



VacCAP

IMPROVING FRUIT QUALITY

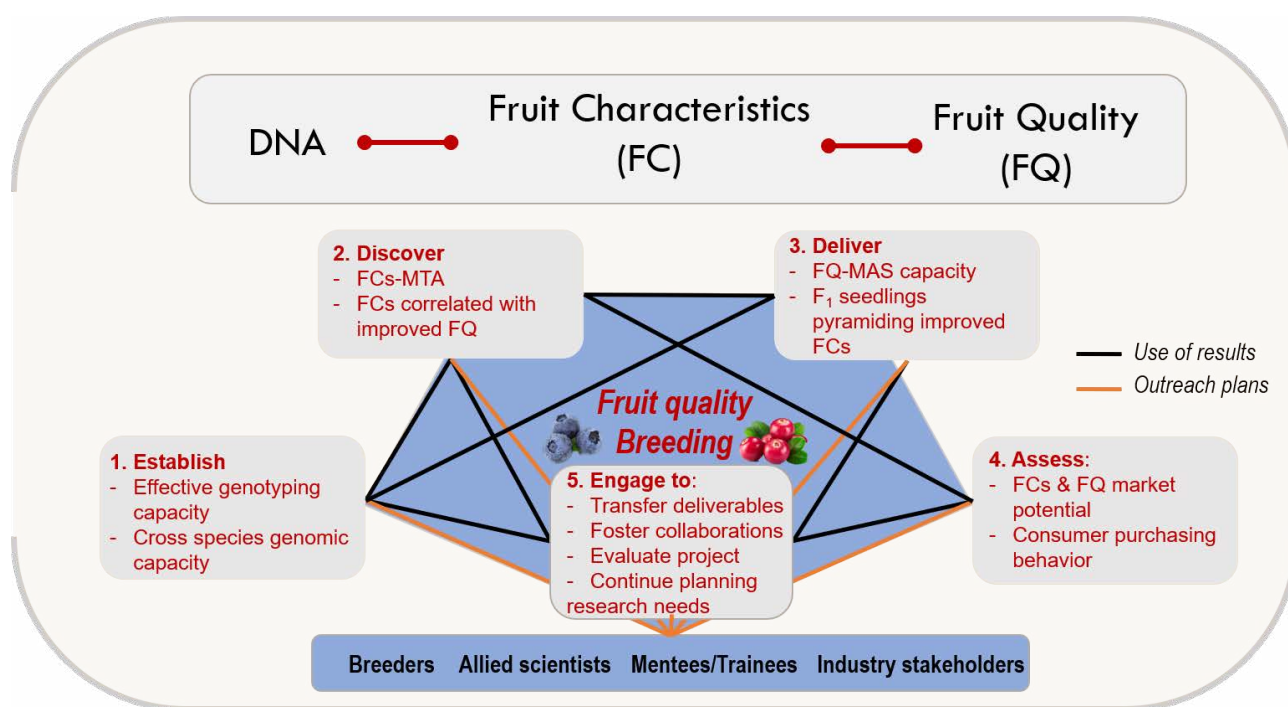
Annual Project Report and Plan

Annual Meeting III | November 5, 2021 | Online



VacCAP Objectives

The Vaccinium Coordinated Agricultural Project (VacCAP) aims at leveraging genetic and genomic resources to enable development of blueberry and cranberry cultivars with improved fruit quality attributes.



United States Department of Agriculture
National Institute of Food and Agriculture
Award #: 2019-51181-30015

Agenda

Advisory Board Meeting | Friday, November 5, 2021 | Online

Presentations Playlist Link: <https://www.youtube.com/playlist?list=PLo3BW8AtfTdNyGWKczp1Lo92IsFZI3cLs>

Presentations were **pre-recorded** and are being shared with AP members. Content of each video align with the summary reported here. AP members can select video based on specific expertise/interest. **Only a short overview of each Objective will be presented during the annual meeting**, leaving more time for discussion.

If you have any questions or discussion point that want to share prior the annual meeting please add to this shared document: https://docs.google.com/document/d/1t9KUCKkeYMGbN_ZatSwdnHuyJA_54Xo/edit

Time	Section 1 Plan
12:00-12:10	Introduction with Iorizzo
12:10-12:30	<p>Objective 1 Brief Overview and Discussion</p> <p>For content of this session watch pre-recorded presentations:</p> <ul style="list-style-type: none"> • Edger – VacCAPIII Genomics and Pangenome • Bassil - VacCAPIII VacGenotyping Platform • Chagné – VacCAPIII Int. Team – Blueberry Genomics and FQ Genetics
12:30-1:10	<p>Objective 2 Brief Overview and Discussion</p> <p>For content of this session watch pre-recorded presentations:</p> <ul style="list-style-type: none"> • Iorizzo - VacCAPIII - Blueberry FC Genetic discovery • Zalapa - VacCAPIII - Cranberry FC Genetic discovery • Perkins - VacCAPIII - Blueberry FQ shelf life and bruising • Giongo - VacCAPIII – International Team Blueberry texture and sensory
1:10-1:30	<p>Objective 3 Brief Overview and Discussion</p> <p>For content of this session watch pre-recorded presentations:</p> <ul style="list-style-type: none"> • Munoz - VacCAPIII – Blueberry FC/FQ breeding and DNA assays • Polashock - VacCAPIII - Cranberry FC/FQ breeding and DNA assays
1:30-1:40	Break
	Section 2 Plan
1:40-1:50	<p>Objective 4 Brief Overview and Discussion</p> <p>For content of this session watch pre-recorded presentations:</p> <ul style="list-style-type: none"> • Canales/Gallardo – VacCAPIII – Cranberry added sugar survey • Sims – VacCAPIII – Blueberry sensory and Willingness-to-Pay (WTP)
1:50-2:20	<p>Objective 5 Brief Overview and Discussion</p> <p>For content of this session watch pre-recorded presentations:</p> <ul style="list-style-type: none"> • Main – VacCAPIII GDV and data management • Atucha – VacCAP III Extension and evaluation
2:20-3:30	Final discussion and plans for administrative activities

*See next page for time conversion table

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Time zone	Session	I	Break	I	Discussion
PT (-3)	9	10:30	10:40	11:30	12:30
CT (-1)	11	12:30	12:40	13:30	14:30
ET	12	13:30	13:40	14:30	3:30
IT (+6)	18	19:30	19:40	20:30	21:30
NZ (+17)	5	6:30	6:40	7:30	8:30

Agenda

Working Group Meeting | Tuesday, September 28, 2021 | Online

9:30 AM Session 1. Objective 2a-b cranberry.

- Objective: Discuss Y2 activities and plan Y3 activities
- Presenters: Zalapa, Polashock
- Coordinator: Zalapa

10:30 AM Session 2. Objective 2a-c blueberry.

- Objective: Discuss Y2 activities and plan Y3 activities
- Participants: Iorizzo, Pottorff, Bassil, Lila, Perkins-Veazie, Munoz, Luby, Mackey, Ferrao, Li, Sims, Giongo, Coe
- Presenters: Iorizzo, Perkins-Veazie, Munoz, Li, Giongo
- Coordinator Obj 2a-b: Iorizzo
- Coordinator Obj 2c: Perkins-Veazie

12 PM Session 3. Objective 3 Cranberry and blueberry.

- Objective: Plan Y3 activities
- Participants: Munoz, Vorsa, Polashock, Zalapa, Iorizzo, Pottorff, Bassil, Ferrao, Mackey, Luby, Coe
- Presenters: Munoz, Luby, Polashock, Vorsa
- Coordinator blueberry team: Munoz
- Coordinator cranberry team: Polashock

1:00 PM Session 4. Objective 5 Extension.

- Objective: Discuss Y2 activities and plan Y3 activities
- Participants: All PIs
- Presenters: Main, Atucha
- Coordinator Data Management Team: Main
- Coordinator Extension Team: Atucha

2:30 PM Session 5. Objective 4. Socio Economics

- Objective: Discuss Y2 activities and plan Y3 activities
- Participants: Gallardo, Canales, Perkins-Veazie, Iorizzo, Pottorff, Munoz, Bassil, Mackey, Luby, Colonna, Sandefur, Giongo, Ferrao
- Presenters: Gallardo, Sims
- Coordinator Socio Economic Team: Gallardo

3:30 - 5:00 PM Session 6. Objective 1

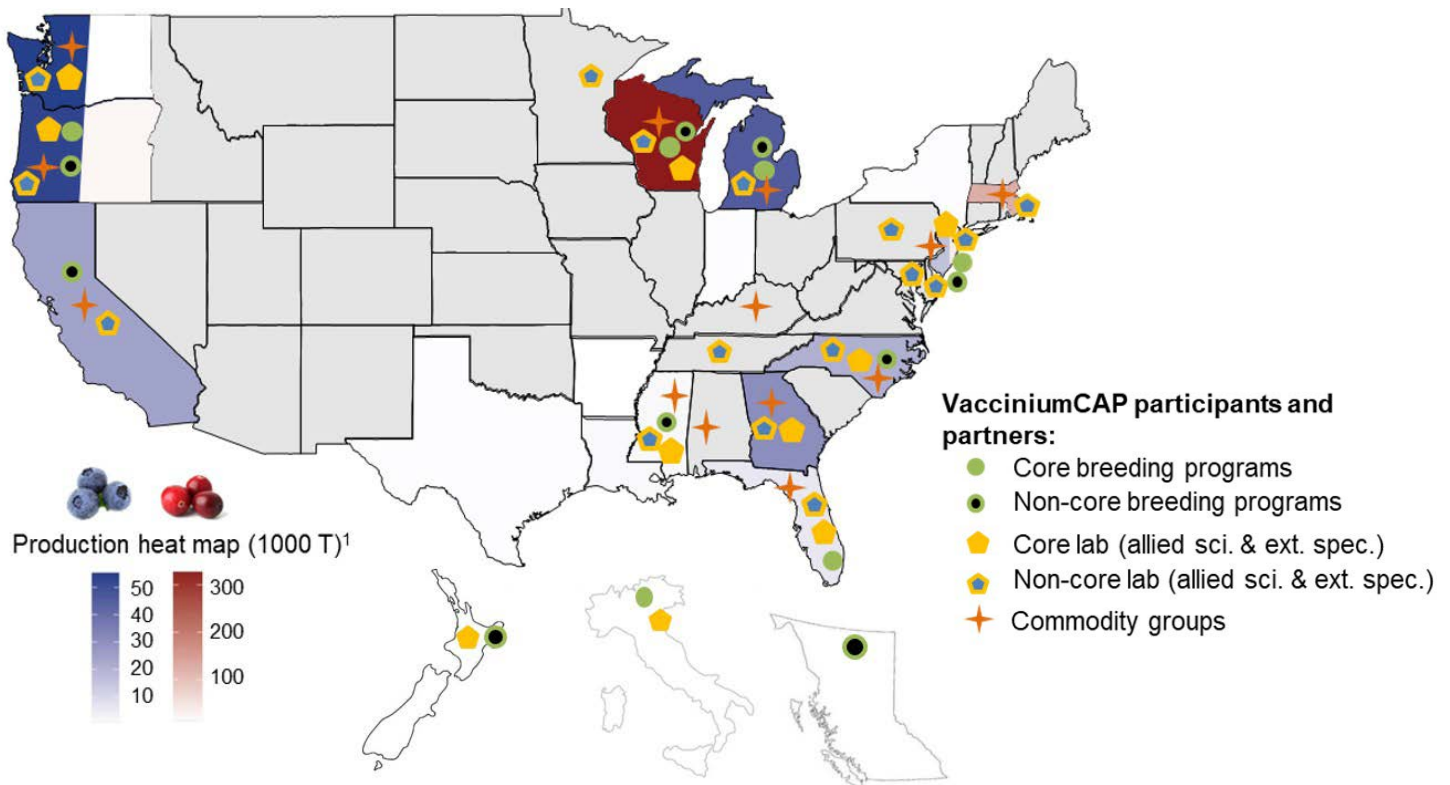
- Objective: Discuss Y2 activities and plan Y3 activities
- Participants: Edger, Bassil, Chagne, Espley, Iorizzo, Munoz, Pottorff, Zalapa, Polashock, Vorsa, Luby
- Presenters: Edger, Bassil, Espley/Dare
- Coordinator Obj 1a: Edger
- Coordinator Obj 1b-c: Bassil

Meeting Attendees

Iorizzo, Zalapa, Polashock, Perkins-Veazie, Lila, Fiente, Trandel, DeVetter, Atucha, Coe, Giongo, Russo, Oh, Vorsa, Pottorff, Bassil, Luby, Munoz, Benevenuto, Ferraro, Li, Gallardo, Kawash, Vorsa, Humman, Colonna, Canales, Pmperkins, Catrina, Ma, Edger, Dare, Espley, Phillips, King

Time zone	Session	I	II	III	IV	V	VI
PT (-3)	6:30	7:30	9	10	11:30	12:30	14
CT (-1)	8:30	9:30	11	12	13:30	14:30	16
ET	9:30	10:30	12	13	14:30	15:30	17
IT (+6)	15:30	16:30	18	19	20:30	21:30	23
NZ (+17)	2:30	3:30	5	6	7:30	8:30	10

VacCAP PIs and Partners



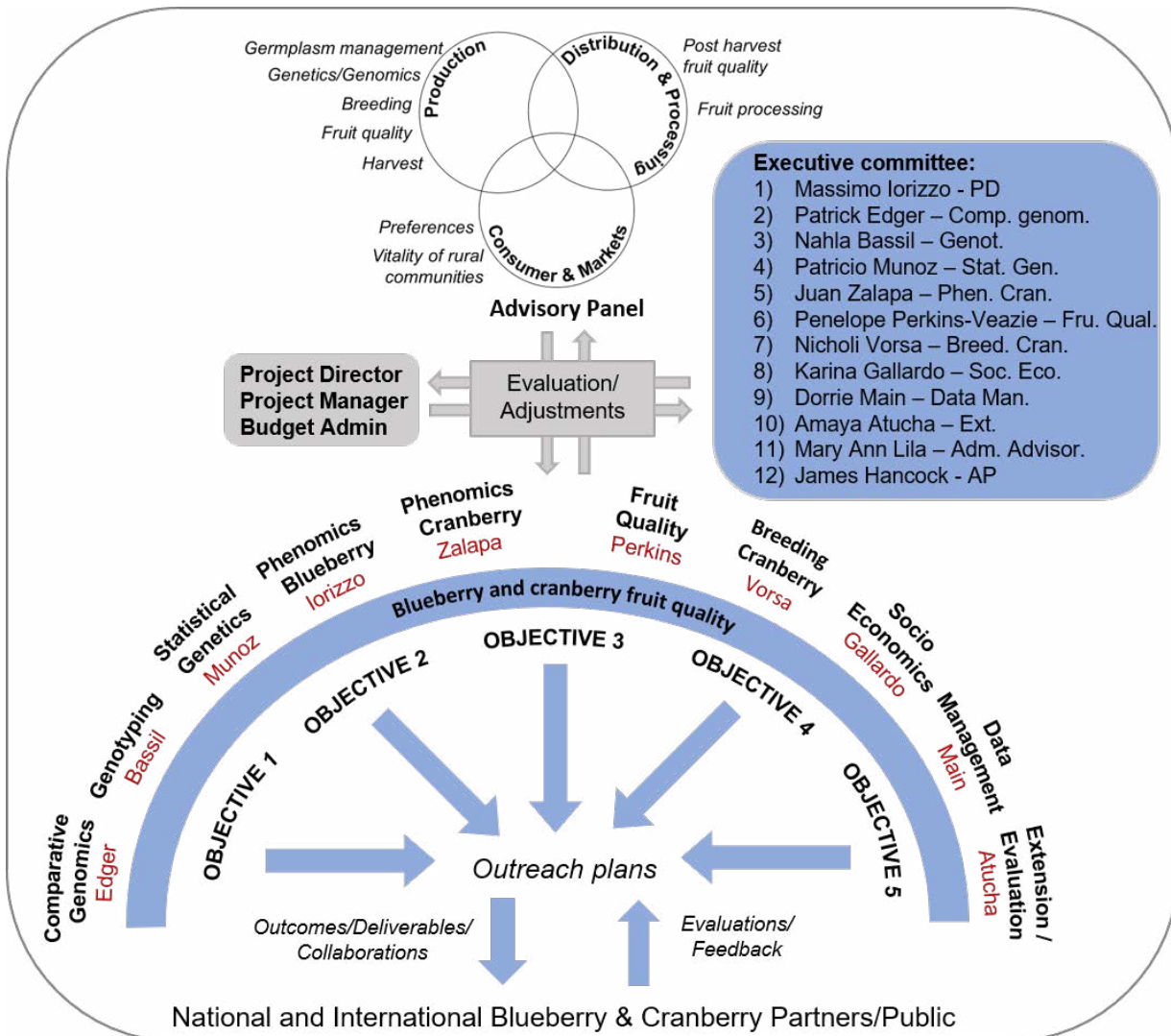
- Massimo Iorizzo, Project Director, North Carolina State University
- Amaya Atucha, Co-PD, University of Wisconsin-Madison
- Nahla Bassil, Co-PD, USDA-ARS and NCGR
- Patrick Edger, Co-PD, Michigan State University
- Karina Gallardo, Co-PD, Washington State University
- Mary Ann Lila, Co-PD, North Carolina State University
- Dorrie Main, Co-PD, Washington State University
- Patricio Munoz, Co-PD, University of Florida
- Claire Luby, Co-PD, USDA-ARS and HCRL
- Penelope Perkins-Veazie, Co-PD, North Carolina State University
- Nicholi Vorsa, Co-PD, Rutgers University

- Juan Zalapa, Co-PD, University of Wisconsin-Madison
- Elizabeth Canales, Co-PI, Mississippi State University
- David Chagné, Co-PI, Plant and Food Research Ltd
- Lisa Wasko DeVetter, Co-PI, Washington State University
- Richard Espley, Co-PI, Plant and Food Research Ltd
- Lara Giongo, Co-PI, Fondazione Edmund Mach
- Changying Li, Co-PI, University of Georgia
- James Polashock, Co-PI, USDA-ARS and GIFVL
- Charles Sims, Co-PI, University of Florida
- Michael Coe, Key Personnel, Cedar Lake Research Group



How VacCAP Is Managed

Our VacCAP team is listed below and organized by project objective. However, nine project teams manage the project and work collaboratively across project objectives and with our advisory panel.



Breeding Teams

- Team Leaders: TBD (Blueberry), N. Vorsa (Cranberry)
- Team members: P. Munoz, P. Edger, N. Vorsa, L. Giongo, J. Zalapa, M. Iorizzo, C. Luby
- Tasks: Finalize the list and maintain the material to use in this project. Collect and distribute blueberry and cranberry materials to be used in this project. Develop blueberry/cranberry F1 progenies to pyramid multiple superior fruit quality traits (Obj. 3). Coordinate the implementation of standardized phenotyping protocols for on-site fruit quality assessment in collaboration with the blueberry/cranberry Phenomic Teams (Obj. 2-3). Serve as a training group to use the data collection software (e.g. Field Book) and BIMS system in collaboration with the Data Management Team. Act as liaison between the Extension Team, the PD, and blueberry and cranberry industry organizations.

Comparative Genomics Team

- Team Leader: P. Edger
- Team members: N. Bassil, P. Munoz, J. Zalapa, N. Vorsa, J. Polashock, D. Chagné, R. Espley.
- Tasks: Coordinate the activities required to develop and characterize the *Vaccinium* pangenome (Obj. 1a) that will be used to develop the SNP catalog. Collaborate with Genotyping Team to establish the SNP catalog (Obj. 1b) and provide bioinformatic analysis needed to select the final set of SNPs used to validate the *Vaccinium* Genotyping Platform (Obj. 1c). Liaise with Breeding and Statistical Genetic Teams to finalize the list of blueberry and cranberry material needed for the comparative genomic analysis. Liaise with Data Management and Extension Teams to deliver genomic resources through the Genome Database for *Vaccinium* (GDV) and to provide content about the comparative genomic outcomes (Obj. 1a).

How VacCAP Is Managed

Genotyping Team

- Team Leader: N. Bassil
- Team members: P. Edger, P. Munoz, J. Zalapa, N. Vorsa, L. Giongo, D. Main, M. Iorizzo, J. Polashock, D. Chagné
- Tasks: Coordinate activities to develop the SNP catalog, validate the SNP set, organize the *Vaccinium* Genotyping forum, and finalize selection and validation of the *Vaccinium* Genotyping Platform (Obj. 1b-c). Collaborate with the Breeding and Statistical Genetics Teams to finalize the list of blueberry and cranberry germplasm needed for the genotyping platform validation. Extract DNA and genotype the blueberry and cranberry samples for the genetic and validation studies (Obj. 2b, 3b). Will provide content for Extension efforts on outcomes.

Statistical Genetics Team

- Team Leader: P. Munoz
- Team members: J. Zalapa, M. Iorizzo, N. Vorsa, L. Giongo, N. Bassil, P. Edger, J. Polashock
- Tasks: Coordinate activities required to perform marker-trait association analysis and QTL/marker validation (Obj. 2b, 3b). Will collaborate with Breeding Team to finalize the list of germplasm to be used in the project. Provide support for statistical analysis needs. Liaise with the Data Management Team to use the computational infrastructure to store and deliver markers/QTL data through the GDV (public and private partitions). Liaise with the Extension Team to provide content about the marker-trait association analysis and QTL/ marker validation outcomes (Obj. 2b, 3b).

Phenomic Teams

- Team Leaders: M. Iorizzo/L. Giongo (blueberry), J. Zalapa (cranberry)
- Team members: P. Munoz, P. Perkins-Veazie, M.A. Lila, D. Main, J. Polashock, N. Vorsa
- Tasks: Coordinate activities required to implement phenotyping methods (Obj. 2a, 3a). Collaborate with the Statistical Genetics Team on marker trait association analysis (Obj. 2a) and QTL/ marker validation (Obj. 3b). Collaborate with the Fruit Quality and Socio-Economic (Blueberry) Teams to correlate fruit characteristics and their performance for shelf life, resistance to bruising, sensory panel (Obj. 2c) and potential economic value (willingness to pay) (Obj. 4a). Liaise with the Breeding and

Statistical Genetics Teams to finalize the list of blueberry and cranberry germplasm needed for the phenotypic analysis (Obj. 2a-3a). Liaise with the Data Management Team to use the data management computational infrastructure to store and/or deliver phenotypic data through the GDV (public and private partitions). Liaise with the Extension Team to provide content about outcomes delivered through the phenotypic analysis (Obj. 5).

Fruit Quality Team

- Team Leader: P. Perkins-Veazie
- Team members: C. Li, C. Sims, M.A. Lila, L. Giongo, M. Iorizzo, P. Munoz
- Tasks: Coordinate activities required for the blueberry fruit quality studies, which includes the evaluation of shelf-life, sensory panel and bruising damage (Obj. 2c) in collaboration with the Phenomic and Breeding Teams. Will collaborate with the Socio Economic Team to plan and conduct a joint consumer panel analysis. Will liaise with the Extension Team to provide content regarding the outcomes delivered through the fruit quality studies (Obj. 2c) and the Extension outreach plans.

Socio-Economics Team

- Team Leader: K.R. Gallardo
- Team members: E. Canales, C. Sims
- Tasks: Coordinate activities required to perform the socio-economic studies (Obj. 4). Collaborate with the Breeding, Phenomic and Fruit Quality Teams to acquire blueberry plant material, phenotyping and sensory data. Work with Extension Team to provide content about the outcomes delivered through the socio-economic studies and to develop outreach activities planned by the Extension Team (Obj. 5).

Extension Team

- Team Leader: A. Atucha
- Team members: D. Main, L. De Vetter, M. Coe (External Evaluator)
- Tasks: Coordinate activities and provide content for the extension and outreach activities/resources (Obj. 5). Will collaborate with the Data Management Team to develop and populate the VacCAP extension platform and work with External Evaluator to collect and evaluate feedback from Extension audiences.

National and International Partners

Breeding and Molecular Geneticist Partners

- Hamid Ashrafi, Assistant Professor, North Carolina State University
- Mark K. Ehlenfeldt, Research Geneticist, USDA-ARS
- Stephen Stringer, Research Geneticist, USDA-ARS
- Lisa J. Rowland, Research Geneticist, USDA-ARS
- Jeff Neyhart, Research Geneticist, USDA-ARS
- Kalpalatha Melmaiee, Associate Professor, Delaware State University
- Ebrahim Babiker, Research Geneticist, USDA-ARS
- Moira Sheehan, Director of Breeding Insight, Cornell University
- Jim Hancock, Breeder, Berry Blue, LLC
- Jim Olmstead, Global Breeding Director-Blueberry, Driscoll's, Inc.
- Mathew Kramer, Director of Product Development & Commercialization, Fall Creek Farm & Nursery, Inc.
- Bob Gabriel, President, Oregon Blueberry Farm and Nursery
- Edward Grygleski, Cranberry Breeder and Producer, Valley Corp.
- Rachel Itle, Assistant Research Scientist, University of Georgia
- Michael Hardigan, Research Geneticist, USDA-ARS

Fruit Quality Partners

- Anne Plotto, Research Plant Physiologist, USDA-ARS
- Randy Beaudry, Professor, Michigan State University
- Steve Sargent, Professor and Associate Chair, University of Florida
- Rod Serres, Manager Agricultural Science, Ocean Spray
- Andy Reitz, Director of Grower Relations, Mariani Co., Inc.
- Mike Mainland, Professor Emeritus, North Carolina State University
- Ann Colanna, Sensory Program Director, Food Innovation Center, Oregon State University

Bioinformatics and Biotechnology Partners

- Robert Reid, Assistant Professor Bioinformatics, University of North Carolina-Charlotte
- Margaret Staton, Associate Professor of Bioinformatics, University of Tennessee
- Guo-Qing Song, Associate Director Plant Biotechnology Resource & Outreach Center, Michigan State University

Extension Network

- Ali Sarkhosh, Assistant Professor, University of Florida
- Cassie Bouska, Assistant Professor, Oregon State University
- Carlos Garcia-Salazar, Extension Educator, Michigan State

University

- Kim Patten, Director Pacific Co. and Extension Professor, Washington State University

Extension Network (Continued)

- Cesar Rodriguez-Saona, Extension Specialist, Rutgers
- Renee Allen, Extension Specialist, University of Georgia
- William O. Cline, Research and Extension Specialist, North Carolina State University
- Kathleen Demchak, Senior Extension Associate, Penn State University
- Ben Faber, Advisor, Cooperative Extension Ventura County
- Mary Rogers, Associate Professor, University of Minnesota
- Hilary A. Sandler, Director of UMass Cranberry Station and Extension Associate Professor, University of Massachusetts-Amherst
- Erick D. Smith, Assistant Professor, University of Georgia
- Eric Thomas Stafne, Extension/Research Professor, Mississippi State University
- Wei Qiang Yang, Associate Professor and District Berry Extension Agent, Oregon State University
- Mike Mainland, Professor Emeritus, North Carolina State University

Other Abiotic and Biotic Stresses Partners

- Christelle Guédot, Associate Professor, University of Wisconsin
- Jonathan Oliver, Assistant Professor-Fruit Pathologist, University of Georgia
- David Bryla, Research Horticulturist, USDA-ARS HCRU
- Scott Lukas, Assistant Professor, Oregon State University Hermiston Agricultural Research and Extension Center

International Partners

- Michael Dossett, Research Scientist at BC Berry Cultivar Development Inc., British Columbia
- Susan McCallum, Blueberry Researcher at James Hutton Institute, Scotland
- Luis Diaz Garcia, Instituto Nacional de Investigaciones Forestales y Agrícolas y Pecuarias, Aguascalientes, Mexico
- Paul Sandefur, Manager of Breeding Operations at Fall Creek Farm & Nursery Inc., Oregon, USA
- Simon Bonin, Director of Grower Relations and Agronomy at Fruit d'Or, Quebec, Canada
- Susan Thomson, Bioinformatician at Plant and Food Research Ltd, New Zealand
- Toshi Foster, Senior Scientist, Plant and Food Research Ltd, New Zealand

National and International Partners

Supporting Industry Partners

- British Columbia Blueberries
- Berry Blue, LLC
- California Blueberry Commission
- Cape Cod Cranberry Growers' Association
- The Cranberry Institute
- The Dole Food Company
- Driscoll's, Inc.
- Fall Creek Farm and Nursery, Inc.
- Florida Blueberry Grower's Association
- Georgia Blueberry Commission
- Kentucky Blueberry Growers Association
- Mariani Premium Dried Fruit
- New Jersey Blueberry Cranberry Research Council
- North American Blueberry Council
- North Carolina Blueberry Council, Inc.
- Ocean Spray Cranberries, Inc.
- Oregon Blueberry Commission
- Oregon Blueberry Farms and Nursery
- Oregon Cranberry Grower Association
- U.S. Highbush Blueberry Council
- Valley Corporation
- Washington Blueberry Commission
- Wayne County Blueberry Growers Association
- Wisconsin State Cranberry Growers Association



Advisory Panels

Stakeholder Panel

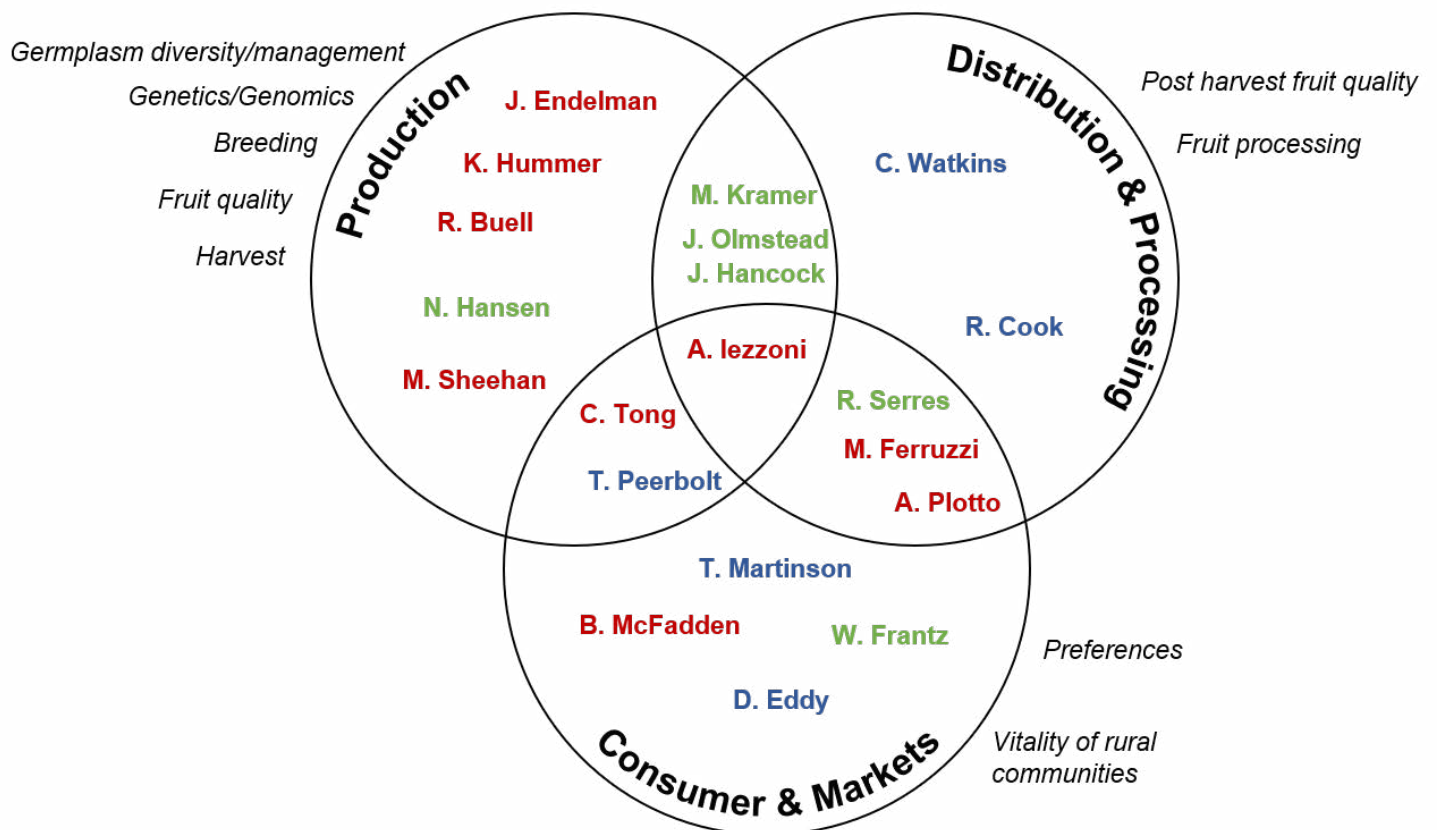
- James Hancock, Berry Blue LLC
- Nicole Hansen, Cranberry Grower
- Matt Kramer, Fall Creek Farm and Nursery
- James Olmstead, Driscoll's, Inc.
- Rod Serres, Ocean Spray Cranberries, Inc.
- William Frantz, The Cranberry Institute

Extension Panel

- Rodney Cook, Ag-View Consulting, Inc.
- David Eddy, Master Media Worldwide
- Tim Martinson, Cornell University
- Tom Peerbolt, The Northwest Berry Foundation
- Christopher Watkins, Cornell University

Scientific Panel

- Robin Buell, Michigan State University
- Jeffrey Endelman, University of Wisconsin-Madison
- Mario Ferruzzi, Arkansas Children's Nutrition Center (ACNC)
- Kim Hummer, USDA-ARS-NCGR (OR)
- Amy Iezzoni, Michigan State University
- Brandon McFadden, University of Delaware
- Anne Plotto, USDA-ARS (FL)
- Moira Sheehan, Cornell University
- Cindy Tong, University of Minnesota



US and Global Coordination Efforts for Improving Cranberry and Blueberry

Misson

Address major bottlenecks for growth of U.S. *Vaccinium* industry, by creating a nationwide coordinated transdisciplinary research approach to develop and implement marker assisted selection (MAS) capacity in *Vaccinium* breeding programs, to enable breeders to select and pyramid fruit characteristics (FCs) that positively contribute to fruit quality and market value. In the long term, this mission will increase production of fruit with improved characteristics that meet the ever-changing industry, market, and consumer preferences.

What Success Would Look Like

Discovery. VacCAP project outputs will increase the knowledge of:

- Cranberry and blueberry genome structure and evolution
- Genetic mechanisms and genes controlling economically important traits including fruit characteristics
- The relationships between fruit characteristics and fruit quality (shelf life, texture, bruising and sensory traits)
- Consumer behavior and interests regarding blueberry fruit quality and cranberry products
- New *Vaccinium* stakeholder priorities for the sustainability and profitability of the industry

Engagement. VacCAP deliverables and outcomes will be utilized by the *Vaccinium* community for the following goals:

- VacCAP DNA tools and phenotyping methods will be utilized by VacCAP PIs and the *Vaccinium* community worldwide to advance breeding and/or research programs
- VacCAP outcomes will be used by growers, processors and distributors to plan production and distribution strategies
- Funding of new off-shoot projects from VacCAP developed tools and deliverables with new collaborative alliances established

Education. The VacCAP team influences the next generation of breeders and scientists, increasing knowledge through collaborations:

- MS/PhD students and post-docs are trained in plant breeding, genetics, fruit phenotyping, postharvest physiology, socio-economics, sensory analysis and extension practices, to become the next generation of breeders, scientists, and agriculture liaisons

Long-Term Impact

- Increased ability of the *Vaccinium* growers, processors and distributors to market a higher percentage of premium fruit through the use of improved varieties without increased production costs
- Increased consumption of blueberry and cranberry products in the US and worldwide due to improved fruit quality
- Increased efficiency of *Vaccinium* breeding programs for selection and improvement of fruit quality traits important to the consumer and industry
- Increased profitability, competitiveness, and sustainability of *Vaccinium* industries

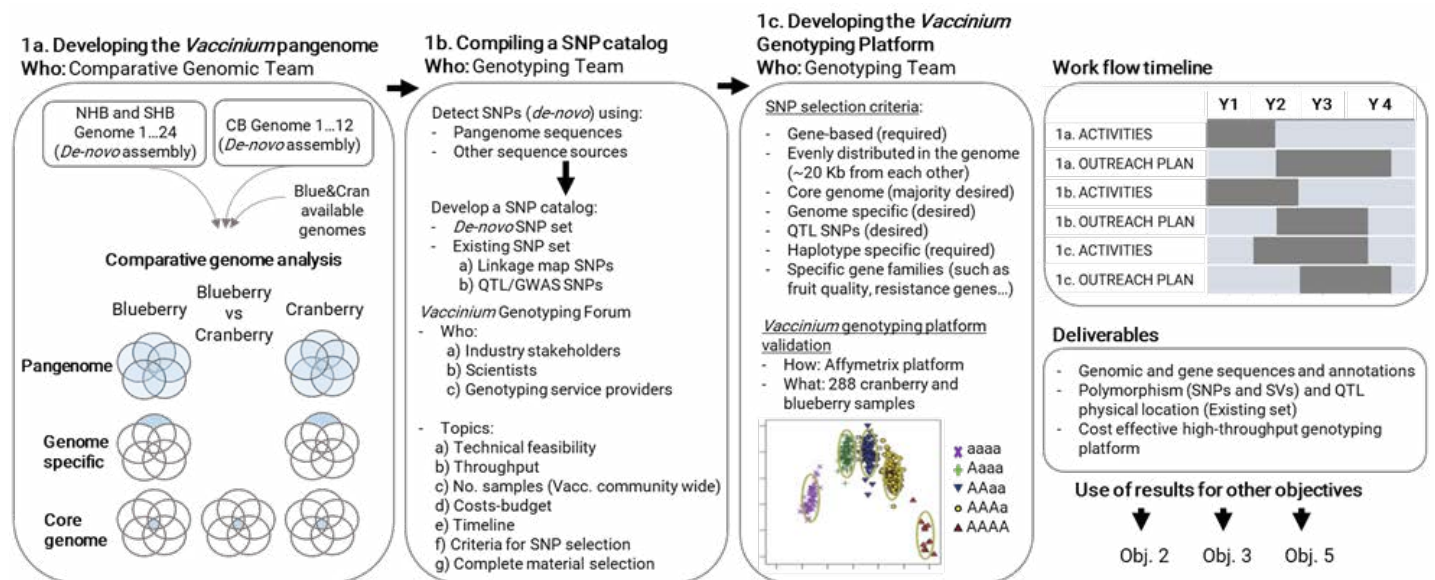
Blueberry Harvest Season

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Objective 1

Establish genomic resources to enable effective association mapping studies in blueberry and cranberry

The Comparative Genomic and Genotyping Teams will develop a cost effective high-density genotyping platform by mining the *Vaccinium* pangenome that represents the genetic diversity of blueberry and cranberry germplasm and their shared ancestry. In the mid and long term, this genotyping platform is expected to significantly expand our capacity to identify and validate DNA markers associated with economically important traits in blueberry and cranberry.



Method Overview

To achieve this objective, the Comparative Genomic and Genotyping Teams will:

- 1a. Develop a *Vaccinium* pangenome. A pangenome for Northern Highbush (NHB), Southern Highbush (SHB), and cranberry (CB) will be assembled. The aim is to identify the core and dispensable portions of the genome. For each species/group, 12 genotypes that are highly representative of the pedigree of NHB, SHB and CB cultivars and that capture the greatest amount of genetic diversity were selected. This approach will limit ascertainment bias in the SNP selection.
- 1b. Compile a SNP catalog. A SNP catalog will combine *de-novo* with existing SNP sets (within linkage maps or representing QTLs). The SNPs catalog will be annotated with SNP location within genes, core or dispensable genes, gene families, alleles and haplotypes. This approach will ensure the identification of highly informative SNPs.
- 1c. Develop the *Vaccinium* Genotyping Platform. DNA regions surrounding highly informative SNPs will be selected to design a genotyping platform. Criteria for SNP selection will aim to maximize the representation of genes, markers associated with QTLs, informative haplotype blocks and to be distributed approximately 20-30Kb apart. *Vaccinium* breeders and geneticist will be engaged to establish a genotyping consortium that will help to lower the genotyping costs per sample, while ensuring application of these new molecular resources.



P. Edger



N. Bassil



D. Chagné



R. Espley

Obj. 1 – VacCAP: Year 2 Progress Summary

Obj. 1. Establish genomic resources to enable effective association mapping studies in blueberry and cranberry	
Reported by: Edger, Chagne, Bassil, Espley, Zalapa, Iorizzo, Polashock, Munoz, Giongo, Vorsa	
Presented by: Edger, Chagne, Bassil	
1a. Developing the <i>Vaccinium</i> pangenome [Comparative Genomic Team]	To whom*
Prepare material for sequencing (12 NHB, 12 SHB and 12 CB). Collected plant material for RNA extractions for remaining cranberry samples (12).	Genotyping and Statistical Genetic Teams, Breeder, Geneticist and Bioinformatic Partners
Whole genome sequencing. Completed sequencing of the transcriptome of all 24 blueberry samples.	
Begin construction of the pan-genome. Assemblies of all blueberry and cranberry cultivars were completed. Completed annotation of blueberry genomes. Initial QC evaluation using BUSCO indicated that the genome assembly captures >96% gene space. In addition, a pangenome graph was constructed, ancestral states estimated and genomic variants identified unique to blueberry, cranberry and individual genomes. The blueberry core genome includes 19,942 genes (on average ~54% of genes in individual genomes) and those data were shared with the Genotyping Team for selecting target SNPs for probe design.	
Development of new genomes and genetic resources for genetic and comparative analysis. Three new genomes and a new high density linkage map (RxA1) were released (Table 1). Analysis of the bilberry and cranberry genomes (completed during year 1), uncovered regulatory genes controlling anthocyanin accumulation in the fruit of these two species, and the history of whole genome duplications in the <i>Ericaceae</i> . This data will facilitate studying divergence and/or conservation of genes controlling fruit quality traits such as anthocaynins across the <i>Vaccinium</i> pangenome. Final data analysis of five additional linkage maps and new genomes are being completed (Table 1). Two bi-parental populations generated from a full diallelic cross between the tetraploid blueberry (<i>V. corymbosum</i>) cultivars ‘Hortblue Petite’ and ‘Nui’ were used to investigate heterochiasmy. Linkage maps were built for ‘Hortblue Petite’ female and ‘Nui’ male (N=150), and for ‘Hortblue Petite’ male and ‘Nui’ female (N=128). For almost all LGs the recombination rate in females was lower than in males. Patterns of recombination rates along the chromosomes are being investigated, as well as regions showing preferential pairing. The consensus linkage map for the population ‘Hortblue Petite’ × ‘Nui’ was also used to detect QTLs for anthocyanin content.	
Impact. The genomic resources developed in the VacCAP project are and will continue to: Expand studies aimed at identifying candidate genes associated with fruit quality and other economically important traits; provide a solid framework to design a genotyping platform that best represents the blueberry and cranberry diversity; enable comparative studies within the <i>Ericaceae</i> and across other plant families.	
1b. Compiling SNP catalog [Genotyping Team]	To whom
Compile existing and De Novo SNP sets from team members and partners. Completed compiling and annotation of the SNP catalog. The catalog includes 5,318,829 SNPs and 2,252,197 INDELS. Out of the 491,588 existing SNPs contributed by collaborators, 444,908 were unique and included in the catalog (Table 2). These Existing SNPs were obtained from 13 studies, nine collaborators/institutions detected in diversity panels (DPs) and mapping populations (MP) and mapped to a single location in the P0 reference blueberry sequence (diploid W85). A total of 1,592,379 structural variants (SVs) were also identified from transcriptome (TR) sequences of 16 cultivars available through NCBI, and 6,446,402 SVs were generated De Novo (D). The De Novo SNPs were detected from a DP consisting of 47 cultivars (25 generated in this project and 22 from Breeding Insight). In total, 4,675 SNPs were associated with known QTLs, representing 23 traits including fruit quality traits (TA, SSC, fruit size, fruit weight, firmness, color, ...)(Table 3). Criteria and parameters to select the 50,000 SNP loci for the testing phase were defined.	Statistical Genetic Team, Breeder and Geneticist Partners

1c. Developing genotyping platform [Genotyping Team]	To whom
Select genotyping platforms and form genotyping consortium. Based on survey outcomes and forum discussions, a target capture-based genotyping technique (Flex-Seq EX-L) was chosen for developing the VacCAP genotyping platform. Compared to other platforms that were evaluated, this platform is cost-effective, delivers more SNPs and more flexibility to redesigning and or updating probe sets. A Vaccinium Consortium was formed which committed to genotyping 7,104 samples (3,744 samples in this project and 3,360 samples by 10 consortium members) in 2022 to secure a genotyping cost of \$49.45 per sample with 20,000 loci for blueberry and 15,000 loci for cranberry using this genotyping platform. This price includes DNA extraction and bioinformatic analyses.	Breeding, Statistical Genetic Teams, Breeder and Geneticist Partners
Initiated testing phase. Leaf samples were collected from 192 blueberry accessions that included representative samples from four industry partners and 10 academic institutions in the US, Canada and New Zealand. DNA was extracted from these samples and submitted to RAPiD Genomics. The SNPs from the SNP catalog are being filtered at this time by RAPiD Genomics to identify 50,000 loci that meet our filtering criteria and those required for the Flex-Seq EX-L target capture-based genotyping platform we chose to genotype this Testing Set of 192 diverse blueberry accessions.	
Expected Impact 1b-c. A cost effective genotyping platform that is useful to the <i>Vaccinium</i> scientific community worldwide that will enable genetic studies to identify loci and markers associated with fruit quality and other important traits.	
Challenges & changes. Due to COVID-19 and delay of funding release from the USDA-NIFA during Year 1 the following activities were not performed as planned: Sequencing of the transcriptome from 12 cranberry samples was delayed resulting in a delay in downstream analysis (finalize the pan-genome construction, Obj. 1a and developing the cranberry SNP catalog was delayed (Obj. 1b-c).	
Addressing challenges and recommendations. A recommendation from the advisory panel was to plan the release of the data used for the pangenome construction. Raw genomic data for all samples included in the pangenome construction were already released on NCBI (see Table 1). This will permit various organizations to utilize that data for sequence alignment, etc. The pangenome, each individual genome, and associated datasets and resources will be made available upon request (under the terms of a Fort Lauderdale Agreement). Activities that were delayed have already restarted and will be completed in Year 3. To compensate for some of the impacted activities, team members completed complementary activities including finalizing linkage maps and new genomes and performing comparative analysis to improve the quality of the available genomes by the Comparative Genomics Team.	

* To whom results are transferred during the project.

Table 1. Summary of genetic and genomic resources developed during year 1 and year 2.

Resource	Lab	Status	Citation	Data release status
Genomes				
Bilberry genome	Chagne D.	Completed*	Wu et al., 2021	Genome assembly, annotation and raw data released in GDV
Blueberry genome diploid (clone W85-23)	Iorizzo M.	Work in progress*		
Cranberry genome (Steven)	Zalapa J	Completed*	Diaz et al., 2021	Genome assembly, annotation and raw data released in GDV
Cranberry genome (Ben Lear inbred line)	Polashock J., Vorsa N.	Completed*	Kawash et al., 2021	Genome assembly, annotation and raw data made available in GDV**
Blueberry pangenome	Edger P.	Work in progress*		Raw data released on NCBI
Cranberry pangenome	Edger P.	Work in progress		Raw data released on NCBI
Linkage maps				
Reveille x Arlen (RxA) 1	Iorizzo M.	Completed*	Mengist et al. 2021	SNPs sequences released in GDV
Reveille x Arlen (RxA) 2	Iorizzo M.	Work in progress*		
Draper Selection 44392 x Jewel	Iorizzo M.	Work in progress*		
Draper x Biloxi	Giongo L.	Work in progress		
Hortblue Petite x Nui	Chagne D.	Work in progress*		
Nui x Hortblue Petite	Chagne D.	Work in progress*		

*indicates resources that were shared and are being used for the comparative genome analysis and SNP catalog development.

** indicates data that were made available in GDV but not released publicly (will be released upon publication and or upon request).

Table 2. Source of data used to compile the existing SNP catalog.

Collaborator	Study Type
Breeding Insight	Diversity Panel
David Change	Map Population
Driscoll's	Diversity Panel
Ebrahiem Babiker	Diversity Panel
Ebrahiem Babiker	Diversity Panel
Hamid Ashrafi	Diversity Panel
Jeannine/Lisa Rowland	Map Population
Kalpalatha Melmaiee	Diversity Panel
Massimo Iorizzo	Map Population
Massimo Iorizzo	Map Population
Massimo Iorizzo	Diversity Panel
Patricio Munoz	Diversity Panel
Patricio Munoz	Map Population
Total	13

Table 3. List of traits represented in the existing SNP-QTL catalog.

20Sfirm(20SFirm)	Scar
3mmfirm(3mmFirm)	Size
BlueFDF	Weight
Firm,Size	Titrate acidity
Firmness	Total soluble solids
Firm	Flavor
DeltaFDF	Fruit weight
DeltaFDF/FirmRetention	Full bloom (FB)
Diameter(Dia)	Early green (EG)
Diameter(Dia),DeltaFDF/FirmRetention	Chilling requirement (ChillReq)
SizeRetention	Cold hardiness (ColdHard)
Color	

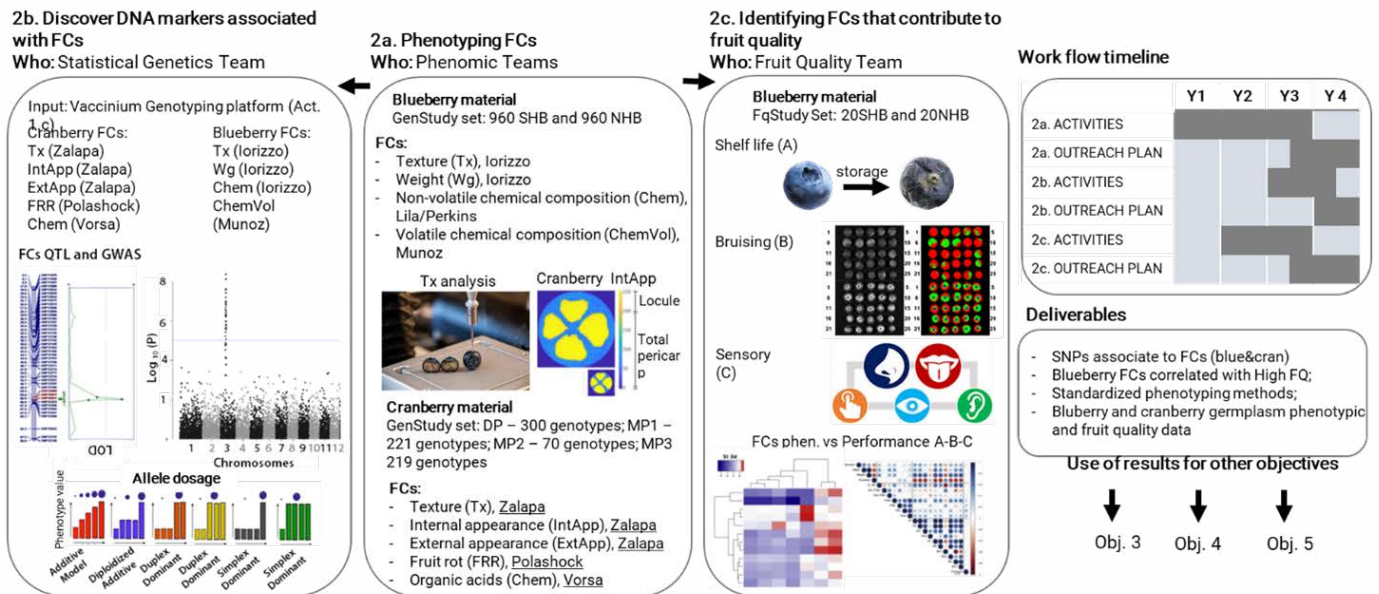
Obj. 1 - VacCAP Plan for Year 3

Obj. 1. Establish genomic resources to enable effective association mapping studies in blueberry and cranberry.
1a. Developing the <i>Vaccinium</i> pangenome [Comparative Genomic Team]
Finalize sequencing of CB transcriptome and genome annotation for all cranberry genotypes
Finalize construction of the pangenome (blueberry-specific, cranberry-specific, and pan- <i>Vaccinium</i>)
Identify cultivar-specific genes associated with fruit quality
1b. Compiling SNP catalog [Genotyping Team]
Finalize compilation of cranberry existing SNPs from team members and partners
Identify locations of existing SNPs in the cranberry pangenome and annotate them
Finalize compilation of existing set of cranberry sequences from public database (GDV) and unpublished data from team members and partners for de-novo SNP identification
Identify and annotate de novo SNPs
1c. Developing genotyping platform [Genotyping Team]
Prepare lyophilized leaf tissue from 192 cranberry accessions to genotype in the testing phase
Finalize selection of 50,000 probes to use in the testing phase for blueberry and cranberry
Complete testing phase for blueberry and initiate genotyping
Initiate and complete testing phase for cranberry
Possible challenges. A bioinformaticist in the Data Management Team will be conducting SNP detection in cranberry and may use a slightly different pipeline than was used in blueberry. Since cranberry is a diploid species, this will most likely not affect the final outcome.

Objective 2

Discover DNA markers and fruit characteristics that maximize industry profitability and match consumer preferences in blueberry and cranberry

The Phenomics, Statistical Genetics and Fruit Quality Teams will identify DNA markers associated with fruit characteristics (FCs) and subcomponents that reduce fruit bruising, contribute to an extension of fruit shelf life, and match consumer preferences. This outcome will establish a link between DNA markers associated with FCs, and fruit quality attributes.



Method Overview

To achieve this objective, the Comparative Genomic and Genotyping Teams will:

- 2a. Phenotype FCs. A blueberry and cranberry Genetic Study set will be phenotyped for texture, weight, appearance and chemical composition (Table 1). The blueberry GenStudy set represents the two predominant cultivated genetic backgrounds, Northern (NHB) and Southern highbush blueberry (SHB), and includes 960 NHB (120 NHB families) and 960 SHB (120 SHB families) individuals. The cranberry GenStudy set includes three bi-parental mapping populations (MP1, 2 and 3) and a diversity panel (DP) set.
- 2b. Discover DNA markers associated with FCs. Individuals will be genotyped using the *Vaccinium* genotyping platform developed in Obj. 1c. Genotyping and phenotypic data from the cranberry DP and all blueberry genotypes in the GenStudy set will be used for GWAS. Genotypic and phenotypic data from cranberry MP 1, 2 and 3 will be used to construct genetic linkage maps and identify Quantitative Trait Loci (QTLs).
- 2c. Three independent experiments will be performed to evaluate which FCs (and sub-components) contribute to the three major indicators of blueberry fruit quality, improved shelf life, matching consumer preferences, and reduction of fruit damage from mechanical harvest (Table 2). These experiments will be performed using the FqStudy set (20 NHB and 20 SHB) which was selected for variation in firmness, shelf life and sensory data; thus, increasing the discriminatory statistical power for these analyses.



M. Iorizzo



J. Zalapa



M. Lila



C. Li



C. Sims



P. Munoz



P. Perkins-Veazie



L. Giongo



N. Vorsa



C. Luby

Obj. 2 Blueberry – VacCAP: Year 2 Progress Summary

Obj. 2. Discover DNA markers and fruit characteristics that improve fruit quality in blueberry.	
Reported by: Perkins-Veazie, Lila, Giongo, Iorizzo, Munoz, Luby, Bassil, Li	
Presented by: Iorizzo, Perkins-Veazie, Giongo	
2a. Phenotyping fruit characteristics (FCs) [Phenomic Teams]	To whom*
Finalize phenotyping protocols, equipment needs and settings. Blueberry: Developed a protocol to use a QR barcode system to collect/store/phenotype and distribute blueberry fruit samples. Barcodes can be directly scanned by phenotyping or processing stations. Some minor adjustments were made to the integrated multi-trait phenotyping collection system that was developed in year 1. The system can simultaneously evaluate 28 fruit traits including texture (Tx), scar diameter (ScD), scar tear (ScT), wet scar (WSc), fruit weight (Wg) and shelf life indicators such as wrinkle/shrivel (Wr/Shr), mold and leakage (Lk) (Table 4). Completed the implementation of this system at three core labs (Perkins-Veazie, Munoz and Luby) where staff and students were trained. Tested the use of Near Infrared Spectrometry (NIR) to quantify sugars and organic acids content and profile. A model for prediction of soluble sugars (fructose, glucose, sucrose) was validated (R=0.8 to 0.9), whereas the model for organic acid was not predictive (R=0.2 to 0.4). The sugar NIR assay is being used to phenotype the GenStudy sets and one mapping population (RxA). For efforts to compare multiple texture analysis methods, a new method was developed and implemented using a needle probe to analyse and capture texture skin subcomponents of blueberry for genetic analysis.	Fruit Quality, Statistical Genetic and Breeding Teams and Partners
Phenotype FCs. Blueberry: Evaluated FCs and shelf life indicators on the NHB GenStudy set (Y1 N=1,032; Y2 N=960), SHB GenStudy set (Y2 N=980), and two mapping populations, DxJ, (Y1-Y2, N=196) and RxA (Y2, N=364) (Table 4). Except for chemical composition all FCs were evaluated at the harvest time and after storage (six or seven weeks under controlled conditions (Table 4). Preliminary analysis indicated a wide range of variation for most of the traits and parameters. As expected, pH and TA were highly correlated while these traits were not correlated with TSS. Preliminary analysis of FC vs shelf life indicators did not identify FC evaluated at T0 that are predictive of extended shelf life. Additional analyses are being performed and other statistical models will need to be evaluated. Fruits for volatile chemistry analysis were frozen and are being phenotyped. Phenotypic data are being analyzed and were used for breeding purposes (see Obj. 3).	Statistical Genetic and Breeding Teams
Expected Impact. The newly developed texture analysis methods measure multiple mechanical parameters that contribute to texture and overcome the limitation of the most commonly used blueberry and cranberry firmness analysis method (Firmtech) that only evaluates external firmness. This will allow us to better understand which texture parameter and/or other FCs contribute to fruit firmness and shelf life, and if it is genetically inherited. Phenotypic data provided information to Oregon and Florida Breeding Programs for advancing selection and selecting parents to use in new crosses to develop cultivars with improved fruit characteristics and shelf life. Development of new methods for evaluating texture and chemical composition will contribute to increasing the number of the potential users of the technique used to phenotype these FCs in blueberry.	
2b. Discover DNA markers associated with FCs [Statistical Genetics Team]	To whom
Collect material, extract DNA from GenStudy set: Completed the collection of leaves from blueberry GenStudy sets.	Breeding and Genotyping Teams, and Geneticist Partners
Perform Genetic analyses. Blueberry: Completed a QTL mapping study for fruit quality traits (pH, TA, TSS, Wg) in a biparental population RxA (Table 5). In total, 18 QTLs were identified, six were stable across three years, two were stable across two years and 10 QTLs were detected in only one year. Stable QTLs across two years for TA and pH overlapped in the same regions. QTLs are being used for validation, candidate gene analysis and evaluated for use in DNA marker assay development. Preliminary evaluation of sugars and organic acid composition in RxA indicated that it is segregating for these traits and most importantly, citric acid was highly correlated with TA and pH. This population is being further evaluated for these metabolites.	
Expected Impact. We expect that FCs-DNA marker associations will be identified. This outcome will provide the fundamental knowledge to understand the position, number of QTLs (as proxy for genes) involved in controlling FCs, and validation of some key FC-QTLs to be used for marker assisted selection.	

2c. Identifying FCs that contribute to essential fruit quality traits [Fruit Quality Team]	To whom
<p>Perform FQ analysis - shelf life: A set of 60 cultivars (named here NC-FqStudy set) were harvested in North Carolina and held up to six weeks at 3°C to follow changes in texture, fruit quality, and fruit chemistry. Completed evaluation of the following FCs include 14 Tx parameters, Weight (Wg), Scar Tear (ScT), Scar Diameter (ScD) and the shelf life indicators: Wrinkle, Leakage, Mold. All FC's were measured at harvest (T0) and followed biweekly for six weeks (T6). Fruit chemistry (pH, TA, SSC, soluble sugars, and organic acids) are being evaluated. Preliminary analyses indicates that fruit stored up to six weeks was found to differ in skin elasticity as well as firmness.</p>	Breeding, Fruit Quality and Other Industry Partners
<p>Analyze cell wall composition: Three cultivars from the NC-FqStudy set were used to determine cell wall components in pulp and peel of crisp, firm, and soft cultivars at harvest. A protocol for cell wall composition analysis was established. Preliminary analysis indicated that blueberry peel has a higher content of arabinogalactan, associated with hemicellulose, than blueberry pulp. Differences in relative amounts of neutral sugars in soft, firm, and crisp types indicate that texture phenotypes may be mirrored in cell wall neutral sugar content and profiles.</p>	Fruit Quality Team and Partners
<p>Perform FQ analysis bruising: A machine learning model based web application software that can automatically determine the blueberry bruise was developed. Bruising was assessed for over 50 images with 50 sliced blueberries within each image using the trained models and the developed website application software. The developed website application is a robust tool for blueberry breeders and farmers to evaluate berry bruising. The method can easily be used on computer and mobile devices and a patent application has been submitted for this tool. The method was applied to evaluate 50 cultivars in the NC-FqStudy set. Bruising was induced by dropping fruit from 45 cm to a steel plate and digital images were collected and analyzed with the new bruising web application software. Preliminary analysis indicated significant variation among cultivars. Bruising data in this set are being used for correlation with all traits measured at harvest time.</p>	Breeding, Fruit Quality and Other Industry Partners
<p>Development of a new method to evaluate blueberry fruit cluster traits. A framework of 3D segmentation was developed for following individual blueberries during cluster development and to extract blueberry cluster traits. An image-capturing system was developed to capture blueberry images to facilitate 3D reconstruction and a 2D-3D projection-based photogrammetric pipeline was proposed to extract berry cluster traits. The reconstruction was performed for four Southern highbush blueberry cultivars ('Emerald', 'Farthing', 'Meadowlark' and 'Star') with 10 cluster samples for each cultivar based on photogrammetry. The final trait extraction algorithm was used to segment individual 3D blueberries to obtain berry number, individual berry volume, and berry maturity. The accuracy of determining the fruit number in a cluster was 97.3% and individual berry volume had an error of less than 0.292 cm³ for clusters with a berry number greater than 10. This study developed an effective method based on 3D photogrammetry and 2D instance segmentation that can determine blueberry cluster traits accurately from a large number of samples and can be used for fruit development monitoring, yield estimation, and harvest time prediction.</p>	Statistical Genetic Team, Breeding, and Other Industry Partners
<p>Texture probes comparison and correlation with sensory. During year 2 the Fruit Quality Team completed a study to evaluate the discriminative power of texture analysis using the following three methods: 1) Penetration flat probe; 2) needle pucture; and 3) double compression. A set of 24 cultivars were evaluated at harvest and postharvest (6 weeks cold storage). The results highlighted: 1) All the methods were able to discriminate cultivars; 2) the needle probe was able to detect difference in skin thickness observed for the different tissue layers (epithelial and hypodermal); 3) the compression test allows for an aggregation of the influence of the mechanical properties of both the skin and pulp of the blueberry and was useful to derive adimensional indexes. This can also be useful if the aim is to identify traits such as springiness, chewiness and resilience, which can be associated with consumer overall liking. In addition, it can be more informative than the other two methods when the blueberries are monitored in postharvest, because the changing dynamics of all the parameters can be observed on the intact tissue structure of the berries.</p>	Breeding, Fruit Quality and Other Industry Partners
<p>Evaluating simple tools that estimate firmness: To address the Advisory Board's recommendations to identify methods that can be easily used in a breeding program for evaluating firmness the following three experiments were performed: 1) OR team evaluated fruit firmness using a Baxlo handheld durometer, FruitFirm 1000, and Firmtech and data will be correlated with mechanical texture data measured with the texture analyzer. 2) FEM team completed a study that demonstrated that firmness measurements from handheld dynamometer were highly correlated with mechanical comparative texture data; 3) UF team measured firmness using a FruitFirm 1000 and data will be correlated with mechanical texture data measured by texture analyzer.</p>	Statistical Genetic Team, Breeding, and Other Industry Partners

Expected Impact. We expect to identify FCs subcomponents that positively correlate with extended shelf life, consumer preferences and reduced internal bruising, contributing to improved fruit quality. This outcome will enable us to establish a link between DNA markers associated with FCs, and high priority fruit quality attributes. Overall, the outcomes of this objective is enabling blueberry breeders and allied scientists to plan development of DNA marker assays to facilitate breeding for FCs and quality attributes that can lead to increased industry profitability in the long term. Phenotypic data are providing information to Oregon and Florida Breeding Programs for advancing breeding selection and about parents to use in new crosses to develop cultivars with improved fruit characteristics and shelf life. Development of new methods to evaluate texture and chemical composition will contribute to increasing the number of the potential users of the technique used to phenotype these FCs in blueberry.

Challenges & Changes. Given that the NIR method for organic acids was not predictive and that evaluating organic acids (OA) by HPLC in very large number of samples is not feasible, the Fruit Quality and Statistical Genetic Teams are considering evaluating OA in a mapping population and a subset of the GenStudy set. To expand our capacity to detect and validate QTLs across studies, additional materials including four mapping populations and a new cranberry diversity panel were added to the project for genetic studies. Due to travel restrictions associated with COVID-19, international teams re-allocated funds to complete complementary projects. For Obj. 2c, the original plan was to use the 20 NHB and 20 SHB cultivars from Florida and Oregon (named OR and FL FqStudy Set) to perform shelf life and bruising analysis in addition to the sensory tests. Difficulties due to COVID-19 to get undamaged fruit led to using NC material instead (NC-FqStudy set). The shelf life experiment was extended to six weeks instead of four weeks based on year 1 preliminary data. As a collaborator from Dr. Li's lab could not travel to NC to perform the bruising analysis, the induced bruising and photos were done by Dr. Perkins' lab. Additional work to evaluate fruit cluster and how it affects mechanical harvest was performed by Dr. Li, complementary with VacCAP objectives.

Addressing challenges and recommendations: New activities were developed to address year 1 challenges and AP recommendations (see Obj. 2c).

* To whom results are transferred during the project.

Table 4. Summary of blueberry phenotyping activities completed or ongoing for the GenStudy sets.

Traits	Phenotyping time	Type and # parameters	Methods	GenStudy set NHB		GenStudy set SHB	Draper x Jewel (DxJ)		Reveille x Arlen (RxA)	New methods reference
FC Traits										
Texture and Appearance FC										
Texture (Txlp) Needle probe	T0* and T1*	4 mechanical parameters	TA.XTPlus***	N=1,032, pheno done	N=960, pheno done	N=1010, pheno done	N=196, pheno done	N=364, pheno done	Giorgio et al. 2021, PBT 183, 111696; Iorizzo et al. 2021, ISHS presentation	
Texture (Txpp) Penetration probe		14 mechanical parameters	TA.XTPlus***							
Weight (Wg) (Size**)		Avg. individual berry weight and avg. 10-12 berry weight	High Precision Mettler Scale***							
Scar (ScD)		Diameter (ScD)	Digital caliper***							
Chemical composition										
Non-Volatile chemical composition (Chem)	T0	Titrateable acidity (TA), pH, Soluble Solid Content (SSC)	Digital refractometers and pH meter	N=814, pheno done	Ongoing	N=965, pheno done	NP	NP	Perkins et al. 2021, Acta Hort.	
		Sugars (SSg): fructose, glucose, sucrose	NIR & HPLC	Ongoing	Ongoing	Ongoing	NP	Ongoing		
		Organic acids (Ac): malic, citric, quinic, shikimic	HPLC	TBD	TBD	NP	NP	Ongoing		
		Volatile organic composition (> 60 volatile compounds)	GC-MS	NP****	Ongoing	NP	NP	NP		
Shelf life indicators										
Wrinkle/Shrivel (Shr)	T0 and T1	Scale 0-3, and % yes/no	Visual***	N=1,032, pheno done	N=960, pheno done	N=980, pheno done	N=196, pheno done	N=364, pheno done	Giorgio et al. 2021, PBT 183, 111696; Iorizzo et al. 2021, Proceeding of XII ISHS Int Vac Symp	
Leakage (Lk)		% yes/no	Visual***							
Mold		% yes/no	Visual***							
Mold color		Visual	Visual***							
Stem scar Tear (ScTr)		% yes/no	Visual***							
Wet stem scar (WSc)		% yes/no	Visual***							
Weight loss (WgLo)		Avg. 10-12 berry weight (T0-T1)	High Precision Mettler Scale***							
Storage Index (SI), (estimated for all Tx and Shelf life indicators)		SI = log2(TiPH/TiH) TiH= Tx at harvest TiPH= Tx post storage	TA.XTPlus							

* T0= 12-24 hours post-harvest; T1= 6 weeks post harvest for Oregon, 7 weeks post harvest for Florida. Material was stored at the following conditions: 3°C and 95% RH in OR; 1°C and 95% RH in FL.

** Fruit weight can be used as a proxy of fruit size in blueberry (Mengist et al. 2020, <https://doi.org/10.3389/fpls.2020.00370>).

*** Phenotypic data collected using the integrated phenotyping system with TA.XTPlus and Exponent software.

**** Phenotyping not planned for this material/ trait

Table 5. Summary of FC QTL studies completed in blueberry.

Crop	FC	Material	FC sub-component	# QTL	# Stable QTL/Years	Reference
Blueberry						
	Chem	RxA	pH	5	1 / 3 Yrs	Mengist et al. 2021, Hort Research 8, 169 (2021). https://doi.org/10.1038/s41438-021-00605-z
		(N=287)	TA	3	2 / 3 Yrs	
			SSC	3	2 / 3 Yrs	
	Appearance		Wg/Size	7	1 / 3 Yrs; 2 / 2 Yrs	

Obj. 2 Blueberry – VacCAP: Plan for Year 3

Obj. 2. Discover DNA markers and fruit characteristics that improve fruit quality in blueberry.
2a. Phenotyping fruit characteristics [Phenomics Team]
<i>Complete analysis of year 1 and 2 data</i>
<i>Continue testing new protocols in GenStudy set:</i> Depending on the analysis of year 1-2 data, the team is considering to use alternative texture analysis probes for NHB GenStudy set and mapping populations.
<i>Complete phenotyping GenStudy set</i>
<i>Evaluate new traits.</i> Consider the possibility to phenotype bruising rates using image analysis tool developed in Obj. 2c, for genetic mapping in DxB population and correlate with the new texture analysis dataset.
2b. Discover DNA markers associated with FCs [Statistical Genetic Team]
<i>Finalize material collection, extract DNA from GenStudy sets</i>
<i>Complete genotyping GenStudy set (blueberry)</i>
<i>Perform preliminary QTL mapping in biparental populations evaluated in year 1-2</i>
<i>Complete QTL analysis for multiple FCs in Draper x Biloxi and Nui x Hortblue Petite populations using existing phenotypic data</i>
2c. Identifying FCs that contribute to essential fruit quality traits [Fruit Quality Team, Penelope Perkins Lead]
<i>Complete Chem (non volatile) on NC-FqStudy set</i>
<i>Analyze y2 data to consider changes and new experiments to set up in year 3 and plan publications.</i>
<i>Consider evaluating the NC-FqStudy set with compression probe for shelf life and bruising experiments.</i> Decision depending on results of year 2 data that is ongoing.
<i>Complete cell wall analysis in post-harvest</i>
<i>Advance new methodology for texture/sensory analysis and shelf life.</i> At FEM, an acoustic implementation of the texture protocols to focus on crispness analysis is planned. The texture profiles will be tested also during postharvest at different conditions, with particular regard to normal cold storage vs modified atmosphere storage.
<i>Excepted challenges.</i> New operational restrictions to prevent the spread of COVID-19 infections could affect the following activities: Harvesting and phenotyping of blueberry and cranberry year 3 materials and delay laboratory work (volatile and non volatile analysis). Some supplies for cell wall analysis has been in backorder and may delay completing this work.

Obj. 2 Cranberry – VacCAP: Year 2 Progress Summary

Obj. 2. Discover DNA markers and fruit characteristics that improve fruit quality in cranberry.	
Reported by: Zalapa, Polashock, Vorsa	
Presented by: Zalapa	
2a. Phenotyping fruit characteristics (FCs) [Phenomic Teams]	To whom*
Finalize phenotyping protocols, equipment needs and settings. Completed testing four texture analysis methods/probes (penetration probe, blade, compression and shear) that measured 37 parameters. Data was collected from samples taken from cultivars in refrigerated storage over a period of six months. Each measurement's ability to detect changes in firmness due to the berries softening over time was compared to the results of The American Society of Agricultural and Biological Engineer's (ASABE) standard (ASAE R2017) for measuring the firmness of convex food items with a compression test performed on the same samples. This test, along with others, was able to identify 10 parameters measured with the double compression method, that were able to perform as well as or better than the standard protocols.	Fruit Quality, Breeding, Statistical Genetic and Breeding Teams and Partners
Phenotype FCs. Completed evaluation of texture (Tx), external appearance (ExtApp), internal appearance (IntApp), Fruit weight and Yield, on 3 mapping populations (MP1, N= 172, MP2, N=71, MP3, N=211)(Table 6). Evaluated % fruit rot in MP3, total anthocyanin, Brix and titratable acidity in MP1-3, and fruit wax in MP4-5 (Table 6). Preliminary analysis indicated a wide range of variation for most of the traits and parameters.	Statistical Genetic and Breeding Teams
Expected Impact. The newly developed texture analysis methods measure multiple mechanical parameters that contribute to texture and overcome the limitation of the most commonly used blueberry and cranberry firmness analysis method that only evaluate external firmness. This will allow a better understanding of which texture parameter and/or other FCs contribute to fruit firmness, and if it is genetically inherited. Phenotypic data are providing information to Breeding Programs for advancing selection and selecting parents to use in new crosses to develop cultivars with improved fruit characteristics. Development of new methods for evaluate texture and other FCs are contributing to increasing the number of the potential users of the technique used to phenotype these FCs in cranberry.	
2b. Discover DNA markers associated with FCs [Statistical Genetics Team]	To whom
Collect material, extract DNA from GenStudy set. Collection of plant material for cranberry GenStudy sets (MP1-3) was completed and collection of plant material for other GenStudy sets is ongoing.	Breeding and Genotyping Teams, Breeders and Geneticist Partners
Perform Genetic analysis. Completed QTL mapping studies for fruit rot resistance (FRR), fruit weight (Wg) and Yield in a Diversity Panel, and for organic acids (citric, malic acid) in two biparental populations (Table 7). Six QTLs for fruit Wg, 5 QTLs for FRR, and 1 QTLs for Yield were identified in the DP. For organic acids, two major QTLs, one in each population, have been identified (overlapping in the same region). These two QTLs segregate as a single Mendelian locus for low vs high MA and were stable across 2 years. Correlation analysis, indicated that citric and malic acids have a positive (moderate) significant correlation with TA. Preliminary QTL mapping in MP3, MP4 and MP5, identified additional QTLs for FRR, wax, TA, TAc, Brix and analysis for these QTLs are being completed.	
Expected Impact. FCs-DNA marker associations are being identified. This outcome is providing the fundamental knowledge to understand the position, number of QTLs (as proxy for genes) involved in controlling FCs, and validation of some key FC-QTLs.	
Challenges & Changes. Due to COVID-19, transplanting of the cranberry diversity panel at Dr. Vorsa location had to be postponed, delaying phenotyping FCs in this material.	
Addressing challenges and recommendations. To ensure success of the genetic studies, additional mapping populations were evaluated for FCs, and an additional diversity panel at the Wisconsin - USDA-ARS (Zalapa) location was used for genetic analysis and is being considered for genotyping and phenotyping in year 3 for texture and appearance traits.	

* To whom results are transferred during the project

Table 6. Summary of cranberry phenotyping activities completed or ongoing for the GenStudy sets.

Table 6. Summary of laboratory phenotyping activities completed or ongoing for the GenStudy sets.																									
Traits		Type and # parameters		Methods		GenStudy MP1 (N=172)				GenStudy MP2 (N=71)				GenStudy MP3 (N=211)				GenStudy MP4 (N= 141)				GenStudy MP5 (N= 68)		New methods reference	
						2019		2020		2019		2020		2019		2020		2019		2020					
FC Traits																									
Texture and Appearance FC																									
Texture (TxCp) compression probe		31 mechanical parameters		TA.XTPlus ***																		Phillips et al., 2021 . Proceeding of XII ISHS Int Vac Symp			
External Appearance (ExtApp)		Length vs Width, Projected Fruit Volume, Eccentricity, Solidity, Black and White Color Scale, Black and White Color Scale Variance, Fruit Length, Fruit Width, Projected Area, Projected Perimeter		Image analysis		Done		Done		Done		Done		Done		Done		NP*		NP*		NP*			
Internal Appearance (IntApp)		Wall Size, Locule Size, Flesh Size, Internal Flesh area, Fruit Width, External to Internal Flesh Ratio, Flesh Area to Locule Area Ratio		Image analysis																		Diaz-Garcia et al., 2020. PlosOne, 14(9): e0222451.			
Weight (Wg)		Avg. Wg		Precision scale																					
Fruit wax				Visual rating		NP*		NP*		NP*		NP*		NP*		NP*		Done		Done		Done			
Fruit Shape				Visual rating																					
Fruit Rot Resistance (FRR)		Sound fruit Wg, % of rot per accession		Precision scale		Done		Done		Done		Done		Done		Done		Done		Done		Done			
Chemical composition																									
Non-Volatile chemical composition (Chem)		Total Anthocaynin Organic acids: quinic, citric, and malic Brix TA		Spectrophotometric HPLC Refractometer Titration		NP**		NP**		NP**		NP**		Ongoing		Ongoing		Ongoing		Ongoing					
Yield (total Wg)		Total Wg/sq. ft.		Precision scale		Done		Done		Done		Done		Done		Done		Done		Done		Done			

NP* indicates that evaluation of the trait was not planned in this set of material
NP** indicates that this trait was already evaluate outside of VacCAP for this plant material

Table 7. Summary of FC QTL studies completed in cranberry.

Crop	FC	Material	FC sub-component	# QTL	# Stable QTL/Years	Reference
Cranberry	Fruit Weight	DP (N=293)	Avg Fruit Weight	6	NA	Diaz-Garcia et al., 2021. Front Plant Sci. 2020;11:607770. https://doi.org/10.3389/fpls.2020.607770
	Fruit rot resistance		Fruit rot	5	NA	
	Yield		Total Yield	1	NA	
	Chem - organic acids	CM151 (N=49)	Low Malic acid (mala) < 2.5 mg/g	1	1 / 2 Yrs	Fong et al., 2020. Tree Genetics & Genomes 17, 4 (2021). https://doi.org/10.1007/s11295-020-01482-8
		CM100 (N=33)	Low Malic acid (mala) < 2.5 mg/g	1	1 / 2 Yrs	

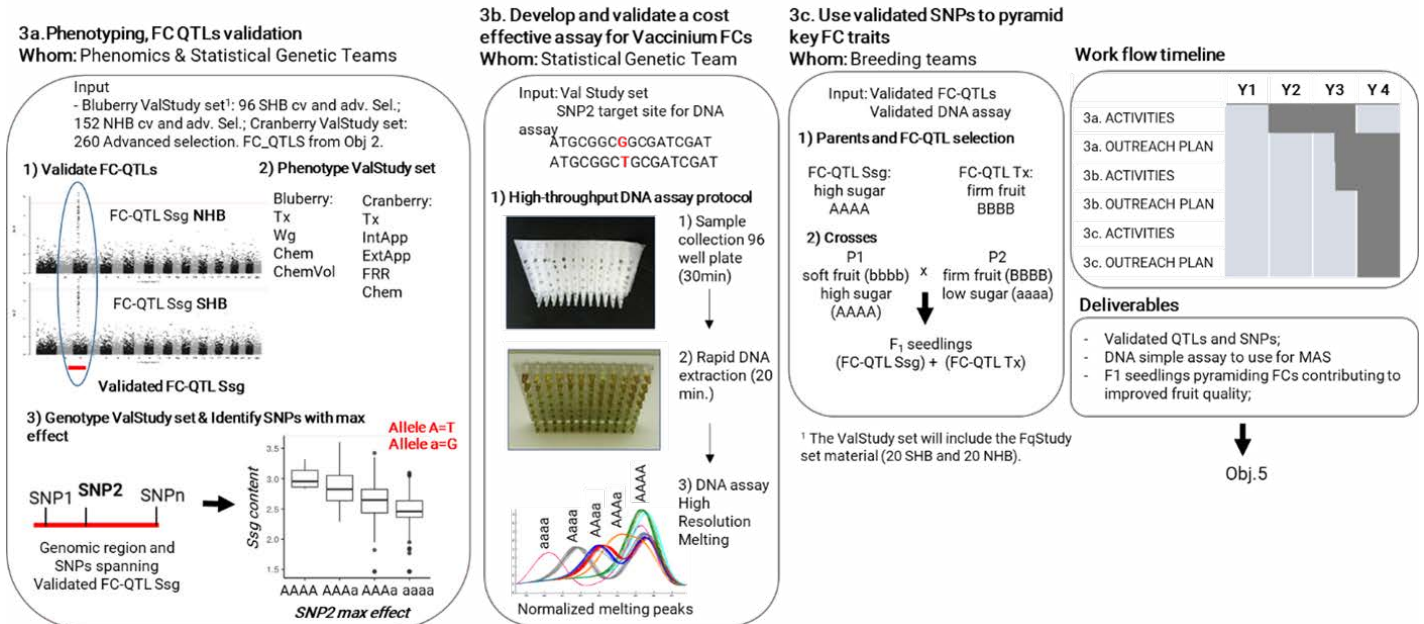
Obj. 2 Cranberry – VacCAP: Plan for Year 3

Obj. 2. Discover DNA markers and fruit characteristics that improve fruit quality in cranberry.
2a. Phenotyping fruit characteristics [Phenomics Team]
<i>Continue testing and developing new methods for phenotyping FC:</i> Considering evaluation of a method to phenotype texture in fresh vs frozen cranberry.
<i>Continue phenotyping FCs.</i> Phenotype an additional mapping population MP6 (N=219), and two DP (Vorsa and Zalapa) (N=200) for texture, chemical composition, appearance, and yield traits.
<i>Complete analysis of year 1-2 data.</i>
2b. Discover DNA markers associated with FCs [Statistical Genetic Team]
<i>Finalize material collection, GenStudy sets.</i>
<i>Perform genotyping GenStudy sets.</i>
<i>Perform QTL mapping analysis for data collected in year 1-2.</i>
Expected challenges. New operational restrictions to prevent the spread of COVID-19 infections could affect the following activities: Harvesting and phenotyping of cranberry year 3 materials; delay laboratory work. Including backorder delays of supplies.

Objective 3

Deliver molecular and genetic resources to improve blueberry and cranberry fruit quality traits that maximize industry profitability and match consumer preferences

The Breeding and Statistical Genetic Teams will validate fruit characteristics (FCs) QTLs in blueberry and cranberry and develop cost effective DNA assays to select parents with FCs that positively contribute to fruit quality and market value. Phenotypic data from Obj 2a-2c, 3a, 4a-b will also be used to select breeding lines with desirable FCs to establish new crosses. These outcomes will provide breeders empirical data to assign a level of importance to FCs relative to consumer preferences, decay during production, processing and distribution, and provide new tools to select for high fruit quality.



Method Overview

To achieve this objective, the Breeding and Statistical Genetic Teams will:

- 3a. Phenotype and validate FC-QTLs. A validation study set (ValStudy set) that will include cultivars used in breeding programs for both blueberry and cranberry, will be phenotyped for all of the FCs evaluated in Obj. 2a and genotyped using the *Vaccinium* genotyping platform. FC-QTLs and existing FC-QTLs from Obj 1b and 2b will be validated in the Validation Set. FC-QTLs will be considered validated when markers show predictive ability for the targeted trait in the ValStudy set. SNPs confirming the genetic association and with max effect will be used as targets to design high-throughput DNA assays.
- 3b. Develop and validate high-throughput cost effective assay for *Vaccinium* FCs. The aim of this activity will be to develop a high-throughput protocol that is fast, cheap and enables the rapid sampling of plant tissue, DNA extraction and precise genotyping. A plate-based DNA assay using the validated FC-SNPs from Obj.3a will be used for targeted genotyping (such as High Resolution Melting, KASP or rhAMP). Markers developed from this objective will be evaluated for consistency to confirm marker-trait associations to be used for marker-assisted selection (MAS) of elite breeding materials.
- 3c. Pyramid key traits using validated SNPs and phenotypic data. Progenies with the desired FC markers or desired FC phenotypic profile evaluated in Obj 2a-b and 3a will be crossed to 'pyramid' multiple positively associated markers into superior genotypes. Breeding priority will be given to genotypes that inherit the highest number of superior FCs, determined by SNP-genotyping and phenotypic data.



P. Munoz



N. Vorsa



M. Iorizzo



J. Zalapa



J. Polashock



C. Luby

Obj. 3 – VacCAP: Year 2 Progress Summary

Obj. 3. Deliver molecular and genetic resources to improve blueberry and cranberry fruit quality traits that maximize industry profitability and match consumer preferences.

Reported by: Munoz, Vorsa, Polashock, Luby, Perkins-Veazie, Iorizzo, Lila

Presented by: Munoz (blueberry), Polashock (cranberry)

3a. Phenotyping, FC QTLs validation and fine mapping [Phenomic and Statistical Genetic Teams]	To whom*
<p>Phenotype ValStudy Set. Blueberry. Completed evaluation of FCs and shelf life indicators (26 parameters), and nonvolatile chemistry (TA, pH, SSC, Sugars) on NHB year 1-2 ValStudy set, and SHB year 2 ValStudy set. Tx and appearance FC and shelf life indicators were evaluated at the harvest time and six (NHB) and seven (SHB) weeks post-harvest (Table 8). Preliminary analysis of NHB year 1 and SHB year 2 data indicate a wide range of variation for most of the traits and parameters. As expected, based on previous studies pH and TA are highly correlated ($R > 0.78$), and no correlation was detected between pH and TA with SSC. Volatile chemistry analysis is ongoing. Cranberry: Phenotyping carried out for several cranberry traits. These include yield, fruit rot, fruit weight, organic acids content, fruit wax, Brix, total anthocyanins and titratable acidity.</p> <p>Collect material, extract DNA from ValStudy set. Blueberry: Collected leaves from blueberry NHB and SHB ValStudy sets (NHB, N=114, SHB, N=109). Samples were stored at -20 °C and ready for submission to RapidGenomics for genotyping. Collected and extracted DNA from additional 384 NHB and SHB samples that were tested with KASP assay. Cranberry: Collected leaves from 196 diverse genotypes from the ValStudy set. Samples were lyophilized and are ready for submission to RapidGenomics for genotyping.</p> <p>Develop list of target existing FC-QTLs. Blueberry: Anchored existing QTLs from six studies (QTL and GWAS) to the Draper reference genome. In total 52 QTLs for FC, FQ, yield and environmental adaption were anchored across all 12 chromosomes. The existing QTLs will represent a framework to identify QTLs for FC-trait in this study that overlap across different blueberry genetic studies. For example, overlapping QTLs for pH on chr 3 were identified across two studies. QTLs detected across multiple studies could be considered as validated QTLs and used to design DNA assays to test their prediction ability and thus their potential for Marker Assisted Selection. Cranberry: Three QTLs, two for malic acid (MA), and one for citric acid (CA) were anchored to the newly developed cranberry genome (Ben Lear inbred line). The QTL for MA (mala/mala) affects CA and quinic acid content and confers a decreased TA to below 1% that is in the range of other fresh fruit such as strawberries. The QTLs for MA overlapped to a region of about 140kb in chr 4. The QTL for CA was anchored to chr 1 and spans a region of about 500kb. These regions were used to validate SSR and SNP assays (KASP and rhAMP) for organic acid content (Table 9). Other QTL are in various stages of SNP, SSR primer design and testing/validation. These include fruit rot resistance (chromosomes 2, 4, 10, 11), fruit wax (chromosome 1), titratable acidity (chromosome 8), fruit anthocyanin content (chromosomes 5, 6, 8), Brix (chromosome 5), color (chromosome 3).</p>	<p>Genotyping and Breeding Teams, Vaccinium Breeders, and Geneticist Partners.</p>
3b. Develop and validate a high-throughput affordable assay for Vaccinium FCs [Statistical Genetic Team]	To whom
<p>Finalize selection and protocol for simple DNA assay. Blueberry: Initiated the development and evaluation of a rapid DNA extraction method and testing two genotyping high-throughput assays including High Resolution Melting (HRM) and Kompetitive allele specific PCR (KASP). For the KASP assay, 48 loci representing QTLs associated with volatiles, hard and soft core genes detected in the pangenome (Obj. 1a) were tested in 384 NHB and SHB genotypes. SNPs data will be used to test their application for MAS and fingerprint analyses. Cranberry: Two SSR markers and 4 SNPs markers were designed and tested for sensitivity and specificity to predict low, moderate and high citric and malic acid content in four biparental populations. All markers were highly specific (> 0.81) and sensitive (> 0.91). SSR scf258d has the highest sensitivity (0.94) and specificity (0.99) for citric acid. SNP MA_476 tested by KASP assay, had the highest sensitivity (0.94) and specificity (1.0) for malic acid. PACE markers have been designed for fruit wax and fruit rot resistance (260 genotypes from different crosses that have fruit rot resistance background). These markers are currently being tested.</p>	<p>Genotyping and Breeding Teams, Vaccinium Breeders, and Geneticist Partners.</p>

3c. Pyramid key traits using validated SNPs and phenotypic data [Breeding Teams]	To whom
<p>Leverage FC and FQ phenotypic data for advancing breeding selections. Blueberry: Based on 2020 and 2021 phenotypic data from the SHB, NHB GenStudy and ValStudy sets, 16 and 29 NHB genotypes and 116 SHB genotypes, respectively, were identified as having berries with good or outstanding flavor and texture at harvest and/or after six/seven weeks in storage. The USDA breeding program (Luby) selected 4 of the genotypes from the 2020 data to replicate and trial as advanced selections and made 7 crosses in 2021 using the phenotypic information gathered through the VacCAP project. The UF/IFAS breeding program used this data to identify parents for the 2022 crosses. Cranberry: Based on the results of the organic acids study, crosses were made to introgress the mala allele into a unrelated high yielding low acid variety to evaluate the major gene's effect in a quantitative low acid background. Individual progeny from over 30 populations, derived from previous crosses from the four genetic sources of FRR, were evaluated for yield, fruit quality and FRR. Selected progeny were used as parents in various crosses, e.g., disassortive and assortive matings (>30 crosses), for the next breeding and selection cycle. These populations will be field planted in 2022 and 2023 and also used for marker validation. Using phenotypic methods and data (Obj. 2) in cranberry for fruit firmness, external appearance, and internal structure, cranberry selections were made in two arenas: in an inbred-hybrid system, 24 selections were made out of 847 plants and 25 selection were made from 300 genotypes representing a diversity panel.</p>	<p>Genotyping and Breeding teams, Vaccinium Breeders, and Geneticist Partners.</p>
<p>Expected Impact 3a-c. Validated QTLs will provide opportunities to evaluate these markers for MAS and also to perform functional characterization of candidate genes. With these validated QTLs we can also plan future ad-hoc experiments to evaluate genotype × environment effects for FCs targeted by validated DNA assays. DNA assays for FCs, and new crosses made based on molecular and phenotypic data will provide foundational work to accelerate breeding for fruit quality in blueberry and cranberry breeding programs.</p>	
<p>Challenges & changes. Due to COVID-19 some activities related to Obj. 3a were delayed. However the team anticipated to year 1-2 beginning of activities associated with the development and validation of DNA assay.</p>	

* To whom results are transferred during the project

Table 8. Summary of blueberry phenotyping activities completed or ongoing for ValStudy sets.

FC Traits	Phenotyping time	Type and # parameters	Methods	ValStudy set	
				NHB	SHB
20202021					
FC Traits					
Texture and Appearance FC					
Texture (TxNp) Needle probe	T0* and T1*	4 mechanical parameters	TA.XTPlus***	N=114, pheno done	N=134, pheno done
Texture (TxPp) Penetration probe		14 mechanical parameters	TA.XTPlus***		
Weight (W g) (Size*)		Avg. individual berry weight and avg. 10-12 berry weight	High Precision Mettler Scale***		
Scar (ScD)		Diameter (ScD)	Digital caliper***		
Chemical composition					
Non-Volatile chemical composition (Chem)	T0	Titratable acidity (TA), pH, Soluble Solid Content (SSC)	Digital refractometers and pH meter	N=114, pheno done	N=109, pheno done
		Sugars (SSg): fructose, glucose, sucrose	NIR & HPLC	Ongoing	Ongoing
		Organic acids (Ac): malic, citric, quinic, shikimic	HPLC	TBD	TBD
Volatile chemical composition (ChemV ol)		Volatile organic composition (>60 volatile compounds)	GC-MS	NP****	N=109, pheno done
Shelf life indicators					
Wrinkle/ShriveI (Shr)	T0 and T1	Scale 0-3, and % yes/no	Visual***	N=114, pheno done	N=134, pheno done
Leakage (Lk)		% yes/no	Visual***		
Mold		% yes/no	Visual***		
Mold color		Visual	Visual***		
Stem scar Tear (ScTr)		% yes/no	Visual***		
Wet stem scar (WSC)		% yes/no	Visual***		
Weight loss (WgLo)	Avg. 10-12 berry weight (T0-T1)	High Precision Mettler Scale***			
Storage Index (SI), (estimated for all Tx and shelf life indicators)		SI= log2(TIPH/TIH) TIH= Tx at harvest TIPH= Tx post storage	TA.XTPlus		

* T0= 12-24 hours post-harvest; T1= 6 weeks post harvest for Oregon, 7 weeks post harvest for Florida. Material was stored at the following conditions: 3°C and 95% RH in OR; 1°C and 95% RH in FL.

** Fruit weight can be used as a proxy of fruit size in blueberry (Mengist et al., 2020, <https://doi.org/10.3389/fpls.2020.00370>).

*** Phenotypic data collected using the integrated phenotyping system with TA.XTPlus and Exponent software.

**** Phenotyping not planned for this material/trait.

Table 9. Summary of DNA assay developed for FCs.

Crop	FC sub-component	Traits	Marker ID	Marker type	Material tested	Sensitivity	Specificity	Reference
Cranberry	Chem - organic acids	Low vs Moderate vs High Citric Acid	scf258d	SSR	Biparental populations	0.94	0.99	Fong et al., 2020. Tree Genetics & Genomes 17, 4 (2021). https://doi.org/10.1007/s11295-020-01482-8
		Low CA < 2.5mg/g; Moderate CA 3-6 mg/g; High CA >6 mg/g	SNP CA_609	KASPs	Biparental populations	0.91	0.98	
		Low vs Moderate vs High Malic Acid	SNP MA_271	KASPs	Biparental populations	0.94	0.81	
		Low MA < 2.5mg/g; Moderate MA 3.5-5 mg/g; High CA >5 mg/g	SNP MA_476	KASPs	Biparental populations	0.94	1	

Obj. 3 – VacCAP: Plan for Year 3

Obj. 3. Deliver molecular and genetic resources to improve blueberry and cranberry fruit quality traits that maximize industry profitability and match consumer preferences.

3a. Phenotyping, FC QTLs validation and fine mapping [Phenomic and Statistical Genetic Teams]

Complete Phenotyping of ValStudy Set

Genotyping needs for ValStudy set

3b. Develop and validate a high-throughput affordable assay for Vaccinium FCs [Statistical Genetic Team]

Finalize protocol for the fast and simple DNA assay

Continue testing protocols for simple DNA assay on a set of existing FC-QTLs

Analyze DNA assay data (blueberry)

Test DNA assays for MA and CA in more diverse populations to evaluate sensitivity and specificity (cranberry)

3c. Pyramid key traits using validated SNPs and phenotypic data [Breeding Teams]

Develop a list of genotypes harboring superior FCs based on phenotypic year 3 data

Validate performance of genotypes selected during year 1-2 for advancing them into trial or to use as a parent in new crosses

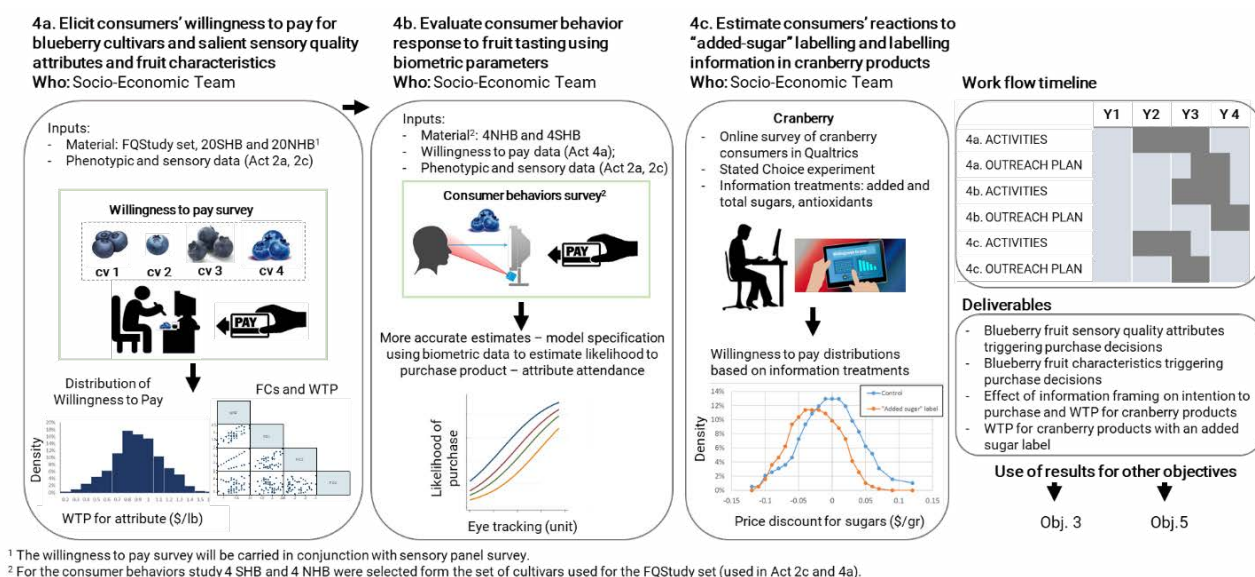
Plant seedlings in nurseries so they are evaluated for their potential contribution to FC

Possible challenges. New operational restrictions to prevent the spread of COVID-19 infections could affect the following activities: Harvesting and phenotyping of blueberry and cranberry year 3 materials; delay laboratory work (volatile and non-volatile analysis); breeding activities.

Objective 4

Assess the potential socio-economic impact of blueberry and cranberry fruit quality improvements on market demand

The Socio-Economic team will evaluate the willingness to pay (WTP) for specific sensory quality characteristics and fruit quality attributes associated with consumer preferences for fresh blueberry and processed cranberry products. Estimates of WTP surveys will inform breeding programs to target the traits of maximum value to consumers. Insights from WTP estimates for cranberry products will inform breeding efforts, specifically to sugar content and acidity in cranberries. Outcomes from this study will provide marketers and stakeholders with insights on new messaging strategies to market blueberry cultivars with improved fruit qualities and cranberry products with nutrition facts.



Method Overview

To achieve this objective, the Socio-Economic team will:

- 4a. Elicit consumers' willingness to pay for blueberry cultivars and salient sensory quality attributes and fruit characteristics. Sensory tasting and willingness to pay elicitation will be combined via the use of sensory taste tests and choice experiments. The study will use 20 NHB and 20 SHB blueberry cultivars, complementing the activities conducted in Act. 2c (fruit quality studies). The selection of these cultivars is based on differences in fruit firmness and sensory profiles that will enable a statistical correlation of sensory characteristics, consumers' preferences and willingness to pay.
- 4b. Evaluate consumer behavior response to fruit tasting using biometric parameters. A subset of four SHB and four NHB cultivars representing a subset of the material used in Obj. 4a, will be used to measure consumers' behavioral reactions to the blueberry sensory quality profile via the collection of biometric data. Respondents will be asked to complete a sensory evaluation questionnaire and their WTP. Data from Obj. 2c, 4a, 4b will be integrated to identify possible FCs that contribute to consumer preferences and WTP.
- 4c. Estimate consumers' reactions to "added-sugar" labeling and labeling information in cranberry products. In this objective an online survey and a choice experiment with a representative sample of U.S. consumers will be conducted to, (a) quantify consumers discount for cranberry products with an "added-sugar" line on the nutrition facts panel label, (b) measure heterogeneity in responses across consumer segments, and (c) evaluate the effect of information framing. The responses will be used to assess consumers' accuracy in evaluating sugar content and evaluate how this affects purchase intent and willingness to pay.



K. Gallardo



E. Canales



C. Sims

Obj. 4 – VacCAP: Year 2 Progress Summary

Obj. 4 Assess the potential socio-economic impact of blueberry and cranberry fruit quality improvements on market demand.	
Reported by: Gallardo, Canales, Sims, Munoz, Iorizzo, Lila, Perkins-Veazie, Bassil, Colonna	
Presented by: Canales/Gallardo and Sims	
4a. Elicit consumers' willingness to pay for blueberry cultivars and salient sensory quality attributes and fruit characteristics [Socio Economic Team]	To whom*
Finalize blueberry WTP and consumer preference survey questionnaires. Completed development of the questionnaire (in Qualtrics), to be used with the sensory taste tests, the logistics to procure fruit and the protocols for the sensory taste tests in conjunction with the sensory scientists CoPI Sims at UF and collaborator Colonna at OSU.	Breeding, Fruit Quality and Statistical Genetic Teams, Blueberry Industry Partners
Perform WTP and consumer preference questionnaire. The team estimated the preferences and willingness to pay for 20 Southern high bush and 20 Northern high bush blueberry varieties (Obj. 2c and 4a Fruit quality set OR and FL) . This was done in conjunction with sensory taste tests conducted in Florida and Oregon. Responses from a total of 240 consumer panelists were collected. Data is being analyzed. In general, there is correlation between the varieties that exhibited the highest ratings of liking scales with the stated willingness to pay. In Florida, the WTP for the 20 Southern high bush varieties range from \$3.51/lb to \$2.99/lb. The following were the highest rated and those who received the highest WTP: 'Colossus', 'Magnus', 'Arcadia', 'Sentinel' and 'Avanti'.	
Integrate WTP, consumer preferences and fruit characteristics measurements. Fruit characteristics (FC) including pH, SSC, TA, texture, sugars, organic acids, volatiles content and profile are measured for integration with survey data. Measurements of texture, pH, SSC, TA for both sets FL and OR were completed. Volatile and sugar analysis are being completed.	
Expected Impact: The willingness to pay for selected fruit sensory quality attributes associated with different cultivars will provide useful information about the attributes that trigger purchase decisions and repeated purchases. The quantitative value (in \$/lb) for each cultivar and quality attribute will inform fruit growers and marketers about the attributes sought by consumers and will also provide information for blueberry breeding programs about fruit traits that maximize consumer value.	
4b. Evaluate consumer behavior response to fruit tasting pricing using biometric parameters [Socio Economics Team]	To whom
Will start on Year 3	Breeding and Fruit Quality Teams, Blueberry Industry Partners
Expected Impact. Information about blueberry quality traits preferred by consumers will be useful for breeders in identifying cultivars with higher consumer acceptance and potential market performance.	
4c. Estimate consumers' reactions to “added-sugar” labelling and labelling information in cranberry products [Socio Economics Team]	To whom
Develop and distribute a U.S. nationwide survey tool. Focused on dried cranberries and cranberry juice. Responses from 4,000 cranberry consumers was collected. Three products were included: dried cranberries, unlabeled cranberry juice (referred to as cranberry juice) and labeled cranberry juice (refers to 100% juice, cocktail, and blend). Preliminary results suggest that consumers are willing to discount for increases in added sugar across all three products. Also the preferred method to achieve the desired sweetness level is the addition of other fruit juice concentrates over adding regular sugar or the combination of regular sugar and fruit juice concentrates. We measured if the disclosure of information on the positive health effects of cranberries can mitigate the discount for added sugars, the results suggest that even receiving positive information on the health benefits, consumers are still willing to discount for added sugar. In general, the study signals that consumers stated a higher discount for unlabeled cranberry juices compared to the labeled counterpart. Consumers were heterogeneous in their perceptions; in general, those attaining college education and with children in the household were the ones consistently discounting for added sugar. Our results offer no conclusive evidence of the effects of self-reported health status (diabetes, cholesterol, heart condition) and trusted sources of information on the discount for added sugars.	Breeding and Statistical Genetics Teams, Cranberry Industry Partners (Nurseries, Processors, Growers, Commodity Group Organizations)
Expected Impact. Information on the potential impact of the added-sugar label on consumers' purchase decisions, the distribution of these impacts across segments of the population, and the impacts of communication strategies (i.e., health benefits messages to counteract the negative effect of “added-sugar” information on the Nutrition facts panel) on consumers will be helpful for cranberry breeding programs and the cranberry industry in formulating targeted marketing strategies for the promotion of cranberry products.	
Challenges & changes: None to report.	

* To whom results are transferred during the project.

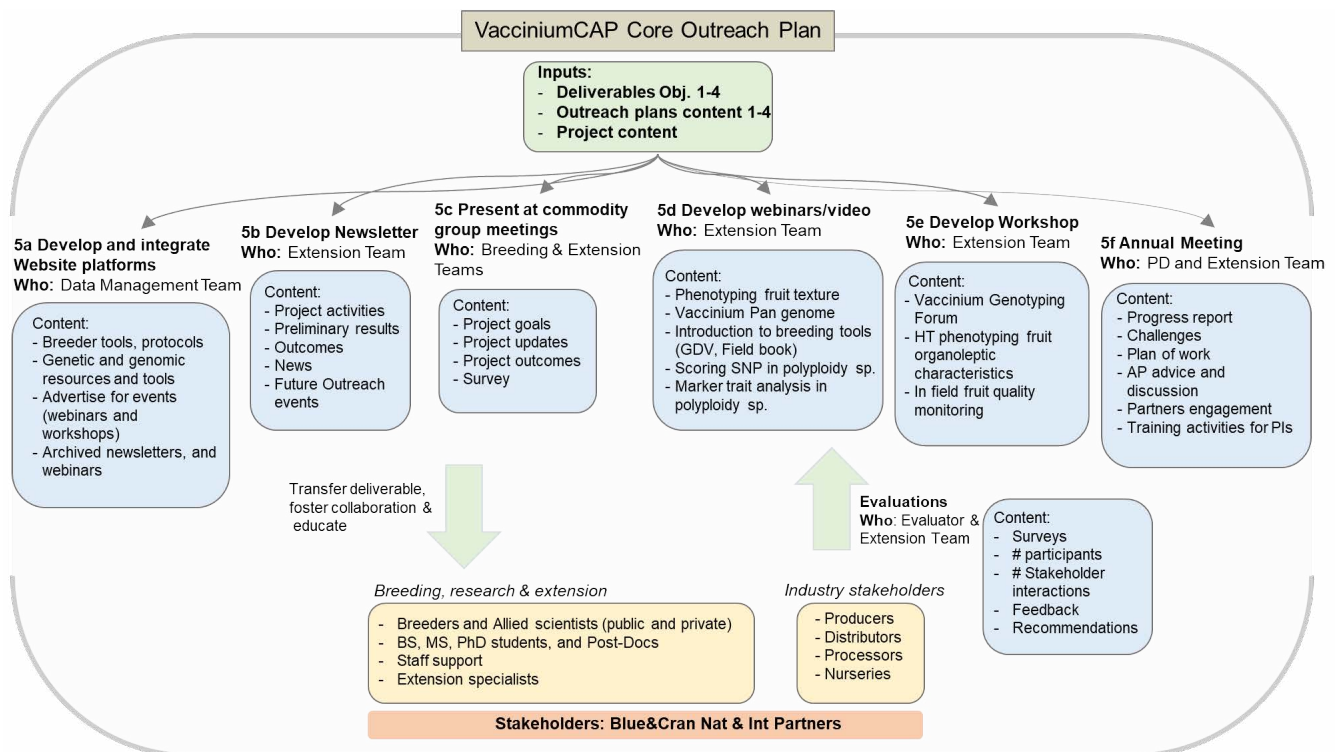
Obj. 4 – VacCAP: Plan for Year 3

Obj. 4 Assess the potential socio-economic impact of blueberry and cranberry fruit quality improvements on market demand.
4a. Elicit consumers' willingness to pay for blueberry cultivars and salient sensory quality attributes and fruit characteristics [Socio Economic Team]
<i>Implement the questionnaire to be used for the blueberry sensory panels that includes WTP elicitation in Year 3.</i>
<i>Finalize the analyses of the data collected in year 2 and prepare manuscripts for dissemination of results.</i>
4b. Evaluate consumer behavior response to fruit tasting pricing using biometric parameters [Socio Economic Team]
<i>Draft questionnaire and experimental design for study</i>
<i>Finalize list of plant materials to be used in study</i>
4c. Estimate consumers' reactions to "added-sugar" labelling and labelling information in cranberry products [Socio Economic Team]
<i>Finalize the data analyses</i>
<i>Outcomes communicated through publications and presentations</i>
Excepted challenges. Obtaining samples of blueberry cultivars on time. Year 2 data analysis is time sensitive, since it will guide year 3 experiments. The team will plan a late fall, early winter meeting to decide how to proceed with year 3 experiments for blueberry (Obj. 4a-b).

Objective 5

Engage U.S. *Vaccinium* stakeholder groups to transfer advanced phenomic and genomic tools to build a more efficient cultivar development system

The Extension and Data Management Teams in collaboration with all VacCAP PIs will use background information and deliverables from the outreach plans of Objectives 1-4 to develop six outreach activities, which will represent our core outreach plan to engage stakeholders. Outreach activities will be developed to engage *Vaccinium* stakeholders by: 1) transferring knowledge and training on project deliverables; 2) educating about project outcomes; 3) fostering new collaborations; and 4) soliciting feedback on the project to fine-tune research and extension activities. Target stakeholders are national and international blueberry and cranberry partners that represent breeders, researchers (breeder's allied scientists), mentees/trainees (staff, mentored students and post docs), extension specialists, and industry stakeholders (producers, processors, distributors, and nurseries), as well as the members of the Advisory Panel (AP) that represent these audiences. Training activities will also target members of VacCAP core labs.



To achieve this objective, the extension and data management teams will:

- 5a. Update and expand online platforms. The existing Genome Database for *Vaccinium* website (GDV, <https://www.vaccinium.org/>) will provide open access to genetic (e.g., all QTLs) and genomic (e.g., pangenome sequences) resources developed in Obj. 1-3. A new VacCAP project website will serve as a repository of information for project participants and *Vaccinium* stakeholders.
- 5b. Develop newsletters. A biannual electronic newsletter will highlight project activities, preliminary results, outcomes, news, and future outreach events.
- 5c. Develop webinars. Webinars will inform breeders, researchers, and mentees/trainees about project results and deliverables, provide technical instructions on how to gain access and use these new resources, which will complement Obj. 5d.
- 5d. Develop workshops and forums. Workshops will train and educate breeders, researchers, mentees/trainees and industry stakeholders on high-throughput phenotyping methods developed and used in the project to evaluate fruit quality traits of blueberry and cranberry.
- 5e. Participate in commodity group meetings. Presentations will be delivered at annual regional grower meetings by members of the Breeding and/or Extension Team and will be facilitated by our extension collaborator network. Presentations will highlight project goals, and relevance to specific industry groups, and will provide opportunities for communication and feedback between the project team and stakeholders.

Objective 5 (Continued)

Engage U.S. *Vaccinium* stakeholder groups to transfer advanced phenomic and genomic tools to build a more efficient cultivar development system

- 5f. Annual meeting. Annual meeting extension activities will include: 1) disseminating project accomplishments to the Advisory Panel and partners; 2) promoting collaborative efforts with partners; 3) hosting workshops specific to the phase of the project.
- 5g. Engage the public (Outreach). To educate the general public about the project outcomes and impacts, team members' will present the VacCAP project and related activities through established outreach activities such as science fairs, guest lectures, and supporting high school science programs.



A. Atucha



L. Wasko DeVetter



D. Main



M. Coe

Obj. 5 – VacCAP: Year 2 Progress Summary

Obj. 5: Engage U.S. Vaccinium stakeholder groups to transfer advanced phenomic and genomic tools to build a more efficient cultivar development system.

Reported by: Atucha, DeVetter, Main and all other PIs

Presented by: Atucha, Main

5.a. Update and expand online platforms [Data Management team]	To whom
<p>VacCAP project website: The project website (https://www.vacciniumcap.org) was launched during YR1 and has been populated with information for project participants, advertisements for events, archives of the newsletter and webinars, and general content about the project and participants. Updates that were done to the VacCAP website in YR2:</p> <ul style="list-style-type: none"> - Website redesign to increase accessibility to project information. - Development of documents summarizing impact of previous planning grant linked to VacCAP Project and Project Overview. - Website stats summary (September 1, 2020–August 31, 2021): 7,230 pageviews; 2,208 users; 1.33 sessions per users. <p>During year 2 we also began the development of the Project Management System.</p>	VacCAP Core Labs and the Vaccinium Genomics, Genetics and Breeding Research Community
<p>Update Genome Database for Vaccinium (GDV): New GDV data updated during year 2 included: 7 genomes and gene models, 466,354 functionally annotated genes; 1,254,515 gene transcripts functionally annotated and mapped to corresponding genomes, 3 genetic maps, 46,261 markers mapped to corresponding genomes; and 62 QTLs as well as functionally annotated orthologs identified through synteny analysis of 9 Vaccinium genomes. GDV was accessed by 5,824 users from 95 countries, over 10,979 visits and 102,278 pages served (Google analytics).</p>	
<p>Develop GDV Newsletter updates: First GDV Newsletter released on 9/9/21 (https://www.vaccinium.org/newsletter).</p>	
5.b. Develop material for newsletters and other social media [Extension Team]	To whom
<p>Develop and release newsletters: Two newsletters were issued during the second year of the project, in May and August 2021. Newsletters are archived on the VacCAP website and are delivered by email to our stakeholders (breeders, researchers, extension specialists, and industry stakeholders, as well as project participants). Newsletter impact: 148 subscribers get the newsletter directly by email, with an opening rate of 48% (compared to an average of 45% among similar email campaigns).</p>	VacCAP Core Labs, Partners, Stakeholders, General Public
<p>Create and maintain VacCAP accounts on social media: A Twitter account (@VacciniumCAP) was created to disseminate information about the project as part of the social media strategy, in conjunction with a YouTube channel. (https://www.youtube.com/channel/UCpAdtvTEebzZjvJ4SJcoXwg). Twitter Stats (August 2020–August 2021): 260 followers; 4,750 (average 365/month) profile visits.</p>	VacCAP Core Labs, Partners, Stakeholders, General Public
5.c. Develop webinars [Extension Team]	To whom
<p>Deliver webinars: The Genome Database for Vaccinium (GDV): A Resource for Vaccinium Genetics, Genomics and Breeding Research on August 19, 2021. Presenters: Dr. Jodi Humann, Dr. Sook Jung and Dr. Dorrie Main (Washington State University) 60 participants.</p>	VacCAP Core Labs, Breeding and Genetic Partners
5d. Protocols and methodology training videos [Extension Team]	To whom
<p>Develop video training. Two instructional videos were produced during Year 2 and posted on the project's YouTube channel. (https://www.youtube.com/channel/UCpAdtvTEebzZjvJ4SJcoXwg). These videos feature the use of new methodology to evaluate fruit quality of blueberries.</p>	VacCAP Core Labs, Partners, and Stakeholders
5e. Develop workshops and forum [Extension Team]	To whom
<p>Deliver workshops: Training and professional development were provided to 500 participants of the American Society for Horticultural Science with two workshops. One workshop covered the collection of fruit chemistry data for large size projects and the other workshop covered ways in which fruit firmness and texture could be measured. Also at the ASHS meeting, as part of the workshop "GWAS and GS in horticultural crops," VacCAP PIs led presentations on computational analysis and tools for genetic studies and genomic selection in polyploid crops.</p>	VacCAP Core Labs, Partners, and Stakeholders

<p>Organize discussion forum/meetings for VacCAP genotyping platform. Two online meetings were organized to discuss parameters and strategy for selecting the final set of 50,000 SNPs to use for testing the genotyping platform. Participants included VacCAP PIs and breeders, and molecular geneticist partners including those that expressed interest in using the platform.</p>	<p>VacCAP Core Labs and the Vaccinium Genomics, Genetics and Breeding Research Community</p>
<p>5f. Participate in commodity group meetings or field days [Extension Team]</p>	<p>To whom</p>
<p>Attend commodity group meetings [meetings in collaboration with industry extension network team]: Delivered 15 presentations at grower association meetings and field days across US states. Developed a handout flyer that summarized the the objectives of the VacCAP project.</p>	<p>Blueberry and Cranberry Growers, Processors, Distributors, Nurseries and Breeding Partners</p>
<p>5g. Annual meeting [Extension Team]</p>	<p>To whom</p>
<p>Facilitate annual meetings targeting project participants, AP members, and national and international partners: Due to the Covid-19 situation and related travel restrictions, the Year 1 annual meeting was conducted online on Nov. 12-13, 2020. An annual report that summarized Year 1 activities and plans for Year 2 was prepared and shared with members of the advisory panel prior to the meeting. Information collected during the annual meeting was used to integrate recommendations from the AP and plan Year 3 activities. The annual report was posted on the VacCAP web site at https://www.vacciniumcap.org/annualreport.</p>	<p>Team Members, AP Members, USDA Program Officers</p>
<p>5h. Engage the public (Outreach) [Extension Team, all teams]</p>	<p>To whom</p>
<p>Perform outreach activities: All outreach activities were suspended during Year 2 due to the COVID-19 pandemic.</p>	<p>General Public, Students</p>
<p>External Evaluation [Extension Evaluator]</p>	<p>To whom</p>
<p>Coordinate the design and implementation of surveys for webinar, workshop and meeting participants and assist with annual report forms for project participants: Participant surveys for the webinar noted in Obj 5c were developed with input from team members; an online survey portal was used to collect feedback from webinar participants. Feedback forms for Advisory Panel members and project team members were developed and deployed at the annual project meeting on November 12, 2020 and the project Working Group meeting on November 13, 2020, to gather systematic feedback from the AP and from project team members on each project objective and on the format of the meetings. An internal survey for team members was developed and implemented in November 2020 to gather feedback aimed at refining project management processes. Assisted with the updated design and implementation of the annual project report form for project Co-PIs and Co-PDs. Worked with genotyping team to develop and implement surveys of stakeholder needs and preferences for genotyping tools, costs and sample size.</p>	<p>Team Members, Extension Audiences</p>
<p>Manage, analyze, interpret and report project evaluation feedback, recommendations, and related data: Feedback and recommendations for the project from webinar participants, Advisory Panel and Working Group participants, and stakeholder genotyping surveys were reported back to the team and are being used to inform future project activities.</p>	<p>Team Members, Extension Audiences, Stakeholders, USDA Program Officers</p>
<p>Assist with integration of evaluation data into project reports, annual meeting and Advisory Panel materials, and outreach materials: Assisted with collation, analysis, and integration of project evaluation data, Advisory Panel and Working Group recommendations, and internal team annual reports into the annual project report for USDA.</p>	<p>Team Members, Extension Audiences, Stakeholders, USDA Program Officers</p>
<p>Expected Impact. Breeders, researchers, trainees/mentees will benefit from the adoption of the genetic and genomic tools developed in this project that will facilitate the application of Marker Trait Association studies, and long-term will result in the next generation of blueberry and cranberry cultivars with improved fruit quality traits. Extension specialists and industry stakeholders will learn and implement accurate high-throughput phenotyping methods to effectively select fruit with desired quality characteristics. Industry stakeholders will learn which fruit quality traits affect market demand, leading to new marketing strategies to increase and sustain consumer demand. The proposed extension activities will also foster team collaboration and new collaborations outside the scope of this project, providing further value to stakeholders by ensuring that related activities continue beyond the length of this project.</p>	
<p>Challenges & changes. Due to COVID-19, some activities were not performed as planned: Workshops that were to be held at the International Vaccinium Symposium (ISHS) were delayed, since the symposium has been postponed to 2024. Due to a logistical delay of the development of the pangenome, the webinar on pangenome was also postponed. The team organized and sponsored a workshop on "Exploring Firmness and Texture in Berry Crops and Peach" at the annual ASHS conferece (in virtual format), presented by Dr.Perkins-Veazie, Dr. Zalapa, and Dr. Giongo. Engagement with the public and commodity groups was limited due to shutdown of physical meetings due to COVID-19.</p>	
<p>Addressing challenges and recommendations. Workshops will be held at our next annual meeting in 2022, and we will continue to develop training videos for new methodologies developed by the team. Grower engagement at meetings and conferences will be done in a hybrid model with both virtual and in-person presentations. Outreach to the general public may simply be limited due to the unique situation from Covid-19, but we will continue to advertise, educate, and promote our project where and when feasible.</p>	

Obj. 5 – VacCAP: Plan for Year 3

Obj. 5: Engage U.S. Vaccinium stakeholder groups to transfer advanced phenomic and genomic tools to build a more efficient cultivar development system.

5.a. Update and expand online platforms [Data Management team]

Update VacCAP: New content will be updated in the VacCAP website monthly including: links to peer reviewed publications, newsletter articles, video recordings of webinars and trainings, and social media updates.

Update GDV: New genomics, genetics and breeding data will be curated and added to GDV as it becomes available from peer-reviewed publications. Other information will include Quarterly GDV newsletters, monthly "how to" videos, training webinars and community events.

Train and update core and non-core labs on data and use of GDV: New training videos on how to use GDV will be posted on our website by the team monthly. Also a second issue of the GDV newsletter will be released to update national and international partners on new tools and resources available on GDV.

5.b. Develop newsletters [Extension Team]

Release additional VacCAP newsletter editions: During Year 3 we will issue 2 editions of our VacCAP Newsletter that will be distributed to our stakeholders and partners.

Prepare article for trade magazines: We plan to release one article in a trade magazine during Year 3.

Maintain VacCAP accounts on social media: The team will continue to actively engage stakeholders and partners through our Twitter account.

5.c. Develop webinars [Extension Team]

Three webinars are planned for Year 3: Blueberry volatiles (Dr. Muñoz); Pangenome (Dr. Edger); Firmness and texture in Vaccinium (Dr. Perkins-Veazie, Dr. Zalapa, and Dr. Giongo).

5d. Develop workshops and forums [Extension Team]

Two in-person workshops/trainings will be held in Year 3 in conjunction with our annual meeting (pandemic conditions permitting).

5e. Protocols and methodology training videos [Extension Team]

Instructional videos will be produced during Year 3, posted on the project website and YouTube channel, and distributed to our audience through our newsletter and social media posts. These videos will feature the use of new methodologies for fruit quality phenotyping developed by our team members, as well as evaluation and comparisons of equipment available to stakeholders. In conjunction with the training videos, and when applicable, PDF files with step-by-step protocols will be available on our website.

5f. Participate in commodity group meetings [Extension Team]

In collaboration with the industry extension network team, presentations will be delivered at commodity group meetings to update stakeholders on project progress.

5g. Annual meeting [Extension Team]

The extension team will facilitate annual meetings targeting project participants, AP members, and national and international partners. The location of the 3rd annual meeting has not yet been determined. At this stage, the plan is to organize it in-person during the late summer or early fall of 2022. Possible timeline and venue will be discussed during the annual meeting.

5h. Engage the public (Outreach) [Extension Team, all teams]

Due to continued restrictions on in-person outreach activities due to Covid-19, the team will continue to engage and educate the general public about the project outcomes and impacts through our website, newsletter, and social media.

Coordinate the design and implementation of surveys for webinar, workshop and meeting participants and assist with annual report forms for project participants. Continue to develop and update surveys, feedback forms, report forms and related instruments and systems as needed during Year 3.

Manage, analyze, interpret and report project evaluation feedback, recommendations, and related data. Continue to report findings and recommendations from stakeholders, advisors and project team members as needed for project management and planning during Year 3.

Assist with integration of evaluation data into project reports, annual meeting and Advisory Panel materials, and outreach materials. Continue to help with the integration of findings and recommendations from stakeholders, advisors and project team members into project reports, meeting materials, and outreach materials.

Expected challenges. Due to Covid-19, public engagement and outreach might be restricted during Year 3. Plans for organizing the Year 3 workshop in-person at the annual meeting may change depending on Covid-19. Possible alternative activities to be performed online will be evaluated during Year 3.

VacCAP Team Publications and Presentations

Journal Article

- Diaz-Garcia¹ L, Garcia-Ortega LF, Rodríguez MG, Delaye L, Iorizzo M and Zalapa J. 2021. Chromosome-level genome assembly of the American cranberry (*Vaccinium macrocarpon* Ait.) and its wild relative *Vaccinium microcarpum*. *Frontiers in Plant Science*, <https://doi.org/10.3389/fpls.2021.633310>
- Diaz-Garcia L, Covarrubias-Pazaran G, Johnson-Cicalese J, Vorsa N, Zalapa J. 2020. Genotyping-by-sequencing identifies historical breeding stages of the recently domesticated American cranberry. *Frontiers in Plant Science*, 11:607770. <https://doi.org/10.3389/fpls.2020.607770>
- Fong SK, Kawash J, Wang Y. et al. 2021. A low malic acid trait in cranberry fruit: genetics, molecular mapping, and interaction with a citric acid locus. *Tree Genetics & Genomes* 17, 4. <https://doi.org/10.1007/s11295-020-01482-8>
- Kawash J, Colt K, Hartwick NT, Abramson BW, Vorsa N, Polashock JJ, Michael TP. 2021. Contrasting a reference cranberry genome to a crop wild relative provides insights into adaptation, domestication, and breeding. *bioRxiv* 2021.06.27.450096. <https://doi.org/10.1101/2021.06.27.450096>
- Giongo L, Matteo A, Pottorff M, Mainland M, Perkins-Veazie P and Iorizzo M. 2021. Comparative multi-parameters approach to dissect texture subcomponents of blueberry at harvest and postharvest. *Postharvest Biology and Technology*, Volume 183, 111696. <https://doi.org/10.1016/j.postharvbio.2021.111696>
- Jung S, Lee T, Gasic K, Campbell BT, Yu J, Humann J, Ru S, Edge-Garza D, Hough H and Main D. 2021. The Breeding Information Management System (BIM): an online resource for crop breeding. *Database*, Volume 2021, 2021, baab054. <https://doi.org/10.1093/database/baab054>
- Jung S, Cheng CH, Buble K, Lee T, Humann J, Yu J, Crabb J, Hough H and Main D. 2021. Tripal MegaSearch: a tool for interactive and customizable query and download of big data. *Database*, Volume 2021, 2021, baab023. <https://doi.org/10.1093/database/baab023>
- Karppinen K, Lafferty DJ, Albert NW, Mikkola N, McGhie T, Allan AC, Afzal BM, Häggman H, Espley RV and Jaakola L. 2021. MYBA and MYBPA transcription factors co-regulate anthocyanin biosynthesis in blue-coloured berries. *New Phytologist*. <https://doi.org/10.1111/nph.17669>
- Mengist MF, Bostan H, Young E, Kay K, Ashrafi H, Gillitt N, Ballington J, Ferruzzi MG, M.A. Lila MA and Iorizzo M. 2021. High density linkage map construction and identification of loci regulating fruit quality traits in blueberry. *Horticulture Research*, 8, 169. <https://doi.org/10.1038/s41438-021-00605-z>
- Samkumar A, Jones D, Karppinen K, Dare AP, Sipari N, Espley RV, Martinussen I and Jaakola L. 2021. Red and blue light treatments of ripening bilberry fruits reveal differences in signalling through abscisic acid-regulated anthocyanin biosynthesis. *Plant, Cell & Environment*, 1–19. <https://doi.org/10.1111/pce.14158>
- Staton M, Cannon E, Sanderson LA, Wegrzyn J, Anderson T, Buehler S, Cobo-Simón I, Faaberg K, Grau E, Guignon V and Gunoskey J. 2021. Tripal, a community update after 10 years of supporting open source, standards-based genetic, genomic and breeding databases. *Briefings in Bioinformatics*, bbab238. <https://doi.org/10.1093/bib/bbab238>
- Wu C, Deng C, Hilario E, Albert NW, Lafferty D, Grierson ERP, Plunkett BJ, Elborough C, Saei A, Günther CS, Ireland H, Yocca A, Edger PP, Jaakola L, Karppinen K, Grande A, Kylli R, Lehtol VP, Allan AC, Espley RV, Chagné D. 2021. A chromosome-scale assembly of the bilberry genome identifies a complex locus controlling berry anthocyanin composition. *Molecular Ecology Resources*, <https://doi.org/10.1111/1755-0998.13467>
- Xueping N, Changying L, Huanyun J and Takeda F. 2021. Three-dimensional photogrammetric reconstruction and deep learning instance segmentation to extract berry fruit harvestability traits. *ISPRS Journal of Photogrammetry and Remote Sensing*. 171: 293-309. <https://doi.org/10.1016/j.isprsjprs.2020.11.010>
- Yocca, AE, Edger, PP. 2021. Machine learning approaches to identify core and dispensable genes in pangenomes. *Plant Genome*, e20135. <https://doi.org/10.1002/tpg2.20135>

Conference Papers and Presentations

- Catrin S, Günther, Dare AP, McGhie TK, Deng C, Jaakola L, Lafferty DJ, Plunkett BJ, Grierson ERP, Turner JL, Albert NW and Espley RV. 2021. Spatiotemporal modulation of flavonoid metabolism in *Vaccinium* berries. *International Conference on Polyphenols (ICP)*, Turku, Finland.

VacCAP Team Publications and Presentations

- Espley RV, Catrin S, Günther, Dare AP, Albert NW, Lafferty DJ, McGhie T, Deng C, Plunkett BJ, Turner JL, Jones L, Grierson ERP, Schwinck KE, Davies KM, Allan AC, Jaakola L. 2021. The spatio-temporal accumulation and regulation of anthocyanin in *Vaccinium*. August 31, 2021, XII International *Vaccinium* Symposium, Dalhousie, Canada.
- Ferraro F. 2021. Genome-wide association of fruit-quality traits reveals the genetic basis for blueberry flavor. In: Application and perspectives of genome wide association studies (GWAS) and genomic selection (GS) in Diploid and Polyploid Horticultural Crops Workshop. ASHS Annual Conference, August 5-9, 2021, Denver, Colorado.
- Giongo L, Ajelli M, Pottorff M, Coe K, Perkins-Veazie P, Bassil N, Hummer K, Farneti B, Iorizzo M. Comparative study on texture: a key for blueberry quality breeding. August 31, 2021, Proceedings of the XII International *Vaccinium* Symposium. Virtual format.
- Giongo L, Ajelli M, Pottorff M, Perkins-Veazie P and Iorizzo M. 2021. Assessing blueberry and texture traits associated with consumer acceptance and shelf life. ASHS Annual Conference, August 5-9, 2021, Denver, Colorado.
- Humann J, Cheng CH, Lee TL, Buble K, Jung S, Yu J, Hough H, Crabb J, Frank M, Scott K, Iorizzo M and Main D. 2021. Using the Genome Database for *Vaccinium* for genetics, genomics, and breeding research. XII International *Vaccinium* Symposium, August 30-September 1, 2021, Halifax, Nova Scotia, Canada. Virtual.
- Iorizzo M, Mengist MF, Bostan H, Curaba J, Pottorff M. 2021. From genome to genes and DNA markers to improve agronomic performance and quality of fruit and vegetables crops. *Genetyka Aplikacyjna Roślin XXI*, September 22-24, 2021, Warsaw, Poland. Virtual.
- Iorizzo M, Lila MA, Perkins-Veazie P, Pottorff M, Finn C, Vorsa N, Edger P, Bassil N, Munoz P, Zalapa J, Gallardo KR, Atucha A, Main D, Giongo L, Li C, Polashock J, Sims C, Canales E, Coe ML, Chagne D, Espley R and DeVetter L. 2021. *VacciniumCAP*, a community-based project to develop advanced genetic tools to improve fruit quality in blueberry and cranberry. Proceedings of the XII International *Vaccinium* Symposium. August 30-September 1, 2021, Halifax, Nova Scotia, Canada. Virtual.
- Jung S, Lee T, Cheng CH, Gasic K, Humann J, Yu J, Hough H and Main D. 2021. An update on the Breeding Information Management System (BIMS). Proceedings of the ASHS Annual Conference; Aug 5-9, 2021; Denver, Colorado.
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- Mengist MF, Burtch H, Debelo H, Pottorff M, Bostan H, Nunn C, Corbin S, Kay CD, Bassil N, Hummer K, Lila MA, Ferruzzi MG, Iorizzo M. 2021. Diversity of phenolic bioaccessibility in blueberry germplasm: towards the development of a genetic framework to improve the efficiency of bioactive delivery. ASHS Annual Conference, August 5-9, 2021, Denver, Colorado.
- Mengist MF, Bostan H, Kay K, Gillitt N, Ashrafi H, Ferruzzi MG, Lila MA and Iorizzo M. High density linkage map construction and identification of loci regulating fruit quality traits in blueberry. 2021. ASHS Annual Conference, August 5-9, 2021, Denver, Colorado.
- Munoz P. 2021. Operational application of genomic selection in tetraploid blueberry. In: Application and perspectives of genome wide association studies (GWAS) and genomic selection (GS) in Diploid and Polyploid Horticultural Crops Workshop. ASHS Annual Conference, August 5-9, 2021, Denver, Colorado.
- Perkins-Veazie P, Ma G, Pottorff M, Lila MA and Iorizzo M. 2021. New tools for rapid fruit quality analysis in blueberry. August 31, 2021, Proceedings of the XII International *Vaccinium* Symposium. Virtual format.
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- Perkins-Veazie P. 2021. Faster Methods for Postharvest Compositional Analysis. *HortScience* 56.
- Perkins-Veazie P. 2021. Introduction and overview of texture and instruments. *HortScience* 56.
- Phillips M, Diaz-Garcia L, Grygleski E, Giongo L, Atucha A, Iorizzo M and Zalapa J. 2021. Cranberry challenges in distinguishing soft and firm berries. ASHS Annual Conference, August 5-9, 2021, Denver, Colorado.
- Samkumar A, Karppinen K, Martinussen I, Espley R, Jaakola L. 2021. Light quality from supplemental light-emitting diodes modulates biosynthesis of secondary metabolites in ripening bilberry (*Vaccinium myrtillus* L.) fruit. August 30, 2021, XII International *Vaccinium* Symposium, Dalhousie, Canada. Virtual format.
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blueberry cultivars. ASHS Annual Conference, August 5-9, 2021, Denver, Colorado.

- Trandel MA, Perkins-Veazie P, Iorizzo M and Johanningsmeier S. 2021. Exploring the plant cell wall in blueberry varieties of different texture. *Acta Horticulturae*.
- Trandel MA, Perkins-Veazie P, Iorizzo M and Johanningsmeier S. 2021. Optimization of blueberry cell wall extraction from grinding through hydrolysis. *HortScience*, 56.
- Yalcin O, Finn C, Mackey T, Pottorff M, Iorizzo M, Hardigan M, Luby C, Bassil NV. 2021. Towards QTL analysis of phenological and fruit quality traits in a tetraploid highbush blueberry population. ASHS Annual Conference, August 5-9, 2021, Denver, Colorado.

Conference Posters

- Humann J and Main M. Genome Database for Vaccinium (<https://vacciniumcap.org>) XII International Vaccinium Symposium, August 30-September 1, 2021, Virtual.
- Mengist MF, Bostan H, Kay K, Gillitt N, Ashrafi H, Ferruzzi MG, Lila MA and Iorizzo M. 2021. High density linkage map construction and identification of loci regulating fruit quality traits in blueberry. XII International Vaccinium Symposium, August 30-September 1, 2021, Virtual.
- Pottorff M, Zielinski K, Mengist M, Honigs D, Grace M, Lila MA and Iorizzo M. 2021. A high-throughput phenotyping method using near infrared spectroscopy to measure anthocyanin content in blueberry. XII International Vaccinium Symposium, August 30-September 1, 2021, Virtual.
- Yalcin O, Finn C, Mackey T, Pottorff M, Iorizzo M, Hardigan M, Luby C, Bassil NV. 2021. Towards QTL analysis of phenological and fruit quality traits in a tetraploid highbush blueberry population. ASHS Annual Conference, August 5-9, 2021, Denver, CO, USA.

Workshops

- Perkins-Veazie P. 2021. "Exploring Firmness and Texture in Berry Crops and Peaches". ASHS Conference, August 7, 2021, Denver, Colorado.
- Perkins-Veazie P. 2021. "Rapid and High-Throughput Methods for Assessing Produce Quality". ASHS Conference, August 6, Denver, Colorado, 2021.

Webinars

- Humann J, Cheng CH, Lee T, Buble K, Jung S, Yu J, Hough H, Crabb J, Frank M, Scott K, Iorizzo M and Main D. 2021. Genome Database for Vaccinium: Genetics Data and Tools. GDV Webinar, August 19, 2021. <https://www.youtube.com/watch?v=ulOakNp-K8g&list=PLMHx16OgbKiMZbXLY8wJXnvAgw7XTiyxU&index=1>
- Jung, S, Lee T, Cheng CH, Gasic K, Humann J, Yu J, Hough H, Iorizzo M and Main D. 2021. Genome Database for Vaccinium: Breeding Data and Tools. GDV Webinar, August 19, 2021.

Instructional videos

- Perkins-Veazie P, Iorizzo M, Oh H. 2021. How to use an acidity meter and the 'offset function' for blueberries. <https://www.youtube.com/watch?v=w1uXscw3jXo>
- Perkins-Veazie P, Iorizzo M, Oh H. 2021. How to use a handheld Brix meter for blueberries. <https://www.youtube.com/watch?v=VP2maYlt0CO>

Presentations for Growers or Other Industry Stakeholders

- Atucha A, and Zalapa J. 2021. Cranberry School. Wisconsin Dells, WI. January 28, 2021.
- DeVetter, L. 2021. Washington Small Fruit Conference. Lynden, WA. December 1, 2021. Virtual format.
- DeVetter L, Bassil N, Luby C. 2021. VacCAP: Improving Fruit Quality. Blueberry Field Day. Oregon State University's North Willamette Research & Extension Center in Canby, Oregon. July 15, 2021.
- Iorizzo, M. VacCAP project update. 2021. North Carolina Blueberry Council Annual Open House. January 13, 2021. Virtual format.
- Iorizzo M. 2021. Leveraging Genetic and Genomic Resources to Enable Development of Blueberry and Cranberry Cultivars with

VacCAP Team Publications and Presentations

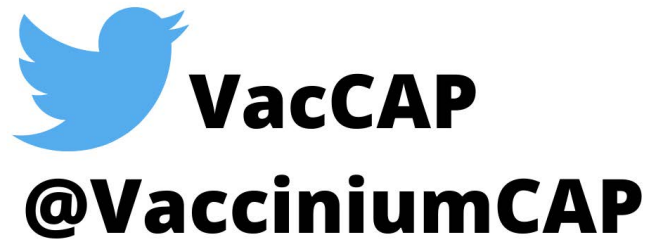
Improved Fruit Quality Attributes. NABC/USHBC Spring meeting. March 18, 2021.

- Iorizzo M, Hancock J, Munoz P. 2021. The Business of Blueberries. Coordinated Effort to Improve Blueberry Genetics" U.S. Highbush Blueberry Council. Episode 31. January 21, 2021. <https://ushbc.blueberry.org/podcast/coordinated-effort-to-improve-blueberry-genetics/>
- Muñoz, P. 2021. Spring Florida Blueberry Growers Association. March 8, 2021. Virtual format.
- Perkins-Veazie P. 2021. Postharvest of Berries. Berry Grower Meeting, Virginia State University. February 18, 2021. Virtual format.
- Phillips M, Diaz-Garcia L, Grygleski E, Atucha A, Iorizzo M, Zalapa J. 2021. An oral presentation on firmness measurement methods in cranberry presented to other researchers in the fruit firmness field. ASHS Annual Conference, August 2, 2021, Denver, Colorado.
- Polaschock, J. 2021. American Cranberry Grower Association Winter meeting, New Jersey. January 23, 2021. Virtual format.
- Polaschock, J. 2021. Blueberry Open House New Jersey. March 9, 2021. Virtual format.
- Polaschock, J. 2021. Cranberry Summer Field day. August 19, 2021.
- Sandler, H. 2021. University of Massachusetts Cranberry Station Annual Extension and Research Update Meeting. January 26, 2021. Virtual Format.
- Zalapa J. 2020. VacCAP project discussion. Wisconsin Cranberry Research Roundtable. Madison, WI. November 9, 2020.
- Iorizzo M. 2021. Blueberry research updates at the Piedmont Research Field Station. August 17, 2021.

VacCAP newsletters

- Atucha A, Russo J, DeVetter L, Gallardo K and Canales E. 2021. Buying Better Berries: What Does Fruit Quality Mean to Consumers? VacCAP Newsletter Issue 2, May 2021. <https://www.vacciniumcap.org/buyingbetterberries>
- Atucha A, Russo J, DeVetter L and Zalapa J. 2021. Piecing Together the Cranberry Genome Puzzle. VacCAP Newsletter Issue 2, May 2021. <https://www.vacciniumcap.org/cranberrygenomepuzzle>
- Atucha A, Russo J, DeVetter L, Bocock B and Serres R. 2021. Fruit Quality: An Industry Perspective. VacCAP Newsletter Issue 2, May 2021. <https://www.vacciniumcap.org/fruitqualityindustryperspective>
- Atucha A Russo J, DeVetter L, Sims C, Gallardo K and Canales E. 2021. Sensory Tasting Survey Helps Connect Fruit Quality Traits and Consumer Preferences in Blueberry. VacCAP Newsletter Issue 3, August 2021. <https://www.vacciniumcap.org/sensorytastingsurvey>
- Atucha A, Russo J, DeVetter L and Vorsa N. 2021. Fighting Fruit Rot: Dr. Nicholi Vorsa's Breeding Program Focuses on Developing Resistant Cranberry Cultivars. VacCAP Newsletter Issue 3, August 2021. <https://www.vacciniumcap.org/fruitrot>
- Atucha A, Russo J, DeVetter L and Li C. 2021. New Imaging Methods Help Researchers Find Blueberry Cultivars Resistant to Bruising. VacCAP Newsletter Issue 3, August 2021. <https://www.vacciniumcap.org/imagingmethods>
- Humann J and Main D. 2021. Genome Database for Vaccinium Newsletter, Issue 1, September 2021. <https://www.vaccinium.org/newsletter>

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- Get the latest articles and resources
- Find webinar announcements and registration links
- Check out photos from the field
- And so much more!

Also check out our new website www.vacciniumcap.org and [YouTube channel](#) to see articles and videos as they go live.

Partners: Do You Want to Contribute to the VacCAP Newsletter?

If you are interested in contributing or have announcements for events, publications, or another other initiative that you want to share, contact Josie Russo at jrusso2@wisc.edu with the subject line "VacCAP Newsletter Contribution"



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www.vacciniumcap.org
vaccapsocial@gmail.com | [@VacciniumCAP](https://twitter.com/VacciniumCAP)