SWD control as pyrethroids (Fig. 9). Nevertheless alternating malathion with zeta-cypermethrin provides excellent control of SWD due to the lingering pyrethroid sublethal residues which will boost the effect of malathion. Malathion MRLs are compatible at 8 ppm for Australia (10ppm) Canada and Japan but require 5 days for Korea (0.5). Residue levels of 0.01 are detectable past 22 DAT therefore shipments to Taiwan (0.01) are not advisable.

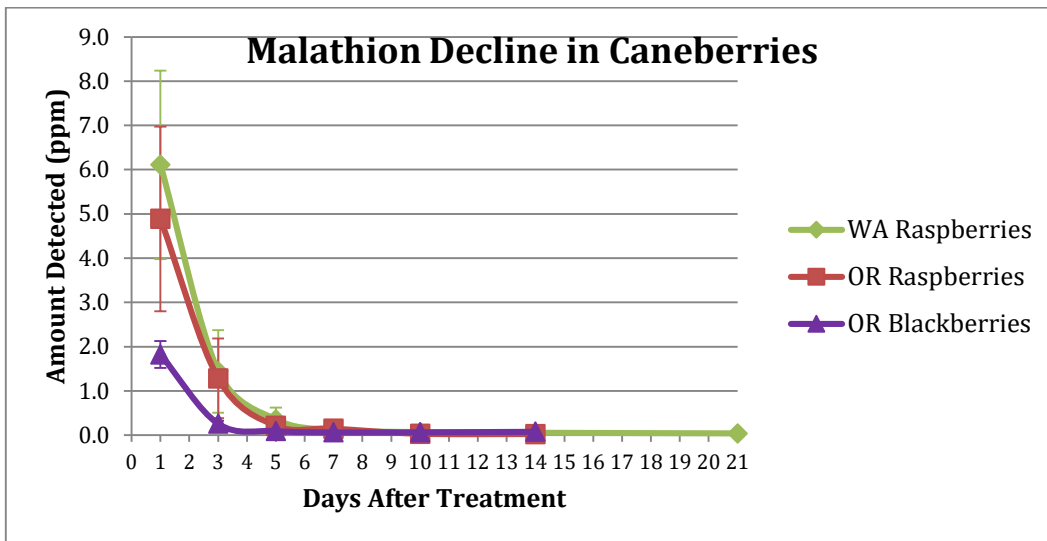


Fig. 9. Malathion decline in caneberries.

Imidacloprid is a neonicotinoid, a systemic product. The US has a tolerance set at 2.5 at 3-day PHI. Tolerances are compatible for all except Korea (0.5) and Taiwan (1.0) but meet all export market tolerances by 2-day PHI for red raspberry and between 3-4 days for blackberry (Fig. 10).

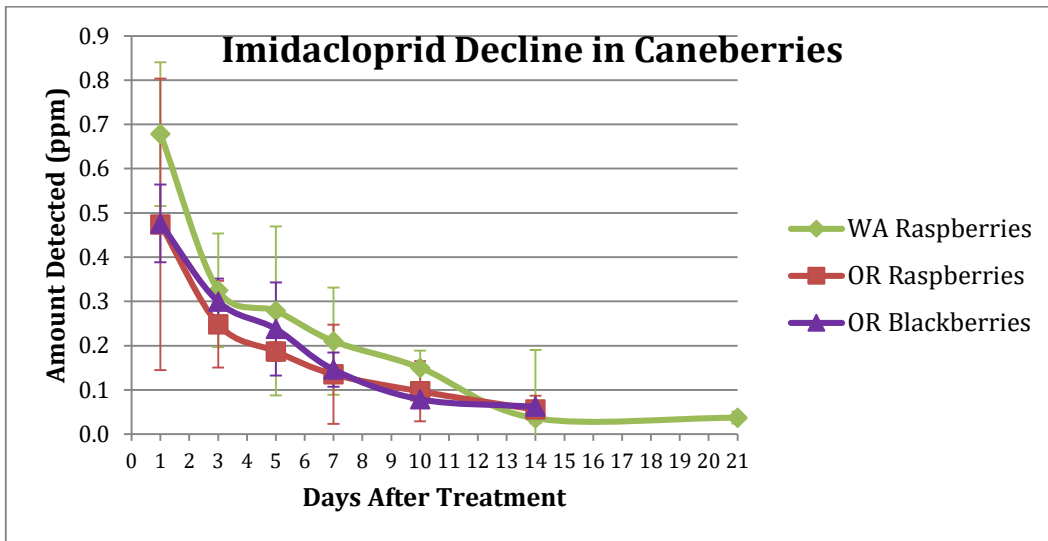


Fig. 10. Imidacloprid decline in caneberries.

Spinosad MRL is set at 1 at 1-day PHI for the US. All residues were within MRLs for all countries after 1 day PHI (Fig. 11). Korea has a lower MRL set for red raspberry (0.5 ppm) than blackberry (1 ppm).

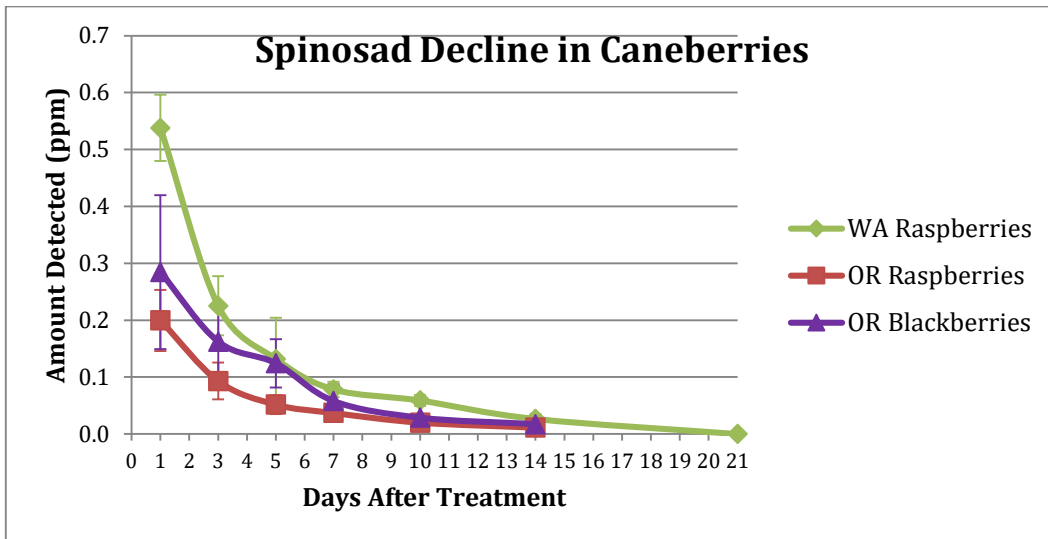


Fig. 11. Spinosad decline in caneberries.

Spinetoram residues are set at 0.8 at 1-day PHI. Both Taiwan and Japan's MRL's are lower for red raspberry than blackberry. All residues are less than 0.5 ppm at 1-day PHI and meet red raspberry residues for Korea after 11 days and fall below 0.01 in blackberry after 14 days for Taiwan (Fig. 12).

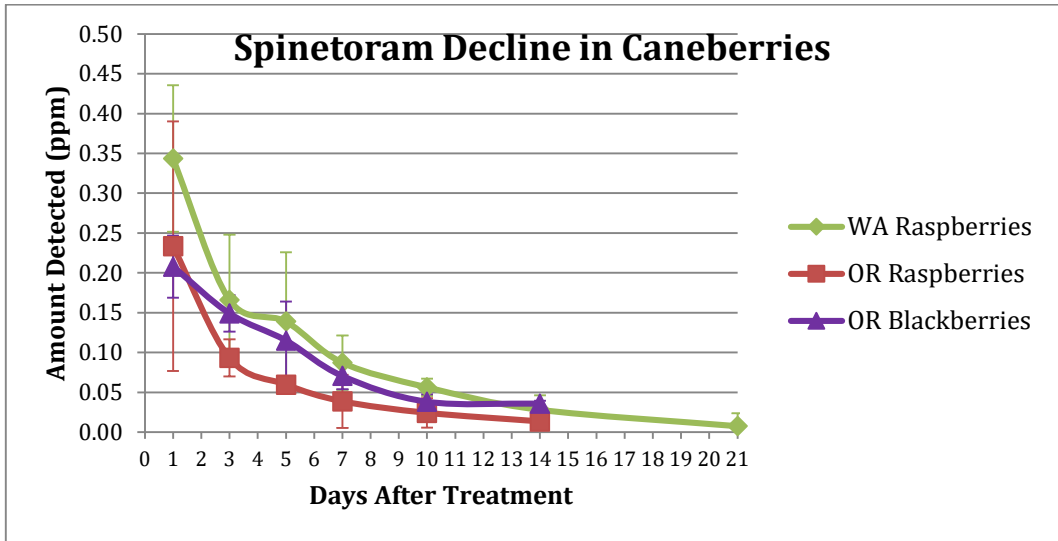


Fig. 12. Spinetoram decline in caneberries.

Results of these decline studies represent 2 locations in a single year. Multiple years are necessary to develop representative residue declines. The USDA FAS TASC grant will provide funding to perform insecticide, miticide and fungicide studies from 2017 – 2019. These analyses will provide preliminary data by representing additional data points for the upcoming study.

Insecticide/Miticide Decline in PNW Caneberries

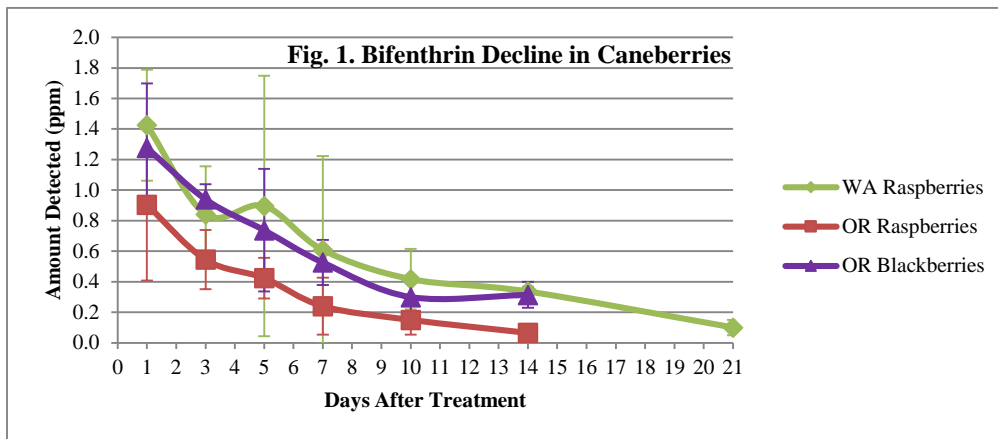
Bev Gerdeman, Joe DeFrancesco, Camille Holladay and Hollis Spitler

bgerdeman@wsu.edu, defrancj@science.oregonstate.edu, cholladay@synpestlab.com,
spitler@wsu.edu

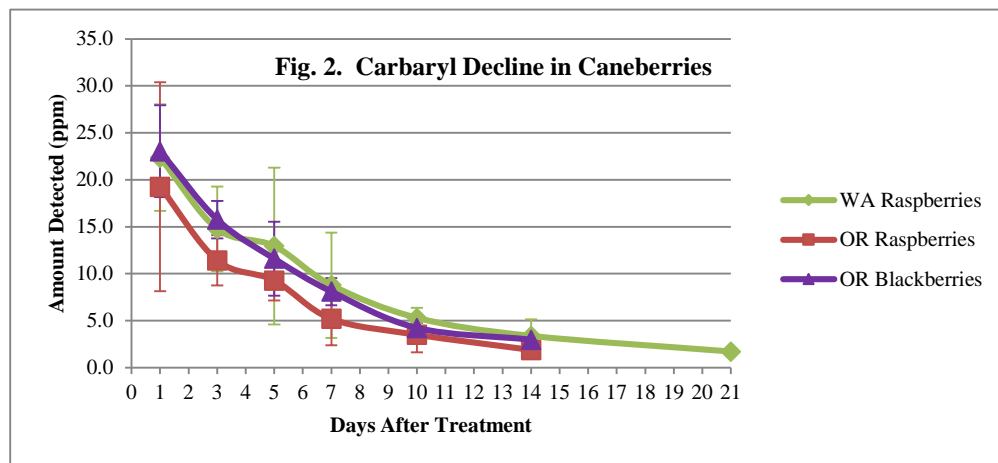
Spotted wing drosophila, *Drosophila suzukii* continues to show preference for all caneberries and weekly insecticide applications necessary to protect berries may put growers at risk for residue violations. This study provides information on insecticide/miticide degradation curves for the PNW region representing a single season. Field sites included: raspberry - Lynden, WA and raspberry and blackberry - Aurora, OR. Washington treatments were replicated 3X with an over-the-row boom and 4X in Oregon using a backpack sprayer. Analyses were performed by Synergistic Pesticide Lab in Portland, OR. Target export countries include Australia (AU), Canada (CA), Japan (JA), Korea (KO) and Taiwan (TA) and MRLs based on www.globalmrl.com database are current as of 11/29/16. (PHI are in parentheses). RR = red raspberry and BB = blackberry

Results

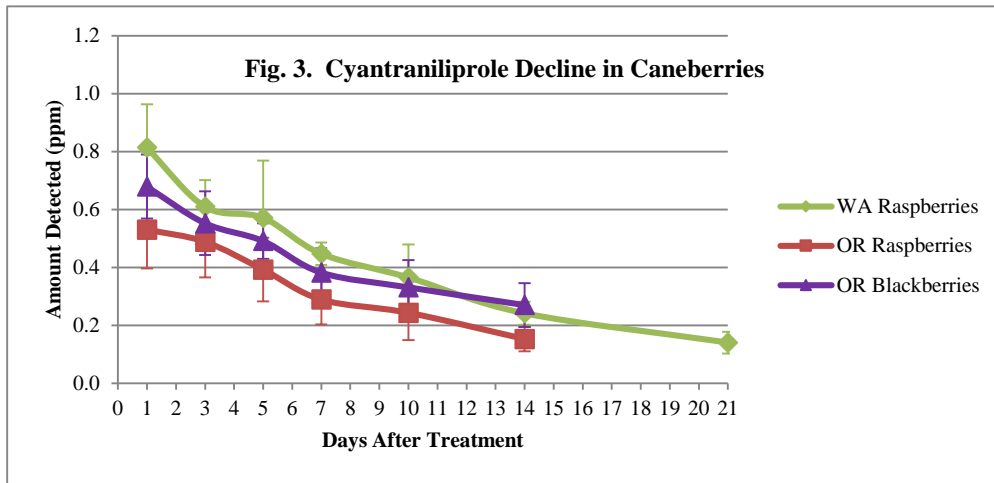
US tolerances for bifenthrin are set at 1 ppm. Bifenthrin meets MRLs for US (3) and all other countries (Fig.1).



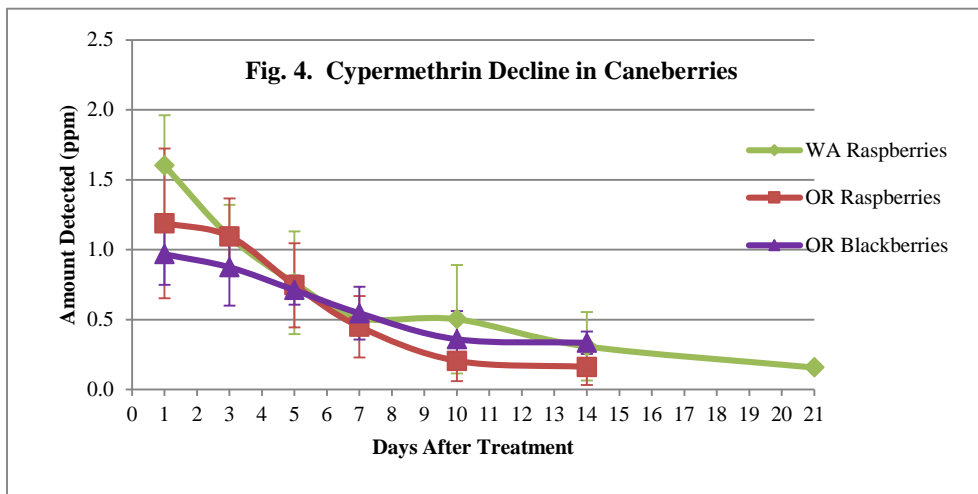
US tolerances for carbaryl are set at 12 ppm. Carbaryl meets MRLs for US (7) and all countries except KO and TA set at 0.5; need to wait 21 days for KO & TA. (Fig. 2)



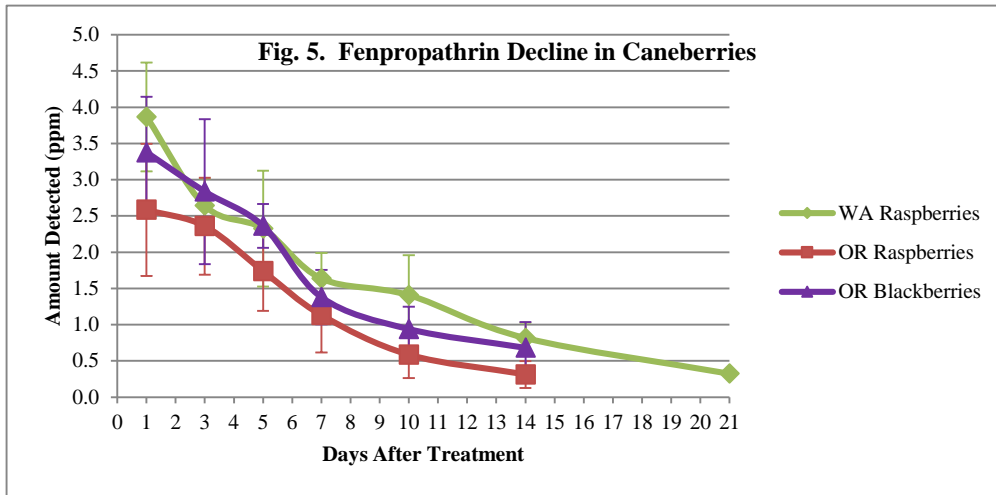
Cyantraniliprole is not registered with no MRLs set. For reference, US tolerances are set at 4.0 ppm for highbush blueberry (3) while caneberry ppm in this study, were < 0.6 at 3-day PHI (Fig. 3).



US tolerances for cypermethrin and zeta-cypermethrin, analytically indistinguishable, are set at 0.8 ppm. Cypermethrin is OK for TA but excessive residues were detected for US (1) & other countries requiring waiting 5 days (Fig. 4). These results will be reviewed.

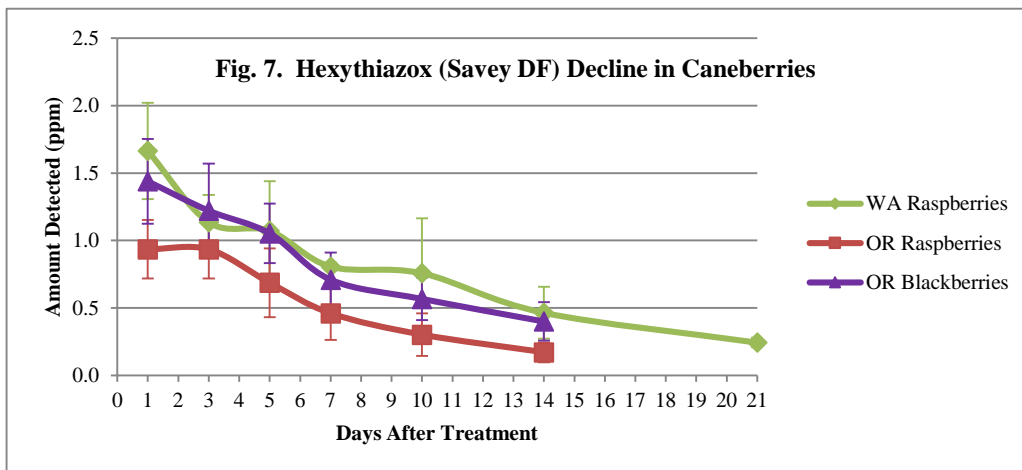
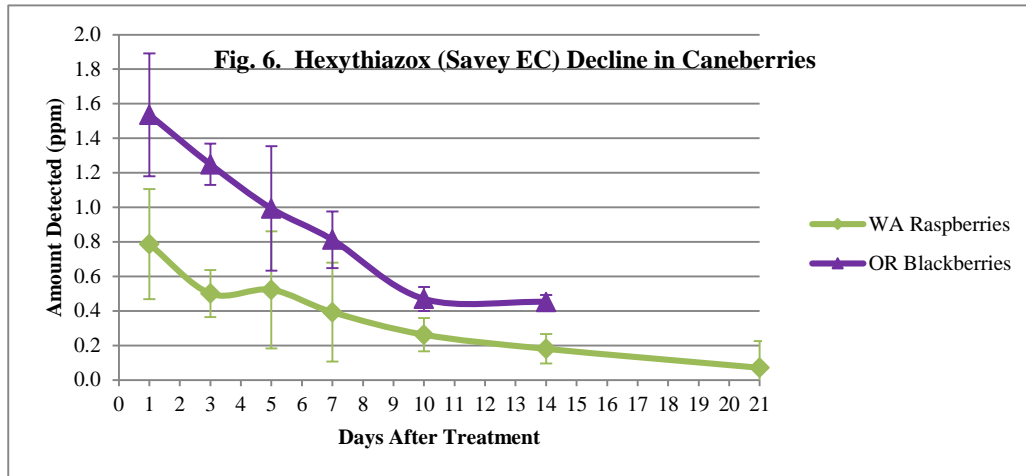


US tolerances for fenpropathrin are set at 12 ppm. Fenpropathrin meets MRLs for the US (3) and all countries except AU (0 ppm) and KO (0.5 ppm); wait 18 days for KO (Fig. 5).

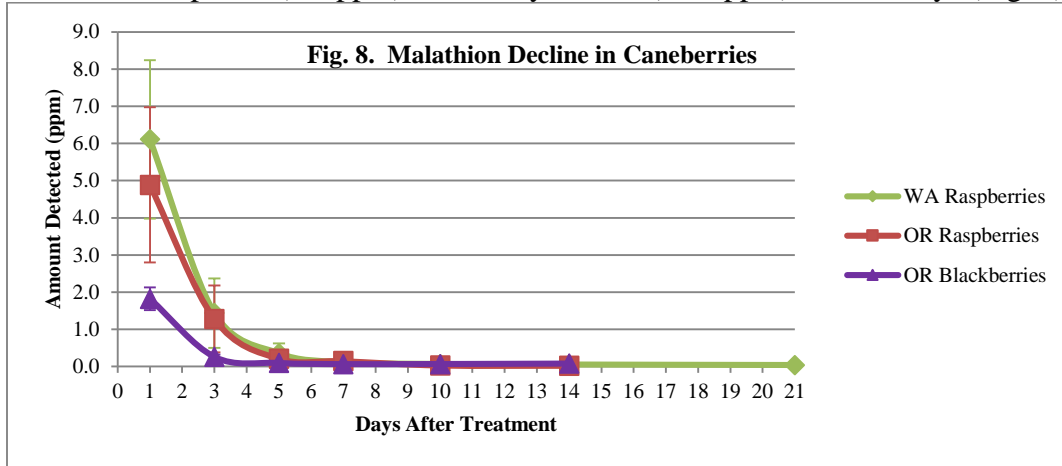


US tolerances for hexythiazox are set at 1 ppm. Hexythiazox

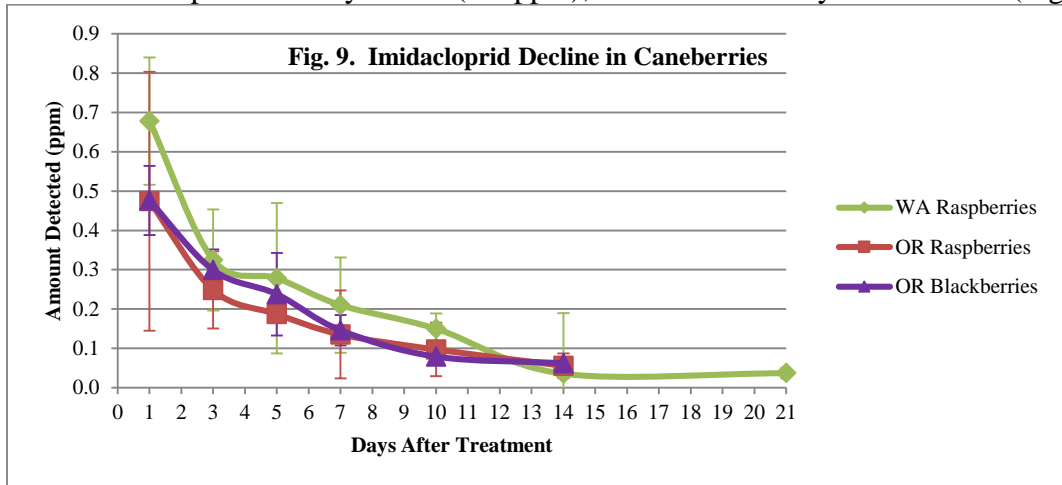
MRLs were OK for CA but excessive residues were detected for US (3) and all other countries (Figs. 6 & 7). This will be reviewed in the coming 3-year study. Decline rates for both formulations did not vary.



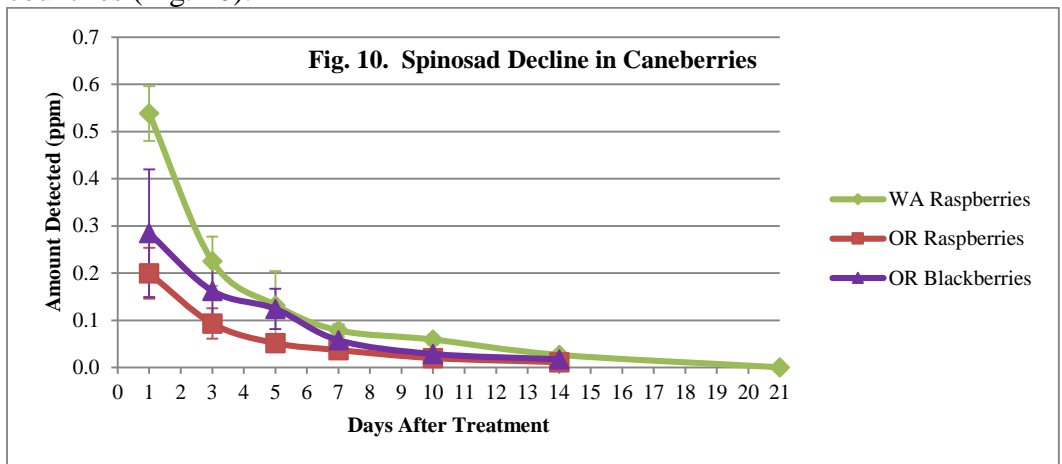
US tolerances for malathion are set at 8 ppm: Malathion meets US (1) and MRLs for all countries except KO (0.5 ppm) wait 5 days & TA (0.01 ppm) wait 21 days (Fig. 8).



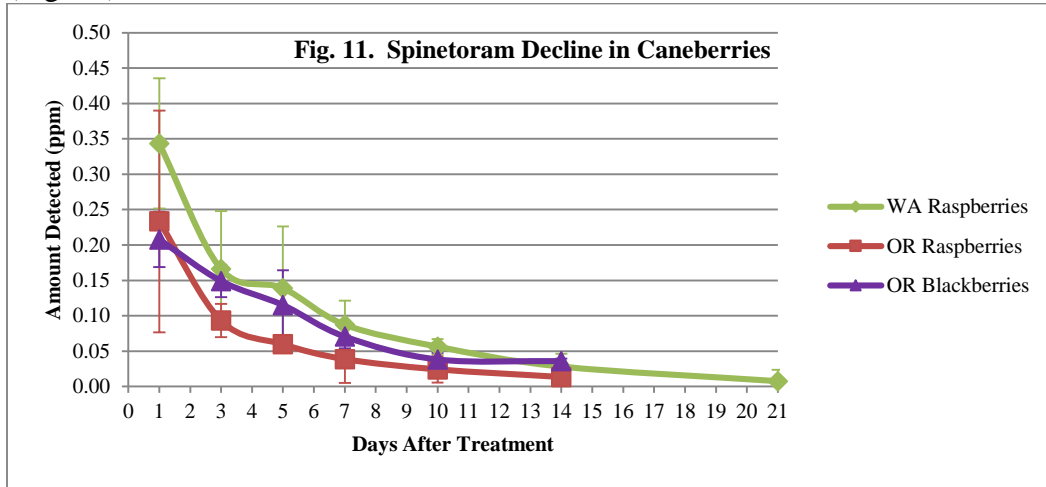
US tolerances for imidacloprid are set at 2.5 ppm. Imidacloprid meets US (3) and MRLs for all countries except blackberry in KO (0.3 ppm); need to wait 4 days instead of 3 (Fig. 9).



US tolerances for spinosad are set at 1 ppm. Spinosad meets MRLs for the US (1) and all countries (Fig. 10).



US tolerances for spinetoram are set at 0.8 ppm. Spinetoram meets MRLs for the US (1) and all countries except KO (0.05 ppm) wait 10+ days & TA (0.5 ppm RR, 0.01 ppm BB) wait 14 days (Fig. 11).



WEEDS

Project Number: 13C-3419-7297

Title: Weed Control in Red Raspberries

Personnel: Timothy W. Miller, WSU Mount Vernon NWREC
Carl R. Libbey, WSU Mount Vernon NWREC

Reporting Period: 2015-16

Accomplishments: A total of three raspberry trials were conducted during 2015-16: an IR-4 trial, an FMC Zeus Prime herbicide (these both had separate funding) trial, and a baby raspberry trial funded by the Washington Red Raspberry Commission. The first two trial was conducted at was conducted at the Purewal Farm near Laurel, WA, the second at WSU NWREC. Data for the baby raspberry trial are reported here and was presented at the Northwestern Washington Small Fruit Conference in Lynden in December, 2016.

Results:

Baby Raspberry Trial. Tissue-culture ‘Cascade Harvest’, ‘Chemainus’, ‘Squamish’, and ‘Wakefield’ red raspberry plugs were obtained from Northwest Plant Company and were transplanted by hand at WSU NWREC May 16, 2016. Three plants of each cultivar were planted sequentially into a single row in each plot (12 plants total/plot). Herbicides were applied post-transplant over the top of each row in the two trials May 18. Weed control and crop injury was estimated on May 23, June 17, July 26, and September 12, and crop injury was also recorded at the May and June evaluations. Length of the longest cane on each plant was measured at the July and September ratings. The experiments were randomized complete block designs, each with three replicates. Means were separated using Tukey’s Honestly Significant Difference statistic ($P \leq 0.05$).

Weed control was excellent for most treatments for the first two months (Table 1). However, July weed control with Devrinol and Prowl H2O was only 40 and 60%, respectively, while control with Zeus, Trellis, and Sandea had fallen into the 70% range. By September, only Fierce was still providing an acceptable level of weed control (87%), although control ratings were quite variable among the plots.

Average raspberry plug response to certain herbicides was rapid. Crop injury was noticeable at one week after treatment (May 23) with a foliar injury rating of 35% with Chateau at 12 oz/a (Table 2). It should be noted that nontreated raspberry was displaying 26% chlorosis at the same rating. Nontreated plant injury was a similar 27% by June, but greater injury was noted with Chateau at both rates, Fierce, and Sandea (38 to 62% injury). It is suspected that the injury from Chateau and Fierce was primarily due to postemergence activity of the flumioxazin in these formulations applied to nonhardened raspberry foliage, and that injury would likely have been different had these products been applied pretransplant.

Raspberry plant survival was essentially the same at the July and September ratings, so plug loss occurred shortly after transplanting/herbicide application (Table 2). The lowest survival rate was

recorded from Chateau and Fierce, with 2.1 to 2.4 plants/plot surviving out of 3 planted at both evaluations. Living nontreated plugs numbered 2.9/plot at both ratings. Cane length was reduced by Chateau at both rates, Fierce, and Matrix in both July and September (Table 2).

Cultivars differed significantly in their response to herbicide treatments, but not to specific treatments. This may indicate that cultivars were more sensitive to the herbicides, or were differentially injured by transplanting operations from greenhouse flats to the field. In either case, initial injury (May) was greatest with ‘Cascade Harvest’ and ‘Wakefield’, with 31% chlorosis/leaf death displayed (Table 3). By June, ‘Wakefield’ plants showed 52% injury, similar to the 48% seen with ‘Cascade Harvest’, contrasted with 41% injury with ‘Squamish’ and 20% injury with ‘Meeker’. Survival was similar for all cultivars except ‘Wakefield’ (2.2 plants/plot of 3 planted). Cane growth was by July was greatest with ‘Meeker’, followed by ‘Squamish’ (36.7 and 15.8 cm/cane, respectively). Growth was rapid during the next two months, especially for ‘Meeker’ and ‘Wakefield’, with canes measuring 121.8 and 77.8 cm/cane, respectively.

A final cane number and length will be determined in December, 2016, after which plots will be mowed. Plots will be treated with glyphosate in February to control emerged weeds, and then retreated with the same herbicides prior to shoot emergence, 2017. Weed control and final growth numbers will be evaluated through summer, 2017.

Table 1. Weed control in newly-planted red raspberry after post-transplant treatment with several herbicides (2016).

Treatment ^a	Rate product/a	Weed control			
		May 23 %	Jun 17 %	Jul 26 %	Sep 12 %
Zeus	8 fl.oz	100 a	97 a-d	77 bcd	12 de
Chateau	6 oz	100 a	100 ab	95 abc	65 abc
Chateau	12 oz	100 a	100 a	95 abc	70 ab
Fierce	6 oz	100 a	100 a	98 ab	87 a
Devrinol	8 lb	98 ab	80 f	40 e	37 b-d
Prowl H2O	3 pt	99 a	91 e	60 de	22 cde
Surflan	6 qt	98 ab	95 bcd	92 abc	48 a-d
Trellis	1.5 lb	100 a	93 de	75 cd	50 a-d
Matrix	4 oz	98 ab	95 cde	83 abc	75 ab
Sandea	2 oz	98 ab	95 cde	78 a-d	67 ab
Simazine	4 lb	96 b	99 abc	100 a	50 a-d

Means within a column and followed by the same letter, or not followed by a letter, are not significantly different ($P \leq 0.05$).

^aRaspberries were transplanted May 16, 2016; herbicides were applied May 18, 2016.

Table 2. Raspberry plant injury, plant survival, and cane length in newly-planted red raspberry after post-transplant treatment with several herbicides (2016).

Treatment ^a	Rate product/a	Crop injury		Survival		Cane length	
		May 23	Jun 17	Jul 26	Sep 12	Jul 26	Sep 12
		%	%	%	%	cm	cm
Zeus	8 fl.oz	29 abc	36 bc	2.8 abc	2.7 abc	22.1 ab	78.8 abc
Chateau	6 oz	34 ab	62 a	2.1 d	2.1 d	12.3 d	65.2 cd
Chateau	12 oz	35 a	69 a	2.3 cd	2.3 cd	11.6 d	61.0 d
Fierce	6 oz	34 ab	58 a	2.4 bcd	2.4 bcd	13.5 cd	59.0 d
Devrinol	8 lb	22 cd	27 bc	3.0 a	3.0 a	26.0 a	81.8 abc
Prowl H2O	3 pt	21 cd	35 bc	2.8 abc	2.8 abc	22.4 ab	84.8 ab
Surflan	6 qt	26 bc	32 bc	2.9 ab	2.9 ab	21.4 ab	89.5 ab
Trellis	1.5 lb	25 bcd	30 bc	3.0 a	3.0 a	23.0 ab	89.7 ab
Matrix	4 oz	20 cd	37 bc	2.9 ab	2.9 ab	17.6 bcd	75.2 bcd
Sandea	2 oz	23 cd	38 b	3.0 a	3.0 a	22.3 ab	87.0 ab
Simazine	4 lb	17 d	32 bc	2.8 abc	2.8 abc	19.8 abc	84.1 ab
Nontreated	---	26 bcd	27 c	2.9 ab	2.9 ab	26.2 a	95.9 a

Means within a column and followed by the same letter, or not followed by a letter, are not significantly different ($P \leq 0.05$).

^aRaspberries were transplanted May 16, 2016; herbicides were applied May 18, 2016. Table 3. Raspberry cultivar injury, plant survival, and cane length in newly-planted red raspberry after post-transplant treatment with several herbicides (2016).

Table 3. Raspberry cultivar injury, plant survival, and cane length in newly-planted red raspberry after post-transplant treatment with several herbicides (2016).

Treatment ^a	Crop injury ^b		Survival ^b		Cane length ^b	
	May 23	Jun 17	Jul 26	Sep 12	Jul 26	Sep 12
	%	%	%	%	cm	cm
'Cascade Harvest'	31 a	48 a	2.9 a	2.9 a	12.0 c	55.7 d
'Meeker'	22 b	20 c	3.0 a	3.0 a	36.7 a	121.8 a
'Squamish'	20 b	41 b	2.8 a	2.8 a	15.8 b	64.2 c
'Wakefield'	31 a	52 a	2.2 b	2.2 b	14.8 bc	77.8 b

Means within a column and followed by the same letter, or not followed by a letter, are not significantly different ($P \leq 0.05$).

^aRaspberries were transplanted May 16, 2016; herbicides were applied May 18, 2016.

^bNontreated raspberry injury was rated at 26 and 27% on May 23 and Jun 17, respectively; nontreated raspberry survival was 2.9 and 2.9 plants/plot, respectively (3 plants were transplanted in each plot); and nontreated raspberry cane length was 26.2 and 95.9 cm on July 26 and September 12, respectively.

Project Number: 13C-3419-5746

Title: Effects of Caneburning on Red Raspberry Quality and Cane Carbohydrates

Personnel: Timothy W. Miller, WSU Mount Vernon NWREC
Lisa Wasko DeVetter, WSU Mount Vernon NWREC

Reporting Period: 2015-16

Accomplishments: The trial was conducted at two sites during 2015-16: one at WSU Northwestern Washington Research and Extension Center (NWREC) in Mount Vernon and the second at WSU Irrigated Agriculture Research and Extension Center (IAREC) in Prosser. Data from both sites are reported here and were presented at the Northwestern Washington Small Fruit Conference in Lynden in December, 2016.

Results:

NWREC Trial. The trial was conducted on five-year-old ‘Cascade Bounty’, ‘Chemainus’, ‘Meeker’, and ‘Saanich’ red raspberry. The four cultivars were planted in 30-ft sections with cultivar order randomly assigned to each of 6 rows. Row spacing was 10 ft. Plots consisted of 15-ft sections of each cultivar with herbicide treatments randomly assigned to all four cultivars. Herbicides tested were Aim (carfentrazone), Goal (oxyfluorfen), and Treevix (saflufenacil) applied April 5, 2016 (when the first primocanes were at 4 to 6 inches tall) as a directed spray to 1.5 ft of the bed on either side of the row using a CO₂-pressurized backpack sprayer. Herbicides and rates are provided in appropriate tables. One average primocane and floricanes were collected from each plot June 13, 2015 when the first berries were just beginning to turn red. Canes were then dried at 75 C for three days and weights recorded. Fruiting laterals were separated from the clipped floricanes prior to drying and were dried separately. Dried primocanes and fruiting laterals were finely ground using a spice grinder and are being analyzed for carbohydrate (structural and nonstructural) content by Dr. John Fellman’s lab in Pullman. Ripe berries were sampled from each plot June 15, 2016 and frozen immediately after collection. Samples consisted of 50 berries collected on the east and west side of plants in each plot. Frozen fruit is being processed and analyzed for pH, titratable acidity, °Brix, and anthocyanin content by Dr. Joan Davenport’s lab in Prosser. The statistical design of this trial was a randomized complete block with three replicates. There was not an interaction between cultivar and herbicide treatment, so data were pooled prior to analysis. Means were separated using Tukey’s Honestly Significant Difference statistic ($P \leq 0.05$).

Berry size was not affected by herbicide treatment (Table 1). Fruit size ranged from 3.3 to 3.4 g/berry when analyzed across cultivars. There was a trend toward larger fruit following caneburning applications when compared to nontreated raspberry. Fruit size did, however, differ among the four cultivars tested (Table 2). ‘Saanich’ and ‘Chemainus’ berries were up to 28% larger than ‘Meeker’ or ‘Cascade Bounty’ fruit.

Cane dry weight did not significantly differ among the herbicide treatments (Table 1). Primocane biomass tended to be greater after treatment with Aim or Treevix compared to

nontreated raspberry, while treatment with Goal generally resulted in less primocane biomass. Nontreated floricanes tended to be the heaviest, while Treevix tended to reduce floricanes biomass. Fruiting laterals tended to be greatest with Goal and least with Treevix. Because these differences were not statistically significant, however, no definitive conclusions should be taken from these data. Primocane and floricanes biomass also did not differ by cultivar (Table 2), although ‘Cascade Bounty’ tended to produce the lightest primocanes while ‘Meeker’ floricanes tended to be the lowest in biomass. But again, these distinctions are mainly speculative. Fruiting laterals, however, were significantly greater in ‘Saanich’ than in ‘Cascade Bounty’ or ‘Chemainus’, while ‘Meeker’ produced heavier laterals than did ‘Chemainus’.

IAREC Trial. The trial was conducted on third-year ‘Chemainus’, ‘Meeker’, and ‘Wakefield’ red raspberry. Plots consisted of 5 plants of each cultivar planted within a 15-ft section of row; cultivar order was randomly assigned within the row, with 10 ft between each cultivar. Row spacing was 10 ft. Herbicides tested were Aim (carfentrazone) applied once or twice and Treevix (saflufenacil) applied once. Products were applied April 4 (when the first primocanes were at 4 to 6 inches tall) and May 3, 2016 as a directed spray to 1.5 ft of the bed on either side of the row using a CO₂-pressurized backpack sprayer. Herbicides and rates are provided in appropriate tables. One average primocane and floricanes were collected from each plot June 10, 2016 when the first berries were just beginning to turn red. Canes were then dried at 75 C for three days and weights recorded. Fruiting laterals were separated from the clipped floricanes prior to drying and were dried separately. Dried primocanes and fruiting laterals were finely ground using a spice grinder and are being analyzed for carbohydrate (structural and nonstructural) content by Dr. John Fellman’s lab in Pullman. Berries were picked weekly when ripe and frozen immediately after collection. Frozen fruit is being processed and analyzed for pH, titratable acidity, °Brix, and anthocyanin content by Dr. Joan Davenport’s lab in Prosser. The statistical design of this trial was a randomized complete block with four replicates. Means were separated using Tukey’s Honestly Significant Difference statistic ($P \leq 0.05$).

Cane dry weight did not significantly differ among the herbicide treatments (Table 3). Primocane and floricanes biomass tended to be less in herbicide-treated raspberry than in nontreated raspberry. Treatment with Treevix generally resulted in more floricanes biomass and less primocane and fruiting lateral biomass than what was sampled after treatment with other herbicides or in nontreated raspberry. There was generally only a slight response of these raspberries between Aim applied once or twice. But because these differences were not statistically significant, no definitive conclusions should be taken from these data. Cane biomass did differ among the tested cultivars (Table 4), with ‘Wakefield’ producing the greatest primocane and fruiting lateral biomass. Floricanes of ‘Wakefield’ also tended to be the largest, although this difference was not significant. ‘Chemainus’ produced more primocane biomass than ‘Meeker’, although these differences were not statistically significant.

Combined data from the two trials. The two datasets had a few points of intersection. Both included the raspberry cultivars ‘Chemainus’ and ‘Meeker’, and both had applications of the cane burning herbicides Aim and Treevix. When these selected data were pooled for the two sites, cane biomass was significantly affected by the trial location (Table 5). Raspberry floricanes and fruiting lateral biomass was less at IAREC than at NWREC, while primocane biomass was greater at IAREC. There were no clear trends between ‘Chemainus’ and ‘Meeker’ primocane

biomass, but ‘Chemainus’ floricanes tended to be heavier than ‘Meeker’ floricanes. While treatment with Treevix generally resulted in lower biomass of all cane types, this difference was not statistically significant. It will be interesting to see if fruit quality or cane carbohydrate content differs by herbicide treatment, cultivar, or trial location. These data should become available later this winter.

Data Tables:

Table 1. Raspberry fruit size and cane dry weight after different cane burning herbicide applications (WSU NWREC, 2016).

Treatment ^a	Rate product/a	Berry size ^b g/berry	Cane dry weight ^c		
			Primocane g/cane	Floricanes g/cane	Fruiting laterals g/cane
Goal	2 pt	3.4	19.9	79.5	69.1
Aim	6.4 fl.oz	3.4	26.3	80.0	65.3
Treevix	1 oz	3.4	25.2	70.7	51.6
Nontreated	---	3.3	24.3	85.5	56.1

Means within a column and followed by the same letter, or not followed by a letter, are not significantly different ($P \leq 0.05$).

^aHerbicides applied April 5, 2016, when first primocanes were 4 to 6 inches tall.

^bFifty fully ripe fruit were systematically collected by hand in each plot (25 on the east side of the row and 25 on the west side of the row) on June 15, 2016.

^cOne average primocane and floricanes were collected per plot for analysis June 13, 2016, when first berries were just turning red.

Table 2. Raspberry fruit size and cane dry weight among different raspberry cultivars (WSU NWREC, 2016).

Cultivar	Berry size ^a g/berry	Cane dry weight ^b		
		Primocane g/cane	Floricanes g/cane	Fruiting laterals g/cane
Cascade Bounty	2.8 b	21.2	83.3	46.9 bc
Chemainus	3.6 a	24.3	79.7	35.7 c
Meeker	3.2 b	24.8	68.7	72.2 ab
Saanich	3.9 a	25.4	84.0	87.3 a

Means within a column and followed by the same letter, or not followed by a letter, are not significantly different ($P \leq 0.05$).

^aFifty fully ripe fruit were systematically collected by hand in each plot (25 on the east side of the row and 25 on the west side of the row) on June 15, 2016.

^bOne average primocane and floricanes per plot were collected for analysis June 13, 2016, when first berries were just turning red.

Table 3. Raspberry cane and lateral weights after different cane burning herbicide applications (WSU IAREC, 2016).

Treatment ^a	Rate	Primocane ^b	Floricanes ^b	Fruiting laterals ^b
	product/a	g/cane	g/cane	g/cane
Treevix	1 oz	29.3	38.3	39.8
Aim once	6.4 fl.oz	31.6	34.7	47.6
Aim twice	6.4 fl.oz fb 6.4 fl.oz	36.6	36.8	46.7
Nontreated	---	39.5	47.9	45.9

Means within a column and followed by the same letter, or not followed by a letter, are not significantly different ($P \leq 0.05$).

^aHerbicides applied April 4 and May 3, 2016 (first primocanes 4 to 6 inches tall); “fb” = followed by.

^bOne average primocane and floricanes were collected per plot for analysis June 10, 2016, when first berries were just turning red.

Table 4. Raspberry cane and lateral weights for three raspberry cultivars (WSU IAREC, 2016).

Cultivar	Primocane ^a	Floricanes ^a	Fruiting laterals ^a
	g/cane	g/cane	g/cane
Chemainus	32.4 ab	38.1	29.6 b
Meeker	30.9 b	36.4	28.6 b
Wakefield	39.4 a	43.8	76.9 a

Means within a column and followed by the same letter, or not followed by a letter, are not significantly different ($P \leq 0.05$).

^aOne average primocane and floricanes were collected per plot for analysis June 10, 2016, when first berries were just turning red.

Table 5. Raspberry cane and lateral weights for the main effects: two raspberry cultivars, three herbicide treatments, and two locations (2016).

Parameter	Primocane ^b	Floricanes ^b	Fruiting laterals ^b
	g/cane	g/cane	g/cane
Cultivar, Chemainus	28.9	55.9	32.2
Cultivar, Meeker	28.3	50.2	47.3
Location, IAREC	34.2 a	39.4 b	45.0 b
Location, NWREC	23.9 b	78.9 a	60.5 a
Treatment ^a , Aim	28.9	57.3	56.4
Treatment ^a , Treevix	27.8	54.5	45.7
Treatment ^a , Nontreated	31.9	66.7	51.0

Means within a column for a given parameter and followed by the same letter, or not followed by a letter, are not significantly different ($P \leq 0.05$).

^aHerbicides applied at NWREC April 5, 2016 and at IAREC April 4 and May 3, 2016 (first primocanes at both locations were 4 to 6 inches tall at time of application).

^bOne average primocane and floricanes were collected per plot for analysis at IAREC June 10, 2016 and at NWREC June 13, 2016, when first berries were just turning red.

2017 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

New Project Proposal

Proposed Duration: (1 year)

Project Title: Determining whether raspberry plants should be caneburned

PI: Timothy W. Miller

Organization: Washington State University

Title: Extension Weed Scientist

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Address: 16650 State Route 536

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Cooperators: These trials will be conducted on grower fields near Lynden, WA

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

Total Project Request: **Year 1** \$3,221 **Year 2** \$0 **Year 3** \$0

Other funding sources:

Agency Name: British Columbia Raspberry Industry Development Council

Amt. Requested: \$3,000

Notes: Additional support for this project includes the herbicides, which are generally provided by the manufacturer.

Description:

Caneburning is the practice of removal of the first flush of primocanes of established raspberry plants in the spring using a postemergence herbicide in effort to increase yield. Approximately 95% of raspberry growers conduct caneburning annually on at least some of their fields, so the practice is almost universally used throughout the PNW region. It is known, however, that caneburning nonvigorous raspberry plants can result in decline or death of those plants, depending on the degree of plant health at the time of application. Such compromised plants normally consist of only a few overwintering floricanes, and it may be possible to gauge their ability to respond positively to caneburning herbicide application based on floricanes counts. Therefore, the objective of this proposed research is to experimentally group raspberry plants by their floricanes number and correlate their growth and yield to applications of various caneburning herbicides. This should provide valuable information to raspberry producers deciding whether to caneburn their raspberries.

Justification and Background:

Two excellent herbicides are currently available for caneburning operations: Goal (oxyfluorfen) and Aim (carfentrazone). Two other products are good candidates for future registration: Treevix (saflufenacil) and Rely (glufosinate). These may also be augmented with Gramoxone

(paraquat) in mixture, and may also be used in combination or sequential with residual herbicides. All quickly remove foliage from primocanes less than about 6 inches in height, and usually result in the complete removal of treated canes (a desirable outcome). It is known that Goal delays primocane regrowth longer than does Aim, while Treevix and Rely will fall between the two extremes.

While caneburning effects on healthy raspberry plants are fairly well understood, there remains much uncertainty as to whether to apply caneburning herbicides to raspberries of lower vigor. Raspberry plants not displaying vigorous growth can be severely injured by removal of the first flush of primocanes, which is the objective of caneburning treatments. What is needed is an evaluation of raspberry plant vigor, from which the expected response from the application of various caneburning herbicides can be estimated. Low vigor plants may be those displaying symptoms of Phytophthora root rot or viruses, presence of perennial weeds such as horsetail (*Equisetum* spp.), quackgrass (*Elymus repens*), or Canada thistle (*Cirsium arvense*), or simply plants in an older raspberry block. Such nonvigorous raspberry plants normally consist of only a few overwintering floricanes, which can easily be counted following dormant-season pruning and training. It may be possible to gauge the ability of these plants to respond positively to caneburning herbicide application based on floricanes counts in winter. Therefore, the objective of this proposed research is to experimentally group raspberry plants by floricanes number and correlate their growth and yield to applications of various caneburning herbicides. The ultimate goal of this research is to produce guidelines for growers to use when deciding whether to caneburn their raspberries.

Relationship to WRRRC Research Priority(s): #3 Priority, Cane Management (including suppression). I am unaware of any other raspberry herbicide projects currently being conducted in Oregon, Idaho, or British Columbia.

Objectives: To determine the ability of nonvigorous raspberry plants to respond positively to application of caneburning herbicides through the use of floricanes counts.

Procedures:

Plots will be established in January-March, 2017 in grower fields at three locations near Lynden, Washington (at least two raspberry cultivars, if possible). Vigorous and nonvigorous raspberry plants in these fields will be identified in January and February, preferably within a single row on the edge of the raspberry block to lower the impact on grower operations. Floricanes will be counted during dormancy and 3-meter (15-foot) sections assigned to one of four groups based on floricanes/meter (3 feet) of row. These groups will be constructed after actual cane counts are obtained from the field, but groups may be something like 1 to 4 floricanes/m, 5 to 9 floricanes/m, 10 to 15 floricanes/m, and 15 or more floricanes/m. These sections will be marked prior to application of caneburning herbicides.

Caneburning will be accomplished when the first-emerging primocanes are 4 to 6 inches tall (late March, early April). Products to be tested in each of the floricanes-groups will be Goal and Aim, applied at two rates each; other plots in each floricanes-group will not be caneburned. All plots will also receive treatment with a residual herbicide, including noncaneburned plots. Weed

control and crop injury will be evaluated periodically through the growing season, with primocane growth noted in particular. Plots will be sampled for berry production to determine caneburning effects on berry yield and fruit size. At the end of the growing season, raspberry primocanes will be counted.

Anticipated Benefits and Information Transfer:

If positive, data from this experiment will be used in a decision model for growers to use when determining whether to apply the caneburning herbicides Goal or Aim. Additional years of testing may be necessary for model construction and validation. 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:

	2017	2017	2018
Salaries¹	\$ 1,000	\$ 0	\$ 0
Time-Slip	\$ 750	\$ 0	\$ 0
Operations (goods & services)	\$ 250	\$ 0	\$ 0
Travel²	\$ 250	\$ 0	\$ 0
Meetings	\$ 0	\$ 0	\$ 0
Other	\$ 0	\$ 0	\$ 0
Equipment	\$ 0	\$ 0	\$ 0
Benefits³	\$ 971	\$ 0	\$ 0
Total	\$ 3,221	\$ 0	\$ 0

Budget Details

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

²Travel is for plot establishment, maintenance, and data collection.

³Benefits (39.85% for A/P scientific assistant, \$399; 76.3% for time-slip help, \$572; total \$971).

PHYSIOLOGY

**2017 WASHINGTON RED RASPBERRY COMMISSION
RESEARCH PROPOSAL**

New Project Proposal

Proposed Duration: 3 years

Project Title: Application of Biodegradable Mulches in Tissue Culture Red Raspberry: Impacts on Weed Control, Parasitic Nematodes, and Crop Growth

PI: Lisa W. DeVetter

Organization: WSU NWREC

Title: Assistant Professor, Small Fruit Horticulture

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Co – PIs:

- Carol Miles, Professor of Vegetable Horticulture, WSU-NWREC, 16650 State Route 536, Mount Vernon, WA 98273, phone: 360-848-6150, milesc@wsu.edu
- Shuresh Ghimire, PhD Student in Vegetable Horticulture, WSU-NWREC, 16650 State Route 536, Mount Vernon, WA 98273, phone: 360-848-6136, shuresh.ghimire@wsu.edu
- Chris Benedict, Extension Educator, WSU Extension Whatcom County, 1000 N. Forest St. Ste. 201, Bellingham, WA 98225, phone: 360-676-673, chrisbenedict@wsu.edu
- Inga Zasada, USDA-ARS Plant Pathologist, 3420 NW Orchard Avenue, Corvallis, OR 97330, phone: 541-738-4051, Inga.Zasada@ars.usda.gov

Cooperators: Enfield Farms has indicated they would be willing to serve as a cooperating farm if this project is funded.

Year Initiated: 2017

Current Year: 2017

Terminating Year: 2019

Total Project Request: \$68,690 **Year 1:** \$10,457 **Year 2:** \$27,825 **Year 3:** \$28,408

Other funding sources: Yes, pending

Agency: Washington State Commission of Pesticide Registration (WSCPR) and Western Sustainable Agriculture Research & Extension (WSARE)

Amount Requested: WSCPR: \$17,312 (for Year 1); WSARE: \$49,133 (for Years 1-3)

Notes: We have applied for matching funds to WSCPR and for graduate student support to WSARE. If successful with WSARE, this will reduce overall project costs for the WRRC, which is high due to graduate student salary.

Description:

Washington State leads national production of processed red raspberries, producing ~73.3 million pounds with a value of \$89 million in 2015 (NASS, 2016). Weed management, especially during establishment, has become a critical issue for growers and is a greater challenge for delicate tissue culture (TC) plugs, which have become increasingly popular within

the industry due to cultivars exclusively produced through TC. Black polyethylene (PE) mulch is used to control weeds in many annual crops, but the difficulty of removal leaves it with limited practical application in perennial crop production. Biodegradable plastic mulches (BDMs) may be a viable alternative for weed control and may also increase plant growth and development. However, BDMs have the potential to increase soil temperatures and subsequent activity of parasitic nematodes, a major pest of raspberry. We propose to evaluate four different BDMs relative to PE and bare-soil (herbicide) controls in on-farm experiments established in Lynden, Washington. We will study effects of BDMs on weed control, nematode populations, and growth/establishment of TC raspberry in both spring and fall planted systems. Additionally, we will monitor mulch intactness and degradation in order to evaluate the suitability of these agricultural tools in red raspberry production in Northwest Washington.

Justification and Background:

Tissue culture (TC) plugs have become increasingly popular within the red raspberry industry. This is largely due to increased plantings of the new cultivar ‘Wakefield’, which is exclusively produced through TC. Other commercial propagators are increasingly moving towards TC production due to the ability to rapidly propagate plant material in smaller spaces and the ability to reduce pathogens and viruses through aseptic techniques. However, plugs resulting from TC are more delicate and difficult to establish, making weed management using herbicides more challenging.

Polyethylene (PE) mulch is widely used to control weeds in annual crops, but is not extensively used in perennial cropping systems. Gerbrandt (2015) found improved growth and establishment of TC raspberry under plasticulture in British Columbia. Yet, PE mulch removal and disposal can be both difficult and costly. Biodegradable plastic mulch (BDM) could be a suitable alternative for weed control in TC plantings if the BDM controls weeds and biodegrades into the soil, thereby avoiding removal, disposal, and soil ecological issues. There has been limited research testing the efficacy of BDMs in raspberry. Król-Dyrek and Siwek (2015) compared three mulches [polypropylene (PLP; non-biodegradable), photo-degradable PP, and poly lactic acid (PLA; biodegradable)] to bare soil cultivation in Poland and found raspberry yield was greater for the three mulch treatments as compared to bare ground. A non-replicated trial in 2015 in NW Washington compared non-mulched TC raspberry to plants grown with a BDM. Ten months after transplanting, plants grown with a BDM were ~25.4 cm taller and produced 7 more canes, demonstrating a potential for increased yields (DeVetter, unpublished).

Plant-parasitic nematodes are another major pest of raspberry, particularly root lesion nematode (*Pratylenchus penetrans*; RLN). Gerbrandt (2015) reported increased nematode populations in a PE mulched raspberry plot, indicating mulches may encourage nematode activity. No studies to our knowledge have explored the impacts of mulches, including BDMs, on RLN activity in raspberry.

This project will address the problem of poor plant establishment and weed management in raspberry systems that use TC plugs at planting, while also evaluating the impact on RLN. We propose to address this problem by studying the impacts of PE mulch and BDMs in both spring and fall planted systems and measuring how they impact weed management, populations of

RLN, plant growth, and crop productivity. Overall, this project will contribute to discovering new labor-saving techniques to improve establishment of TC raspberry in NW Washington.

Relationship to WRRRC Research Priorities:

This project is related to priority #1, labor saving practices, as mulches have the potential to reduce labor associated with weeding. Additionally, this project is related to priority #3, weed management, as mulches are a physical form of weed control.

Objectives:

1. Evaluate weed incidence with BDMs in comparison to bare ground (standard, herbicide treated control) and PE mulch (control) in establishing TC red raspberry in both spring and fall planted systems in NW Washington.
2. Monitor surface and in-soil degradation of BDMs in spring and fall planted raspberry systems.
3. Assess populations of RLNs in the soils and roots of raspberry before and after using biodegradable and PE mulches; populations will also be compared across treatments.
4. Evaluate growth and establishment of raspberry grown with BDM and PE mulch in comparison with bare ground.

All objectives will be addressed within the first year of the project (2017). In 2018, we will collect yield data from the spring planted trial only (the fall planted trial will not yield until 2019) and continue measuring mulch biodegradation, populations of RLNs, and plant growth in both the spring and fall planted trials.

Procedures:

The experiment will be carried out in two separate field trials established on commercial farms with grower cooperators in Lynden, WA. The spring planted trial will be established in March 2017 and the fall planted trial will be established in Aug. 2017. Experimental design will be a randomized complete block, with each plot being one row length (~700 ft). Six treatments will be applied per site, replicated five times, and will include four commercially available BDMs [NatuREcycle 1, NatuREcycle 2, Organix A.G. Film (0.5 mil), and Organix A.G. Film (0.6 mil)], a non-degradable PE mulch, and a bare ground control. Investigators will assist with machine laying, hole punching, and planting (DeVetter's mulch layer can be used). TC plugs of 'Wakefield' will be planted after mulch application. Six and 12-18 months after mulch application for spring and fall planted trials, respectively, PE will be removed by the investigators, whereas all BDMs will be covered with 2-3 in. thickness of soil from the alleyways to initiate mulch degradation.

The following data will be collected:

- Annual cumulative plant growth - measured monthly from 10 plants per plot.
- Percent soil exposure (PSE) - recorded twice per month after treatment application for 6 and 18 months in spring and fall plantings, respectively, to assess the rate of above-soil mulch deterioration.
- Weed pressure – collected monthly during the 2017 and 2018 growing season; will include weed counts and biomass (fresh and dry).

- Soil temperature - recorded hourly using data loggers from planting to Nov. 2017; probes will be reinstalled March 2018 and remain in the field until Nov. 2018.
- RLN - baseline pre-plant populations will be determined in March and Aug. 2017 for spring and fall plantings, respectively. RLN will subsequently be determined from soil and root samples collected annually in Sept. 2017-2019.
- In-soil mulch degradation - prior to PE removal and BDM covering, the area of mulch from six locations per plot will be determined and re-placed in the field. These samples will be marked and re-visited 6 and 12 months after covering BDMs with soil. Mulch samples will be excavated, cleaned, weighed, and the area determined before replacement.
- Yield and fruit quality - Yield and fruit quality (berry size, °Brix, and pH) will be determined in the spring planted in 2018 and both plantings in 2019.

Anticipated Benefits and Information Transfer:

BDMs are a promising tool to enhance establishment and productivity of TC plants, control weeds, and reduce labor associated with weeding, thereby promoting on-farm efficiencies. We will present project information at annual small fruit field days and the Small Fruit Conference in Lynden, WA, in 2017-2019. Additionally, we will post project results on WSU Small Fruit Horticulture website (<http://smallfruits.wsu.edu/>) and the webpage <http://vegetables.wsu.edu/AltMulch.html>. Final results will be shared through the following mechanisms: *Whatcom Ag Monthly*, Peerbolt Small Fruit Update, WSU Extension Fact Sheet, industry trade journals, and scientific publications.

References:

Gerbrandt, E. 2015. New techniques for getting raspberries and strawberries off to a better start. Proceedings from the 2015 Lower Mainland Horticulture Improvement Association Horticulture Growers’ Short Course. Available at: <http://www.agricultureshow.net/horticulture-growers-short-course>.

Król-Dyrek, K. and P Siwek. 2015. The influence of biodegradable mulches on the yielding of autumn raspberry (*Rubus idaeus* L.). *Folia Horticulturae* 27(1): 15-20.

Budget:

	2017	2018	2019
Salaries ^{1/}	\$4,005	\$15,027	\$15,628
Timeslip ^{2/}	\$2,600	\$2,704	\$2,812
Operations (goods & services) ^{3/}	\$2,260	\$760	\$260
Travel ^{4/}	\$0	\$0	\$0
Equipment	\$0	\$0	\$0
Benefits ^{5/}	\$1,592	\$9,334	\$9,708
Total	\$10,457	\$27,825	\$28,408

¹Salary - 50% FTE for a MS student starting spring 2018 and 2019 (1 semester/year) = \$15,027 in 2018 and \$15,628 in 2019; figures include 4% annual increase; Research associate (Ed Scheenstra) for 0.5 month @ \$4,037/m = \$2,019 in 2017 only; Scientific assistant (Sean Watkinson) for 0.5 month @ 3,971/m = \$1,986 for 2017 only.

²Partial timeslip support for summer graduate student in 2017-2019: \$10/hour x 40 hours/week x 6.5 weeks = \$2,600; figures include 4% annual increase.

³Partial support for supplies in 2017: soil temperature loggers = \$100/probe x 12 = \$1,200 (Year 1 only); Shuttle for logger = \$300 (Year 1 only); \$200 for outreach publications and materials (Years 1-3); \$60 for bags, flags, and other field supplies (Years 1-3); \$500 for test mulch (Years 1 & 2).

⁵Benefits for graduate student: QTR, health, and benefits at 60.32% when salaried and 10.00% when on summer timeslip = \$9,064 + 270 = \$9,334 in 2018 and \$9,427 + \$281 = \$9,708 in 2019; Benefits for research associate and scientific assistant in 2017 at 44% and 35.44%, respectively 888

Budget Approved on 12/07/16 by Jeanne Burritt.

**2017 WASHINGTON RED RASPBERRY COMMISSION
RESEARCH PROPOSAL**

New Project Proposal

Proposed Duration: 2 years

Project Title: Impact of Nitrogen on Nematode Parasitism of Red Raspberry

PI: Lisa W. DeVetter

Organization: WSU NWREC

Title: Assistant Professor, Small Fruit Horticulture

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Address: 16650 State Route 536

City/State/Zip: Mount Vernon, WA 98273

Co-PI: Inga Zasada

Organization: USDA-ARS

Title: Research Plant Pathologist

Phone: 541-738-4051

Email: Inga.Zasada@ars.usda.gov

Address: 3420 NW Orchard Avenue

City/State/Zip: Corvallis, OR 97330

Cooperators: None. We are proposing to conduct this study using microplots at WSU NWREC.

Year Initiated: 2017

Current Year: 2017

Terminating Year: 2018

Total Project Request: \$20,730

Year 1: \$10,182

Year 2: \$10,548

Other funding sources: None

Description:

The objective of this project is to explore if different nitrogen rates during red raspberry establishment influences plant and root growth, root lesion nematode (*Pratylenchus penetrans*; RLN) populations, and subsequent damage to plants. We have observed high RLN populations in raspberry roots collected in Whatcom County on plants that display few symptoms of infestation. Many of these plantings are high input systems, leading us to wonder if nitrogen influences RLN infestations and subsequent impacts on crop growth and yield. Through this study, we will evaluate if modification of nitrogen rates during establishment impacts damage due to RLN parasitism. The end goal is to understand if nitrogen rate can be used as a post-fumigation cultural management tool in plantings with high RLN pressure or in instances where fumigation results are poor.

Justification and Background:

Root lesion nematode (*Pratylenchus penetrans*; RLN) is a migratory endoparasite that feeds on plant roots, including red raspberry (*Rubus idaeus*). RLN feeding damages roots, which reduces root functioning (water and nutrient transport), plant growth, and subsequent yields. RLN is one of the key pests in red raspberry systems in Northwest Washington. Most growers utilize pre-plant fumigation using Telone C-35[®] and/or Vapam[®] for RLN management. While in some fields RLN suppression using pre-plant fumigation has been observed to be variable to poor, there is currently a strong collaborative effort to improve pre-plant fumigation techniques. Yet, there are few tools to manage RLN in a post plant situation. This project explores how different nitrogen rates during raspberry establishment impacts RLN populations and subsequent damage to plants. We have consistently observed high RLN populations in raspberry roots collected

from plants that display few symptoms of infestation. Many of these systems are well managed and high input systems, leading us to question if plant growth as impacted by nitrogen applications will enable raspberry plants to “outgrow” high RLN populations and escape damage. This project explores this question using microplots previously established at the WSU NWREC.

Relationship to WRRRC Research Priorities:

This project is related to the following #2 priorities: 1) Soil fumigation techniques and alternatives to control soil pathogens, nematodes, and weeds; 2) Understanding soil ecology and soil borne pathogens and their effects on plant health and crop yields; 3) Nutrient management.

Objectives:

The primary objective will be to explore if different nitrogen rates during red raspberry establishment influences plant and root growth, RLN population densities, and subsequent damage to plants. We will measure these parameter during the first year of establishment in 2017. In 2018, we will continue to monitor RLN populations, plant growth, and damage, as well as yield and fruit quality.

Procedures:

The experiment will be established in field microplots located at WSU NWREC in Mount Vernon, WA. There is a total of 100 individual microplots spanning five rows that were constructed in 2010 by burying 3 ft diameter polypropylene weed mat cylinders into the soil. The experimental design will be a randomized complete block with individual treatments applied to a five-microplot section per block, replicated five times (25 microplots per treatment; 1 plant per microplot). Individual microplots will be planted with tissue culture ‘Meeker’ and inoculated with RLN (approximately 250 RLN/250 g soil) at planting to simulate a field situation with moderate RLN pressure. RLN used for inoculation will be collected from root samples harvested from field sites in Whatcom County.

Four treatments differing only in total nitrogen rates will be applied to the plants, including: 0 lbs N/acre (negative control), 30 lbs N/acre, 60 lbs N/acre, and 100 lbs N/acre. Nitrogen fertilizers will include a mixture of pre-plant and liquid fertilizers. Liquid fertilizers will be applied weekly from mid-April to mid-July. We are receptive to feedback if the review committee feels the nitrogen program should be augmented so it is more representative of commercial production in Whatcom County.

Data to be collected include:

- Populations of RLN in raspberry roots and soil; determined Sept. 2017 and April and Sept. 2018
- Soil chemistry (pH, macro- and micro-nutrients) determined Sept. 2017 and 2018
- Raspberry tissue nutrient concentrations (macro- and micro- nutrients) determined the first week of Aug. 2017 and 2018
- Cumulative plant growth measured from three tagged plants per treatment section; growth will be measured monthly from March – Nov. 2017 and 2018

- Raspberry yield and fruit quality (average berry size, °Brix, and pH); measured in 2018 only
- Visual assessments of plant health and vigor, measured once per month in June, July, and Aug. 2017 and 2018.
- Root and shoot dry weight biomass (divided by floricanes and primocanes) from one plant per treatment plot in 2017 and 2018.

Anticipated Benefits and Information Transfer:

This project will provide growers, crop consultants, and researchers with potential post-fumigation cultural management practices that can mitigate problems due to RLN. Project results will be shared at the 2018 small fruit field day and Small Fruit Conference in Lynden, WA. Additionally, project results will be posted on the WSU Small Fruit Horticulture website (<http://smallfruits.wsu.edu/>) and shared through in the WSU *Whatcom Ag Monthly* and Peerbolt Small Fruit Update in 2018. Final results will be published in a scientific journal.

References: None.

Budget:

	2017	2018
Salaries ^{1/}	\$3,971	\$4,130
Timeslip ^{2/}	\$1,440	\$1,440
Operations (goods & services) ^{3/}	\$2,590	\$2,740
Travel ^{4/}	\$630	\$630
Equipment	\$0	\$0
Benefits ^{5/}	\$1,551	\$1,608
Total	\$10,182	\$10,548

¹Salary for Scientific Assistant (Sean Watkinson) for 1 month/year; figures include 4% annual increase.

²Timeslip at for 40 hr/week x 3 weeks =120 hours @ \$12/hr = \$1,440/year.

³RLN extraction by Zasada @ \$25/sample x 20 samples/year in 2017 and 40 samples/year in 2018= \$500 in 2017 and \$1,000 in 2018; soil samples @ \$10/sample x 20 samples/year = \$200/year; tissue tests @ \$17/sample x 20 samples/year = \$340/year; Land use fees at WSU-NWREC = \$1,000/year; Tissue culture plants, 100 plants x \$2/plant = \$200 for 2017 only; Fertilizer, drip tape, emitters, etc. = \$300 in 2017 and \$150 in 2018; shipping costs = \$50/year.

⁴Travel for Zasada to visit site 1/year, 640 RT @ \$0.54/mile = \$346; per diem rate (lodging, meals, incidentals) for 2 days in Mount Vernon, WA = 142 x 2 days = \$284.

⁵Benefits for Scientific Assistant (Sean Watkinson) at 35.44%; Timeslip at 10%.

Budget Approved on 12/7/16 by Jeanne Burritt

**Washington Red Raspberry Commission
Progress Report for 2015 – 2016 Projects**

Project number: 3455-6640

Title: Comparison of Alternate- and Every-Year Production in Summer-Bearing Red Raspberry

Personnel:

PI: Lisa Wasko DeVetter

Co-PI: Suzette Galinato

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

Title: Assistant Professor, Small Fruits

Title: Research Associate, Economics

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City/State/Zip: Mount Vernon/WA/98273 **City/State/Zip:** Pullman/WA/99164

Cooperators/Co-PI: Jonathan Maberry, Maberry Packing LLC

Reporting Period: This report presents data from 2016, one year after the project was initiated.

Accomplishments:

- Treatments were established in Mr. Jon Maberry's field in Lynden, WA in 2015 (please see the proposal for more details); 2016 was an "off production year" for the AY treatment, so only yield from EY treatments were collected.
- A focus-group led by Dr. DeVetter and Mrs. Galinato was held in Lynden, WA, in Sept. 2015. Red raspberry growers and crop consultants attended the meeting and provided cost of production data for the revision of the red raspberry enterprise budget initially published by MacConnell and Kangiser in 2007. A revision of this enterprise budget was published in 2016. These data will be used as a benchmark for the alternate-year production study.
- Data collected in 2016 include: primocane height, primocane number, primocane diameter, estimated yield (EY plots only), berry size (EY plots only), and foliar leaf nutrients (tissue collected July 21, 2016).

Results:

- Plots were harvested 19 times between June 11 and July 19, 2016. Estimated and actual yields and berry weight were compared across blocks, showing no difference due to block.
- There were no treatment effects for primocane number, height, nor diameter, although there was numerically more primocanes in the EY treatment (17.8 versus 15 primocanes/hill in EY versus AY treatments, respectively; P -value = 0.06)
- Tissue nutrient concentrations were not different across treatments except for iron and aluminum, with concentrations being greater and above sufficiency for iron in EY treated plots (P -values = 0.02 and 0.01 for iron and aluminum, respectively).

Publications:

Galinato, S.P. and L.W. DeVetter. 2016. 2015 Cost Estimates for Establishing and Producing Red Raspberries in Washington State. WSU Enterprise Budget. TB21.

<<http://pubs.wpdev.cahnrs.wsu.edu/pubs/tb21/>>.

Acknowledgements: Many thanks to the WRRRC and red raspberry grower cooperators. We look forward to continuing this project. We'd be happy to provide additional data upon request.

2017 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

Project number: 3455-6640

Proposed Duration: 6 years

Project Title: Comparison of Alternate- and Every-Year Production in Summer-Bearing Red Raspberry

PI: Lisa Wasko DeVetter

Co-PI: Suzette Galinato

Organization: Washington State University

Title: Assistant Professor, Small Fruits

Title: Research Associate, Economics

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City/State/Zip: Pullman/WA/99164

Cooperators/Co-PI: Jonathan Maberry, Maberry Packing LLC

Year Initiated 2015 **Current Year** 2016 **Terminating Year** 2020

Total Project Request: **Year 1** \$8,958 **Year 2** \$8,277 **Year 3** \$6,635 **Year 4** \$6,848
Year 5 \$9,050 **Year 6** \$16,030

Other funding sources: *None at this time.*

Description:

Increasing costs and decreasing availability of labor are compromising the economic viability of commercial red raspberry production in western Washington. The grower community is in need of alternative production systems that minimize labor needs, maintain productivity, and are economically viable. This project addresses that need by evaluating the economic viability of alternate-year production relative to traditional every-year production systems. Specific sub-objectives of this projects are to: 1) Evaluate differences in plant productivity and yield between alternate- and every-year production systems; and 2) Complete a benefit-cost analysis to assess the on-farm net benefits of alternate-year production relative to traditional every-year production systems. Results of this project will be disseminated at conferences, field days, and through a Washington State University extension publication. Overall, this long-term project will provide valuable information regarding potential labor savings and the economic feasibility of this alternative system of red raspberry production.

Justification and Background:

The increasing cost of labor has become prohibitive for many growers of horticultural crops, including red raspberry (*Rubus idaeus*). Summer-bearing raspberry is particularly labor intensive, with annual pruning and tying of canes representing approximately 10% of total annual costs during established bearing years (personal communication with grower). Access to labor is also extremely challenging for growers. These issues demonstrate a need to investigate alternative production systems that reduce growers' dependency on labor. Alternate-year

production, which entails removal of spent floricanes and producing fruit on an every-other-year cropping cycle, represents one potential system that reduces labor associated with pruning and tying.

In Oregon, alternate-year production is practiced in 20-55% of 'Marion' blackberry fields (Strik, 1996). Average two-year yields are reduced by 10-30% relative to every-year production, but several advantages contribute to its adoption (Bullock, 1963; Martin and Nelson, 1979). Decreased labor costs, primary due to reduced pruning and training needs, as well as reduced pesticide usage and improved cold hardiness, are several of the advantages that contribute to the persistence of alternate-year production in blackberry (Bell et al., 1992). Minimal research on alternate-year production systems have been completed in red raspberry. Furthermore, no published research has been conducted in Whatcom County, which contributes approximately 93% of total production in Washington State (WRRRC, 2014). In a six-year study performed in Vancouver, Washington, with 'Meeker' and 'Willamette', investigators found yield was reduced by 60% in an alternate-year system (Barney and Miles, 2007). However, it was not articulated if primocane suppression occurred during the course of the study, which can impact yield potential. Studies in New York have found yield reductions of only 30% over the long-term and these reductions can be partially offset through suppression of the first flushes of primocanes during fruiting years (Pritts, 2009).

Despite potential yield reductions, these systems may be economically viable given the current scenario of high labor costs and reduced labor availability. The increasing problems related to costs and availability of labor need to be addressed and this project proposes to address this need by systematically evaluating the costs, potential savings, and yield of summer-bearing raspberries produced using an alternate-year production system.

Relationship to WRRRC Research Priority(s):

This project directly addresses the priority labor saving cultural practices, including A/Y systems and mechanical pruning.

Objectives:

The overall objective of this project is to evaluate the economic viability of alternate-year production for summer-bearing red raspberries growers in western Washington. Specific sub-objectives include: 1) Evaluate differences in plant productivity and yield between alternate- and every-year production systems; and 2) Complete a benefit-cost analysis to assess the on-farm net benefits of alternate-year production relative to traditional every-year production systems.

Procedures:

Treatment plots of 'Meeker' red raspberry were established in the spring of 2015 with Mr. Jon Maberry in Whatcom County, Washington. The experimental design is a randomized complete block, with two treatments (alternate- and every-year production) replicated three times. Experimental units will be two rows randomized within a block (**Fig. 1**). In 2015, primocanes were suppressed in the alternate-year treatment plots and an initial crop was harvested from all treatments. In Winter 2015/2016, all canes will be removed from the alternate-year treatment plots, while the every-year treatment plots will be pruned and caned per industry standard. Fruit production in alternate-year treatment plots will be prevented in 2016, but primocanes will be

grown for fruit production in 2017. Fall mowing of spent floricanes in the alternate-year treatment plots will be repeated in 2017 and 2019, preceded by three-to-four spring applications of primocane suppressive herbicides during bearing years. Every-year treatment plots will be managed according to commercial standards throughout the duration of the project, which will entail annual pruning and tying.

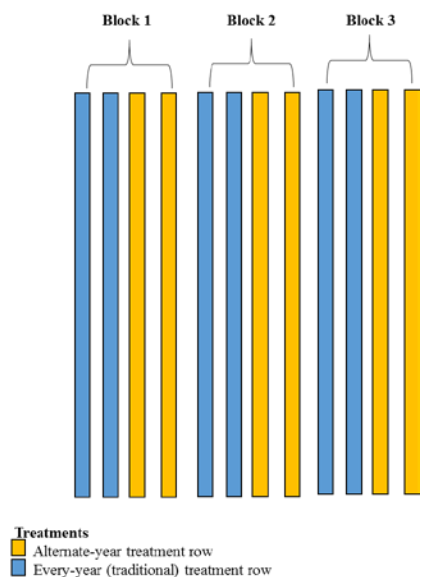


Figure 1. Experimental design comparing alternate- and every-year production systems in summer-bearing red raspberry. Two rows per experimental unit within a block are required for equipment operation.

Data collection began in 2015, in which a baseline enterprise budget was developed through a focus group with growers. These data will be used to update the raspberry production cost study completed by MacConnell and Kangiser (2007). This budget will be used as benchmark for assessing and estimating changes in net profit due to alternate-year production. Supplementary information, such as differences in number of pesticide and fertilizer applications between the two treatments, labor requirements, as well as yield and productivity, will be incorporated in these budgets. Plant growth and productivity will be measured from ten plants randomly selected within each treatment plot. Cane numbers and height will be measured to assess establishment and growth between the two treatments. Yield (both estimated and actual) and average berry size will also be determined in order to assess how the treatments impact fruit production. Foliar tissue samples will also be collected to evaluate treatment impact on plant nutrient status. Overall, these data will be utilized to evaluate the economic viability of alternate year-production, as well as impacts on plant growth and yields.

Given the proposed objectives, this will be a long-term project that will collect harvest data from alternate-year treatment plots for three cropping seasons. This translates into a six-year project, with alternate-year production occurring in 2015, 2017, and 2019, and years of strictly primocane production in 2016, 2018, and 2020. Six years of data collection is warranted to study the impacts of these treatments on a perennial plant like raspberry. A table describing the timeline of the project is provided in **Table 1** (revised from 2014 because we collected production data in 2015).

Table 1. Timeline of crop production for project comparing alternate- and every-year production of red raspberry.

Treatments	2015	2016	2017	2018	2019	2020
Alternate-year production	Initial harvest (First crop)	No crop	Second crop	No crop	Third crop	No crop
Every-year production	Initial harvest (First crop)	Second crop	Third crop	Fourth crop	Fifth crop	Sixth crop

Anticipated Benefits and Information Transfer:

Completion of this project will provide growers relevant information about the potential cost savings of alternate-year production relative to traditional every-year production. This project will also provide baseline information about implementation of this system in summer-bearing red raspberry grown in Washington. Both information derived from the benefit-cost analysis and evaluations of plant growth and productivity will be shared at grower conferences and through

two WSU Extension Publication (Fact Sheet and Excel Workbook). Results will also be available on the WSU Small Fruits Horticulture website (<http://smallfruits.cahnr.wsu.edu/>) and be published in a peer-reviewed research publication.

References:

Barney, D.L. and C. Miles (eds.). 2007. Commercial Red Raspberry Production in the Pacific Northwest. PNW 598.
 Bell, N., E. Nelson, B. Strik, and L. Martin. 1992. Assessment of winter injury to berry crops in Oregon, 1991. Oregon State University Agricultural Experiment Station, Special Report 902, July, 1992. 23 pp.
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 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.
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 Pritts, M., 2009. Pruning Raspberries and Blackberries. New York Berry News. 8(4): 7 pp. Accessed 5 Nov. 2014 at: < <http://www.fruit.cornell.edu/berry/production/pdfs/rasppruning.pdf>>.
 Strik, B. 1996. Blackberry Production in Oregon. 11th Annual Conference of the North American Bramble Growers Association. Accessed 5 Nov. 2014 at: < <http://berrygrape.org/blackberry-production-in-oregon/>>.
 Washington Red Raspberry Commission (WRRC). 2014. 2014 Pacific Northwest Raspberry Assessment Report. WRRC. Accessed Dec. 5 2015 at: <<http://www.red-raspberry.org/>>.

Budget and Justification:

	2017	2018	2019	2020
Salaries^{1/}	\$3,932	\$4,089	\$5,743	\$9,070
Time-Slip^{2/}	\$800	\$800	\$800	\$800
Operations (goods & services)^{3/}	\$50	\$50	\$50	\$1,050
Travel^{4/}	\$380	\$380	\$380	\$1,935
Meetings	\$	\$	\$	\$
Other	\$	\$	\$	\$
Equipment^{4/}	\$	\$	\$	\$
Benefits^{5/}	\$1,473	\$1,529	\$2,077	\$3,175
Total	\$6,635	\$6,848	\$9,050	\$16,030

^{1/} Research Associate (co-PI Mrs. Suzette Galinato) at the WSU School of Economic Sciences [4.17% FTE in 2016 (0.5 month at \$2,648); 2.08% FTE in 2019 (0.25 month at \$1,490); and 6.25% FTE in 2020 (0.75 month at \$4,647)]; Scientific assistant in Small Fruit Horticulture program (Mr. Sean Watkinson) at 5% FTE per year from 2016 to 2020 (\$2,318 in 2016; \$3,932 in 2017; \$4,089 in 2018; \$4,253 in 2019, and \$4,423 in 2020); yearly salaries include 4% inflation.

^{2/}Timeslip in 2017-2020 for plant growth and fruit quality data collection: 40 hr/week x 2 weeks = 80 hours @ \$800.

^{3/}General office supplies (2015); incentives to participants who will help develop and review the enterprise budgets (2016); field supplies (e.g. sample bags, flagging tape, etc.) for 2016 to 2020; journal publication charge (2020).

^{4/} Research Associate will meet with growers in order to collect and validate data for the every-year raspberry enterprise budget (2015 and 2016) and the alternate-year raspberry enterprise budget (2020). Research associate will also co-present with PI key results of the study at a grower conference in 2020 (e.g., Washington Small Fruit Conference); travel for PI to commute from Mount Vernon, WA, to field site in Lynden, WA approximately three times in 2015 and eight times per year from 2016-2020 (88 mi/roundtrip at 0.54 cents/mi) .

^{5/}No equipment funding requests.

^{6/}Benefits are calculated at 32.86% of monthly salary for Research Associate (\$870 in 2016; \$490 in 2019; and \$1,527 in 2020); Benefits for Scientific Assistant is 35.44% (\$1,393 in 2017; \$1,449 in 2018; \$1,507 in 2019, and \$1,568 in 2020). Benefits for timeslip at 10%.

Budget reviewed and approved by: Jeanne Burritt 12/5/2016 .

Washington Red Raspberry Commission Progress Report for 2016 Projects

Project No:

Title: Mechanizing red raspberry pruning and cane tying

Personnel: Manoj Karkee; Joan Davenport

Reporting Period: Nov 2015 – Oct 2016

Accomplishments:

In 2016, 0.5-acre plot of raspberries established in 2013 and 2014 in Prosser, WA was managed following common commercial practices. Weed control was achieved by a combination of herbicide, mowing, and hand weeding. The plot was pruned and tied after leaf fall. Irrigation was achieved with a drip system.

In 2016, a cane tying mechanism was fabricated and evaluated in the lab and in the field. A circular gear-teeth end-effector was designed to wrap an adhesive tape around bundled canes (Fig. 1 and 2). Canes enter into the wrapper through an opening of 45 degree in the wrapper. The circular wrapper was driven by using two pinion gears connected through the belt. The pinion gear was motorized using a stepper motor. A half-scale prototype was fabricated using 3D printer. The tape tying end-effector prototype was evaluated in the field along with the bundling mechanism developed and evaluated earlier in this project. Based on the field evaluation, the prototype was improved using a rack and pinion mechanism for the linear motion of the tape cutting and holding component. This mechanism works in three steps. Initially, open end of tape is fixed in the holding mechanism which is connected to the stationary section. The pinions drive the wrapper in which the tape is placed. The wrapper then wraps an adhesive tape for a specified number of rotations. In the final rotation, the tape is cut and detached from the plant. Along with the cutting, the mechanism holds the tape as the initial position for next tying cycle. The improved prototype was also fabricated using 3D printer and was evaluated in the laboratory environment earlier this fall (Fall 2016). Scotch Filament tape (Fig. 2) was selected to wrap the canes due to the presence of a thin filament in it.



Fig. 1: A set of raspberry canes bundled and wrapped with a Scotch Filament tape.



Fig. 2: A mechanism to tie adhesive tape around a bundle of canes; Left - Earlier version of the prototype being evaluated along with a bundling mechanism. Right – Improved version of the mechanism being evaluated in the lab.

A robotic end-effector or a hand was designed and fabricated using a scissor mechanism (Fig. 3). The end-effector was then integrated with a robotic manipulator or arm and was automatically controlled to achieve a pruning cut in the laboratory environment.

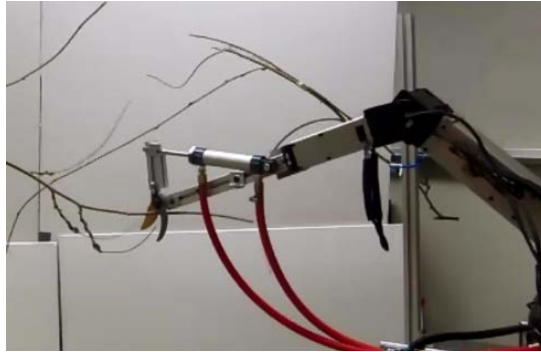


Fig. 3: A manipulator and a pruning end-effector in lab testing

Results and Discussion:

The first prototype bundling and tying mechanism accommodated cane bundles with a maximum of 7 canes. After this field test, the tying mechanism has been improved by adding a rack and pinion-based arm, which can cut the tape as well as grab and hold the tape for next plant to be bundled. The following are the key features of the improved prototype machine.

- *Newly added tape cutting and grabbing mechanism is fully automated and is controlled by a single dc motor.*
- *The tape wrapper design has been improved by adding supports for guiding the tape for grabbing and cutting mechanism. Opening in the new tape wrapper has been increased to let up-to nine canes enter inside it.*
- *Special attention has been given to synchronize the movement of wrapping and cutting mechanisms.*
- *Special attention was given in selection of the tape for long durability and strength. This filament helps in improve the strength, and adhesiveness with the plant for long period, which achieved better tying than the normal tape.*

Publications:

Shrestha, A., Karkee, M. and Zhang, Q., 2016. Mechanism for Bundling and Tying of Red Raspberry Primocanes. *IFAC-PapersOnLine*, 49(16), pp.166-170.

2016 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

New Project Proposal

Proposed Duration: One year

Project Title: Mechanizing red raspberry pruning and cane tying

PI: Manoj Karkee

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Address 2:

City/State/Zip: Prosser, WA 99350

Co-PI: Joan Davenport

Organization: WSU-IAREC

Title: Research Horticulturist

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Address: 24106 N. Bunn Rd.

Address 2:

City/State/Zip: Prosser, WA 99350

Cooperators: Qin Zhang, WSU-CPAAS

Year Initiated 2017 **Current Year** 2017

Terminating Year 2017

Total Project Request: Year 1 \$9,832

Other funding sources: Last year, \$54,188 was funded as a sub-contract to WSU from the funding that WRRC and WSU scientists received through the WA Specialty Crop Block Grant program. Part of this funding remains to be spent in 2017. An additional \$9,832 is requested from WRRC to complement engineering research activities under this grant.

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. Over the last three years in this project, we planned to develop a systematic approach for cane management through horticultural modifications and engineering solutions. A red raspberry plot was developed with three different types of red raspberries in eastern WA for their feasibility in mechanized pruning of two-year old canes. In addition, we have been developing techniques to bundle one-year old canes together and tie them to the trellis wires. Current accomplishment from this work has been discussed in the progress report submitted with this proposal. In the next year of this project, we will complete field evaluation of the second version of tape tying mechanism developed in Fall 2016. We expect that the successful completion of the proposed work 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



Fig. 1: Red-raspberry pruning and tying

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 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 have been systematically evaluating horticultural 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.

“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.

This project will impact all red raspberry growers in WA who use the floricane 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 is expected to generate industry-applicable 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 WRRRC Research Priority(s): This project directly addresses priority #1: “Labor saving practices.”

Objectives to be accomplished in 2017:

The primary goal of the proposed work is to minimize labor demand in red raspberry pruning through integrated horticultural and mechanization, or automation solutions. To achieve the overall goal, we have been particularly focusing on the following objectives over the last three years 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. 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 floriculture identification and a floriculture pruning mechanism

Please refer to the progress report submitted along with this funding proposal for the accomplishments made in 2016. Particularly in the Year 2016, progress will be made in the following research activities.

1. Continue to manage the raspberry plot in Prosser, WA
2. Complete the fabrication of improved prototype for cane bundling and tying
3. Evaluate the improved prototype in red-raspberry field
4. Outreach activities

Procedures:

Objective #1 - Horticultural Management of Red Raspberry Plot (Lead –Davenport): All cultural practices will be according to commercial standards. 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 floriculture canes.

Objective #2 - Engineering Approaches (Lead – Karkee): We will complete the development and evaluation of the improved prototype to bundling and tying mechanism primocanes. The prototype was evaluated in the lab and field in 2016 and will be further evaluated in the field. This year, we will also continue to investigate the method to identify and locate floriculture canes for pruning. A spectrometer will be used to identify and locate floriculture canes for pruning. We will also continue the investigation of the use of food-grade paints as well as red string-tying to provide the additional information for the image processing system.

Anticipated Benefits and Information Transfer:

This project will evaluate different mechanisms for mechanizing both training and pruning of various cultivars of red-raspberry, 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.

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

	2017
Salaries	\$23,993
Time-Slip	\$8,000
Operations (goods & services)	
Travel	\$1,000
Other/Miscellaneous	
Benefits	\$832
Total	\$9,832

Budget Justification

Wages (Sub-Total: \$8,000) – 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 is \$8,000.

Travel (Sub-Total: \$1,000) – Each year, one graduate student will travel to Lynden, WA to conduct field experiments in collaboration with grower collaborators. A part of the travel cost will also be used in extension activities.

Benefits (Sub-Total: \$832) – All values are in accordance with Washington State University's mandated rates for benefits and benefit inflation according to staff classification. It is calculated @10.4% of the wages requested.

Current & Pending Support-Karkee

NAME (List/PD #1 first)	SUPPORTING AGENCY AND AGENCY ACTIVE AWARD/PENDING PROPOSAL NUMBER	TOTAL \$ AMOUNT	EFFECTIVE AND EXPIRATION DATES	% OF TIME COMMITTE D	TITLE OF PROJECT
ACTIVE					
Slaughter (PD); Fennimore; Giles; Karkee; Siemens; Smith; Tourte; Upadhyaya; Voigioukas; Zhang	USDA-NIFA-SCRI	\$2,715,901	09/14 to 08/18	10%	Crop Signaling for Automated Weed/Crop Differentiation and Mechanized Weed Control in Vegetable Crops
Karkee (PD); Whiting; Zhang	USDA-NIFA-AFRI	\$495,480	12/14 to 11/17	10%	Shake and Catch Harvesting for Fresh Market Apples
Hashimoto (PD), Chiang; Cooper; Eggeman; Karkee; Vaugh; Yanagida; Zhang; etc.	USDA-NIFA-BRDI	\$6,000,000	04/12 to 03/17	12%	Conversion of High-Yield Tropical Biomass into Sustainable Biofuels
Bierlink and Karkee (PD); Tarara	WSDA Special Crop Block Grant	\$199,926	02/14 to 01/18	8%	Mechanizing Red Raspberry Pruning and Tying
Karkee (PI) ; Lewis ; Mo; Zhang	National Robotics Initiative - NSF and USDA-NIFA	\$548,735	09/13 to 08/17	10%	Human machine collaboration for automated harvesting of tree fruit

PENDING: N/A

PATHOLOGY

VIROLOGY

Raspberry Botrytis Program Trial 2016

During the summer of 2016, a research trial was conducted investigating the efficacy of various conventional fungicide programs for botrytis control within an 11 year old stand of “*Meeker*” raspberries grown of Whatcom county of western Washington. The experimental design for this trial was a RCB with 4 replications and plot sizes of 10ft x 30ft. Applications for this trial were made with an Over the Row sprayer calibrated to apply treatment sprays at 90 gallons per acre. Both sides of each plot’s raspberry canes were simultaneously sprayed to ensure complete coverage with the experimental products used. The treatment applications for this trial began on May 5th (10% Bloom), and continued at ~10 day intervals until June 29th (mid harvest). In order to increase disease pressure an overhead misting system was installed and the misting system was turned on for several hours a day when conditions were most conducive for disease development.

The first disease evaluation was conducted on July 9th, ten days after the last application and 65 days after the first application (data column 1). A second evaluation was done three days later. Of the two ratings, the first rating is considered to be the more accurate assessment of disease control. Botrytis was rated by counting the entire number of infected berries in a plot. This was done by going from the beginning to the end of the plot (30 feet) on both sides for a total of 60 row feet.

Raspberry Botrytis Program Trial 2016- Results:

Crop Name					Red raspberry		Red raspberry						
Description					# w/ Botrytis		# w/ Botrytis						
Rating Type					COUNT		COUNT						
Sample Size, Unit					1 PLOT		1 PLOT						
Rating Date					Jul-9-2016		Jul-12-2016						
Days After First/Last Applic.					65 10		68 13						
Trt No.	Treatment Name	Form Conc	Form Unit	Rate Rate	Appl Code								
						1		2					
1	Untreated Check					15.0a		23.3a					
2	CAPTAN	80%W/W		2lb/a	A	3.8cd		10.8bc					
	SWITCH	62.5%W/W		14oz/a	A								
	CAPTAN	80%W/W		2lb/a	B								
	PRISTINE	38%W/W		23oz/a	B								
	CAPTAN	80%W/W		2.5lb/a	C								
	Rovral	4LBA/GAL		32fl oz/a	C								
	CAPTAN	80%W/W		2lb/a	D								
	SWITCH	62.5%W/W		14oz/a	D								
	CAPTAN	80%W/W		2lb/a	E								
	PhD	11.3%W/W		6.2oz/a	E								
	CAPTAN	80%W/W		2lb/a	F								
	SWITCH	62.5%W/W		14oz/a	F								
3	CAPTAN	80%W/W		2.5lb/a	A					3.0cd		6.8bc	
	SWITCH	62.5%W/W		14oz/a	A								
	CAPTAN	80%W/W		2.5lb/a	B								
	PRISTINE	38%W/W		23oz/a	B								
	CAPTAN	80%W/W		2.5lb/a	C								
	Rovral	4LBA/GAL		32fl oz/a	C								
	CAPTAN	80%W/W		2.5lb/a	D								
	SWITCH	62.5%W/W		14oz/a	D								
	CAPTAN	80%W/W		2.5lb/a	E								
	PhD	11.3%W/W		6.2oz/a	E								
	SWITCH	62.5%W/W		14oz/a	F								
4	CAPTAN	80%W/W		2.5lb/a	A	10.5ab		22.0a					
	CAPTAN	80%W/W		2lb/a	B								
	PhD	11.3%W/W		6.2oz/a	B								
	CAPTAN	80%W/W		2.5lb/a	C								
	SWITCH	62.5%W/W		14oz/a	C								
	CAPTAN	80%W/W		2.5lb/a	D								
	SWITCH	62.5%W/W		14oz/a	D								
5	CAPTAN	80%W/W		2.5lb/a	A	12.8a		15.3ab					
	SWITCH	62.5%W/W		14oz/a	A								
	CAPTAN	80%W/W		2.5lb/a	B								
	CAPTAN	80%W/W		2.5lb/a	C								
	CAPTAN	80%W/W		2.5lb/a	D								
	SWITCH	62.5%W/W		14oz/a	D								

Raspberry Botrytis Program Trial 2016 – Results:

6CAPTAN	80%W/W	1.5lb/a	A	9.0abc	19.8a
CAPTAN	80%W/W	1.5lb/a	B		
CAPTAN	80%W/W	1.5lb/a	C		
CAPTAN	80%W/W	1.5lb/a	D		
SWITCH	62.5%W/W	14oz/a	D		
7CAPTAN	80%W/W	1.25lb/a	A	3.8cd	8.0bc
SWITCH	62.5%W/W	14oz/a	A		
CAPTAN	80%W/W	1.25lb/a	B		
PRISTINE	38%W/W	23oz/a	B		
CAPTAN	80%W/W	2.5lb/a	C		
Kenja	3.3LBA/GAL	15.5fl oz/a	C		
CAPTAN	80%W/W	1.25lb/a	D		
SWITCH	62.5%W/W	14oz/a	D		
PhD	11.3%W/W	6.2oz/a	E		
SWITCH	62.5%W/W	14oz/a	E		
PhD	11.3%W/W	6.2oz/a	F		
SWITCH	62.5%W/W	14oz/a	F		
8CAPTAN	80%W/W	2lb/a	A	3.0cd	6.3c
SWITCH	62.5%W/W	11.2oz/a	A		
CAPTAN	80%W/W	2lb/a	B		
PRISTINE	38%W/W	20oz/a	B		
CAPTAN	80%W/W	2.5lb/a	C		
Rovral	4LBA/GAL	32fl oz/a	C		
CAPTAN	80%W/W	2lb/a	D		
SWITCH	62.5%W/W	11.2oz/a	D		
CAPTAN	80%W/W	2lb/a	E		
PhD	11.3%W/W	6.2oz/a	E		
CAPTAN	80%W/W	2lb/a	F		
SWITCH	62.5%W/W	11.2oz/a	F		
9Kenja	3.3LBA/GAL	15.5fl oz/a	A		
SWITCH	62.5%W/W	14oz/a	B		
Kenja	3.3LBA/GAL	15.5fl oz/a	C		
PRISTINE	38%W/W	20oz/a	D		
Kenja	3.3LBA/GAL	15.5fl oz/a	E		
SWITCH	62.5%W/W	14oz/a	F		
10ELEVATE	50%W/W	1.5lb/a	A	5.3bcd	8.0bc
Rovral	4LBA/GAL	32fl oz/a	B		
ELEVATE	50%W/W	1.5lb/a	C		
PRISTINE	38%W/W	20oz/a	D		
ELEVATE	50%W/W	1.5lb/a	E		
SWITCH	62.5%W/W	14oz/a	F		
LSD P=.05				6.59	8.82
Standard Deviation				4.54	6.08
CV				67.02	49.53
Bartlett's X2				22.356	11.563

Agriculture Development Group, Inc

Raspberry Botrytis Field Efficacy Program - 2016

Trial ID: Rasp.Botry.Efficacy.WS-2016 Location: Western WA Trial Year: 2016
 Protocol ID: Rasp.Botry.Efficacy.WS Investigator: Alan Schreiber
 Project ID: Study Director: Tom Walters
 Sponsor Contact: WA Red Raspberry Commission, several others

General Trial Information

Study Director: Tom Walters **Title:** Ag Researcher
Investigator: Alan Schreiber **Title:** Research Director

Discipline: F fungicide
Trial Status: F one-year/final **Trial Reliability:** GOOD
Initiation Date: Mar-10-2016 **Planned Completion Date:** Oct-30-2016
Completion Date: Oct-15-2016

Trial Location

City: Everson **Country:** USA
State/Prov.: Whatcom County, WA **Climate Zone:** PNW

Objectives:

To test fungicide products for control of botrytis in raspberries of western Washington state.

Contacts

Study Director: Tom Walters **Title:** Ag Researcher
Organization: Walters Ag Research
Address: 2117 Meadows Lane
City+State/Prov: Anacortes, WA 98221 **Mobile No.:** 360-420-2776
E-mail: waltersagresearch@frontier.com

Investigator: Alan Schreiber **Title:** Research Director
Organization: Agriculture Development Group, Inc
Address: 2621 Ringold Road **Phone No.:** 509-266-4348
City+State/Prov: Eltopia, Washington **Mobile No.:** 509-539-4537
Postal Code: 99330 **E-mail:** aschreib@centurytel.net
Country: USA United States

Crop Description

Crop 1: RUBID Rubus idaeus Red raspberry
Variety: Meeker

Agriculture Development Group, Inc

Raspberry Botrytis Field Efficacy Program - 2016

Trial ID: Rasp.Botry.Efficacy.WS-2016 Location: Western WA Trial Year: 2016
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 Sponsor Contact: WA Raspberry Commission

Pest Description

Pest 1 Type: D **Code:** BOTRSP Botrytis sp.

Common Name: Botrytis sp.

		Site and Design	
Treated Plot Width:	10 FT	Site	
Type:	FIELD	Experimental	
Treated Plot Length:	30 FT		
Unit:	1 PLOT	Treatments:	23
Treated Plot Area:	300 FT ²		Tillage Type:
NOTILL			
Replications:	4	Study Design:	Randomized Complete Block (RCB)

Moisture and Weather Conditions
Overall Moisture Conditions: NORMAL

Application Description						
	A	B	C	D	E	F
Application Date:	May-5-2016	May-16-2016	May-25-2016	Jun-7-2016	Jun-17-2016	Jun-29-2016
Appl. Start Time:	12:00 PM	3:00 PM	9:00 AM	8:00 AM	6:00 AM	8:00 AM
Appl. Stop Time:	3:00 PM	6:00 PM	11:30 AM	12:00 PM	11:00 AM	11:00 AM
Application Method:	SPRAY	SPRAY	SPRAY	SPRAY	SPRAY	SPRAY
Application Timing:	10% Bloom	30% Bloom	50% Bloom	1st Harvest		Mid Harvest
Application Placement:	FOLIAR	FOLIAR	FOLIAR	FOLIAR	FOLIAR	FOLIAR
Applied By:	TOM W	TOM W	TOM W	TOM W	TOM W	TOM W
Air Temperature, Unit:	62 F	60 F	50 F	65 F	55 F	64 F
Wind Velocity, Unit:	4 MPH	2 MPH	2 SE	0 MPH	0 MPH	0 MPH
Wind Direction:	W	SW	S			

Crop Stage At Each Application						
	A	B	C	D	E	F
Crop 1 Code, BBCH Scale:	RUBID BPER	RUBID BPER	RUBID BPER	RUBID BPER	RUBID BPER	RUBID BPER

Agriculture Development Group, Inc

Raspberry Botrytis Field Efficacy Program - 2016

Trial ID: Rasp.Botry.Efficacy.WS-2016 Location: Western WA Trial Year: 2016
 Protocol ID: Rasp.Botry.Efficacy.WS Investigator: Alan Schreiber
 Project ID: Study Director: Tom Walters
 Sponsor Contact: WA Raspberry Commission

Pest Stage At Each Application

	A	B	C	D	E	F
Pest 1 Code, Type, Scale:	BOTRSP D	BOTRSP D	BOTRSP D	BOTRSP D	BOTRSP D	BOTRSP D

Application Equipment

	A	B	C	D	E	F
Appl. Equipment:	Rears OverRo	Rears OverRo	Rears OverRo	Rears OverRo	Rears OverRo	Rears OverRo
Equipment Type:	3POINT	3POINT	3POINT	3POINT	3POINT	3POINT
Operation Pressure, Unit:	130 psi	130 psi	130 psi	130 psi	130 psi	130 psi
Nozzle Type:	Tee-Jet	Tee-Jet	Tee-Jet	Tee-Jet	Tee-Jet	Tee-Jet
Nozzle Size:	D-3	D-3	D-3	D-3	D-3	D-3
Nozzle Spacing, Unit:	12 in	12 in	12 in	12 in	12 in	12 in
Nozzles/Row:	4	4	4	4	4	4
Band Width, Unit:	6 ft	6 ft	6 ft	6 ft	6 ft	6 ft
Boom Length, Unit:	6 FT	6 FT	6 FT	6 FT	6 FT	6 FT
Ground Speed, Unit:	3.2 mph	3.2 mph	3.2 mph	3.2 mph	3.2 mph	3.2 mph
Carrier:	WATER	WATER	WATER	WATER	WATER	WATER
Spray Volume, Unit:	100 gal/ac	100 gal/ac	100 gal/ac	100 gal/ac	100 gal/ac	100 gal/ac
Mix Size, Unit:	10.5 liters	10.5 liters	10.5 liters	10.5 liters	10.5 liters	10.5 liters

Agriculture Development Group, Inc

Raspberry Botrytis Field Efficacy Program - 2016

Trial ID: Rasp.Botry.Efficacy.WS-2016 Location: Western WA Trial Year: 2016
 Protocol ID: Rasp.Botry.Efficacy.WS Investigator: Alan Schreiber
 Project ID: Study Director: Tom Walters
 Sponsor Contact: WA Raspberry Commission

Reps: 4

Plots: 10 by 30 feet

Spray vol: 100 GAL/AC

Mix Size: 12.514 liters (12.514 liters calculated mix size)

Trt No.	Treatment Name	Form Conc	Form Unit	Form Type	Rate Rate	Rate Unit	Amt Product to Measure	Rep 1	2	3	4
1	Untreated Check							101	223	311	409
2	PhD	11.3%	W/W	WG	6.2oz/a		4.842 g/4 pl	102	215	307	411
3	OMEGA	4.17	LB/GAL	EC	1.25pt/a		16.29 ml/4 pl	103	217	312	418
4	Luna Tranquility	45%	W/W	L	18fl oz/a		14.66 ml/4 pl	104	207	317	401
5	SCALA	54.6%	W/W	SC	18fl oz/a		14.66 ml/4 pl	105	208	313	412
6	SWITCH	62.5%	W/W	WG	14oz/a		10.93 g/4 pl	106	221	322	406
7	CAPTAN	80%	W/W	WG	2.5lb/a		31.24 g/4 pl	107	205	302	421
8	ELEVATE	50%	W/W	WG	1.5lb/a		18.74 g/4 pl	108	209	314	420
9	PRISTINE	38%	W/W	WG	23oz/a		17.96 g/4 pl	109	203	320	410
10	IPRODIONE	4%		SL	1pt/a		13.04 ml/4 pl	110	213	304	417
11	Oxidate	100%		L	32fl oz/100 gal		31.28 ml/mx	111	212	310	415
12	PROLINE	4	LB/GAL	EC	5fl oz/a		4.073 ml/4 pl	112	219	315	414
13	OSO	5%		SC	12fl oz/a		9.776 ml/4 pl	113	206	301	402
14	Topsin M	70%		WP	1.5lb/a		18.74 g/4 pl	114	201	319	422
15	NeoBoost	100%		WG	3lb/a		37.49 g/4 pl	115	222	305	423
16	Jet Ag	100%		L	1% v/v		125.1 ml/mx	116	211	309	416
17	Fontelis	1.67	LB/GAL	SC	20fl oz/a		16.29 ml/4 pl	117	218	303	404
18	Kenja	400	GAL	SC	15.5fl oz/a		12.63 ml/4 pl	118	202	306	413
19	Kenja	400	GAL	SC	13.5fl oz/a		11.0 ml/4 pl	119	220	316	419
20	EXP # 101	100%		L	1.5pt/a		19.55 ml/4 pl	120	216	318	407
21	EXP # 101	100%		L	2pt/a		26.07 ml/4 pl	121	204	321	408
22	EXP # 101	100%		L	3pt/a		39.11 ml/4 pl	122	214	323	403
23	Untreated							123	210	308	405

Sort Order: Treatment

Agriculture Development Group, Inc

Raspberry Botrytis Field Efficacy Program - 2016

Trial ID: Rasp.Botry.Efficacy.WS-2016 Location: Western WA Trial Year: 2016
 Protocol ID: Rasp.Botry.Efficacy.WS Investigator: Alan Schreiber
 Project ID: Study Director: Tom Walters
 Sponsor Contact: WA Raspberry
 Commission

Product quantities required for listed treatments and applications of trials included in this table:

Amount*	Unit	Treatment Name	Form Conc	Form Unit	Form Type	Lot Code
34.863g		PhD	11.3	%W/W	WG	
117.316ml		OMEGA	4.17	LB/GAL	EC	
105.585ml		Luna Tranquility	45	%W/W	L	
105.585ml		SCALA	54.6	%W/W	SC	
78.723g		SWITCH	62.5	%W/W	WG	
224.921g		CAPTAN	80	%W/W	WG	
134.952g		ELEVATE	50	%W/W	WG	
129.330g		PRISTINE	38	%W/W	WG	
93.853ml		IPRODIONE	4	%	SL	
225.252ml		Oxidate	100	%	L	
29.329ml		PROLINE	4	LB/GAL	EC	
70.390ml		OSO	5	%	SC	
134.952g		Topsin M	70	%	WP	
269.905g		NeoBoost	100	%	WG	
900.910ml		Jet Ag	100	%	L	
19.553ml		Fontelis	1.67	LB/GAL	SC	
170.109ml		Kenja	400	GA/L	SC	
610.045ml		EXP # 101	100	%	L	

* 'Per area' calculations based on 4 replicates of 10 by 30 feet 'Plot' experimental units (area of one treatment).

* 'Per area' calculations based on spray volume= 100 GAL/AC, mix size= 12.514 liters (mix size basis).

* Product amount calculations increased 20 % for overage adjustment.

* 'Per volume' calculations use spray volume= 100 GAL/AC, mix size= 12.514 liters.

* Adjusted for multiple applications in treatment list.

* One application was assumed for blank application codes or multipliers.

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Raspberry Botrytis Field Efficacy Program - 2016							
Crop Name				Red raspberry		Red raspberry	
Pest Name				Botrytis sp.		Botrytis sp.	
Rating Type				COUNT		COUNT	
Description				# w/ Botrytis		# w/ Botrytis	
Sample Size, Unit				1 PLOT		1 PLOT	
Rating Date				Jul-9-2016		Jul-12-2016	
Days After First/Last Applic.				65 10		68 13	
Trt No.	Treatment Name	Form Conc	Form Unit	Rate Rate	Rate Unit	Appl Code	
						1	2
1	Untreated Check					13.0a	21.0abc
2	PhD	11.3%	W/W	6.2oz/a		ABCDEF	4.3cde
3	OMEGA	4.17LB/GAL		1.25pt/a		ABCDEF	0.8e
4	Luna Tranquility	45%	W/W	18fl oz/a		ABCDEF	2.0de
5	SCALA	54.6%	W/W	18fl oz/a		ABCDEF	5.8b-e
6	SWITCH	62.5%	W/W	14oz/a		ABCDEF	7.3a-d
7	CAPTAN	80%	W/W	2.5lb/a		ABCDEF	5.3b-e
8	ELEVATE	50%	W/W	1.5lb/a		ABCDEF	6.0b-e
9	PRISTINE	38%	W/W	23oz/a		ABCDEF	4.0cde
10	PRODIONE	4%		1pt/a		ABCDEF	7.0a-e
11	Oxidate	100%		32fl oz/100 gal		ABCDEF	11.0ab
12	PROLINE	4LB/GAL		5fl oz/a		ABCDEF	5.0b-e
13	OSO	5%		12fl oz/a		ABCDEF	5.8b-e
14	Topsin M	70%		1.5lb/a		ABCDEF	5.8b-e
15	NeoBoost	100%		3lb/a		ABCDEF	7.0a-e
16	Jet Ag	100%		1% v/v		ABCDEF	7.0a-e
17	Fontelis	1.67LB/GAL		20fl oz/a		ABCDEF	4.0cde
18	Kenja	400GA/L		15.5fl oz/a		ABCDEF	3.5cde
19	Kenja	400GA/L		13.5fl oz/a		ABCDEF	5.5b-e
20	EXP # 101	100%		1.5pt/a		ABCDEF	8.0a-d
21	EXP # 101	100%		2pt/a		ABCDEF	6.8a-e
22	EXP # 101	100%		3pt/a		ABCDEF	8.5abc
23	Untreated					11.0ab	17.3abc
LSD P=.05						6.47	13.85
Standard Deviation						4.58	9.81
CV						73.18	57.42
Bartlett's X2						21.441	36.086
P(Bartlett's X2)						0.494	0.03*
Skewness						0.7171*	1.324*
Kurtosis						-0.0314	1.3067*

Trial Comments. The trial was located in Whatcom County, near Everson Washington. The variety was Meeker and the stand is 11 years old. There were a total of 23 treatments with two untreated checks. The first application was made on May 5th with five subsequent applications made at roughly ten day intervals as weather and local conditions permitted for about two months. Weather conditions during the trial could be described as above average in temperatures and below average in precipitation. In order to increase disease pressure an overhead misting system was installed. The misting system was turned on for several hours a day when conditions were most conducive for disease development. Due to the unusually warm temperatures and low overall humidity, there was virtually no disease pressure until the end of the trial.

The first disease evaluation was conducted on July 9th, ten days after the last application and 65 days after the first application (data column 1). A second evaluation was done three days later. Of the two ratings, the first rating is considered to be the more accurate assessment of disease control. Botrytis was rated by counting the entire number of infected berries in a plot. This was done by going from the beginning to the end of the plot (30 feet) on both sides for a total of 60 row feet. Each berry in each plot was evaluated. A team of four people evaluated the trial. A total of 5,520 row feet (more than a mile) were examined at each evaluation.

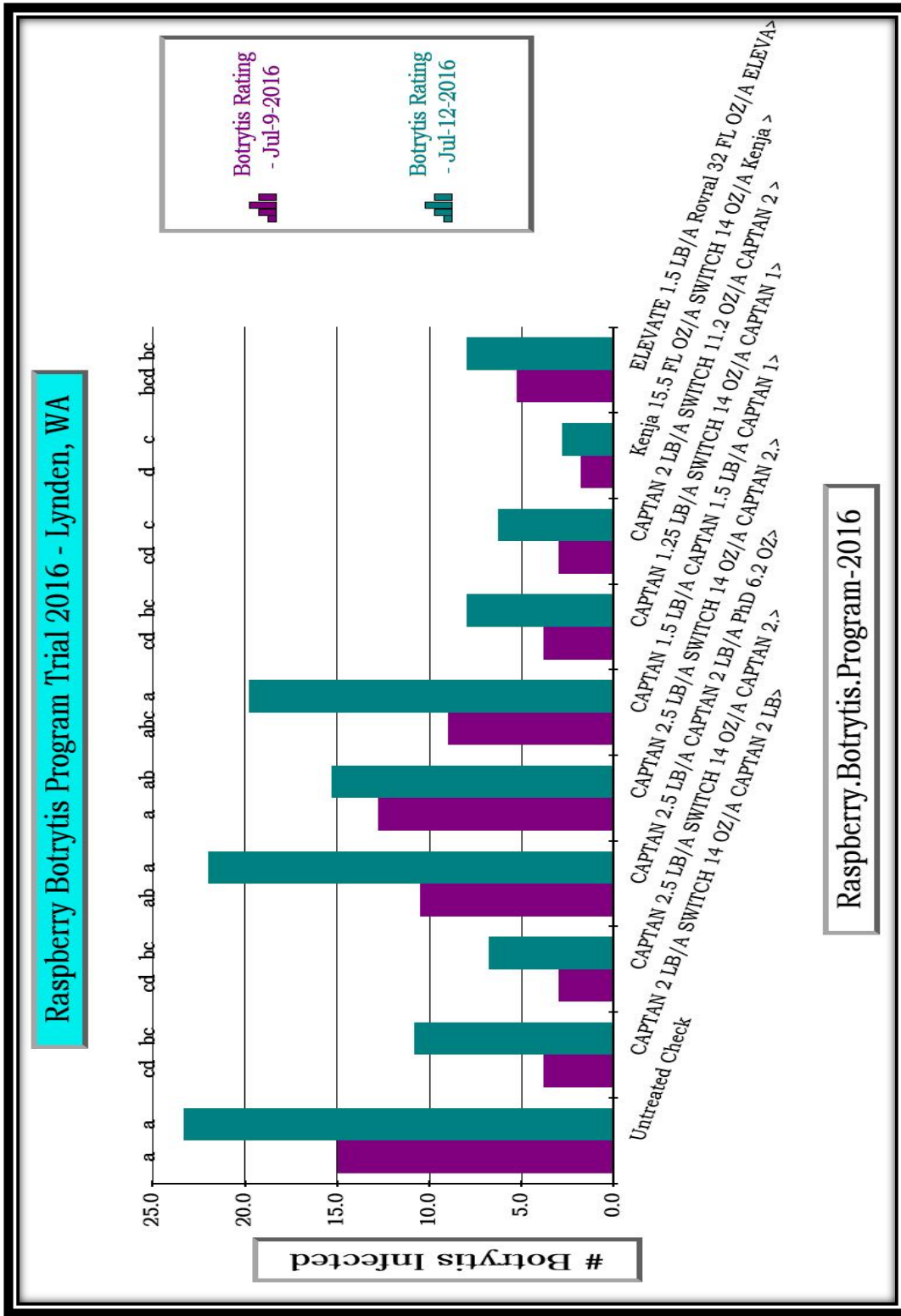
The first untreated check had a total of 13 infected fruit and the second untreated check had a total of 11 infected fruit at the first evaluation. One treatment, Oxidate, had a similar level of infection as the second untreated check, with 11 infected fruit. Treatments with a lower number of infected fruit but were statistically not different from the untreated checks were all three of the experimental #101, Jet Ag, NeoBoost, iprodione and Switch. Iprodione was applied at the 1 pint rate which is the lower rate and may explain its reduced efficacy. Switch has two constituent products, one of which is cyprodinil. Resistance to cyprodinil has been documented as occurring at this trial location and may explain the poor showing of Switch. Oxidate, Jet Ag and NeoBoost all have relatively short periods of residual control and may not demonstrate efficacy ten days after application.

The most effective products in the trial were, ranked in terms of increasing number of infected fruit are Omega with 0.8 infected fruit, Luna Tranquility with 2.0 infected fruit, Kenja at the high rate of 15.5 oz with 3.5 infected fruit, followed by Fontelis and Pristine with 4.0 infected fruit each, followed by Proline at 5 infected fruit, followed by Captan at 5.3 infected fruit, followed by Kenja at the low rate of 13.5 oz with 5.5 infected fruit, and Scala, Oso and Topsin M with 5.8 infected fruit. It is our belief that any of these products would provide commercially acceptable levels of control in normal botrytis conditions.

Three days later and 12 days after the last application and at which time harvested was ending, botrytis has become more common throughout the trial with 14 of the fungicidal treatments being not statistically different from the untreated check. The treatments that significantly reduced botrytis levels as compared to untreated check were a subset of the products listed previously. The products are presented in increasing levels of infected berries per plot; Captan (7.8), Luna Tranquility (9.8), Oso (10), Omega (10.5), Kenja (11.8) and Pristine (13). There was no statistical difference between these treatments in efficacy.

Based on the results of this trial, during a year of low botrytis pressure as seen in 2016, it would appear that the following products could provide commercially acceptable levels of control Omega, Luna Tranquility, Kenja, Fontelis, Pristine, Proline, Captan, Kenja, Scala, Oso and Topsin M. Please note that several of the products are not currently registered on caneberries.

Raspberry Botrytis Program Trial 2016



Raspberry Botrytis Program Trial 2016

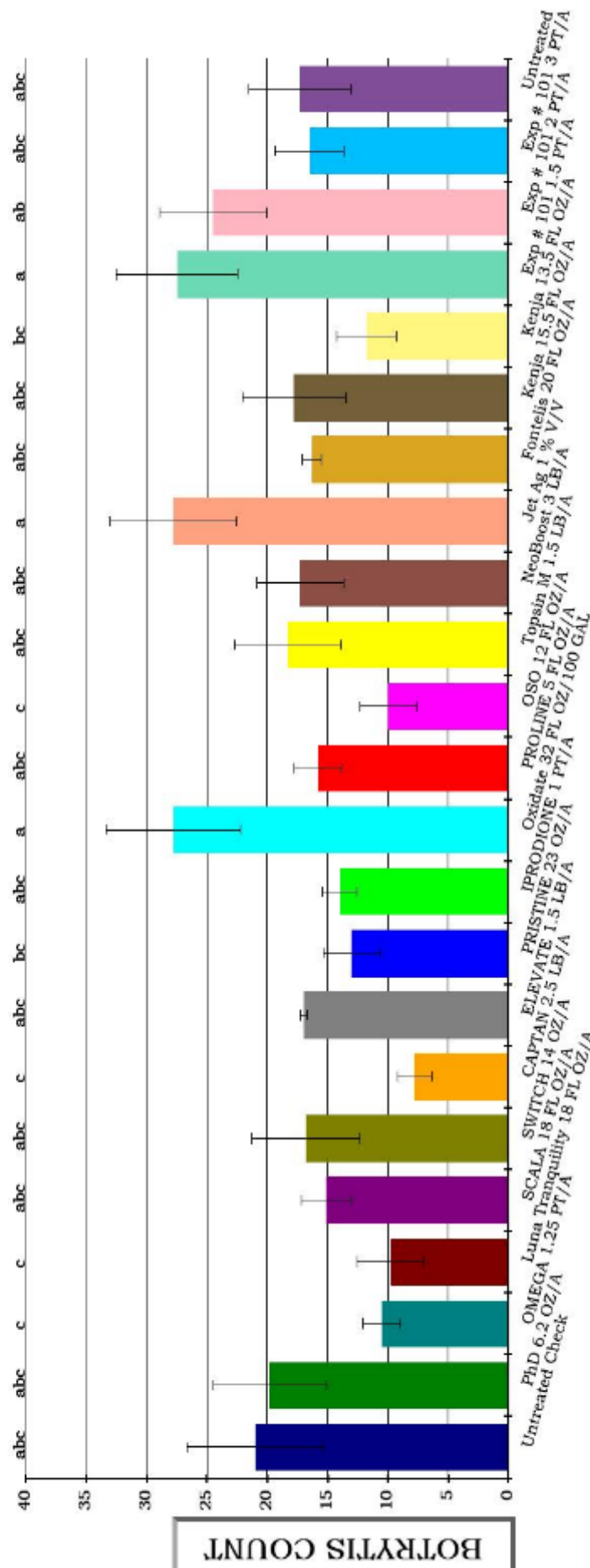


Photo #1 (left) The over the row sprayer used to make treatment application to the “*Meeker*” raspberries of the trial

Graph #1

(above) – Summary of botrytis infected raspberries counted within each plot during the July 12th evaluation date.

23 TRT - Raspberry Botrytis Field Efficacy Program - 2016



Trial ID: Rasp.Botry.Efficacy.WS-2016

Project Proposal to WRRC

Proposed Duration: 3 Years

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), 509 539 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: 2016

Current Year: 2016

Terminating Year: 2018

Total Project Request: Year 1 \$12,000 Year 2 \$12,000 Year 3 \$12,000

Other Funding Sources: I have submitted a parallel proposal to the Washington State Commission on Pesticide Registration for \$22,500. I expect that registrants will be involved in this project and will contribute, but how much is not known.

Description: Resistance has been documented to three of five active ingredients used for control of botrytis in 2012. Dr. Peever and I differ on the degree to which resistance to a fifth product, iprodione exists. Based on Dr. Peever's work, it is clear that there is widespread resistance to Elevate, Pristine and Switch and the level of resistance appears to have increased in the short time that he has started monitoring resistance. This 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 standard efficacy trial that is modeled after the 2014 trial, but with some improvements based on what was learned during the course of the previous trial. Data generated supported a Section 18 for a new fungicide that was shown to be more effective than any currently available product used for botrytis control. This project involved three trials; an efficacy program screening several fungicides, a program trial that evaluates all major raspberry botrytis programs and a third trial on blackberry where disease pressure is higher than on raspberry.

Justification and Background: This project will generate data on which fungicidal products are effective for controlling botrytis and which products are not. Dr. Peever will take the lead on berry pathology and biological work, but at this time he does not plan to take the lead on efficacy trials in raspberries. Dr. Peever will work cooperatively with this project. 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.

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 as mycelia on dead leaves and mummified fruit. These sclerotia will produce conidia in spring, when a moist, humid environment provides the ideal conditions for the spread and sporulation of this pathogen. 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 economic or action threshold for this disease. If you find it, think you have it, or are at risk of having it, then you have to start a treatment program. The PNW Small Fruit Research Center ranks it as the number one priority for research in blueberry and raspberry. Raspberry, blueberry, blackberry and strawberry fundamentally have the same disease issues, and are often planted adjacent to each other, using the same fungicides and creating similar fungicide resistance issues. Raspberry has fruit that is susceptible earlier than blueberry and has heavier selection pressure. It is likely that spores which survive a raspberry fungicide programs will infect blueberry fields that mature later in the season, and are subsequently subjected to another fungicide program within the same year.

Despite aggressive treatment programs, growers will incur annual losses to this pest. *Botrytis* is well known for developing resistance to fungicides. Growers, crop advisors, researchers and extension representatives are concerned that genetic mutations facilitating resistance may be developing faster than new fungicide products can be developed. 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.*”

Growers try using all four modes of action during a season for resistance management (although some can only use three products due to MRLs). Other issues occur due to label restrictions such as number of application restrictions, REI and PHIs. The loss of even one product could mean a significant problem; the loss of two products would cause a crisis in the industry. We coordinate our efforts with OSU, USDA ARS and BC disease research programs.

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

Objectives: Our objective is to generate botrytis efficacy data for new products labeled for red raspberry. This information is very important to support the Section 18 for Kenja. A secondary

objective is to use this data and information provided by Dr. Peever to develop better botrytis control recommendations for raspberry.

Procedures: We plan to conduct efficacy trials in 2017 that are similar to the trials done in 2016. The testing techniques would be similar to what we have used in past years, with some improvements. Although testing details have not been finalized, we would like to use the same site as in 2016. One trial looked primarily at single ingredient programs to ascertain how that particular product worked against botrytis. The second trial evaluated 12 different programs used by the Whatcom County raspberry industry. The 12 programs covered the breadth of contract strategies used by growers. The second trial looked at 19 different active ingredients. The trials took place in a location that had documented fungicidal resistant botrytis.

We propose to conduct three trials in 2017, one that would screen for new products and a second trial that would evaluate season long programs that are currently being used by growers. A third trial would be conducted in blackberry. The reason we are targeting blackberry is because it appears to have a higher likelihood of developing botrytis. Conducting this third trial in blackberry is an insurance policy to increase the likelihood that we would generate useful data for raspberry growers. A commercial style applicator would be used and each treatment would be replicated four times.

Applications would start prebloom and would continue into harvest. The start and end dates, and number of application depends on environmental / 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 Schreiber would oversee the trial, collect and analyze the data to generate research reports.

The experimental design, including products and treatments, used in the previous trials will serve as the base for the 2017 trial. Scientists involved in project will meet with raspberry industry members and discuss what adjustments should be made to improve the trial.

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:	2017	2018	201
Salaries	6,000	6,000	8,000
Operations	3,000	3,000	3,000
Travel	1,500	1,500	1,500
Benefits	1,500	1,500	1,500

Total	\$12,000	\$12,000	\$14,000
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These funds would be primarily be used to cover the time of Schreiber and Walters spent on the project. It would cover the applicator's time, tractor/equipment usage, product purchases and other costs. WSCPR funds would be used to fund the effort to make applications and collect data. All travel costs are related to traveling to the site and/or meeting with industry representatives.

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

Related Information.

Results from 2014. Unfortunately, disease pressure in 2014 was low. Disease was not detected in the trials until very late in the season and even then it was quite low. Virtually every fungicide provided a significant level of control. The one product that did not provide control, Regalia, should probably be considered a marginally effective product at best, and probably not considered an option by conventional growers. Some new active ingredients were identified that had activity against botrytis in a very low pressure situation. These products have new modes of action so if they are proven efficacious, they may be of great interest to the industry. We feel that the products, treatments and experimental design of 2014 would be suitable for the 2015 trial. The principal investigators of this trial would meet with industry representatives to review the 2015 treatments to ascertain if any adjustments or improvements could be made.

Results from 2015. Based on significant feedback from the raspberry industry and knowledge of potential fungicides that are becoming available, a very good set of fungicide trials was set up and deployed. Disease pressure in 2015 was low despite use of an overhead irrigation system to foment disease development. Despite the low pressure, some data were generated that showed existing registered fungicides were limited in the ability to control botrytis, and at least one product was very effective against fungicide resistant botrytis. A parallel, similar and smaller trial on blackberry with fungicide resistant botrytis showed excellent efficacy against botrytis by the ingredient for which we are pursuing a Section 18 in 2016.

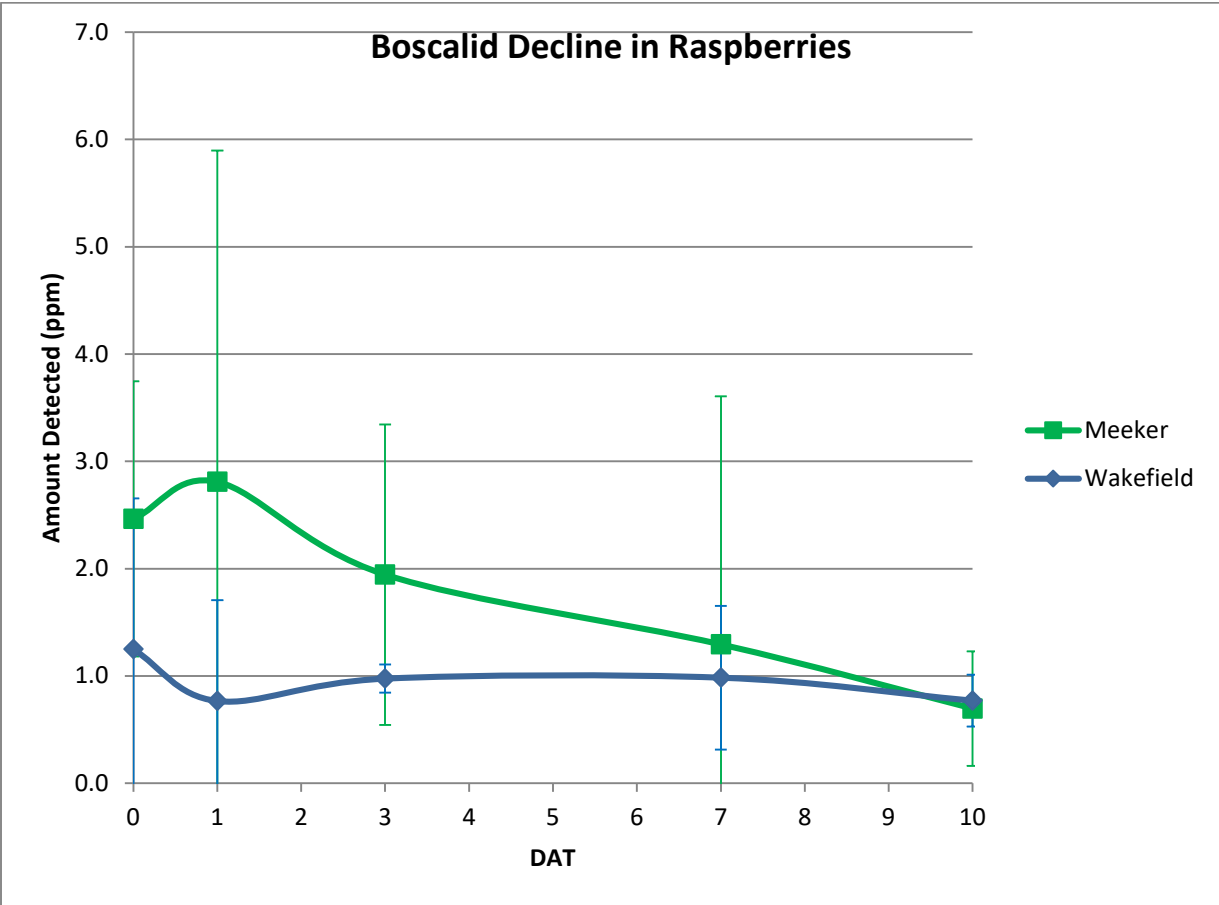
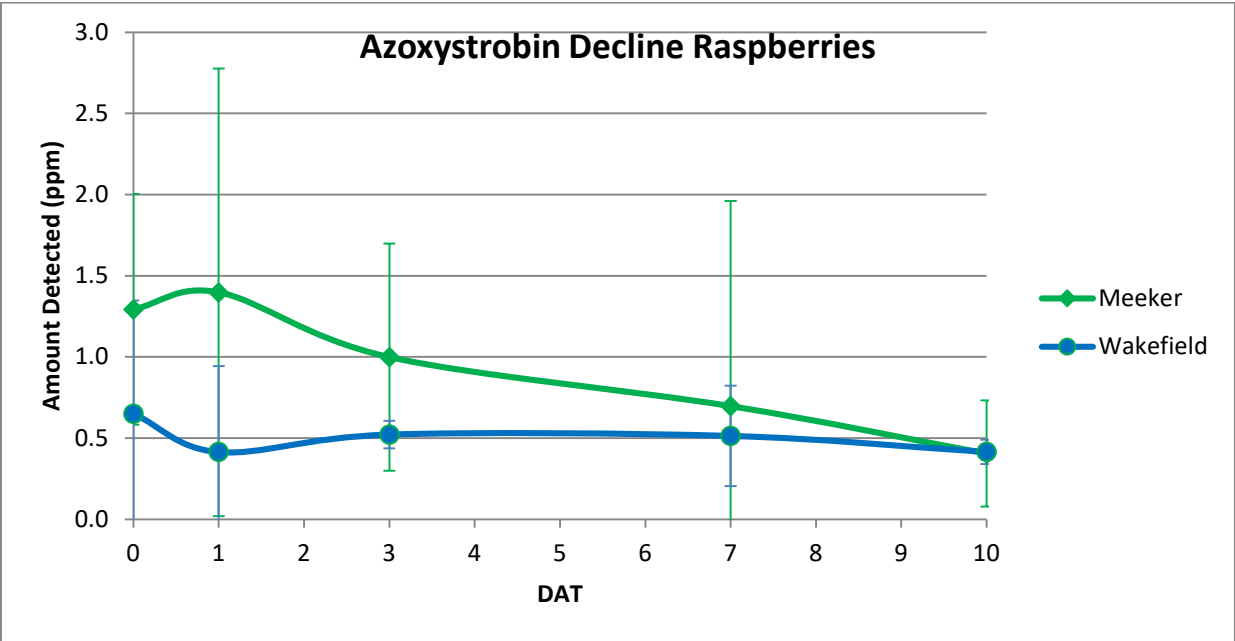
Results from 2016. The blackberry trial had heavy disease pressure and iprodione, Switch, Pristine and Elevate failed. Kenja, Luna Tranquility and Fontelis significantly reduced disease pressure. Disease pressure was moderate and late in the 2016 raspberry botrytis trial. The most effective products in the trial were, ranked in terms of increasing number of infected fruit are Omega with 0.8 infected fruit, Luna Tranquility with 2.0 infected fruit, Kenja at the high rate of 15.5 oz with 3.5 infected fruit, followed by Fontelis and Pristine with 4.0 infected fruit each, followed by Proline at 5 infected fruit, followed by Captan at 5.3 infected fruit, followed by Kenja at the low rate of 13.5 oz with 5.5 infected fruit, and Scala, Oso and Topsin M with 5.8 infected fruit. It is our belief that any of these products would provide commercially acceptable levels of control in normal botrytis conditions.

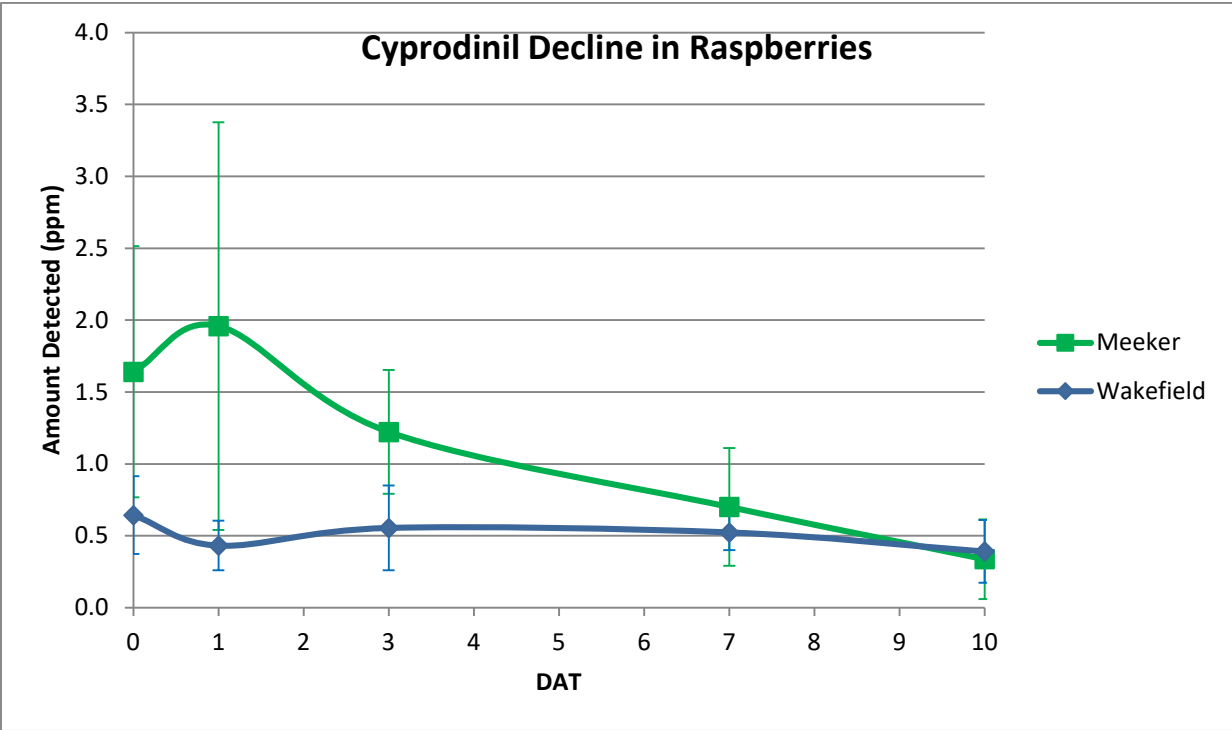
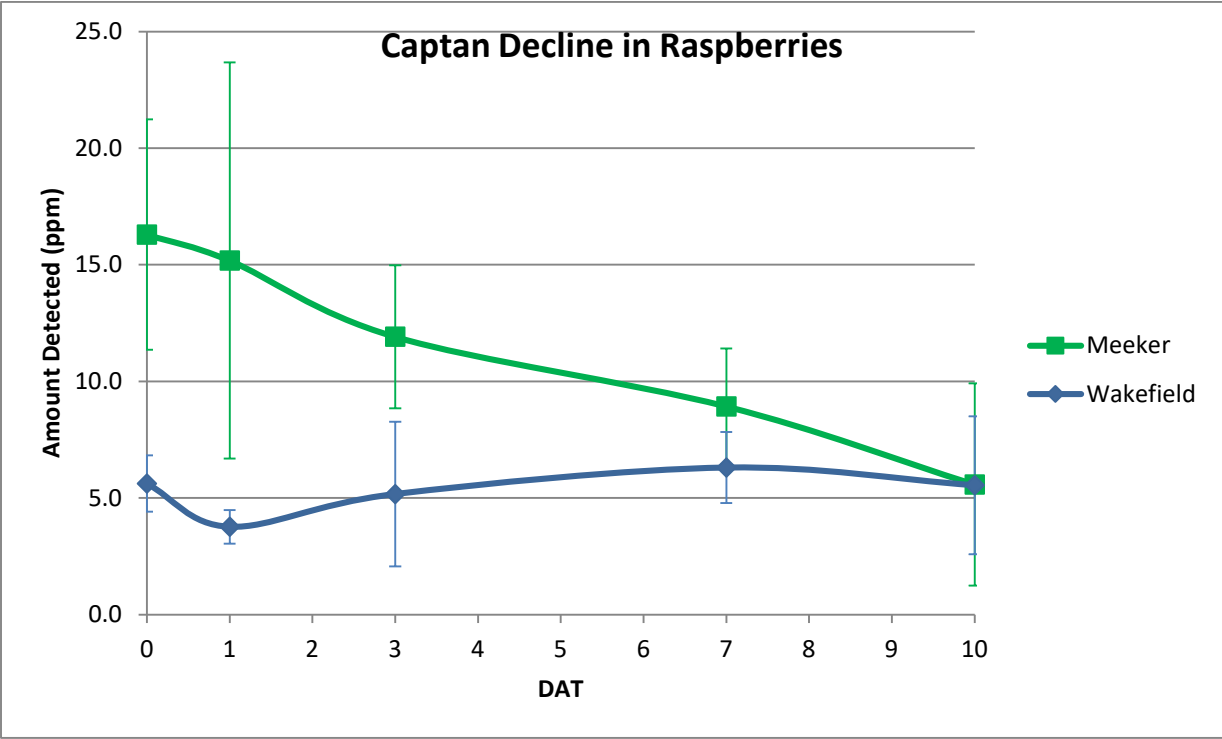
Project Proposal to WRRC**Proposed Duration: 2 - 3 Years****Project Title: Fungicide Decline Curves for meeting MRLs for Raspberry****PI:** Alan Schreiber**Organization:** Agriculture Development Group, Inc.**Title:** Researcher**Phone:** 509 266 4348 (office), 509 539 4537 (cell)**Email:** aschreib@centurytel.net**Address:** 2621 Ringold Road, Eltopia, WA 99330**Cooperators:** Tom Walters-Walters Ag Research**Year Initiated:** 2016**Current Year:** 2016**Total Project Request:** Year 1 \$14,000 Year 2 \$14,000 (not applying)

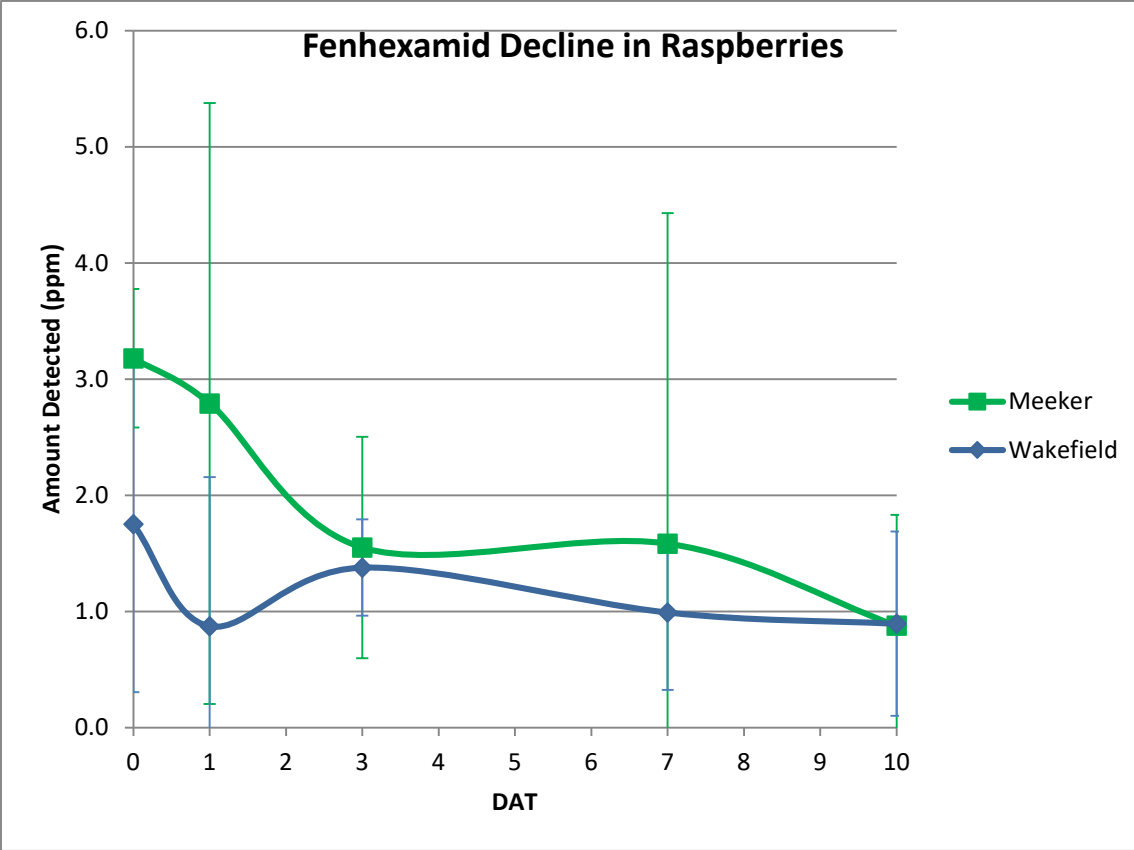
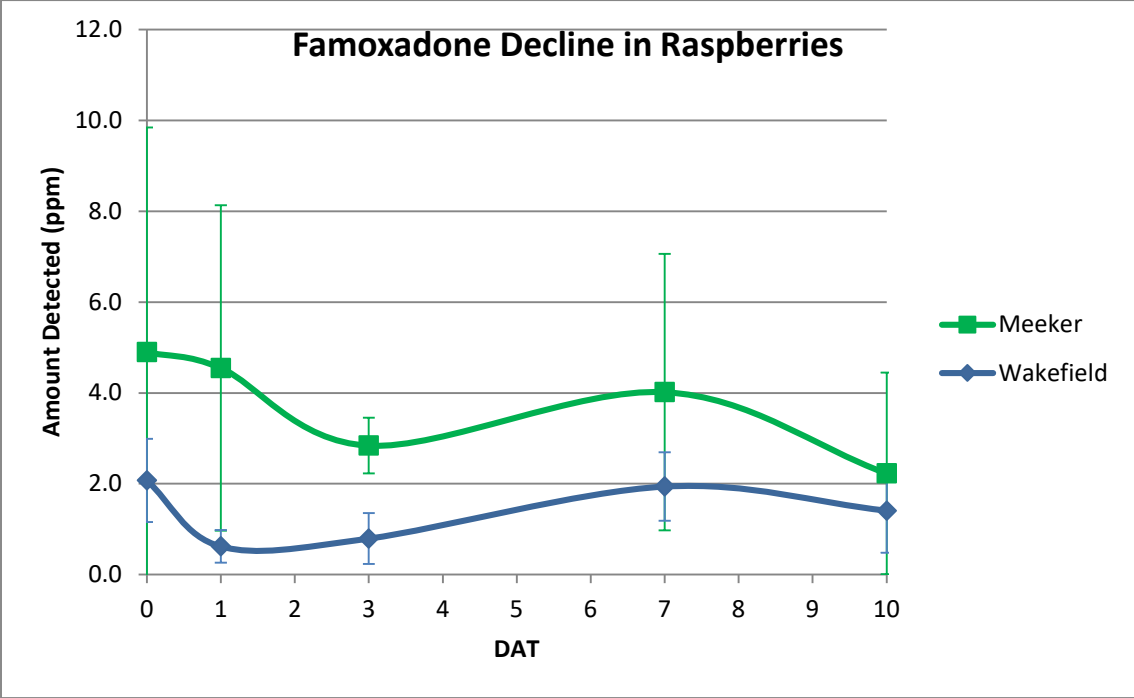
This is the first year of this project. All applications were made, one set to a Meeker planting and one set to a Wakefield planting. Both sets were in Whatcom County (west of Everson, and west of Lynden), were made on the same day, with same application equipment. Every thing possible was done to keep all variables to a minimum in order to have the only significant variable is variety. There were no problems with the applications. Products included in the trial were captan, iprodione, Pristine (boscalid, pyraclostobin), Switch (cyprodinil, fludioxinil), Tanos (famoxadone), Kenja (isofetamid), Luna Tranquility (fluopyram, pyrimethanil), Abound (anazoxystrobin), Elevate (fenhexamid) and Protexio (fenpyrazimine). Because Pristine, Switch and Luna Tranquility are dual package mixes residue decline curves for thirteen active ingredients are generated. Fruit samples were collected and shipped to the analytical lab with experience in doing this type of research. It is important to conduct this work for subsequent years and to try and do the work as similarly as possible between years.

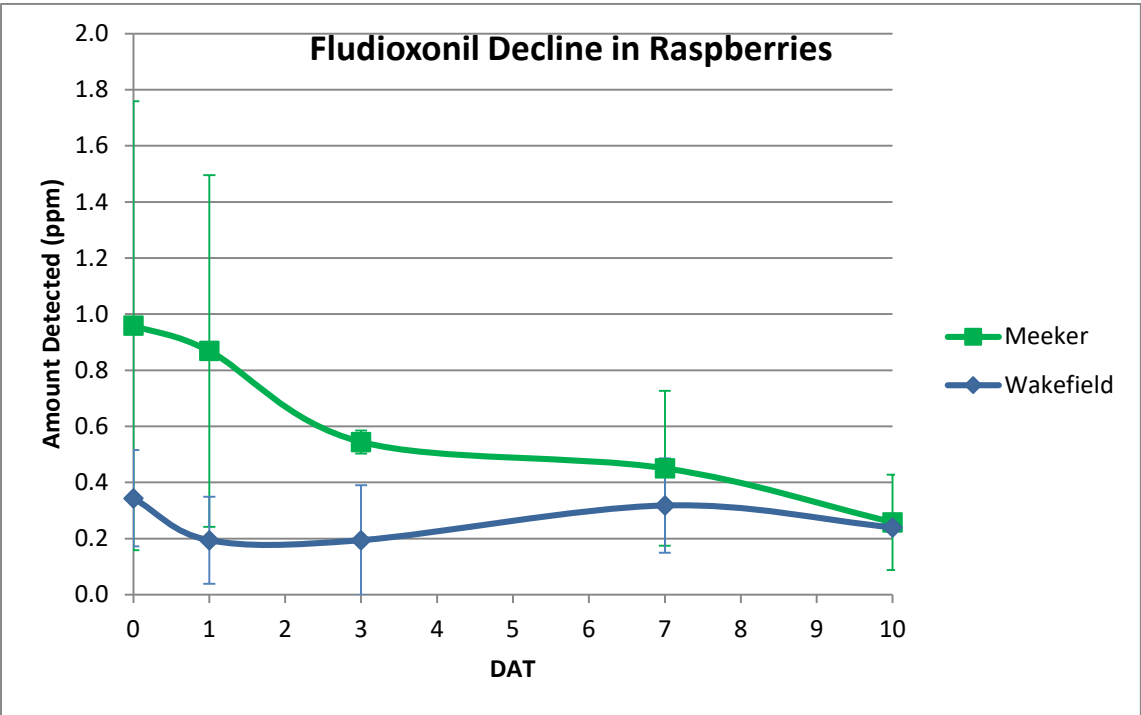
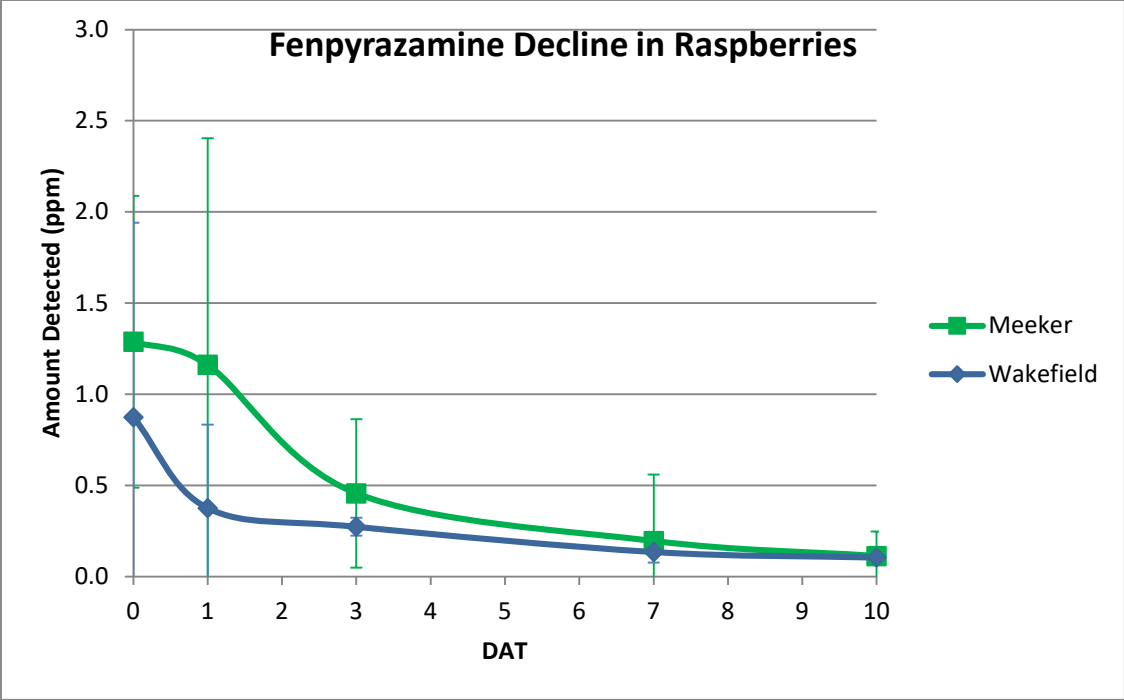
These results are quite remarkable. There is striking consistency in the relative slopes of residue decline curves for each active ingredients and for each variety. The consistency is something not seen in other residue decline curve projects and are likely due to minimizing variables. The second striking finding is that Meeker residues are always higher than Wakefield, for every fungicides. This difference is thought to be due to the way the fruit presents itself in each variety. Wakefield canopy is thought to be more pronounced than Meeker allowing less fungicides to reach the fruit. A second related possibility is that the Meeker site has much older plants (over 10 years) and has a weaker canopy. Thus the higher residues at the Meeker site could be more related to age of the canopy than the plant architecture. It is quite possible that both are factors.

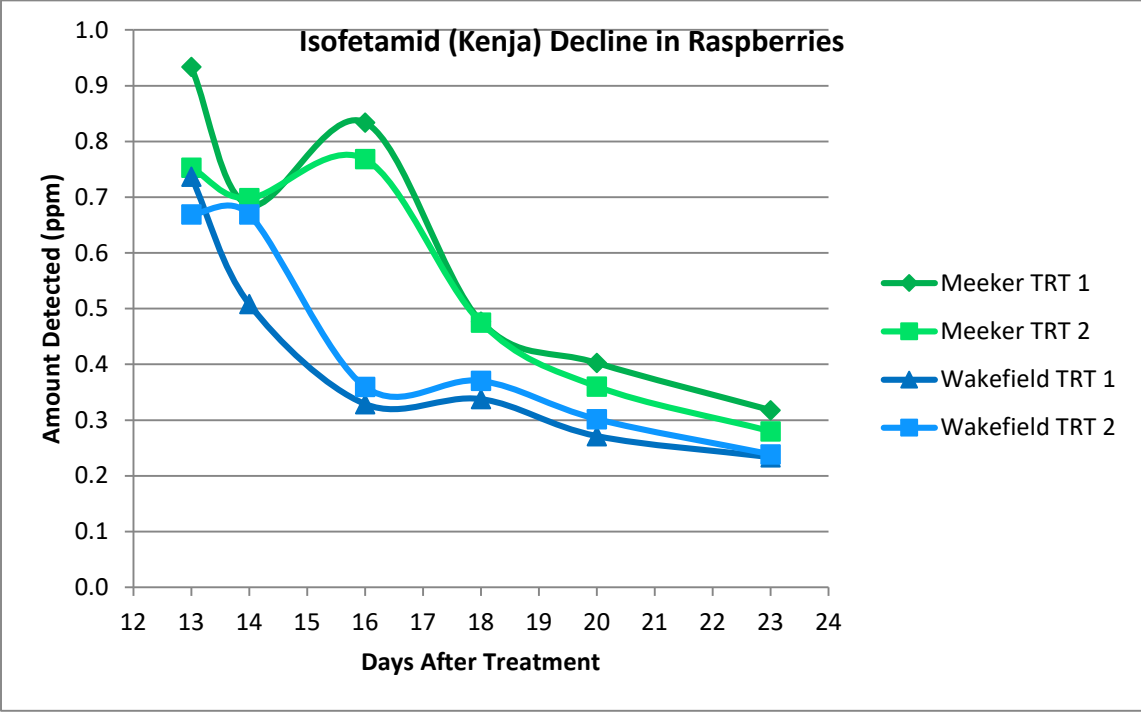
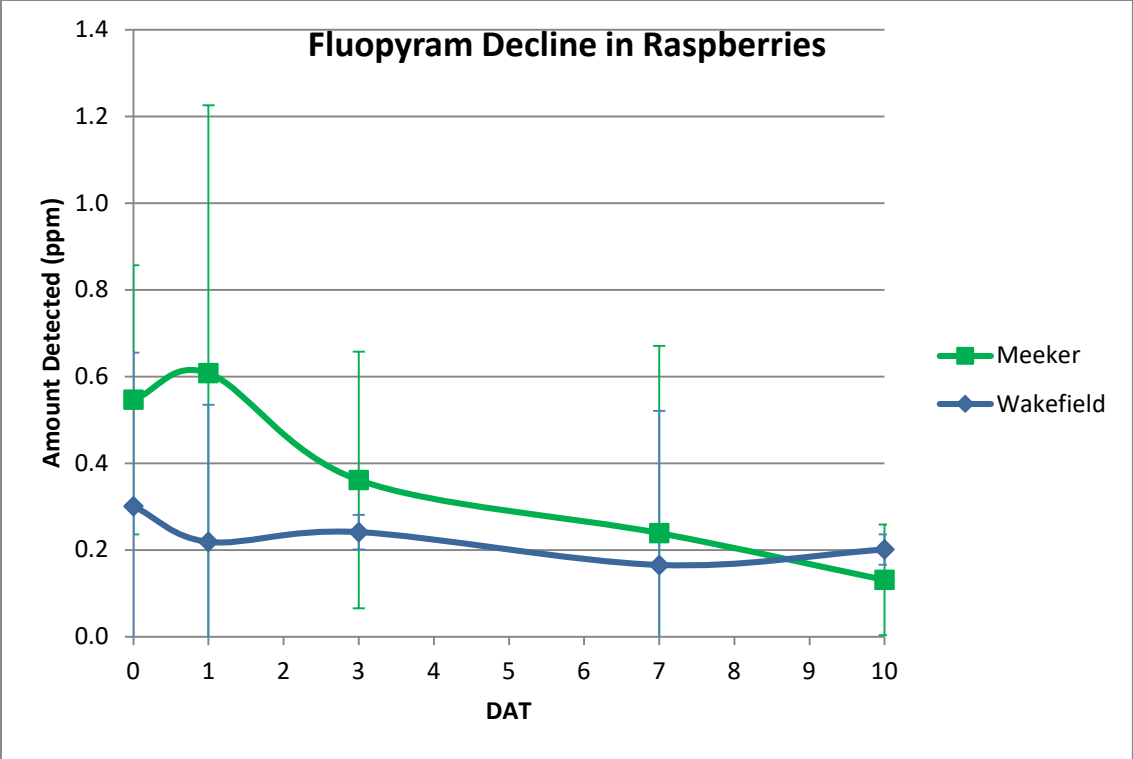
The value of these data are limited due to a single year's work has been completed. In order to use this data, an additional year's work is needed. If a second year's data is consistent with year one data, then it may be possible to conclude this project. However, if resources permit, a third year's work would increase the precision and confidence of the data set. The country MRL's used in this report were selected by industry representatives.

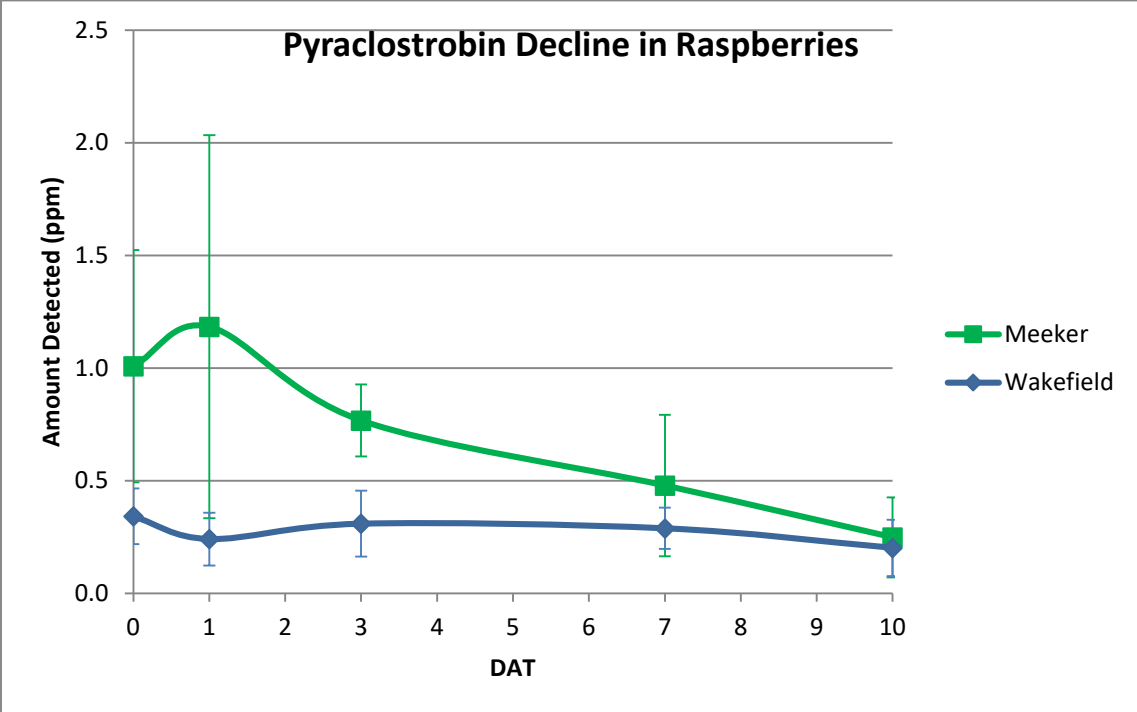
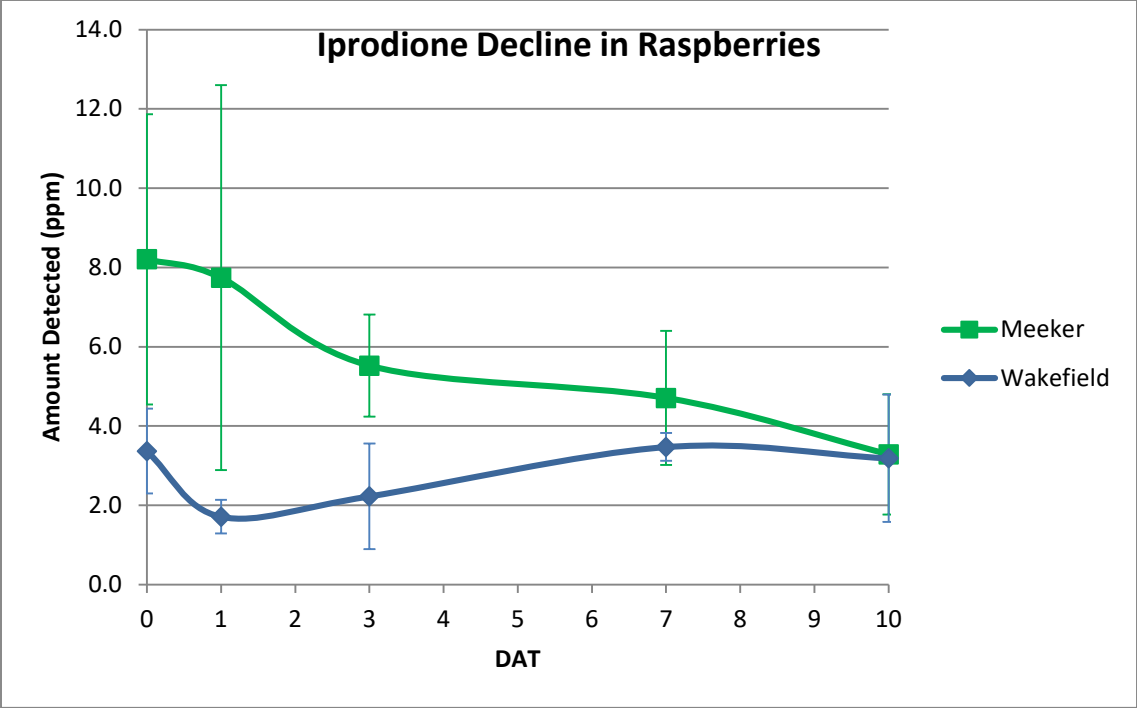


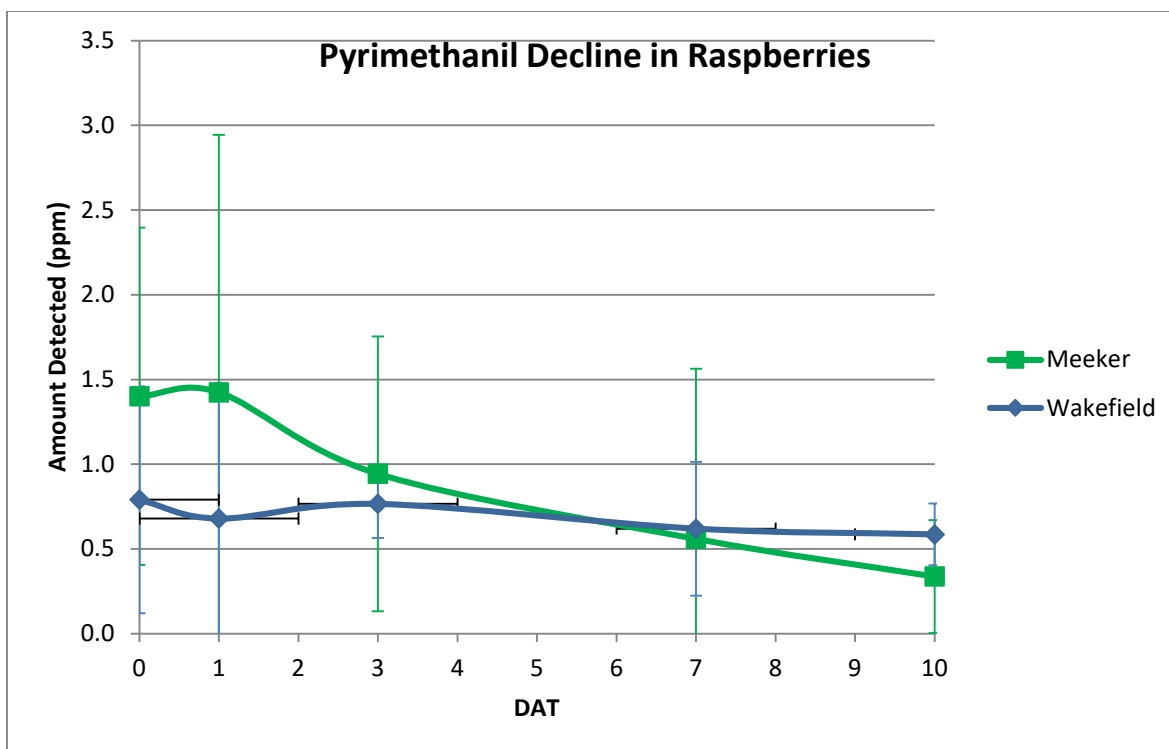












RASPBERRY	US	AU	CA	JA	KO	TA
Azoxystrobin	5	5	5	5	3	5
Boscalid	10	10	6	10	5	6
Captan	25	30	5	20	5	20
Cymoxanil	4	NT=0	4	4	0.5	1
Cyprodinil	10	10	10	10	1	3
Famoxadone	10	NT=0	10	10	2	0.01
Fenhexamid	20	20	20	15	15	0.01
Fenpyrazamine	5	NT=0	5	NT=0.01	2	NT=0
Fludioxonil	5	5	7	5	5	5
Fluopyram	5	NT=0	5	3	3	NT=0
Iprodione	15	12	25	5	30	5
Isofetamid	4*	NT=0	NT=0.1	NT=0.01	7	NT=0
Pyraclostrobin	4	4	3.5	3	1	3
Pyrimethanil	15	5	15	10	15	0.02

Project Proposal to WRRC

Proposed Duration: 3 Years

Project Title: Managing SWD in Red Raspberry with Reduced Insecticide Residues

PI: Alan Schreiber

Organization: Agriculture Development Group, Inc.

Title: Researcher

Phone: 509 266 4348 (office), 509 539 4537 (cell)

Email: aschreib@centurytel.net

Address: 2621 Ringold Road, Eltopia, WA 99330

Cooperators: Tom Walters-Walters Ag Research

Year Initiated: 2017

Current Year: 2017

Terminating Year: 2019

Total Project Request: Year 1 \$15,000 Year 2 \$15,000 Year 3 \$17,000

Other Funding Sources: At this time, there are no other funding sources for this project.

Description: Approximately 20% of Washington’s raspberry production is exported. The primary export markets are Canada and Japan, with smaller amounts going to other Pacific Rim countries. Not only is Washington red raspberry production increasing, the export of raspberry products are also increasing. On a per pound basis, exported raspberries have a higher value than domestic markets, making raspberry exports an attractive market. The Washington red raspberry industry has had shipments rejected due to MRL issues, however the bigger problem is that growers/exporters are being shut out of markets because they cannot meet MRL requirements of foreign markets. This project focuses on both of these problems.

The goal of this project is to develop spotted wing drosophila (SWD) management programs in red raspberry that will have reduced insecticide residues without a reduction in efficacy. SWD management programs would be front loaded with “harder” conventional insecticides and would switch to products that are exempted from tolerance or have residues that degrade more quickly. A successful outcome of this program would allow fruit produced under the low residue programs to be exported to markets that are currently challenged by low MRLs. Programs entirely composed of tolerance exempted products or products with residues that quickly decline have been developed for blueberries and blackberries. This project would take elements from those programs and combine them with elements of existing conventional raspberry SWD management programs.

Justification and Background: Most of the insecticides used for SWD by the Washington raspberry industry have longer preharvest intervals (Asana (7), carbaryl (7), diazinon (7), Danitol (3), Success (3), Delegate (3) and/or residues that do not degrade quickly such as malathion and cabaryl. The blueberry industry has developed an organic program for control of SWD that

appears to provide a level of control that is comparable to a conventional program. The level of control for this program is sufficient to produce large volumes (over 20 million pounds) of fresh early and mid season blueberries that have a SWD tolerance level that is lower than for processed raspberry. Based on last minute feedback from a raspberry industry, export quality processed raspberries may have a tolerance similar to that of fresh market blueberries (meaning a very low tolerance.) The organic blueberry program was developed in eastern Washington in later season blackberry, a crop that has high SWD pressure. Our thinking is that the program developed for blackberry could be adapted to red raspberry. The goal of this program is not to develop an organic program for raspberry but rather to use some of the products in that program that have proven more effective for SWD control and that are either exempt for tolerance or have residues that decline more quickly than the insecticides that are currently being used in raspberry SWD programs.

Based on the SWD efficacy program developed in caneberry for the organic blueberry industry, Entrust (which contains the same active ingredient as Success), Grandevo, Venerate, Veratran and Jet Ag have all shown significant efficacy against SWD. Grandevo, Venerate and Jet Ag are exempt from tolerances. Entrust/Success have tolerances that degrade quickly. Delegate, which is very closely related to the active ingredient in Entrust/Success, has not been included in the organic blueberry project as it is a conventional product but its residues are known to decline relatively quickly and could be included in this program. Delegate residues do not degrade as quickly as Success, but it has higher efficacy.

In this project, insecticides with longer PHIs and or having residues that do not decline sufficiently to meet MRLs would be used earlier in the program. In addition to giving these products time for their residues to decline and to come into compliance with PHI requirements, these products are thought to have greater efficacy and would “knock down” SWD populations.

This proposal was circulated among some members of the raspberry industry and received some “critical” reviews. The idea that this type of a program having export permissible insecticide residues and a level of control comparable to existing programs that rely on highly effective insecticides but that have MRL issues was challenged by members of the industry (i.e. Bajema, Berendsun and Midboe). I believe that a program can be developed that provides export quality processed raspberries without a significant sacrifice of efficacy. It took four to five years to do this for organic blueberries. I believe such a program could be developed in three years.

Relationship to WRRC Research Priority: This directly addresses the priorities “Management options for control of Spotted Wing Drosophila” and maximum residue limits.

Objectives: Develop SWD management options that will meet MRLs of key trading partners without reducing efficacy.

Procedures: A randomized complete block designed trial, with four replications will be overlaid the botrytis efficacy program. We would be using exclusively or almost exclusively products that have existing tolerances or are exempt from tolerance so this would not be crop destruct trial. There will be 8 treatments developed in consultation with raspberry industry representatives.

Examples of potential programs

1. untreated check.
2. malathion, Asana Danitol, Success+Grandevo, Success+Grandevo, Grandevo+Venerate , Grandevo+Venerate.
3. standard 1, standard 2, standard 3, Success+Venerate, Success+Venerate, Venerate+Veratran, Venerate+Veratran
4. standard 1, standard 2, standard 3, Delegate+Grandevo, Success+Grandevo, Venerate+Veratran
5. standard 1, standard 2, standard 3, Delegate, Venerate+Jet Ag, Grandevo+Jet Ag, Venerate+Jet Ag
6. Entrust, Grandevo + Jet Ag, Entrust, Grandevo+Venerate, Veratran+Jet Ag, Grandevo+Jet Ag
7. Standard program 1 – to be selected by the industry.
8. Delegate, Malathion, Actara/Tundra, Malathion, Malathion, Mustang Max, Mustang Max

Applications would be made roughly every seven days or when conditions or pest pressure would dictate. Prior to each application and seven days after the last application, a berry sample would be collected from each plot and analyzed for SWD larvae. Just prior to harvests late and at the end of the control program, samples would be collected and sent to an analytical lab for testing for pesticide residues.

It is noteworthy there is no evaluation of products novel to the berry industry being conducted on raspberries in the Pacific Northwest. If so directed by the WRRC, this program could be modified to include evaluating new conventional insecticides. This could include new modes of action, products considered more bee safe, shorter pre harvest intervals, lower residues or other components of an SWD use pattern that may be of value to the industry.

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:	2017	2018	2019
Salaries	6,000	6,000	8,000
Operations	6,000	6,000	6,000
Travel	1,500	1,500	1,500
Benefits	1,500	1,500	1,500
Total	\$15,000	\$15,000	\$17,000

These funds would be primarily be used to cover the time of Schreiber and Walters spent on the project. It would cover the applicator’s time, tractor/equipment usage, product purchases and other costs. An estimated \$3,000 of operations would be used to cover the cost of laboratory analyses. All travel costs are related to traveling to the site and/or meeting with industry representatives.

**Washington Red Raspberry Commission
Progress Report 2016**

Project No: 3061-4303

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

Personnel: Tobin L. Peever, Associate Professor, Dalphy Harteveld, Post-Doctoral Associate, Olga Kozhar, PhD student

Reporting Period: January 2016 to November 2016

Project objectives:

- Determine the dynamics of flower and fruit infection by *Botrytis cinerea* throughout the growing season
- Relate infection events to environmental conditions
- Population genetics study of *Botrytis* populations on different small fruit crops

Accomplishments: Completed a second season of sampling of raspberry flowers and fruit during April-August 2016 in order to assess timing of *Botrytis cinerea* infection. Determined relationship between *Botrytis* infection and weather conditions (clear relation to rainfall events and air temperature) and raspberry flower and fruit development. Sampled and isolated approximately 1000 *B. cinerea* isolates from different geographic regions and small fruit hosts for population genetics study. Preliminary population genetics analysis of 4 populations of *B. cinerea* from raspberry and blueberry fields.

Progress report

1. Dynamics of flower and fruit infection by *Botrytis cinerea* throughout the growing season

Eight developmental stages of raspberry flowers and fruit were identified and sampled throughout the two seasons (2015 and 2016) for studying the infection process of *Botrytis* and disease development (Fig. 1).



Fig. 1. Developmental stage of flowers and fruit

Raspberry flowers and fruit were sampled from four raspberry fields during April-August 2015 and 2016 to assess the timing of *B. cinerea* infection. In 2015, both fields were untreated with fungicides and were located in different areas (one in Whatcom County, one in Skagit County). In 2016 the same untreated field in Skagit County was sampled again and one conventional raspberry field treated with fungicides was sampled in Skagit County. Overall 5360 (1440 in 2015, and 3920 in 2016) flowers and fruit were sampled from fields untreated with fungicides, and 940 flowers and fruit were sampled from fungicide treated field in 2016 (Table 1).

Table 1. Sampling locations and total amount of samples used for *B. cinerea* recovery

Year	County	Field ID	Fungicides sprays	Total # of samples	Mistigation Yes/No	Surface disinfestation*	Medium**	# of samples
2015	Skagit, WA	F1.1	No	720	No	+	1	210
					No	-	1	210
					Yes	+	1	150
					Yes	-	1	150
2015	Whatcom, WA	F2	No	720	No	+	1	180
					No	-	1	180
					Yes	+	1	180
					Yes	-	1	180
2016	Skagit, WA	F1.2	No	3920	No	-	2	490
					No	-	1	490
					No	+	2	490
					No	+	1	490
					Yes	-	2	490
					Yes	-	1	490
					Yes	+	2	490
2016	Skagit, WA	F3	Yes	940	No	+	2	245
					No	-	2	245
					No	-	1	245
					No	+	1	245

* "+" - surface disinfested (10s 70% ethanol, 1m 1% bleach, 1 m rinse with water - 3 times), "-" - non-surface disinfested

** "1" - 1.5% water agar; "2" - Botrytis selective medium modified from Kritzman and Netzer (1987)

Percent *B. cinerea* recovery was not significantly different between two years and locations of untreated fields. Due to the nature of culturing assays recovery of fungi from stage 8 (overripe fruit) was not sampled in 2016. In 2015 high level of colonization of raspberry flowers by other fungi was observed. To eliminate potential competition from other fast growing fungi that might affect the level of *Botrytis* recovery, the use of a *Botrytis* selective medium was added in 2016.

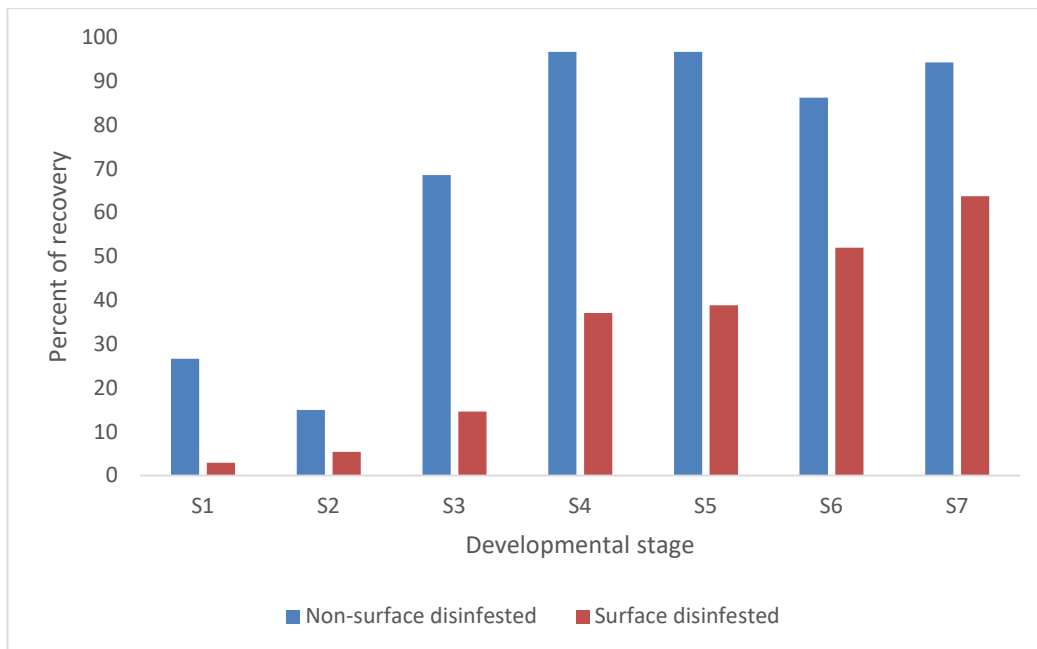


Figure 1. *B. cinerea* recovery from non surface-disinfested and surface-disinfested samples by developmental stage.

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). This suggests that infection of flowers may be the most important source of fruit rot later in the season. *B. cinerea* recovery from surface disinfested samples increased through the season with a significant increase at the open flower stage (S3) relative to closed green buds (S1) or half-open flowers (S2) (Fig. 1). Recovery from surface-disinfested (SD) open flowers (S3) remained low throughout the season, and *Botrytis* was recovered from only 15% of SD open flowers during an 8 week flowering period. Recovery from surface-disinfested samples increased as the fruit developed and ripened and reached average of 64% at the mature fruit stage (S7). Recovery of *Botrytis* from stage S3 onwards throughout the season was above 60% and reached close to 100% from green to mature fruit (S4-S7)). These results do not support the hypothesis that flower infection and latent infections by *Botrytis* are the sole route of raspberry infection and responsible for the majority of raspberry fruit rot. To eliminate the potential effect of the host defense system at flower stages S1 to S3 preventing *Botrytis* recovery from these samples, samples from stages S1-S3 were surface disinfested, air dried in a laminar flow hood, and fresh frozen at -80C. Frozen samples were cultured on modified *Botrytis* selective medium (Kritzman and Netzer 1987). No differences in rates of recovery of *B. cinerea* recovery from frozen flowers comparing to fresh were observed (Fig.1). This suggests that host defenses system did not influence our rates of recovery of *B. cinerea* from flowers and that internal infection of flowers is low. The significant difference in rate of recovery of *Botrytis* between flowers and fruit suggests that infection occurs continuously throughout the season rather than only at flowering. We hypothesize that these secondary infection events that are occurring throughout the season might be more important in inciting fruit rot later on the season compared to latent infection of flowers.

In 2016, total of 940 samples from conventional field were sampled throughout the season and cultured on two media as described above (Table 1). Results showed high levels of *Botrytis* recovery from samples obtained from conventionally managed (ie. fungicides applied) field during the season, and at some developmental stages appeared to be higher than rates of recovery from the untreated plot (Fig. 2).

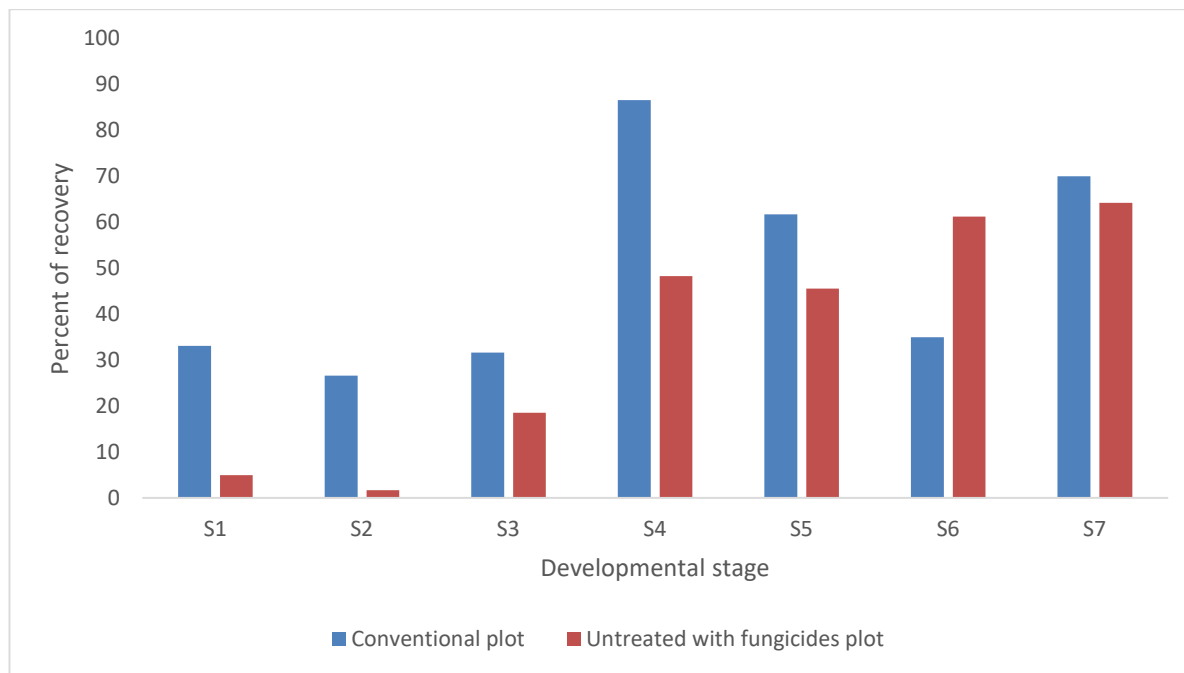


Figure 2. Percent *B. cinerea* recovery from samples from fungicide-treated and non fungicide-treated plots in 2016.

Based on the information from the grower, the conventional field wasn't heavily managed during the sampling season 2016, with only a few fungicide sprays being applied based on calendar schedule. Lack of significant difference of the fungal recovery from treated and untreated plots proves the importance of scheduling the fungicide treatments based on the pathogen biology and ecology rather than on calendar basis.

To further investigate the role of flower infection in fruit rot development, each sampling event, in addition to 10 sampled flowers used for culturing and recovery of fungi in the laboratory, 5 newly opened flowers were marked and their development was monitored in the field throughout the season. This allowed us to track the timing of fruit development of each cohort of flowers and relate *B. cinerea* recovery from samples in the laboratory to the phenology of raspberry plant. Figure 2 shows a timeline of floral development over 13 sampling events with each developmental stage shown in colored bars. Numbers on bars indicate percent of *B. cinerea* recovery from carpels of surface disinfested samples. Newly opened flowers were sampled and tracked for 8 weeks (represented by 8 multicolored bars). When the developmental stages of samples from each sampling event were aligned with the phenology of the plant, the data showed no correlation was observed between *Botrytis* recovery from flowers at the beginning of the season and from fruit that developed from these flowers few weeks later. This provides another piece of evidence to suggest that *Botrytis* colonization of ripe fruit does not originate exclusively from floral infections.

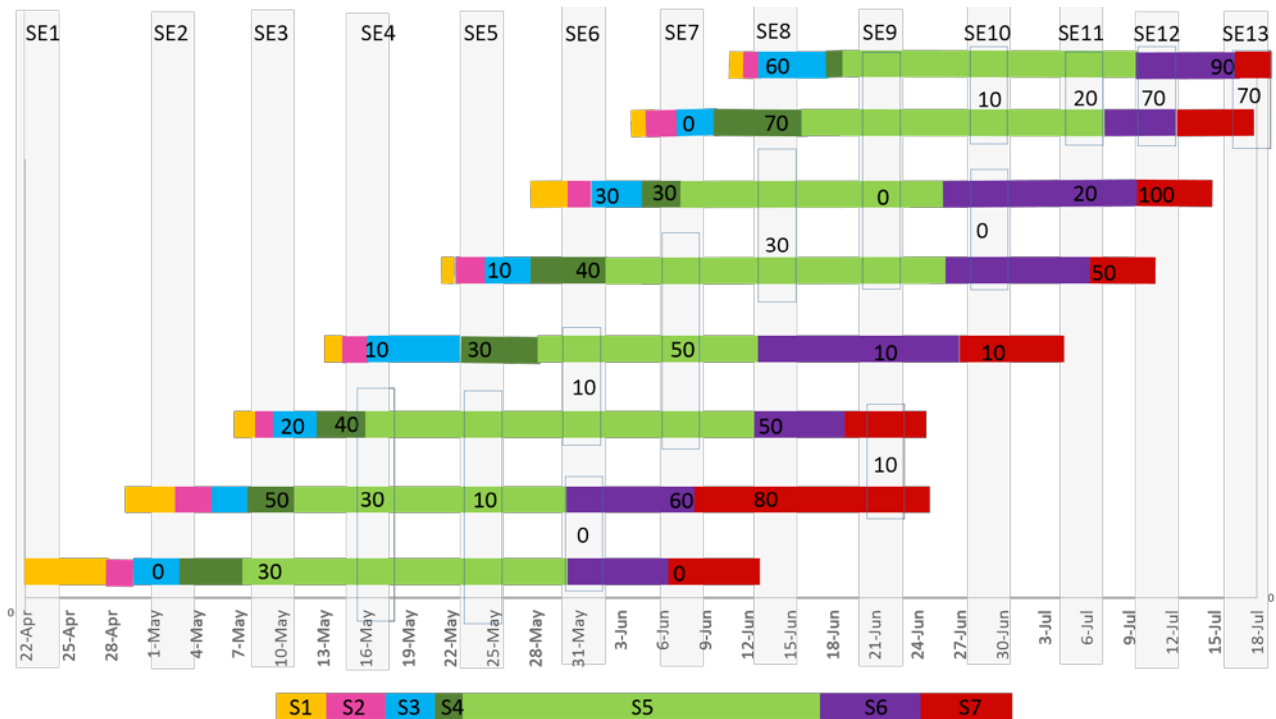


Figure 3. Phenology of raspberry observed in 2016. Each color represents a different developmental stage (shown on legend across the bottom). Numbers indicate percent of *B. cinerea* recovery from the samples obtained at each sampling event. Sampling events (SE) are indicated across the top.

Other researchers have suggested that flowers and immature fruit were less susceptible to *B. cinerea* infection compared mature fruit due to the presence of high levels of phytoalexins (Langcake 1981, Bavaresco et al. 1997), proteinaceous inhibitors of fungal cell wall degrading enzymes, and polygalacturonase inhibiting proteins (PGIPs) in the host tissue that restrict ability of the fungus to infect the host (De Lorenzo et al., 2001). Increased defense responses of the plant may explain the low level of colonization of flowers of red raspberry. Additional factors such as low level of inoculum (Bristow et al. 1986) and of external fungal colonization in the beginning of the season (low recovery from NSD samples, Fig.1), and relatively short period of flowering (2-3 days, blue bars in Fig. 3) may also play role in poor flower infection. Taken together, these results suggest that flowers may not be the primary infection route of *Botrytis* leading to Botrytis fruit rot of raspberry in the Pacific Northwest. Rather it appears that *Botrytis* colonization of raspberry is a continuous process starting with the female parts of the flowers but progressing on to other floral parts throughout the season and possibly also direct secondary infection of ripe fruit later in the season.

Colonization by other fungi

Based on morphological identification in culture, the majority of fungi recovered on water agar (WA) belonged to four fungal genera. These included *B. cinerea*, *Trichoderma* spp., *Cladosporium* spp., *Phomopsis* spp. The identification to genus was confirmed by sequencing of the nuclear ribosomal internal transcribed spacer region (ITS) of two representatives of each morphological group. Percentage of recovery of these fungal genera fluctuated through the season (Fig.4, Fig.5), and also differed between surface-disinfested and non surface-disinfested samples. In spring and early summer *Trichoderma* spp. had the highest level of recovery from non-surface disinfested samples (Fig.4), and the percent of recovery declined while the season progressed. Colonization of non-surface disinfested

samples by *B. cinerea* increased though time and reached nearly 100% starting from week 7. *Trichoderma* spp. were reported as an important biological control of plant pathogens (Vos et al. 2014), including control of *B. cinerea* on strawberry (Freeman et al. 2004). Such a negative correlation between recovery of *B. cinerea* and *Trichoderma* spp. (Fig. 4) may indicate competitive relationships between these fungi on raspberry flowers and the potential significance of *Trichoderma* spp. for biological control of *B. cinerea* early in the season. The recovery level of *Cladosporium* spp. from non-surface disinfested samples remained low reaching the highest 40% in week 7. Higher recovery of *Cladosporium* spp. was indicated from surface-disinfested samples, reaching 60% in week 7 and week 12. From week 9 to 11 the recovery of *Phomopsis* spp. from surface-disinfested samples increased to 60-80%, while recovery of other fungi decreased during that period.

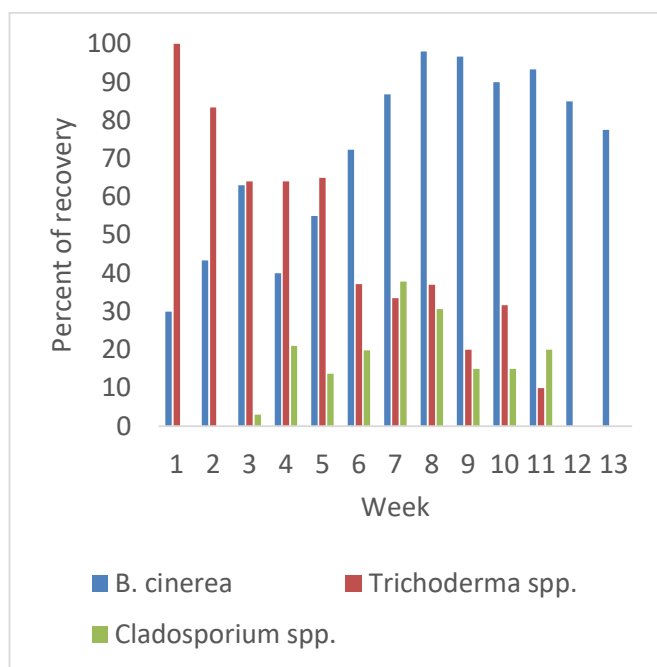


Figure 4. Fungal recovery from non-surface disinfested samples on water agar

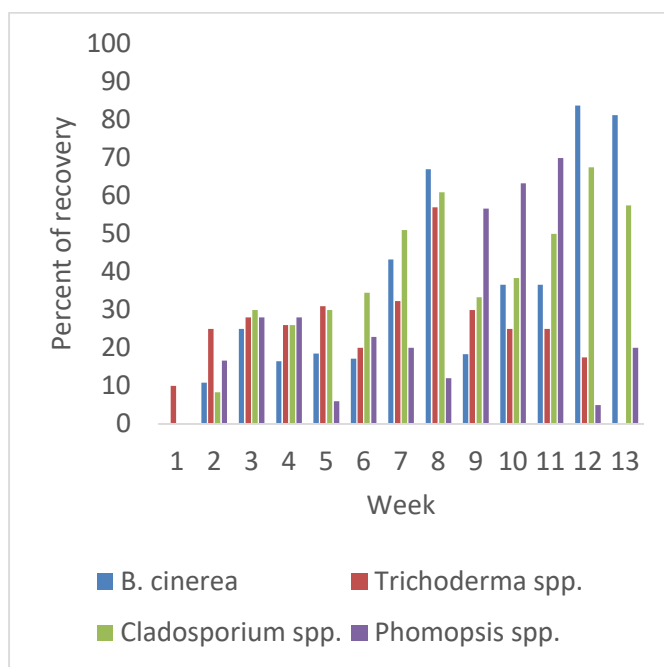


Figure 5. Fungal recovery from surface disinfested samples on water agar

This may also indicate competitive relationships between these fungi. *Phomopsis* spp. were not recovered from non-surface disinfested samples.

Potential route of *B. cinerea* infection of red raspberry

In order to analyze potential pathways of *B. cinerea* infection of red raspberry flowers and fruit, all surface disinfested samples were also divided into morphological parts (Fig. 6) and each part was separately incubated on water agar (WA) or Botrytis selective medium for analysis of fungal recovery.

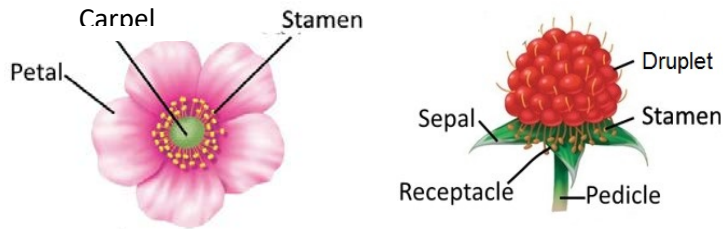


Figure 6. Morphology of raspberry flowers and fruit

Among 14% of sampled flowers (S3) that had recovery of *B. cinerea*, female part of the flower (carpel) was most frequently colonized by the fungus (Fig.7). This finding supports earlier observations and attempts to connect fungal infection of flowers to pollination (Viret et al. 2004, McClellan & Hewitt 1973, Pezet & Pont 1986, Dashwood & Fox 1988, McNicol et al. 1985). Pollination results in the flower's ovaries becoming part of each drupelet with drupelets forming the compound fruit of red raspberry (Fig. 6). As fruit develops through the season, other flower parts such as stamens sepals senesce but remain attached to the fruit (Fig. 6). Senescent tissue is hypothesized to be a suitable substrate for colonization of a necrotrophic fungus such as *B. cinerea*. As flower and fruit development proceeded in our experiment stamens, sepals and receptacles became increasingly infected by *Botrytis* while rates of recovery from female flower parts decreased (Fig. 7).

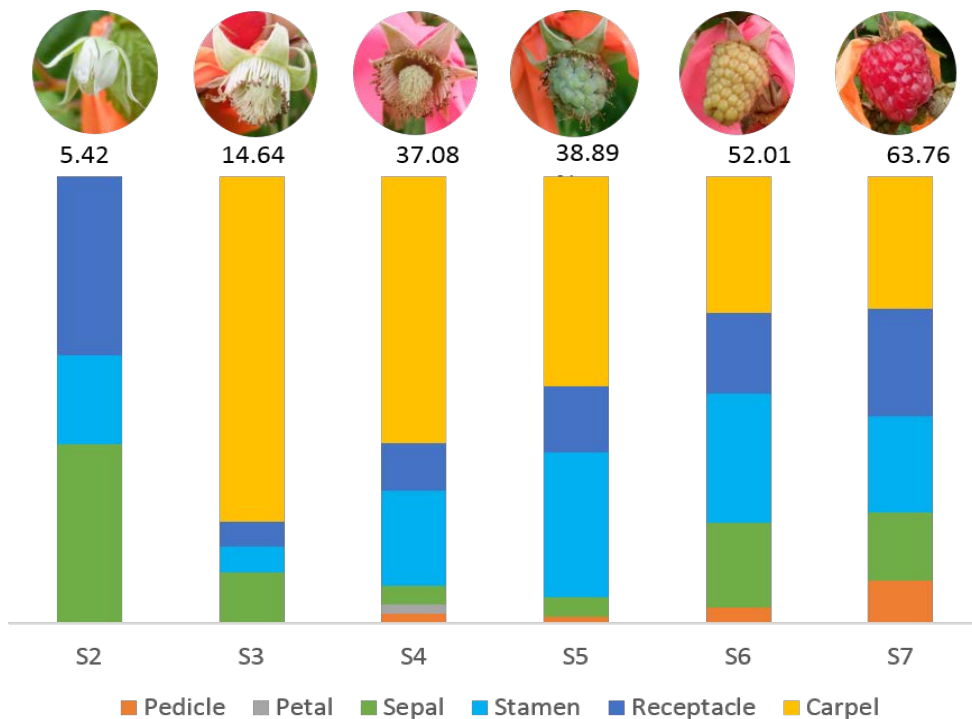


Figure 7. Level of *B. cinerea* recovery from different floral parts and fruit of red raspberry at each stage of floral/fruit development. Vertical bars represent the proportion of recovery of *Botrytis* from each part among samples where *Botrytis* was recovered. Photos along top show developmental stages while numbers show proportion *Botrytis* recovery

Figure 8 shows percent recovery of *B. cinerea* from the female parts (carpels) of sampled flowers compared to other floral parts and fruit during the growing season. At the beginning of the season

when there was no fruit present yet, 100% of the fungal recovery that was obtained from samples appeared from carpels of open flowers (S3), half-developed green fruit (S4) and green fruit (S5). Only after week 5 did we start to recover *Botrytis* from other morphological parts (shown in dashed bars in Fig. 8). Recovery of *B. cinerea* from floral and fruit parts in addition to carpels increased as the season progressed and fruit started to ripen.

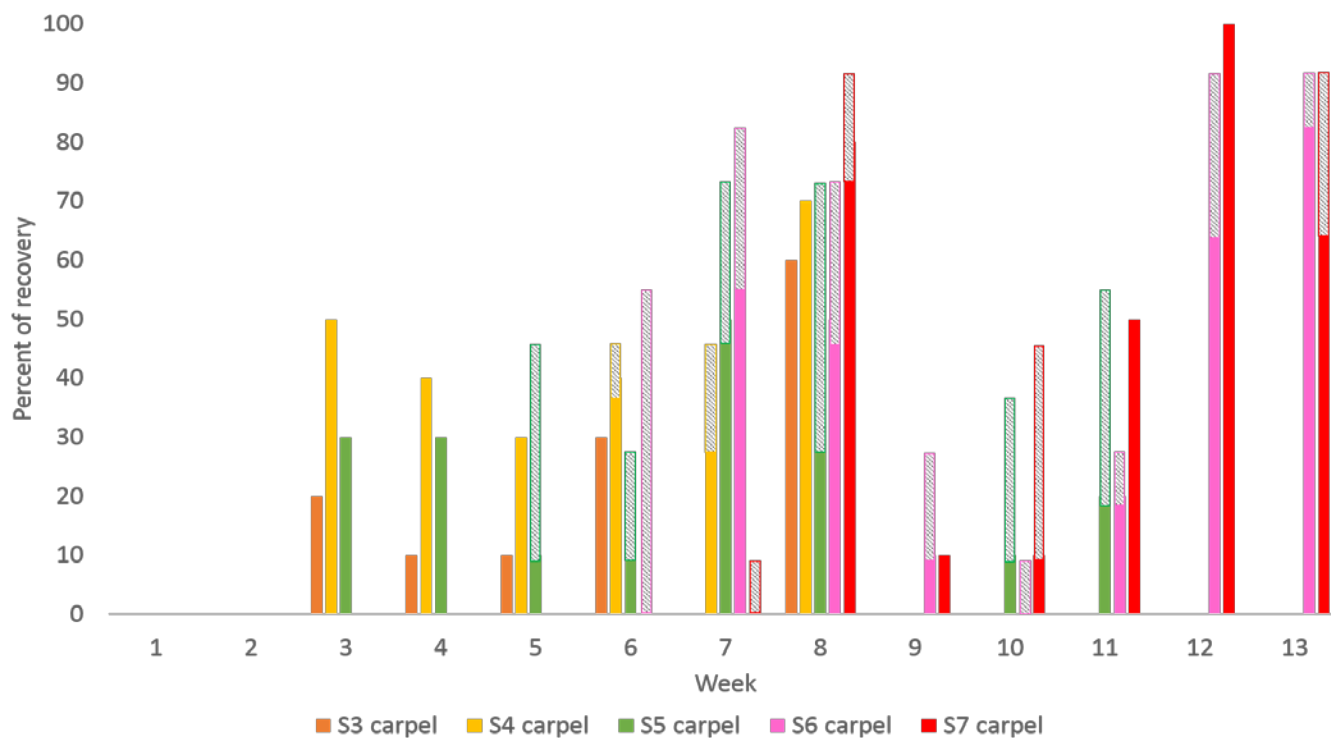


Figure 8. *B. cinerea* recovery from carpels (solid color bars) and other flower/fruit morphological parts (hatched bars), related to developmental stage of raspberry flowers and fruit during the growing season. Stages S1 and S2 are not shown due to absence of fungal recovery from carpels and overall low *Botrytis* recovery from these stages.

Monitoring of *B. cinerea* recovery during season showed the overall increase of the fungal recovery level from carpels in later stages of fruit (S6, S7) that does not appear to be related the recovery level from carpels earlier in the season (Fig. 3, Fig.8). We would expect to see a strong correlation between *Botrytis* recovery from flowers and the fruit that develops from those flowers if all or the majority of fruit infection was initiated from early flower infection. Our data does not support this hypothesis as we saw colonization of additional floral parts later in the season that remain attached to fruit and an overall increase on *Botrytis* colonization as the season progressed. Therefore, it appears that early flower infection by *Botrytis* may not be the most likely major source of infection for subsequent *Botrytis* fruit rot infections of red raspberry.

2. Effect of environment on *Botrytis* infection events

Botrytis spp. require free water for conidial germination, germ tube growth and penetration of host

tissue and the duration of surface moisture is widely used as a factor in modeling disease epidemics (Williamson et al, 1995). In order to study the effect of the environment on the infection process, each non fungicide-treated field was divided into two plots – one with mistigation system imitating rain and one without artificial mistigation. Four environmental data loggers were placed in fields (one per each plot with or without mistigation system) and measured air temperature, relative humidity and leaf wetness duration. Rainfall data was obtained from Agweathernet stations 48 (Lynden) and 49 (WSU Mount Vernon). To identify influence of environmental factors on infection incidence and development, the daily averages of each climatic factor were calculated for the period starting from April, 28 till August, 15 in 2015 and from April, 22 till July, 18 in 2016.

Mistigation was started at 4-6pm and run until 8-10am the following day. This provided leaf wetness periods of approximately 14-16 hours per day which should have been sufficient for *B. cinerea* infection and disease development (16 hours of leaf wetness was previously suggested by Bulger et.al 1987 using artificial inoculation of strawberry flowers and fruit as minimum for *B. cinerea* infection). However in two years of experiment there was no significant difference in recovery of *B. cinerea* between mistigated and non-mistigated plots. This suggests that under field conditions presence of optimal leaf wetness duration is not enough for the disease development and indicates presence of confound effects of several environmental factors on Botrytis fruit rot incidence and severity.

In order to better understand the influence of weather and environment on *B. cinerea*, weather data from May to July 2012 was compared with weather data from May to July 2015. A high incidence of gray mold was observed in 2012 while 2015 was a year of very low disease pressure. The two data sets showed no significant differences in relative humidity (average 80%). They did, however, show a significant difference in overall rainfall as well as frequency of rainfall events (Fig. 9). Both data sets indicated air temperature beyond the optimal level (optimal level for *B. cinerea* infection is suggested 20C, Bulger et al. 1987). However in 2012 air temperature was still lower (average 25C) than in 2015 (average 30C). This comparison supports observations on strawberry flowers reported earlier (Jarvis 1964, Xu et al. 2000). It also suggests a difference in behavior of *B. cinerea* in the laboratory and in the field as previous laboratory experiments showed a decrease in infection with temperature above 25C and the same prolonged leaf wetness duration (Ciliberti et al. 2015), suggesting that temperature and moisture are both crucial for infection process.

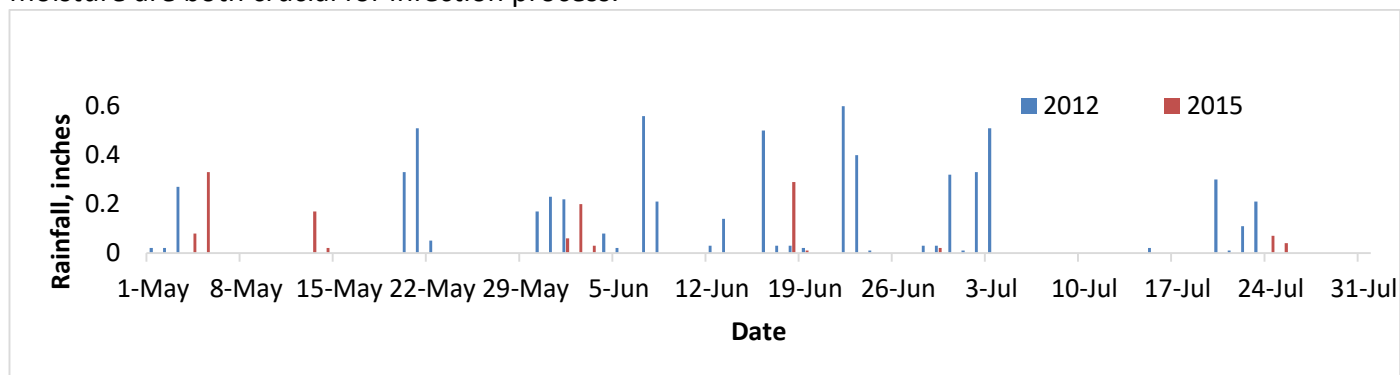


Figure 9. Rainfall level comparison between 2012 and 2015

Analysis of rainfall data during sampling period in 2016 resulted in discovery of more clear relationship between rain events, developmental stage of the host, and level of *B. cinerea* recovery from flowers and fruit (Fig. 10). The level of the fungal recovery from non-surface disinfested samples increased with

fruit development (stages S5, S6 and S7). From week 3 rainfall slightly increased that might have favored *B. cinerea* infection of red raspberry fruit (S4, S5), however level of recovery from flowers remained low. Starting from week 6 recovery from fruit S6 and S7 was higher than from other developmental stages suggesting that under the same rain conditions immature and mature fruit are more susceptible to the fungal infection than other stages. On week 8 the highest level of *B. cinerea* recovery was observed at all stages present in the field at that time. That week had the largest amount of rainfall (0.24 inches) that together with high colonization of flowers and fruit on the surface (100% of recovery from NSD samples) created favorable conditions for the infection.

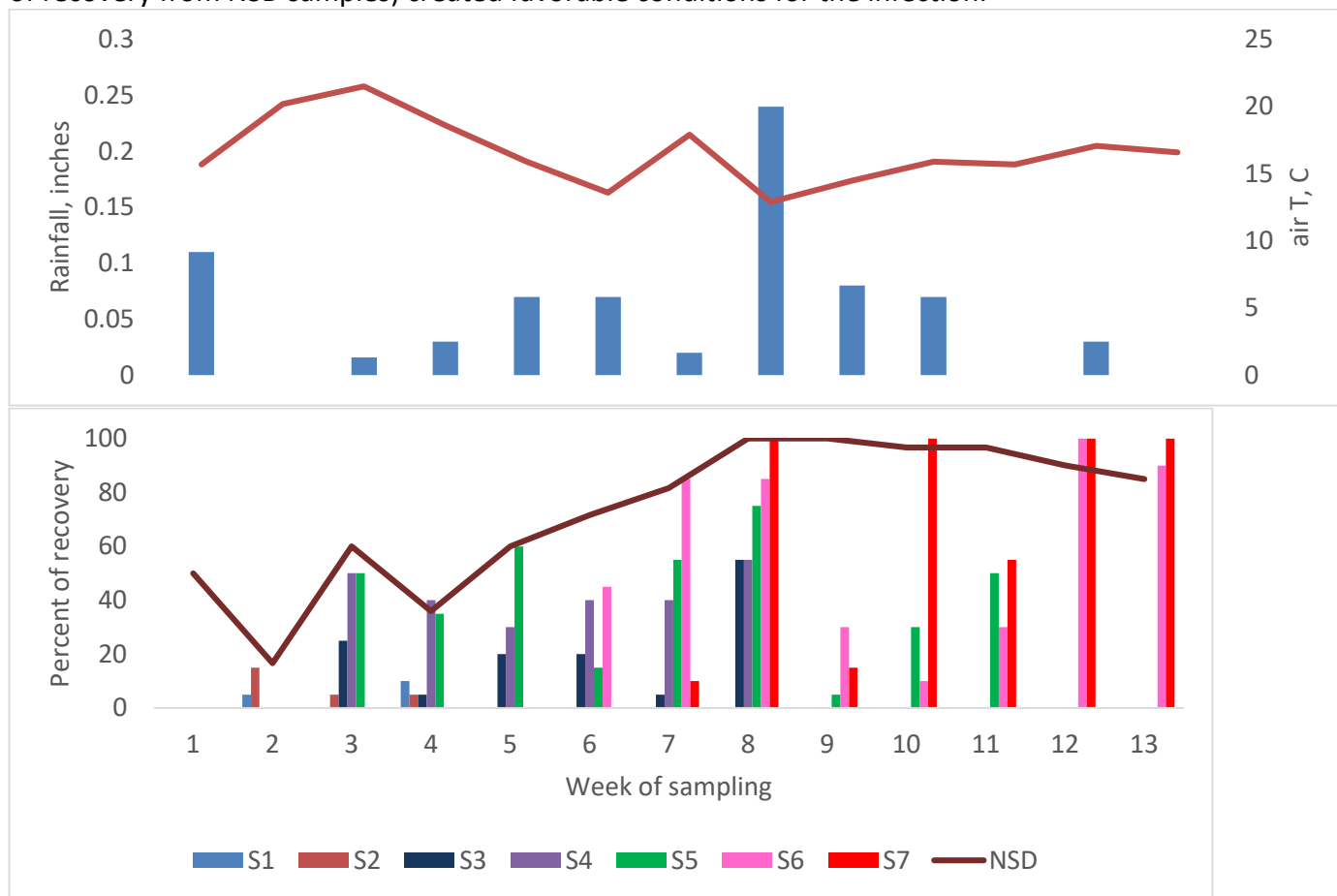


Figure 10. Association of *B. cinerea* recovery with rainfall events during season 2016. Colored bars represent different developmental stages of raspberry flowers and fruit. NSD line shows recovery level from non-surface disinfested samples. Top graph – bars represent rainfall, line shows maximum air temperature in Celsius.

Interestingly, that level of *B. cinerea* recovery on week 9 rapidly decreased from surface-disinfested samples. That might have happened due to decrease of flower/fruit developmental stages in the field (the plants stopped flowering) or inability of the fungus to infect the fruit or recover from the samples under the excess of free water. For the fungus to be able to sporulate a rapid drop in relative humidity and increase in temperature in early morning are required for releasing conidia from conidiophores into air (Jarvis 1962). On week 9, 100% leaf wetness period extended till 19 hours per day keeping plants wet almost all the time.

Principal component analysis of environmental factors indicated confound effects of some factors on

the level of *B. cinerea* recovery. Leaf wetness duration and air temperature are significant for both colonization of non-surface disinfested and surface disinfested samples. For surface disinfested samples relative humidity is also significant (Fig. 11 and Fig. 12).

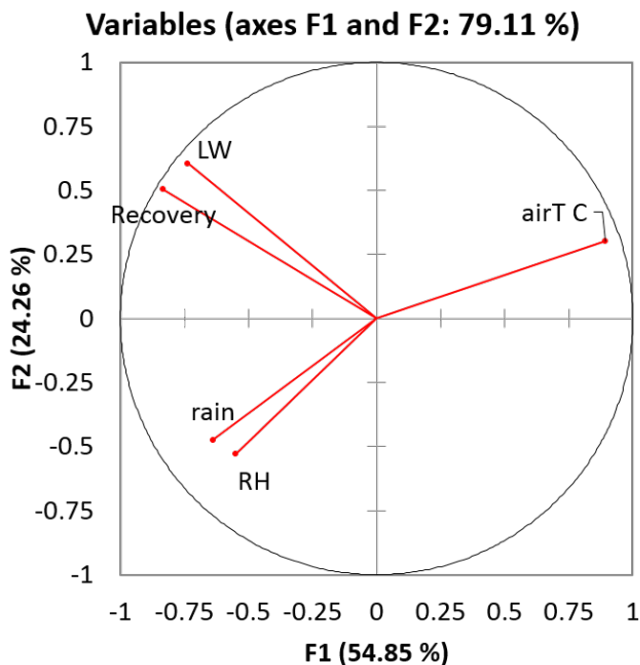


Figure 11. Principal component analysis of weather factors affecting percent of recovery of *B. cinerea* from **non-surface disinfested samples**

Significant factors:

- Leaf Wetness (LW)
- Air temperature (airT C)

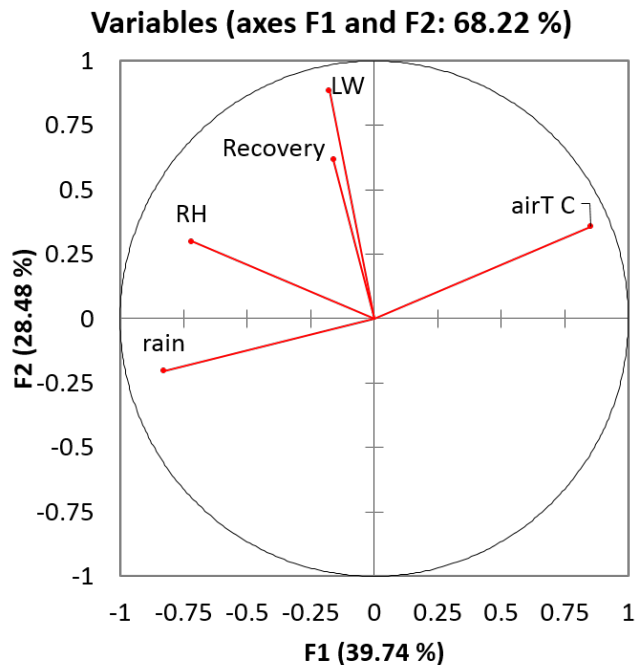


Figure 12. Principal component analysis of weather factors affecting percent of recovery of *B. cinerea* from **surface disinfested samples**

Significant factors:

- Leaf wetness (LW)
- Air temperature (airT C)
- Relative humidity (RH)

Conclusions

A low overall level of colonization of raspberry flowers by *Botrytis cinerea* was observed during flowering and early fruit development. The level of *B. cinerea* recovery increased through time as fruit developed and peaked in mature fruit. This increased recovery of *B. cinerea* from mature fruit may be due to increased susceptibility of ripe fruit to *B. cinerea* and secondary infection events later in the season. Alternatively, the low recovery of *Botrytis* early in the season may have been due to our inability to recover the pathogen even though present in a latent stage. In the early stages of flower and fruit development, carpels showed the highest level of *B. cinerea* recovery. As the season progressed, other floral parts such as sepals and stamens became colonized by *Botrytis* and these infections may play role in secondary infections of fruit later in the season. Analysis of weather data and its influence on the recovery of *B. cinerea* from the samples showed that rainfall events play an important role for the fungal infection, especially when fruit is present. Leaf wetness duration and air temperature were both significant factors for *B. cinerea* infection under field conditions as has been shown previously. A high level of colonization of raspberry flowers and fruit by fungi other than

Botrytis was observed indicating that *Botrytis* may compete with these other fungi for infection courts. This information may be useful in identifying members of the raspberry fungal flora with potential as biological control agents for *B. cinerea* on red raspberry.

3. Population genetics of *B. cinerea*

Botrytis cinerea was sampled from 43 commercial fields of five small fruit crops (raspberry, blueberry, strawberry, blackberry, currants) as well as from several wild blackberry populations during 2015 and 2016. Sampling was carried out in Skagit, Island, and Whatcom counties of WA, Linn, Clackamas, OR as well as the Fraser Valley of British Columbia, Canada from June to October 2015 and 2016. Attempts were made to identify fields of the same crop found at different spatial scales (i.e., meters apart to miles apart) as well as adjacent fields of different small fruit crops in more than one location. Adjacent fields of different small fruit crops were identified in order to determine host specificity of *B. cinerea* within the dispersal distance of the pathogen.

During fall 2015, several DNA extraction protocols (phenol/chloroform, CTAB, MOBio PowerSoil DNA Isolation Kit, MP Bio Quick DNA Isolation Kit) were tested for their ability to extract high quality DNA from fresh mycelium of *B. cinerea*. Mycelium of *B. cinerea* was grown on potato dextrose agar (PDA) plates overlain with cellophane in order to eliminate liquid culture and lyophilization steps used previously. Taking into account DNA yield, purity and time needed for performing the extractions, the MOBio Power Soil DNA isolation Kit used with a tissue disruptor was chosen. Modifications made to the Power Soil DNA Isolation Kit extraction method included:

- Growing mycelium on an agar medium with lower levels of carbon sources such as half-strength PDA) increased yield of DNA
- Decreasing the wet weight of mycelium used to 40 micrograms
- Adding an additional washing step with ice cold 70% ethanol to prevent adherence of polysaccharides to spin column substrate

A total of 12 microsatellite (SSR) markers were used to study the genetic diversity of *B. cinerea* populations. The markers set included 8 SSR markers that were developed and published by Fournier et al., 2002, and 4 new SSR markers that were developed by colleagues from the USDA Horticultural Crops Research Unit in Oregon State University. SSR markers were combined into three multiplex PCR reactions and genotyped using AB 3730 capillary DNA sequencer and analyzed with GeneMarker® software.

A minimum of 20 isolates was obtained from each of the 43 locations and approximately 1000 *B. cinerea* isolates were sampled in total. A preliminary estimate of genetic diversity of *B. cinerea* populations infecting different small fruit hosts was obtained by genotyping isolates with the SSR markers. DNA was extracted from 20 isolates of *B. cinerea* sampled from each of two blueberry, one raspberry field and 19 isolates from another raspberry field in Island and Whatcom counties (Table 2). The raspberry and blueberry fields were located in a close proximity to each other in each county. Preliminary data analysis revealed that all 12 loci were polymorphic in all populations. The number of alleles per locus varied from 4 to 22 with 61 private alleles among 12 loci (from 1 to 7 per locus). Among 79 isolates examined from the four populations, 66 multilocus haplotypes were identified (Blueb_I – 18, Raspb_I – 20, Blueb_W – 12, Raspb_W – 16). The highest genotypic diversity (based on

H, G and lambda values, Table 2) was observed in population Raspb_I, while population Blueb_W had the lowest genotypic diversity. Population Blueb_W also had the lowest value of evenness that can be explained by presence of 8 isolates with the same multilocus genotype among observed isolates. Gene diversity Hexp within populations varied from 0.45 in blueb_W (blueberry population in Whatcom County) to 0.86 in Raspb_I (raspberry population in Island County) (Table 2).

Table 2. Genotypic diversity of *B. cinerea* populations

Population	N	MLG	G	A	Lambda	E ₅	Hexp	r _D
Blueb_I	20	18	16.67	23	0.94	0.96	0.83	0.12*
Blueb_W	20	12	5.13	5	0.81	0.58	0.45	0.48*
Raspb_I	20	20	20.00	38	0.95	1.00	0.86	0.11*
Raspb_W	19	16	15.70	8	0.94	0.95	0.75	0.05*
Total	79	66	42.46	61	0.98	0.73	0.82	0.09*

N – number of individuals, MLG – number of multilocus genotypes, G - Stoddart and Taylor’s Index of MLG diversity, A – number of private alleles, lambda - Simpson’s Index , E₅ - Evenness, Hexp - Nei’s unbiased gene diversity, r_D – Index of association (*P<0.001) (Calculated in PoppR 2.2.0, Kamvar et al. 2014)

The index of association test (r_D) (Table 2) allowed us to reject the hypothesis of random mating in every population sample suggesting that *B. cinerea* populations from these four fields are clonally reproducing and the sexual stage of the fungus is absent.

Genetic distance between populations based on estimation of Nei’s genetic distance (Table 3) showed greater differentiation between *B. cinerea* populations between fields located in different counties despite the host type. *B. cinerea* populations from the two different hosts located next to each other had much lower genetic differentiation (0.217 between Blueb_I and Raspb_I and 0.351 between Blueb_W and Raspb_W) (Table 3, highlighted in yellow). Lower genetic differentiation was observed between populations from adjacent fields possibly due to migration of *B. cinerea* between fields or to a common recent origin of both populations.

Table 3. Pairwise genetic differentiation among populations using Nei’s genetic distance

blueb_I	raspb_I	blueb_W	raspb_W	
0.000				blueb_I
0.217	0.000			raspb_I
0.661	0.878	0.000		blueb_W
0.267	0.394	0.351	0.000	raspb_W

Analysis of Molecular Variance (AMOVA, Table 4) supported the results of the pairwise genetic differentiation showing that these four populations are significantly genetically differentiated by geography rather than by host type suggesting a lack of host specificity.

Table 4. Pairwise genetic differentiation (φst) among populations from different counties based on 999 permutations (*P <0.05).

Blueb_I	Raspb_I	Blueb_W	Raspb_W	
				Blueb_I
0.02				Raspb_I
0.09*	0.12*			Blueb_W
0.21*	0.09*	0.02		Raspb_W

The *Erg27* gene is the molecular target of the fungicide fenhexamid (tradename “Elevate”) used for gray mold control in small fruit crops and mutations in this gene confer resistance to fenhexamid (Fillinger et al. 2008, Amiri, Peres, 2014; Grabke 2013; Albertini, Leroux, 2004,). Approximately 1000 base pairs of *Erg27* was amplified and sequenced for 40 *B. cinerea* isolates that were both sensitive and resistant to fenhexamid. These isolates were obtained from the fields described above and previously screened for fenhexamid sensitivity *in vitro*. Analyses of these sequence data are in process. Amino acid substitutions in 30 different codons were observed. Among them, 18 substitutions were previously reported with different levels of sensitivity to fenhexamid. Fifteen of these were associated with a high level of insensitivity and 3 were associated with moderate level of insensitivity (Amiri, Peres, 2014; Grabke 2013; Albertini, Leroux, 2004). Thirteen of the isolates examined had an amino acid substitution from serine to phenylalanine in the 1st position of codon 412 that was previously demonstrated to confer a high level of resistance to fenhexamid (Albertini, Leroux, 2004).

Conclusions

Preliminary analysis of the population structure of *B. cinerea* isolates sampled from four small fruit fields, including isolates from adjacent fields of raspberry and blueberry collected in two different counties (Island and Whatcom, WA), showed that populations are genetically diverse, clonal, and have a high number of private alleles (alleles unique to a location). Significant genetic differentiation was detected between populations from different counties, but not between populations from different hosts located next to each other suggesting lack of host specificity. Local differentiation of populations based on neutral markers suggests restricted mobility of the pathogen in space. Preliminary analysis of *Erg27* target gene for fungicide fenhexamid showed high level of polymorphism in these populations and contains mutations previously reported in other areas, as well as polymorphic sites that have not been described previously.

Publications: No publications have resulted from this work to date.

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2017 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

New Project Proposal

Proposed Duration: 1 year

Project Title: Evaluation of FRAC Group 7 (SDHI) fungicides for control of *Botrytis* fruit rot of red raspberry in WA

PI: Tobin L. Peever

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Address: P.O. Box 646430

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

Year Initiated: 2017 **Current Year:** 2017 **Terminating Year:** 2018

Total Project Request: \$20,610

Other funding sources: Washington State Commission on Pesticide Registration, Northwest Center for Small Fruits Research, WA Specialty Crop Block Grant

Description

Several recent studies have demonstrated that newly developed FRAC group 7 (SDHI) fungicides may provide control of boscalid-resistant strains even though these fungicides have similar chemistry and mode of action to boscalid (Amiri et al. 2014, Olaya et al. 2016, Sierotzki and Scalliet 2013). Specific outputs of this project will include a detailed study of the sensitivity of WA *Botrytis* isolates from raspberry in WA to six new SDHI fungicides, determination of cross resistance relationships among these fungicides and to boscalid, and determination of the ability of field rates of these fungicides to control boscalid-resistant strains using a recently developed fruit protection assay (D. Dutton and T.L. Peever, *unpublished*). We are specifically interested in determining if these new SDHI fungicides are able to control *Botrytis* isolates that are resistant to boscalid in WA and thus provide growers with new disease control options. This study, coupled with in-field efficacy testing of the same products by other scientists, will provide important baseline data necessary for the future registration of these fungicides for use in WA raspberry production and provide WA raspberry growers with additional and critically needed disease control options.

Justification and Background

Washington produces 90% of the US processed raspberry supply with approximately 66 million lbs harvested and a farm gate value of 80 million dollars in 2015. About 10,000 acres are currently in raspberry production and yields average approximately 4 tons per acre. Despite intensive fungicide applications used to control gray mold, caused by *Botrytis cinerea*, it is estimated that fruit losses and downgrades in fruit quality exceed 25% of the harvestable fruit due to incomplete disease control in disease-conducive years. This represents an average loss of 1 ton per acre equating to approximately \$1500 per acre. Gray mold is the most economically

significant disease affecting raspberry production in WA and aggregate losses are approximately 15 million dollars per year. Of the five major fungicides registered for raspberry gray mold control in WA, resistance has been documented to three of them (boscalid, fenhexamid, and cyprodinil) severely compromising disease control options for WA raspberry growers (T.L. Peever, *unpublished*). New fungicides with different modes of action are urgently needed in WA raspberry production.

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) Determine the ability of new FRAC group 7 (SDHI) fungicides to control WA *Botrytis cinerea* strains resistant to boscalid using laboratory sensitivity assays
- 2) Determine cross resistance relationships among the new FRAC Group 7 (SDHI) fungicides and boscalid in WA strains of *B. cinerea*
- 3) Determine the ability of the new FRAC Group 7 (SDHI) fungicides to control boscalid-resistant isolates using a fruit protection assay

Procedures

- 1) **Determine the ability of new FRAC group 7 (SDHI) fungicides to control WA *Botrytis cinerea* strains resistant to boscalid using laboratory assays**

To date, approximately 600 isolates of *Botrytis cinerea* have been sampled from WA raspberry from 2014-2016 and these isolates are currently in long-term storage in my laboratory. All of these isolates have been screened for sensitivity to five fungicides commonly used to control gray mold of raspberry in WA including fenhexamid, cyprodinil, boscalid, fludioxonil, and iprodione. Approximately 70% of these isolates are resistant to boscalid. Mycelial growth assays on discriminatory concentrations of technical grade or formulated fungicides is a common method to assay fungicide sensitivity in *B. cinerea* (Leroch et al 2013, Weber 2011) and this method has been used to evaluate fungicide sensitivity in *B. cinerea* in WA in my lab since 2014. Quantitative estimates of sensitivity and the frequencies of isolates in each sensitivity category will be obtained for six new SDHI fungicides including adepidyn, isofetamid, fluopyram, penthiopyrad, fluxapyroxad, and solatenol. We will initially estimate EC₅₀ (effective concentration to inhibit 50% growth) values for randomly selected isolates from different raspberry fields in WA on agar amended with different concentrations of technical grade fungicide. Once a mean EC₅₀ estimate for this sample is obtained, a single discriminatory dose of each fungicide near the population mean EC₅₀ will be used to estimate sensitivity phenotypes among a much larger sample of isolates. Approximately 200 isolates sampled from raspberry in WA during 2015 and 2016 that are sensitive and resistant to boscalid will be screened against six SDHI fungicides to provide an estimate of baseline sensitivity to each of the new SDHI fungicides and to determine if any of these new fungicides are able to inhibit boscalid-resistant isolates in WA.

1) Determine cross resistance relationships among the new FRAC Group 7 (SDHI) fungicides in WA strains of *B. cinerea*

Although the newly developed SDHI fungicides mentioned above are in the same chemical class as boscalid (one of the components of “Pristine” along with pyraclostrobin), and have the same mode of action, recent studies (Olaya et al. 2016, Amiri et al. 2014) have suggested that these new SDHIs may be able to control boscalid-resistant isolates due to differences in their molecular targets. Resistance to boscalid is conferred by several mutations in the target site molecule, succinate dehydrogenase in the fungal mitochondrion (Sierotzki and Scalliet 2013). The effectiveness of the new SDHIs in controlling boscalid-resistant isolates will depend upon the types of mutations present in *Botrytis* populations where the fungicides are used (Sierotzki and Scalliet 2013). In order to predict the potential effectiveness of these new SDHI fungicides for use against gray mold in WA raspberry, we need to understand if they are able to control boscalid-resistant isolates that are currently common in WA raspberry fields. We will also determine the cross-resistance relationships among the six new SDHI fungicides and boscalid by growing selected isolates on agar medium amended with each fungicide as described above. Sensitivity to each fungicide will be estimated as proportion growth of the same isolates on un-amended agar as described above. Such cross-resistance data will be critical for the design of effective resistance management strategies to extend the useful life of these new fungicides once they are registered.

2) Determine the ability of the new FRAC Group 7 (SDHI) fungicides to control boscalid-resistant isolates using a fruit protection assay

The mycelial growth assays described above allow an estimate of the *in vitro* sensitivity to each fungicide but do not allow us to predict whether an isolate that is less sensitive to a particular fungicide is resistant to field rates of that fungicide under field conditions. In order to relate fungicide sensitivity phenotypes that we observe on agar medium to the predicted field performance of each fungicide, a raspberry fruit protection assay has been developed (D. Dutton and T.L. Peever, *unpublished*). This assay has allowed us to determine that the isolates that are highly insensitive to boscalid, cyprodinil and fenhexamid in WA are also resistant to field rates of fungicide under field conditions. This gives us confidence in classifying isolates as sensitive or resistant. *Botrytis* isolates displaying different fungicide sensitivities to the new SDHIs will be selected for testing on raspberry fruit treated with each fungicide. Fruit will be treated with field rates of formulated fungicides, then inoculated with a known quantity of *Botrytis* inoculum, incubated and disease allowed to develop. Quantitative estimates of gray mold affecting the fruit will be recorded for 5 days after inoculation allowing us to determine if isolates with different fungicide sensitivities are controlled by field rates of each fungicide. We will be particularly interested in determining which, if any, of the new SDHIs are able to control boscalid-resistant strains on fruit when applied at field rates.

Anticipated Benefits and Information Transfer

This research addresses a critical need in the raspberry industry for effective disease control options for *Botrytis* gray mold of raspberry in WA. Widespread resistance to three of five main fungicides exists in *Botrytis cinerea* populations infecting small fruit in WA and this has severely compromised disease control options available to growers. Screening several new Group 7 fungicides against the boscalid-resistant strains currently found in WA will allow us to assess the potential effectiveness of these chemicals for use in controlling gray mold in

raspberry. Should we find that some or all of these chemicals are effective, this data will then be used to support new registrations of these fungicides for use in raspberry. The availability of additional new disease control options for WA raspberry growers will reduce reliance on a limited number of chemistries and allow the implementation of more effective resistance management strategies.

References

Amiri, A, S.M. Heath and N. Peres. 2014. Resistance to fluopyram, fluxapyroxad, and penthiopyrad in *Botrytis cinerea* from strawberry. *Plant Disease* 98: 532-539.

Leroch, M., C. Plesken, R.W.S. Weber, F. Kauff, G. Scalliet and M. Hahn. 2013. Gray mold populations in German strawberry fields are resistant to multiple fungicides and dominated by a novel clade closely related to *Botrytis cinerea*. *Applied and Environmental Microbiology* 79: 159-167.

Olaya, G., R. Linley, K. Edlebeck and T. Harp. 2016. ADEPIDYN fungicide: Cross resistance patterns in *Alternaria solani*. Abstract presented at the 2016 Annual Meeting of the American Phytopathological Society, July 30 to August 3, Tampa, FL.

Sierotzki, H. and G. Scalliet. 2013. A review of the current knowledge of resistance aspects for the next-generation succinate dehydrogenase inhibitor fungicides. *Phytopathology* 103: 880-887.

Weber, R.S. 2011. Resistance of *Botrytis cinerea* to multiple fungicides in Northern German small fruit production. *Plant Disease* 95: 1263-1269.

Budget:

Salaries ¹	12,216
Time-slip	0
Operations (Goods & Services) ²	2,500
Travel ³	1,500
Meetings	0
Other	0
Equipment	0
Benefits ⁴	4,394
Total	\$20,610

*Budget approved by Julie Lang at WSU Johnson Hall Business Center

Budget Justification:

¹ 0.25 FTE post-doctoral salary - Dr. Dalphy Harteveld
² Travel of post-doc to professional meeting
³ Lab supplies including petri dishes and agar
⁴ Benefits rate = 35.06%

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:				
Peever	WA Raspberry	24207	01/01/16 to 12/31/16	15	Biology and Control of Botrytis fruit rot of red raspberry
Peever	WA Blueberry	22984	01/01/16 to 12/31/16	15	Biology and Control of Mummy Berry and Botrytis fruit rot of blueberry
Peever	WSCPR	28353	01/01/16 to 12/31/16	15	Biology and Control of Mummy Berry and Botrytis fruit rot of blueberry
Peever	WSCPR	23780	01/01/16 to 12/31/16	15	Biology and Control of Botrytis fruit rot of red raspberry
Peever and Grunwald	Northwest Center for Small Fruits Research	104738	10/1/204 to 09/30/2017	15	Host specificity and gene flow of fungicide resistance alleles among <i>Botrytis cinerea</i> populations infecting small fruit in the US Pacific Northwest
Peever	BC Blueberry Council	54238	05/15/2014 to 03/15/2017	10	Fungicide resistance of <i>Botrytis cinerea</i> infecting raspberry and blueberry in BC

Pending:					
Peever	WA Raspberry	20230	01/01/17 to 12/31/17	15	Evaluation of FRAC Group 7 (SDHI) fungicides for control of Botrytis fruit rot of red raspberry in WA (this proposal)
Harteveld & Peever	WA Blueberry	21892	01/01/17 to 12/31/17	15	Host resistance of blueberry mummy berry
Harteveld & Peever	WSCPR	23989	01/01/17 to 12/31/17	15	Host resistance of blueberry mummy berry
Peever	WSCPR	26442	01/01/17 to 12/31/17	15	Evaluation of FRAC Group 7 (SDHI) fungicides for control of Botrytis fruit rot of red raspberry in WA
Peever, Harteveld & Dossett	BioAg	20200	01/01/17 to 12/31/17	10	Resistance to Blueberry Mummy Berry
Schilder & Peever et al.	USDA-SCRI	2800000	09/01/17 to 08/30/21	10	Developing a Weather-based Decision Support Tool for Management of Blueberry Fruit Diseases

2017 WASHINGTON BLUEBERRY COMMISSION RESEARCH PROPOSAL

Project Title: Laboratory Equipment for Small Fruit Pathology at NWREC

Year Initiated 2016 **Current Year** 2016 **Terminating Year** 2017

Principal Investigator: Tobin L. Peever
Organization: Department of Plant Pathology, Washington State University
Phone: 509-335-3754
Email: tpeever@wsu.edu
Address: P.O. Box 646430
City/State/Zip: Pullman, WA 99164

Co - PI: Dalphy O.C. Hartevelde
Organization: Department of Plant Pathology, Washington State University
Phone: 360-848-6157
Email: doc.hartevelde@wsu.edu
Address: 16650 State Route 536
City/State/Zip: Mt Vernon, WA 98273

Total Project Request: \$ 42,250

Justification and Background:

Blueberry production in the Pacific Northwest (PNW) is challenged by several fungal diseases that cause significant economic damage including mummy berry, Botrytis gray mold, anthracnose, and Alternaria fruit rot. Emerging diseases are a constant threat to the industry and threaten the economic viability of blueberry production in WA. Over the past three years, WSU's Small Fruit Pathology program has addressed several of these blueberry disease research priorities as well as research priorities of the WA Red Raspberry Commission. In order to continue to address research questions critical to the industry, enhanced research capacity is required. For the past two years, WSU's Small Fruit Horticulture program under the leadership of Dr. Lisa DeVetter has graciously shared her research lab and research equipment with our research program. A new lab has recently been opened up for the Small Fruit Pathology Program at NWREC and we began the process of moving in last week (Dec 1, 2016). Critical pieces of equipment, identified in this proposal, are needed for this space and are required for the WSU's Small Fruit Pathology program to meet the research needs of the industry.

Relationship to WBC Research Priorities:

Grant proposals submitted to the WBC over the past three years from WSU's Small Fruit Pathology Program have addressed several of the industry's top priorities.

Objective:

- 1) Equip newly acquired laboratory facilities at WSU-NWREC in Mount Vernon with state-of-the-art equipment to facilitate small fruit pathology research

Procedures: NA

Describe how this research will benefit Washington blueberry growers:

Purchase of the laboratory equipment proposed here will be of direct benefit to WA blueberry growers because it will build research capacity in small fruit pathology at the NWREC and allow the small fruit pathology program to continue to deliver research relevant to grower priorities.

References: NA

Budget:

	2017
Salaries	
Time-Slip	0
Operations (goods & services)	0
Travel	0
Meetings	0
Other	0
Equipment ^a	42,250
Benefits	0
Total	42,250

^a See table below for detailed breakdown of cost for each item

Detailed Budget:

Item	Cost
Culture incubator (light/temp)	\$7,000 ^a
Laminar flow (sterile) hood	\$10,000
Analytical scale	\$1,500
Dissecting microscope	\$7,000 ^b
Compound microscope	\$17,000 ^b

^a Estimate provided by Percival, USA

^b estimates provided by Nikon, USA

Washington Red Raspberry Commission Progress Report Format for 2016 Projects

Project No: MA-WRRC2016-001

Title: Development of novel disease management methods for fruit rots of raspberry

Personnel: Virginia Stockwell with Gayle McGhee and Brenda Shaffer. USDA-ARS HCRU, Corvallis, Oregon

Reporting Period: Year 1: 2/13/2016 to 2/14/2017

Accomplishments:

- We found that the composition of microbial communities (bacteria, yeasts, and fungi) isolated from raspberry floral buds and open flowers was very diverse and represented several genera, whereas microbial communities isolated from green fruit and ripe fruit were much less diverse. It is unclear if the observed decrease in diversity is due to fruit tissues supporting growth of different microorganisms than floral tissues or if the decrease is due to abiotic factors, such as environmental conditions and/or agricultural inputs.
- The patterns in the incidence of colonization of tissues and the succession patterns of microbial communities were similar comparing isolates from 'Meeker' tissues with isolates from 'Wakefield' tissues.
- Yeasts and fungi were isolated from floral tissues and fruit, even though the field sites were treated with fungicides since early bloom.
- We found that yeasts and bacteria reside primarily on the surfaces of fruit and were rarely isolated from internal fruit tissues. This suggests that these microorganisms may be managed by application of materials on fruit surfaces, if there are concerns about yeasts populations or bacterial populations on fruit.

Results:

- We isolated microbial communities from tissues representing different stages in fruit development (floral bud emergence, prebloom, early bloom, full bloom, green fruit, and ripe fruit) from three Meeker and two Wakefield fields in Whatcom County. The incidence of colonization of tissues from bud emergence to early bloom varied among sites from ~30% to 90% of samples harboring populations of yeasts, bacteria or fungi.
- At full bloom, nearly every flower from the majority of sites was colonized by bacteria. Only 30% of flowers at full bloom at each site were colonized by yeasts and fungi. The low incidence of colonization of flowers by yeasts and fungi may be due to the application of fungicides.
- On green fruit and ripe fruit, nearly every sample harbored bacteria and yeasts (primarily on the surfaces of fruits) and fungi (isolated from fruit surfaces and internal tissues). The most abundant microorganisms found on fruit were: *Bacillus* spp. (bacteria), *Aureo-basidium pullulans* (yeast), and *Alternaria* spp, *Botrytis* spp., *Diaporthe* spp., and *Penicillium* spp. (fungi).

Publications:

- No publications. Presentation entitled "Microbes of red raspberry buds, flowers, and fruit" at Washington State Small Fruits Conference, November 2016, Lynden, Washington.

NOTE: Limit annual Progress Report to one page and Termination Report to two pages, except for publications.

2017 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

New Project Proposal

Proposed Duration: Total 2 years

Project Title: Fungicide Sensitivity of Blossom and Cane Disease pathogens of Red Raspberry

PI: Virginia Stockwell

Organization: USDA-ARS, Horticultural Crops Research Unit

Title: Research Plant Pathologist

Phone: 541-738-4078

Email: virginia.stockwell@ars.usda.gov

Address: 3420 NW Orchard Avenue

City/State/Zip: Corvallis, OR 97330

Year Initiated 2016 Current Year 2017 Terminating Year 2018

Total Project Request:

Prior Year 1: \$ 10,000

Current Year 2: \$ 5,919

Other funding sources: None.

Description:

The proposed research for 2017 is an extension of observations and results from the 2016 Red Raspberry microbiome project. In the course of sampling red raspberry floral and fruit tissues, we observed cane blight and in discussions with growers learned that little was known about the fungicide resistance profiles of the cane blight pathogen. This project will embark on examining tolerance of the cane blight pathogen and spur blight pathogen, if isolated, to fungicides. We also noticed an unusual blossom blight of red raspberry. The disease was observed on 'Meeker' and more commonly on 'Wakefield' red raspberry. We isolated a *Monilinia* spp. fungus from diseased blossoms, but would like to conduct a targeted sampling trip to estimate the prevalence of the disease and obtain additional samples to correlate the symptoms with the fungus. Identity of the causal agent of the blossom blight is essential for the development of management programs, if the disease increases in frequency.

Justification and Background:

The damaging disease, cane blight (causal agent *Leptosphaeria coniothyrium*) was observed in fields sampled in 2016. Damage associated with cane blight includes lateral shoot wilt, bud failure and death of the cane (Heidenreich, 2006). The fungicide resistance profile of the cane blight pathogen is not documented and will be investigated in this project. Spur blight (causal agent *Didymella applanata*) is another disease that infects red raspberry leaves and buds, but was not observed during our 2016 project. Canes and leaves will be examined for early symptoms of spur blight, and if present, isolates will be gathered and evaluated for tolerance to fungicides. Information on the fungicide resistance profiles of these pathogens would be valuable for the development of disease control programs.

We found a blossom blight of red raspberry in 2016 (see photo below). The disease was patchy in fields, but on some plants, up to 30% of blossoms were killed on a stem. We isolated a *Monilinia* spp. from a dead green fruit. *Monilinia fructicola* and *M. laxa* are species

that cause blossom blights and brown rot of stone fruits and occasionally pome fruits (Richie, 2000). *M. fructicola* was reported to cause an unusual fruit rot on blackberry in Germany (Hinrichs-Berger & Muller 2010) and on strawberry in Australia (Washington & Pascoe, 2000). Thus, it is possible that the observed blossom blight of Red Raspberry is caused by *Monilinia* spp. At this point, we need to obtain more samples of blossom blight of Red Raspberry to isolate the causal agent from numerous samples, verify pathogenicity, determine its identity, and develop a fungicide resistance profile.



Blossom blight on 'Wakefield' red raspberry. Left photos: Diseased flower (top) and healthy flower (bottom). Right photo: Branch with three of nine flowers diseased. Progression of the disease was limited to the peduncle and did not extend into the main stem.

Relationship of the proposed project to other projects: This project complements, but does not duplicate, the research programs of Dr. Peever (WSU) and Dr. Sabaratnam (AgCanada, Abbotsford BC). The information obtained on fungicide resistance of cane disease pathogens benefits each program as we gather and share knowledge about the emergence of fungicide resistance in *Botrytis* (Peever's research) and other fungal pathogens (this project) in the small fruit growing region of northern Washington and BC.

The blossom blight disease on red raspberry was discussed with Drs. Peever and Sabaratnam at the WA Small Fruit Conference and neither group has a project on this disease.

Relationship to WRRRC Research Priority(s): The proposed research addresses Priority group #1 "Fruit rot, including pre harvest, post harvest, and/or shelf life" and Priority group #3 "Foliar & Cane diseases – i.e. spur blight, yellow rust, cane blight, powdery mildew, etc."

Objectives:

The two objectives would be addressed in this funding year.

- 1) Isolate causal agents of cane diseases and determine fungicide sensitivity profiles.
- 2) Investigate a blossom blight disease of red raspberry.

Procedures:

Research on both objectives will be conducted in 2017.

Objective 1) Isolate causal agents of cane diseases and determine fungicide sensitivity profiles.

Incidence, sampling and isolation of pathogens. Up to 10 red raspberry fields in Whatcom County will be scouted for cane blight and spur blight by walking transects through fields. The incidence of cane blight and spur blight will be estimated by counting the number of plants with and without disease symptoms between posts, which will be repeated several times in each transect. Between 10 to 30 samples of canes with symptoms of cane blight and spur blight will be collected per field. In the lab, cane segments will be surface-disinfested with dilute bleach and ethanol soaks, followed by sterile distilled water rinses. Symptomatic stem sections will be placed on the surface of Potato dextrose agar amended with streptomycin and incubated at 25°C. When fungi emerge from tissues they will be transferred, identified, stored, and evaluated for sensitivity to fungicides.

Fungicide sensitivity profiles. We will estimate the Minimum Inhibitory Concentrations (MIC or EC₅₀) of *L. coniothyrium* and *D. applanata* exposed to a concentration series of iprodione (FRAC 2), boscalid (FRAC 7), cyprodinil (FRAC 9), azoxystrobin and/or pyraclostrobin (FRAC 11), fludioxonil (FRAC 12), and fenhexamid (FRAC 17). The methods assess the MIC of will be adapted from those on the FRAC website (<http://www.frac.info/monitoring-methods>) and in Fillinger and Walker (2016). This information is important to determine concentrations of fungicides that will discriminate between sensitive and resistant isolates of fungi in assays.

Objective 2) Investigate the blossom blight disease of red raspberry.

Incidence, sampling, and isolation: Similar methods to those in Objective 1 will be use to estimate the incidence of blossom blight on red raspberry. Up to 10 red raspberry fields in Whatcom County will be scouted for blossom blight. The proportion of plants with disease symptoms and the incidence of blossom blight on individual plants will be estimated in blocks in each field. Up to 30 samples of blossom blight will be collected per field. In the lab, symptomatic blossoms will be surface-disinfested, halved and placed on Potato dextrose agar amended with streptomycin and tart cherry medium, and incubated at 20°C. Outgrowing fungi will be transferred, identified by morphology and ITS sequence analysis (Borman et al. 2008), and stored. Ability to cause disease on raspberry fruits will be evaluated in laboratory assays. Verified pathogens will be evaluated for fungicide tolerance by the methods above.

Anticipated Benefits and Information Transfer:

This project will provide information about the incidence of cane diseases in red raspberry fields and the fungicide sensitivity profiles of the pathogens. This information could provide management options for growers that have persistent occurrences of cane diseases. The information gained from the investigation of the blossom blight is important to assess if we are observing a potential emerging disease that may increase in incidence in certain years or if it is just a localized minor disease. Project information will be delivered through presentations at grower and commission meetings and through scientific publications.

References:

1. Borman, A.M., Linton, C.J., Miles, S-J., and Johnson, E.M. 2008. Molecular identification of pathogenic fungi. *J Antimicrob. Chemot.* 61: i7-i12.
2. Fillinger, S., and Walker, A-S. 2016. Chemical control and resistance management of *Botrytis* diseases, pp189-216, *In: Botrytis-the Fungus, the Pathogen and its Management in Agricultural Systems*, S. Fillinger and Y. Elad, eds. Springer, New York.
3. Heidenreich, New York Berry News, Vol. 5 No. 3, March 13, 2006
4. Hinrichs-Berger, J., and Müller, G. 2010. First record of *Monilia fructicola* on blackberry fruits. *J. Plant Dis. Protect* 117:110-111.
5. Ritchie, D.F. 2000. Brown rot of stone fruits. *The Plant Health Instructor*. DOI: 10.1094/PHI-I-2000-1025-01
6. Washington, W.S., and Pascoe, I. 2000. First record of *Monilinia fructicola* on strawberry fruit in Victoria, Australia. *Australasian Plant Pathology* 29: 70.

Budget:	2017
Salaries^{1/}	\$ 1,891
Undergraduate pay^{1a/}	\$ 2,460
Operations (goods & services)^{2/}	\$ 1,000
Travel^{3/}	\$ 220
Meetings	\$ 0
Other	\$ 0
Equipment	\$ 0
Benefits^{4/}	\$ 348
Total	\$ 5,919

Budget Justification

^{1/} McGhee, 1 month salary. McGhee is a part-time technician with broad experience with morphological and molecular characterization of microorganisms. She will provide hands-on support for processing samples, clean-up and storage of isolates.

^{1a/} Undergraduate student for three months. Student will assist with media production, sample processing, and development of fungicide resistance assays for fungal isolates that are not *Botrytis* spp.

^{2/} Partial support of materials and supplies for media, petri dishes, molecular reagents, and sequencing.

^{3/} Stockwell, 1 trip Corvallis to Lynden raspberry fields in late May or June; Two nights at a hotel @110/night.

^{4/} Benefits are included for the undergraduate student worker (\$197) and McGhee (\$151).

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
Stockwell	Current: WRRC	\$10,000	2/13/2016 2/14/2017	20%	Development of novel disease management methods for fruit rots of raspberry
Stockwell	Pending: NWCSFR	\$65,996	10/1/2016 to 9/30/2018	35%	Survey of diseases of small fruits and prevalence of fungicide resistance in Oregon
Stockwell & Pscheidt	OR Blueberry Commission	\$10,326	4/1/17 to 3/30/2018	20%	Survey of <i>Botrytis</i> Green Fruit Rot and Silver Leaf in Oregon blueberry fields
Stockwell	WRRC	\$ 5,919	2/14/2017 2/15/2018	15%	Fungicide Sensitivity of Blossom and Cane Disease pathogens of Red Raspberry (this proposal)

Project: 13C-3755-5642
Title: Evaluation of Raspberry Bushy Dwarf Virus strains
Personnel: Patrick P. Moore, Professor and Kara Lanning.
Washington State University Puyallup Research and Extension Center
Cooperator: Bob Martin, USDA-ARS, Corvallis, OR.

Reporting Period: 2016

Accomplishments:

Plants of 23 cultivars in a field planting at WSU Puyallup were virus tested in 2014. All 16 of the resistant and 7 susceptible cultivars had at least one plant test virus positive. This indicates a resistance breaking strain of RBDV is widespread in the breeding plots at WSU Puyallup. At least three strains of RBDV could be distinguished by ELISA and two PCR tests. This has direct implications on breeding for RBDV resistance. At this time, we do not know what effects these different strains have on the plants. This study was designed to determine the effects of these three strains of RBDV on three raspberry cultivars.

Results

Plants of ‘Chief’, ‘Boyne’ and ‘Latham’ all tested positive in a PCR test for the viral polymerase gene, indicating they were infected with RBDV. ‘Chief’ and ‘Boyne’ both tested ELISA positive for the viral coat protein, but ‘Latham’ tested ELISA negative. ‘Chief’ tested positive for the viral coat protein using a PCR test and ‘Boyne’ and ‘Latham’ tested negative. These tests can distinguish between the RBDV strains in these plants. A single plant of each of these three cultivars was used to graft sets of plants of ‘Meeker’, ‘Chemainus’ and ‘Willamette’. These plants will be tested by ELISA and the two PCR tests to confirm that they have the particular strain of RBDV. Virus testing of the grafted plants in 2016 produced ambiguous results and the plants need to be re-tested in early 2017. Plants of each cultivar will be planted in the field in 2017 and fruited in 2018. Enclosures will be constructed and beehives introduced when the plants flower in 2018. Virus free plants will be enclosed in an enclosure and serve as a control. Fruit will be harvested and firmness, fruit weight and drupelet number determined. Fruit of infected plants will be compared to that of virus free plants.

After fruiting in 2018, plants infected with raspberry leaf mottle virus (RLMV) may be introduced into the enclosure along with aphids. Co-infection of RBDV and RLMV results in a much higher level of RBDV in the infected plants. Infection with only RBDV may not result in crumbly fruit, but infection with both RBDV and RLMV more commonly results in crumbly fruit. RBDV and RLMV are common in grower fields in the PNW.

Publications:

Lanning, K.K., P.P. Moore, K.E. Keller and R.R. Martin. First report of a resistance-breaking strain of *Raspberry bushy dwarf virus* in red raspberry (*Rubus idaeus*) in North America. Plant Disease 100:868

Project: 13C-3755-5642

Proposed Duration: 2 year

Project Title: Evaluation of Raspberry Bushy Dwarf Virus strains

PI: Patrick P. Moore

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Co-PI: Wendy Hoashi-Erhardt

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Cooperator: Bob Martin

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Year Initiated 2015

Current Year 2017

Terminating Year 2018

Total Project Request: Year 1 \$4,386

Other funding sources: None

Description: RBDV resistant raspberry cultivars were identified in 2014 that tested RBDV positive. This resistance breaking strain of RBDV was widespread in breeding plots at WSU Puyallup. Raspberry plants were tested using ELISA and primers targeting two viral genes. ELISA and the primer results were used to group the resistance breaking viral strains into three groups. The purpose of this project is to determine the effects of these strains on fruit characteristics. Plants of each viral strain were grafted onto 'Meeker', 'Chemainus' and 'Willamette' to produce plants of each cultivar with each virus strain. These plants along with virus free plants were grown in pots and will be planted in the field and allowed to fruit. Fruit will be weighed, fruit firmness measured and number of drupelets per fruit counted. The effects of each virus strain will be compared with the virus free controls.

Justification and Background: RBDV is a widespread, pollen borne virus with large economic effects. The only control method has been to breed for RBDV resistant cultivars. Studies conducted in 2014 indicated the presence of resistance breaking strains of RBDV. At this time, we do not know what effects these resistance breaking strains have. This study will compare fruit weight, fruit firmness and drupelet number between virus free plants and plants with different strains of RBDV as indications of the severity of the different viral strains.

Relationship to WRRRC Research Priority(s):

This project addresses a first-tier priority and a third-tier priority of the WRRRC:

First-Tier: Develop cultivars that are summer bearing, high yielding, winter hardy, machine-harvestable, disease resistant, **virus resistant** and have superior processed fruit quality

Third-Tier: Viruses/crumble fruit, pollination

Objectives:

This project will determine the impact of resistance breaking strains of RBDV on the fruit of selected raspberry cultivars.

Plants of selected raspberry cultivars were infected with specific strains of RBDV in 2015. Virus testing of the grafted plants produced ambiguous results and the plants need to be re-tested in early 2017. The plants will be planted in the field in 2017 and fruit evaluated in 2018.

Procedures: This project will take 2 years.

Plants with strains of RBDV will graft inoculate ‘Meeker’, ‘Chemainus’ and ‘Willamette’ in 2015.

The strains of RBDV are distinguished by positive or negative results when tested by ELISA for the coat protein, by PCR for the coat protein gene and by PCR for the viral polymerase gene.

RBDV strains	ELISA	Coat Protein gene	Polymerase gene
Resistance Breaking strain 1	+	+	+
Resistance Breaking strain 2	+	-	+
Resistance Breaking strain 3	-	-	+

Based on results from 2014, plants were tested by the three methods and were selected and used to graft inoculate plants of ‘Meeker’, ‘Chemainus’ and ‘Willamette’. ‘Meeker’ and ‘Chemainus’ are susceptible to the common strain and ‘Willamette’ is a RBDV resistant standard. Virus testing of the grafted plants produced ambiguous results and the plants need to be re-tested in early 2017.

The plants will be planted in the field in 2017 and allowed to fruit in 2018. The plants of ‘Meeker’, ‘Chemainus’ and ‘Willamette’ that are positive for a virus strain will be placed in separate netted enclosures and bees introduced. Virus free plants will be enclosed in a separate enclosure and serve as a control. Fruit will be harvested and firmness, fruit weight and drupelet number determined. Fruit of infected plants will be compared to virus free plants.

After fruiting in 2018, plants infected with raspberry leaf mottle virus (RLMV) with aphids on the plants may be introduced into the enclosure. Co-infection of RBDV and RLMV results in a much higher level of RBDV in the infected plants. Infection with only RBDV may not result in crumbly fruit, but infection with both RBDV and RLMV more commonly results in crumbly fruit. RBDV and RLMV are common in grower fields in the PNW.

Anticipated Benefits and Information Transfer:

This project will provide growers with information on the effects of these newly identified strains of RBDV. If there are serious effects of these strains, virus testing procedures may be modified and breeding efforts for RBDV resistance may be modified and include breeding for RBDV tolerance.

Results of this research will be included in the progress report and at commission and other grower meetings.

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

	2016
Salaries	\$2328
Benefits	\$830
Time-slip¹	\$120
Goods and Services³	\$1,096
Travel	\$0
Benefits 10.0%	\$12
Total	\$4,386

¹8 hours @\$15/hour for plot establishment

² K. Coats Molecular lab services .5 FTE + benefits \$3,158

³Laboratory supplies including consumable laboratory supplies and supplies for ELISA, Reverse-transcriptase qPCR and conventional PCR.

2017 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

New Project Proposal

Proposed Duration: (1, 2 or 3 years)

This is a new project proposal with 1 year duration.

Project Title:

Regional Survey of Commercial Red Raspberry Fields in the Pacific Northwest for a Resistance-Breaking Strain of Raspberry bushy dwarf virus

PI: Kara Lanning
Organization: Pacific Lutheran University
Title: Visiting Assistant Professor
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Email: lanninkk@plu.edu
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Address 2:
City/State/Zip: Tacoma, WA 98447

Year Initiated _____ **Current Year** 2017 **Terminating Year** _____

Total Project Request: **Year 1 \$** **15,740.00** **Year 2 \$** **Year 3 \$**

Other funding sources: *(If no other funding sources are anticipated, type in "None" and delete agency name, amt. request and notes)*
None

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

In summer 2016, the PI and two undergraduate researchers began a regional survey in Lynden, WA for a novel, resistance-breaking strain of Raspberry bushy dwarf virus (RBDV). Collecting leaf samples from 8 established red raspberry fields, the researchers identified a viral strain that appears to be more similar to the resistance-breaking strain of RBDV previously isolated to Europe, than the common D-isolates that are known to be established in the Pacific Northwest. The purpose of this project is to continue this work, and confirm the results collected in summer 2016 through further leaf collection, expansion of the survey to the commercial growing regions in Oregon, assessing graft transmissibility of the virus to genetically resistant cultivars, and through genetic sequencing.

Justification and Background: (400 words maximum)

- Provide a statement that clearly defines the issue you plan to address
- Why you plan to address it
- State how this project relates to other projects in British Columbia, Idaho and Oregon.

Raspberry bushy dwarf virus (RBDV) is a globally important, pollen-transmitted virus. The primary symptom of RBDV is crumbly fruit, which has severe impact on crop productivity. In 2015, a resistance-breaking strain of RBDV was identified in Puyallup, WA¹; and in 2016, a regional survey for this resistance-breaking strain in the commercial growing regions in Northern Washington was initiated. RBDV was detected using multiple diagnostic assays. Real-time RT-

PCR primers designed for the European resistance-breaking strain amplified viral RNA in the collected samples while the common strain primers failed to amplify the target sequence. The data indicate that the virus isolated from tissue samples collected is more similar to the resistance-breaking strain that is common in Europe than the common isolate that is well established in North America. To validate the data, further work is needed to investigate the resistance-breaking nature of the virus, including conducting graft transmission assays to see if the virus is able to infect genetically resistant Red Raspberry plants. Additional work is needed to understand the spread of the virus within the Pacific Northwest, which would include the expansion of the survey to Oregon. Lastly, sequence data is needed to compare the isolated virus collected from the regional survey to the known resistance-breaking strain of RBDV established in Europe.

First, this project is in-line with the Dr. Pat Moore's project that sets out to characterize the symptom development and impact of 4 novel viral strains that Kara Lanning identified during her Ph.D. research program. Understanding the regional spread of this novel virus, coupled with the data that Dr. Moore collects in his own project, will provide growers with a robust analysis of the potential impact of this novel RBDV strain on red raspberry production. Second, if the presence of a resistance-breaking strain of RBDV is confirmed, this information will inform breeders to reset their breeding goals to look for selections that are highly tolerant to RBDV infection, rather than identifying selections that are genetically resistant to RBDV.

Relationship to WRRRC Research Priority(s):

This project addresses the first and third priority outlined by the Washington Red Raspberry Commission:

- 1. Develop cultivars that are summer bearing, high yielding, winter hardy, machine-harvestable, disease resistant, virus resistant and have superior processed fruit quality.**

The results of this project will provide evidence to breeders to select for RBDV tolerance, rather than true virus resistance.

- 3. Viruses/crumby fruit, pollination**

This project looks specifically at the regional spread of RBDV. RBDV in mixed viral infection with other *Rubus* viruses results in crumby fruit disease.

Objectives: Provide specific objectives that you will attempt to accomplish during the project period.

1. Confirm diagnostic results collected summer 2016, using three diagnostic methods that detect the viral capsid protein: ELISA, RT-PCR, and Real-Time RT-PCR.
2. To see if the virus identified in Whatcom County is graft transmissible to Willamette; a cultivar that is genetically resistant to the common D-isolates.
3. To obtain a genetic sequence of the virus and compare to the genetic sequence of known RBDV strains.
4. To expand the survey to the commercial growing regions of Oregon.

Which objectives will be addressed this funding year?

All objectives will be addressed this funding year.

Procedures: (400 words maximum)

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

This project is anticipated to take 1 year, with the bulk of the tissue collection, grafting, and laboratory procedures occurring during a summer research program at Pacific Lutheran University. The project will proceed under the following research schedule:

1. Winter 2017

- a. Plant 100 ‘Willamette’ seedlings in the green house

2. Spring 2016

- a. Confirm disease-free status of ‘Willamette’ seedlings
- b. Collect tissue from Whatcom County, graft seedlings with collected tissue

3. Summer 2016

- a. Collect leaf samples from established fields in both Whatcom County and in the Willamette Valley in Oregon.
- b. Perform the following diagnostic procedures to confirm the presence of the virus in **both** grafted seedlings and collected leaf samples: ELISA, RT-PCR, Real-Time RT-PCR
- c. Analyze diagnostic and grafting data
- d. Send isolated viral RNA for genetic analysis and compare sequence data to known strains of RBDV, including D-200 isolates and R-15 (RB-Strain in Europe)

Anticipated Benefits and Information Transfer: (100 words maximum)

- What specific benefits will result from this project for producers and/or the raspberry industry? Be clear and direct.
- How will results be transferred to users?

This research project will directly benefit the raspberry industry as it will answer the question: is there a resistance-breaking strain of RBDV in the Pacific Northwest. If there is, then the data collected will redirect the breeding focus of selecting for a genetically resistant raspberry cultivar to a highly tolerant cultivar of raspberry. The information gained in this project will be made available through presentation of the results at the 2017 Small Fruit Conference, through the WSU extension bulletin, and through publication in an academic journal.

References:

1. Lanning, K. K., Moore, P. P., Keller, K. E., Martin, R. R. (2016, April). First Report of a Resistance-breaking Strain of Raspberry bushy dwarf virus in Red Raspberry (*Rubus idaeus*) in North America. *Plant Disease*, 100(4), 898

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

	2017	2018	2019
Salaries^{1/}	\$ 7,200.00	\$	\$
Time-Slip	\$ 4,000.00	\$	\$
Operations (goods & services)	\$ 2100.00	\$	\$
Travel^{2/}	\$ 1440.00	\$	\$
Meetings	\$	\$	\$
Other	\$	\$	\$
Equipment^{3/}	\$ 1000	\$	\$

Benefits^{4/}	\$ 0	\$	\$
Total	\$ 15,740.00	\$	\$

Budget Justification

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

Funding is requested to cover the salary of the Principle Investigator, Kara Lanning. Pacific Lutheran University does not cover the PI's summer salary per the terms of her current contract. It would cover the salary from the months of June through mid-August, with 10% withheld for taxes.

Additionally funding is requested to cover a \$4,000 stipend for one undergraduate student researcher for the same time period.

^{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.

Funding for travel is requested to support the mileage and housing for the principal investigator and one undergraduate scientist for a total of three trips. Travel is a necessary component of the project as plant tissue will need to be collected from sites in Washington and Oregon. The anticipated cost of travel is broken down as the following line items:

Mileage: \$400

Overnight Accommodations: \$800, at a rate of \$100 per night (2 rooms x 4 nights) with anticipated tax included

Per Diem: \$240.00, at \$30/day per person for a total of 4 travel days.

^{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.

Funding is requested to purchase a new micropipette set that will be used exclusively for the purpose of scientific research, and will not be available for teaching purposes. Common use micropipettes are frequently damaged and contaminated due to the circumstances that surround biology laboratory education. The purchase of a micropipette set will ensure that the molecular procedures carried out in this project are precise, free from contamination and that the data collected will be reproducible. The production of contaminant-free and reproducible results is integral to the integrity of the project.

^{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.

None requested.

SOILS

Year 3 Report, Washington Red Raspberry Commission 2016 funded Project:
“Evaluating soil fumigation alternatives in Washington raspberry fields”, Walters and Zasada.

Dominus/Vapam trial. A trial comparing a conventional Telone C-35 application (shank-injected approximately 16 inches below the soil surface, 35 gallons/Acre) with shallow-applied Vapam (shanks 5 and 10 inches below the soil surface, 75 gallons/Acre) and Dominus (applied same as Vapam, 40 gallons per acre) was established in 2014, and was monitored in 2015 and 2016. Four monitoring sites were established in the Dominus and Vapam plots, and 8 additional monitoring sites were established in the Telone C-35-treated field surrounding the plots. Through 2015, *P. Penetrans* counts were highest in the Telone C-35-fumigated areas, with markedly fewer nematodes in Vapam and Dominus-treated plots (Table 1). In 2016, *P. penetrans* numbers in the Vapam-treated plot were still less than 20% of those in the C-35 treated and Dominus-treated areas.

In 2015, plant growth (primocanes per hill and height of the longest primocanes in a hill) were better in the Vapam and Dominus-treated plots (Table 2). Differences were less pronounced in 2016; C-35 treated areas had slightly (but not significantly) fewer canes per hill and taller primocanes.

Shallow application of Vapam and Dominus markedly improved nematode control in this trial. When Vapam was used, this effect remained visible two years after fumigation. In this trial, shallow-dwelling nematodes could be found on the winter cover crop planted at fumigation and were a major source of infection in the newly planted raspberry crop.

Cover crop trials. Additional trials were established to assess the efficacy of managing *P. penetrans* on the winter cover crop by either killing the cover crop early (thus starving the nematodes in the roots) or by applying a nematicide to the cover crop. Results were not statistically significant, but the Lannate-treated plots in both trials had numerically lower levels of *P. penetrans* than untreated plots. Follow-up trials established in 2015 (funded by this project and a WSDA grant) found no effect of insecticide application or time of cover crop kill, but delaying cover crop planting did reduce *P. penetrans* numbers in the following raspberry crop.

Like the trial above, these trials suggest that shallow-dwelling *P. penetrans* escape fumigation, find refuge in winter-planted cover crops and readily infect newly planted raspberry. We set up another preplant cover crop trial following fumigation in the fall of 2016. In this trial, cover crops identified as poor *P. penetrans* hosts (‘Jessup’ fescue, black oats, ‘Wheeler’ rye) are being compared with winter wheat and a fallow check.

A raspberry field that had been left unplanted for a year instead of being immediately replanted was also monitored this past year. The old raspberry crop was removed from this field in 2013. It was planted with a small grain cover crop in the winter of 2013-14, and white mustard (*Sinapis alba*) was planted, grown and incorporated in 2014. In 2015, we found moderate numbers of *P. penetrans* on the winter cover crop (250/g root, similar to numbers in fumigated fields), and similarly found moderate *P. penetrans* numbers on the raspberry plants in September (667/g root). This grower’s strategy of combining a fallow year with white mustard incorporation in place of fumigation appears successful in this moderately infested field.

2015 fumigation trials. A replicated broadcast fumigation trial established in 2015 was evaluated in 2016. This trial included nontarped C-35 (industry standard), tarped C-35, nontarped C-35 with a Vapam cap and shallow-applied Dominus. The tarped C-35 and

Vapam cap treatments controlled nematodes better on the preplant cover crop than the nontarped C-35 and Dominus treatments did, but no differences were noted on nematodes on the raspberry plants in June, July or October.

A bed fumigation trial was also established in 2015, with adjoining beds fumigated with C-35, Dominus or Dominus:pic (75% Dominus, 25% chloropicrin). In July 2016, plants in the Dominus:pic-treated beds were slightly (but not significantly) taller than those in C-35 or Dominus-treated beds, and there were slightly more primocanes per hill in the Dominus-treated beds (Figure 1). Root and soil samples for nematode analysis were collected in October 2016. In general, samples contained very few *P. penetrans*, and there were no significant differences between treatments. The low *P. penetrans* numbers indicate that these treatments were quite effective in this field. We will continue to follow this trial next year.

A Field day was held September 1, with presentations from Walters, Zasada, Wasco-DeVetter, Weiland and students.

Table 1. *P. penetrans* per g root in cover crop and in spring-planted raspberry following soil fumigation treatments September 2014.

Treatment	Cover crop Nov 2014	Raspberry July 2015	Raspberry Sept 2015	Raspberry Sept 2016
Telone C-35	5142± 919	1130 ± 159	1148 ± 516	4154 ± 974
Dominus	710 ± 380	296 ± 161	148 ± 50	5043 ± 1420
Vapam	626 ± 263	6 ± 6	55 ± 35	762 ± 214

Table 2. Raspberry plant growth in 2015 and 2016 following soil fumigation treatments September 2014.

Treatment	Primocanes per hill¹		Primocane Height, cm²	
	2015	2016	2015	2016
Telone C-35	12.9 ± 0.7	13.3 ± 0.5	32.4 ± 0.7	95.8 ± 0.8
Dominus	17.8 ± 0.7	14.2 ± 0.7	39.1 ± 1.0	89.2 ± 1.2
Vapam	17.5 ± 0.6	14.2 ± 0.8	36.7 ± 1.4	89.2 ± 1.5

¹average of primocanes per hill, counting 10 hills in each of at least 4 locations

²height of the 3 tallest primocanes in each hill, 10 hills in each of at least 4 locations

Figure 1. A) Raspberry primocane height and B) Primocanes per hill in beds fumigated with Telone C-35, Dominus or Dominus:pic

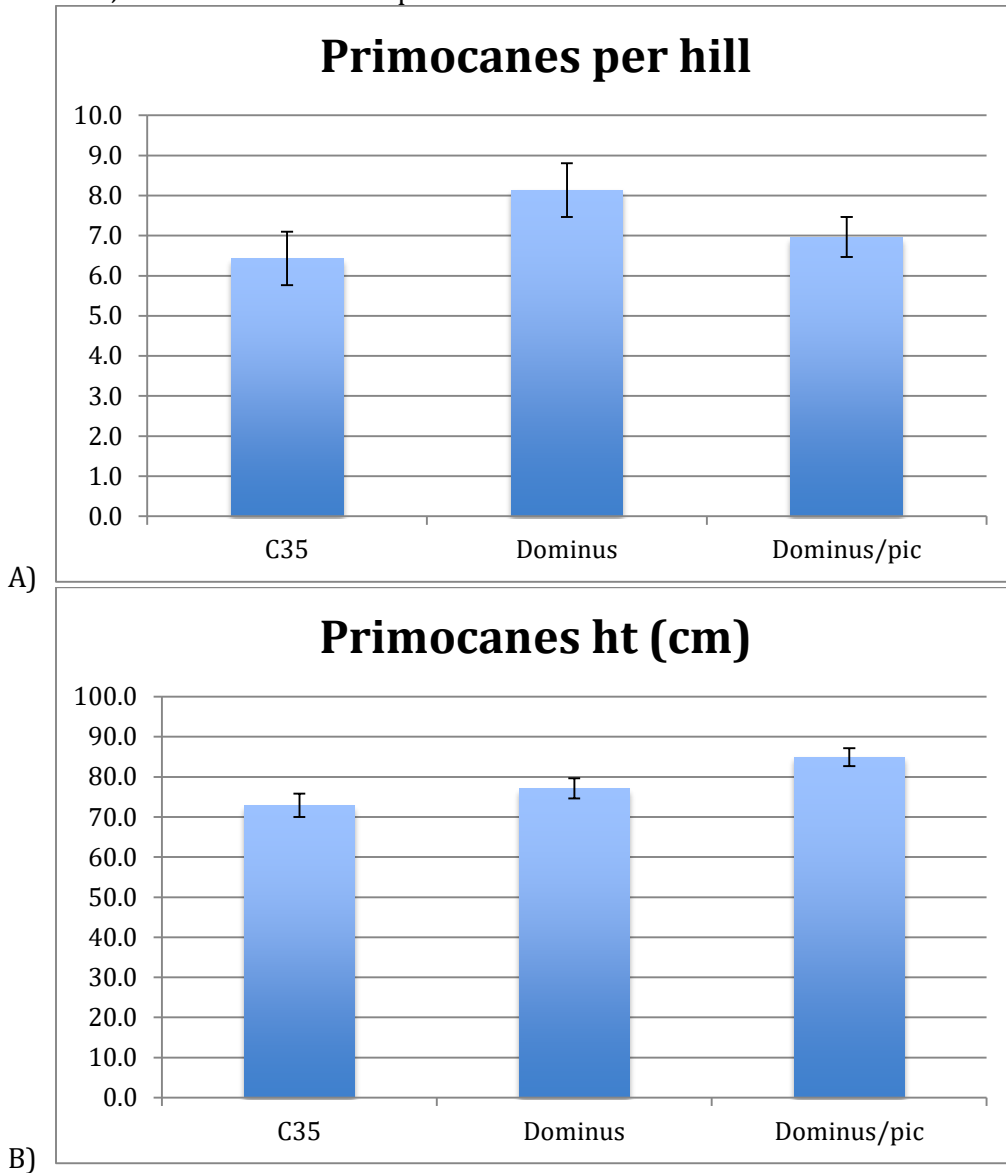


Table 3. Soil and root *P. penetrans* in raspberry beds fumigated with Telone C-35, Dominus or Dominus:pic, October 2016.

Treatment	Pp/50 g soil	Pp/g root
Telone C-35	0 ± 0	6.3 ± 3.5
Dominus	2.8 ± 1.7	4.5 ± 1.9
Dominus:pic	1.4 ± 1.2	11.8 ± 5.1

Fungicide Sensitivity of *Phytophthora rubi* from Washington

Weiland

Progress Report 7-14-2016

We are assessing whether fungicide resistance is present in *Phytophthora rubi* isolates of Washington's red raspberry industry. In 2015, we collected 83 isolates of *P. rubi* from 19 red raspberry fields with symptoms of root rot in Whatcom (16 fields) and Skagit (3 fields) counties. In 2016, we sampled an additional six fields in Clark (2 fields), Puyallup (1 field), Skagit (1 field), and Whatcom (2 fields) counties to obtain isolates from new regions (Clark and Puyallup) and to sample fields where fungicide resistance is suspected (Skagit and Whatcom). So far, we have 48 new isolates from the 2016 fields. However, the isolates from one field do not appear to be *P. rubi* – we are sequencing those isolates to determine what they are. We have screened most of the *Phytophthora rubi* isolates collected in 2015 for resistance to mefenoxam. The majority of the isolates appear to be sensitive to mefenoxam, but there are six isolates that appear to have low to moderate mefenoxam resistance. We are retesting those potentially resistant isolates to make sure the results are correct. Our next step will be to test the isolates collected in 2016 against mefenoxam, then to finish isolate testing against the next fungicide, phosphorous acid. Finally, we will test a subset of isolates against cyazofamid and dimethomorph. Both fungicides have shown promise in protecting against *Phytophthora* root rot in other crop systems. We do not anticipate any issues in completing the project.

We completed the mefenoxam sensitivity testing this month, but are still working on phosphorous acid and the other two fungicides, so we have not sent in a final report. I expect the other three fungicides will be finished by the end of 2017.

Jerry

2017 WASHINGTON RED RASPBERRY COMMISSION RESEARCH PROPOSAL

New Project Proposal

Proposed Duration 3 years

Project Title: Vapam cap, crop termination, and bed fumigation treatments to improve soil fumigation.

PI: Thomas Walters
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Co-PI: Lisa DeVetter
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Co-PI: Jerry Weiland
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Cooperators: Mike Conway and Tim Purcell, Trident Ag Products;
Chris Benedict, WSU Whatcom County Extension

Year Initiated: 2017

Current Year: 2017

Terminating Year: 2019

Total Project Request: Year 1 \$13,407 Year 2 \$14,207 Year 3 \$14,857

Other funding sources: No other cash sources. Trident has indicated willingness to provide fumigant, services and tarps for bed fumigation trials.

Description: We will address the need for affordable, effective preplant soil fumigation in an increasingly challenging regulatory environment. According to our own research and grower reports, Vapam caps significantly improve nematode and disease control in Telone C-35 fumigated fields. In Florida, postharvest crop termination treatments are often used to reduce pests and diseases prior to fumigation. Bed fumigation has proven effective and economical in many settings.

Our objective is to evaluate the effectiveness of a Vapam cap and of a post-harvest termination treatment in reducing nematode and disease carryover in fields of different soil types. We will evaluate this through soil and root nematode assays, and plant growth and disease (root rot) evaluation. We will also evaluate bed-applied fumigants as a cost reduction strategy

The major outcome of this work will be improved grower understanding of how these practices will best work for them. It's already pretty clear that Vapam caps can be effective, but it would help to know which soil types and conditions are the best for them. Crop termination with

Vapam or other products may improve nematode control in and near the root zone, where numbers should be greatest.

Justification and Background

Fumigators must cope with buffer zone and other regulatory limitations. Most raspberry growers have a custom applicator fumigate their fields in blocks with combinations of 1,3-D (Telone) and chloropicrin, and most growers report clear benefits from this practice. However, some plant parasitic nematodes and diseases escape current fumigation procedures, and growers often find there are some nematode, disease and weed problems in newly fumigated fields (Walters et al, submitted). For example, we recently documented substantial numbers of *P. penetrans* on post-fumigation, preplant cover crops in Washington raspberry fields (up to 5100 *P. penetrans*/g root, as presented in 2015 Progress report). These problems can reduce the growth and first-season yields of newly developing plants, and can develop into chronic problems throughout the lifespan of the planting.

Telone prices will increase 10% in January 2017, and may increase again in July. High price and low availability pressures applicators and growers to use products containing less Telone and more chloropicrin. This is risky (chloropicrin is not an effective nematicide), and it makes fumigation more difficult (buffers depend upon the chloropicrin content of the fumigant). Regulations on the use of both fumigants are likely to become more restrictive in the future. Although there is no crisis with Telone or chloropicrin today, we feel this is the time to prepare for a day when we may have less to use.

Raspberry growers often terminate the old crop with herbicides, but this practice does not greatly impact soilborne disease and nematode populations. Crop termination with a drip-applied soil fumigant has been a useful pest management tool in other systems (MacRae et al., 2010), and is used to address carryover disease and nematode problems.

Bed fumigation has been researched in Washington raspberries before; tarped bed fumigation with Telone C-35 was as effective or more effective than nontarped broadcast fumigation with the same product (Walters et al., submitted). Preliminary data with nontarped bed fumigation is promising (Walters and Zasada progress report), but more time and trials are needed to evaluate this practice. Nontarped applications of Telone C-35 can fail to control fungi and *Phytophthora rubi*, but increasing the concentration of chloropicrin with Pic-Clor 60 (1,3-D 40%, chloropicrin 60%) and tarping has been effective against these pathogens in other crops (Weiland et al., 2016). We expect this will also be the case in raspberry.

Relationship to WRRC Research Priorities

This proposal directly addresses the WRRC's #2 priority "Soil fumigation techniques and alternatives to control soil pathogens, nematodes and weeds". This proposal evaluates existing techniques (Vapam cap and bed fumigation) and a novel supplement (crop termination with a fumigant) to address this priority.

Objectives:

- Evaluate crop termination via buried drip and caps (both with Vapam) as ways of reducing nematodes and diseases escaping preplant soil fumigation.

- Evaluate low Telone-use systems (bed fumigation with Telone C-35 and with Pic-Clor 60) using Trident Ag Product's bed fumigation system.
- Estimate the economic costs and benefits of these practices.

Procedures

Crop Termination. With a grower-cooperator (possibly Enfield Farms) we will identify and presample a field to identify an appropriate study area. We will establish eight plots, each 30 ft x 50 ft in four randomized blocks. Root and soil samples for nematode evaluation will be collected after harvest, prior to treatment. If buried drip is not already present, tapes will be buried 2-3" deep in plots to be treated. Soil fumigant (Vapam, up to 74 gal/A) will be injected into the buried tapes with enough water to wet most of the rooted zone (approximately 1 gallon/row foot). Beginning 1 week after treatment, leaf and cane dieback will be monitored weekly. Post-treatment root and soil samples will be collected approximately 2 weeks after treatment. The study area will be fumigated with a Telone:chloropicrin combination along with the rest of the grower's field.

Nematode population densities will be determined: from cover crop roots in Jan 2018; raspberry soil and roots September 2018, Spring 2019 and Fall 2019. Plant growth data will be collected August 2018. If there are differences in nematode population densities or plant growth, plant yield data will be collected in July 2019.

Vapam caps. We will identify two raspberry fields with moderate to high nematode pressure but with different soil types (e.g., a sandy loam, and a heavier silt loam). We will presample to identify study areas in these fields. Each study area will include 12-16 plots, each 30 ft x 50 ft. There will be 4 replicate blocks of 3-4 treatments:

- Untreated check (with grower's permission)
- Deep shank applied C-35, 35 gal/A
- Shallow shank applied Vapam, 74 gal/A
- Both C-35 (deep) and Vapam (shallow)

Before treatment, plots will be cored to determine vertical nematode distribution. A Giding soil corer, 3 ft long, will be driven into the ground. A plastic sheath inside the corer will be removed after sampling and partitioned into 6 inch segments for separate nematode extraction. These plots will be monitored in the same way as crop termination plots described above.

Bed fumigation. Plots will be established in fields with *Phytophthora* pressure and bed fumigated by Trident. Treatments include tarped and nontarped applications of Telone C35 and Pic-Clor 60 in 4 replicate blocks. *Phytophthora* inoculum in bags will be buried to assess treatment efficacy and retrieved 1 month after fumigation. Nematode, pathogen and plant growth will be monitored as described for crop termination

Anticipated Benefits and Information Transfer

We know from other high value production systems that soil plays an important role in fumigant efficacy. This information is not available for soil types in northern WA. We will develop fumigation recommendations specific to the soil types present in NW raspberry fields. It is also necessary to apply and evaluate alternative fumigation application methods that have proven to be successful in other high value crop production systems to the raspberry production system.

Our research will determine whether crop termination with a drip applied fumigant improves nematode and disease management above and beyond broadcast fumigation alone.

References:

Culpepper, A.S., and Smith, J.C. 2015. 2015 Vegetable Fumigant systems for Plasticulture in Georgia. Accessed 12/6/16 at <http://www.gaweed.com/HomepageFiles/MBAlternatives2015-final%20Nov%2017.pdf>

Kroese, D.R., Weiland, J.E., and Zasada, I.A. 2016. Longevity and distribution of *Pratylenchus penetrans* in red raspberry. Journal of Nematology (in press).

MacRae, A., Noling, J., and Snodgrass, C. 2010. Maximizing the efficacy of soil fumigant applications for raised-bed plasticulture systems of Florida. HS1169, Horticultural Sciences Department, Florida Cooperative Extension Service, IFAS, University of Florida.

Walters, T.W., Bolda, M., and Zasada, I.A. Alternative fumigation practices for western states raspberry. Plant Health Progress (submitted)

Weiland, J.E., Littke, W.R., Browning, J.E., Edmonds, R.L., Davis, A., Beck, B.R., and Miller, T.W. 2016. Efficacy of reduced rate fumigant alternatives and methyl bromide against soilborne pathogens and weeds in western forest nurseries. Crop Protection 85: 57-64.

Budget:

	2017	2018	2019
Salaries ^{1/}	\$7,486	\$7,486	\$7,486
Time-Slip			\$500
Operations (goods and Services) ^{2/}	\$4,817	\$5,617	\$5,517
Travel ^{3/}	\$400	\$400	\$650
Meetings			
Other: shipping			
Equipment			
Benefits ^{4/}	\$704	\$704	\$704
Total	\$13,407	\$14,207	\$14,857

Budget Justification:

^{1/} Walters 0.055 FTE, benefits included: coordinate with growers, stake out plots, supervise crop termination and Vapam cap applications, coordinate C-35 applications. Sean Watkinson (technician for DeVetter), 0.042 FTE.

^{2/} Walters: \$400 shipping 2017 and 2018, \$650 shipping 2019. Zasada: \$2800 sample processing 2017, \$3600 2018 and 2019. Weiland: \$1667 sample processing 2017, 2018 and 2019.

^{3/} Walters, 2017: 6 trips Anacortes to Lynden, 2018: 6 trips, 2019 10 trips.

^{4/} Watkinson, 35.44%.