VacCAP

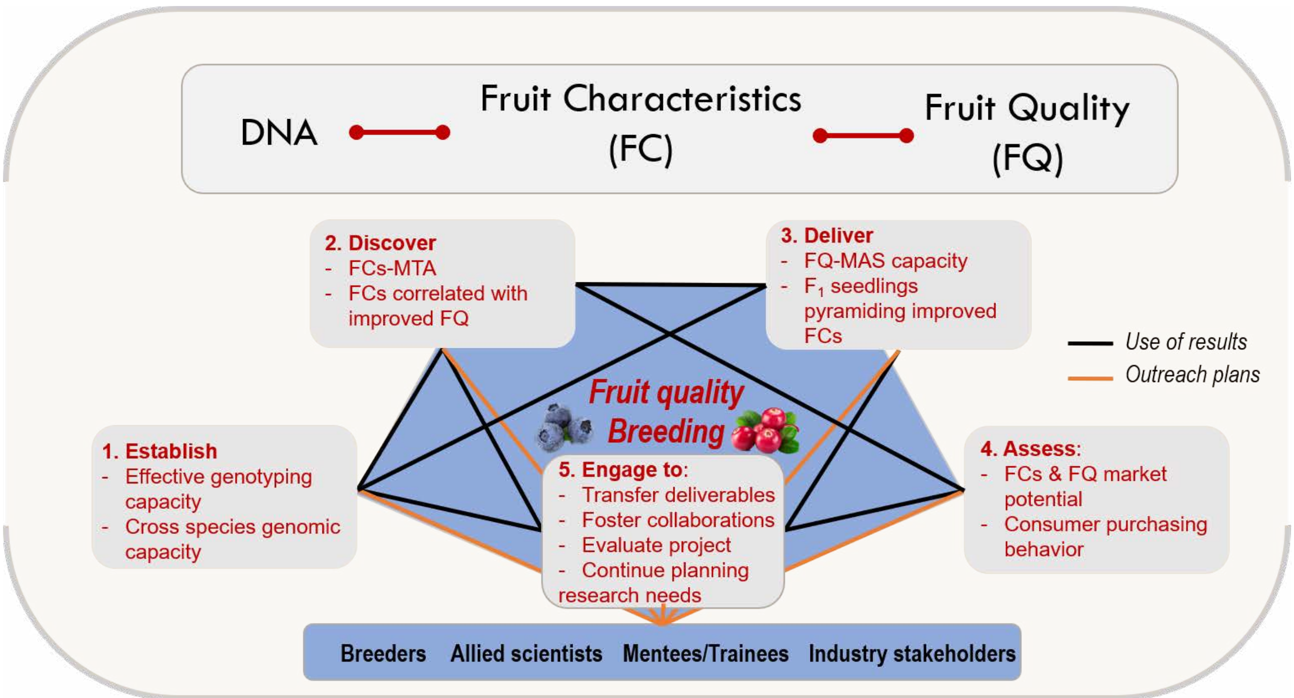
IMPROVING FRUIT QUALITY

Annual Project Report and Plan

Annual Meeting IV | March 2–3, 2023 | Hybrid

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 | Thursday, March 2, and Friday, March 3, 2023 | Hybrid

Time (EST)	March 2
7:30-8:30	Breakfast
8:30-10:00	Session 1. Intro and Obj. 1 Genomics and Genotyping Moderator: Bassil • Iorizzo, Massimo - Intro annual meeting • Bassil, Nahla - Intro Obj. 1 • Edger, Patrick - Goals of PanGenome and future directions • Yocca, Alan - Construction and analysis of pangenome • Bassil, Nahla & Clare, Shaun - Genotyping platform Discussion
10:00-12:00	Session 2. Obj. 2-3 Cranberry: Phenotyping, Genetics, and Marker Development Moderator: Zalapa • Zalapa, Juan - Intro obj. 2, cranberry • Lopez, Moreno Hector - Cranberry texture method • Loarca, Jenyne – Cranberry diversity panel evaluation • Knowles, Sara - Phenotyping mapping population MP3 • Polashock, James - Intro obj. 3, MP3 genetics and DNA assay development Discussion
12:00-1:00	Lunch
1:00-3:00	Session 3. Obj. 2-3 Blueberry: Phenotyping, Genetics, and Marker Development Moderator: Iorizzo • Iorizzo, Massimo - Intro Obj. 2, chem traits update • Ferrao, Felipe - Genetics of volatile and other traits • Oh, Heeduk - Post-harvest study 1 • Mengist-Fentie, Molla - Post-harvest study 2 • Giongo, Lara - Acoustic method • Perkins-Veazie, Penelope & Li, Charlie – Bruising • Munoz, Patricio - Intro Obj. 3, status of DNA marker development Discussion
3:30-4:30	Session 4. Obj. 5 Extension Moderator: Atucha • Atucha, Amaya - Intro Obj. 5 • Humann, Jodi - GDV updates • Atucha, Amaya – VacCAP updates • Coe, Michael - Evaluation summary Discussion
4:30-5:00	International Collborator Updates • NZ team – blueberry genome assembly using trio-mating
6:00	Group Dinner at Boardwalk Billy's Raw Bar and Ribs, walking distance from hotel

**See next page for time conversion table*

Agenda

Time	March 3
8:30-10:00	Session 1. Obj. 4 Blueberry and Cranberry Socio Economic Studies Moderator: Gallardo • Gallardo, Karina - Intro Obj. 4 • Gallardo, Karina - Cranberry consumer studies • Ma, Xueying - Blueberry consumer and grower studies • Sims, Charles - Blueberry sensory and fruit characteristics • Canales, Elizabeth – Willingness To Pay estimates for sensory parameters and fruit characteristics Discussion
10:00-12:00	Session 2. Partners and Advisory Panel Moderator: Iorizzo • Moira, Sheehan - Support for USDA-ARS Blueberry and Cranberry breeding programs • Rife, Trevor - GDV training • Neyhart, Jeff - Cranberry/blueberry pre-breeding • Ru, Sushan - Expanding Southern Highbush Blueberries to Underserved Regions of Southeastern U.S. Questions
12:00-1:00	Lunch
1:00-3:00	Administrative meeting (Only PIs zoom link will be sent by email)

Thursday March 2, 2023

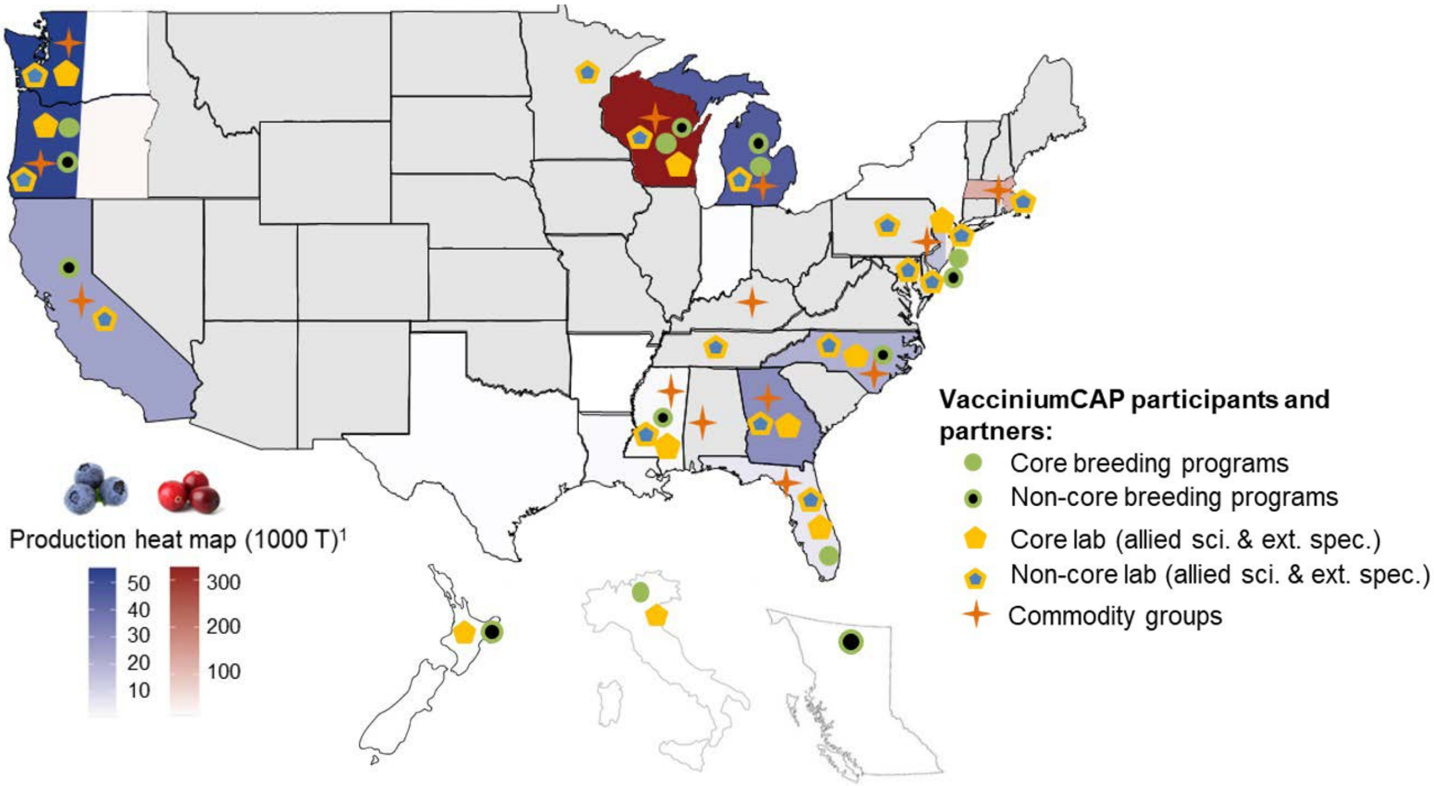
Time zone	Breakfast	Session 1	Session 2	Lunch	Session 3	Session 4	Session 5	Dinner
PT (-3)		5:30AM	7:00AM	9:00AM	10:00AM	12:30PM	1:30PM	
CT (-1)		7:30AM	9:00AM	11:00AM	12:00PM	2:30PM	3:30PM	
ET	7:30AM	*8:30AM*	10:00AM	12:00PM	1:00PM	3:30PM	4:30PM	6:00PM
IT (+6)		2:30PM	4:00PM	6:00PM	7:00PM	9:30PM	10:30PM	
NZ (+17)		2:30AM	4:00AM	6:00AM	7:00AM	9:30AM	10:30AM	

Friday March 3, 2023

Time zone	Breakfast	Session 1	Session 2	Lunch	Administrative meeting
PT (-3)		5:30AM	7:00AM	9:00AM	10:00AM-12:00PM
CT (-1)		7:30AM	9:00AM	11:00AM	12:00PM-2:00PM
ET	7:30AM	*8:30AM*	10:00AM	12:00AM	1:00PM-3:00PM
IT (+6)		2:30PM	4:00PM	6:00PM	7:00PM-9:00PM
NZ (+17)		2:30AM	4:00AM	6:00AM	7:00AM-8:00AM

Table of Contents

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
- Jennifer Johnson-Cicalese, Co-PI, Rutgers University

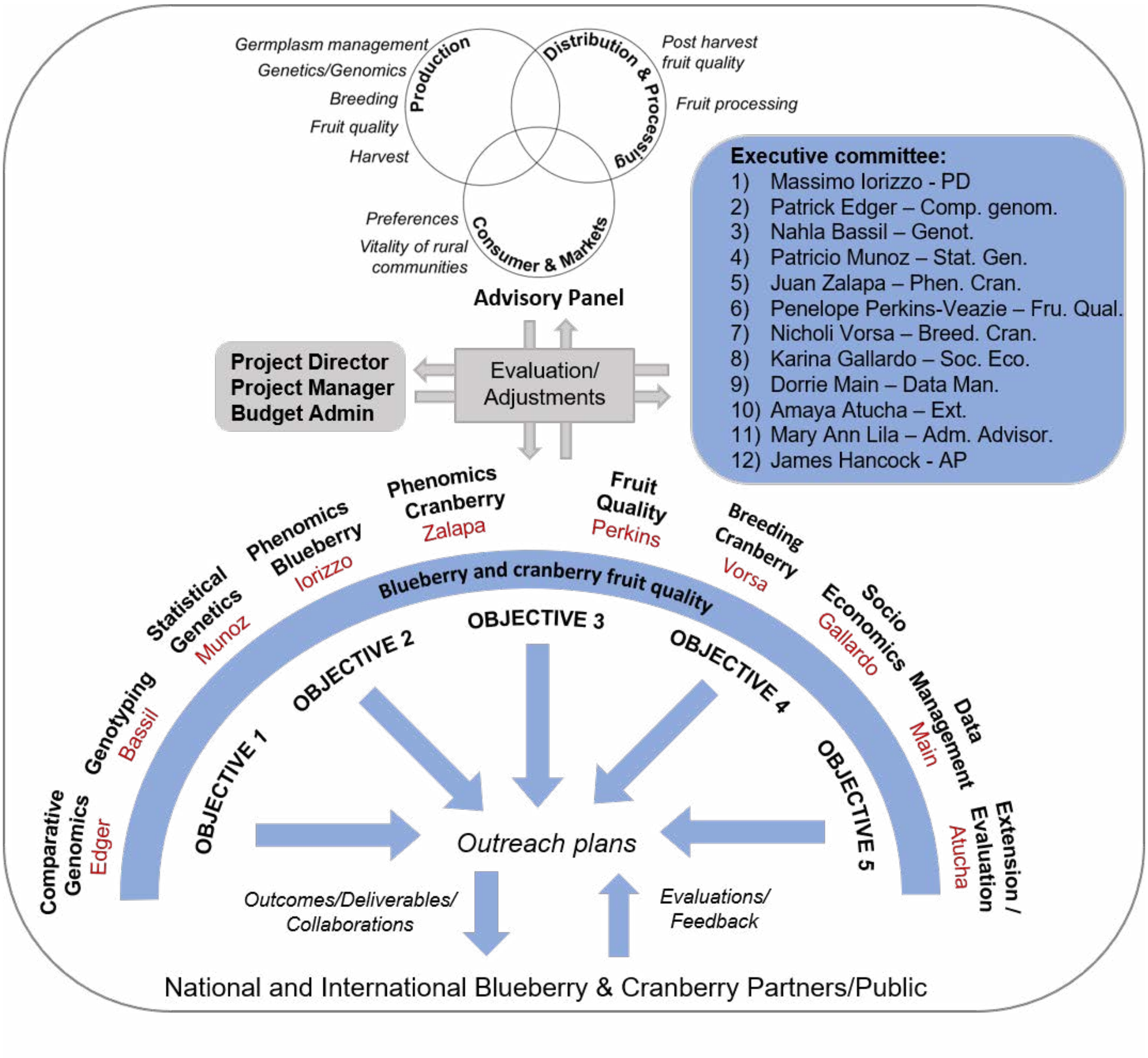
UF UNIVERSITY of FLORIDA | NC STATE UNIVERSITY | MISSISSIPPI STATE UNIVERSITY | FONDAZIONE EDMUND MACH

WISCONSIN UNIVERSITY OF WISCONSIN-MADISON | RUTGERS | WASHINGTON STATE UNIVERSITY | Plant & Food RESEARCH RANGAHAU AHUMARA KAI

UNIVERSITY OF GEORGIA | MICHIGAN STATE UNIVERSITY | USDA ars Agricultural Research Service | CEDAR LAKE RESEARCH GROUP LLC

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, J. Johnson-Cicalese
- 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, J. Johnson-Cicalese
- 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, J. Johnson-Cicalese
- 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

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
- Ebrahiem 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

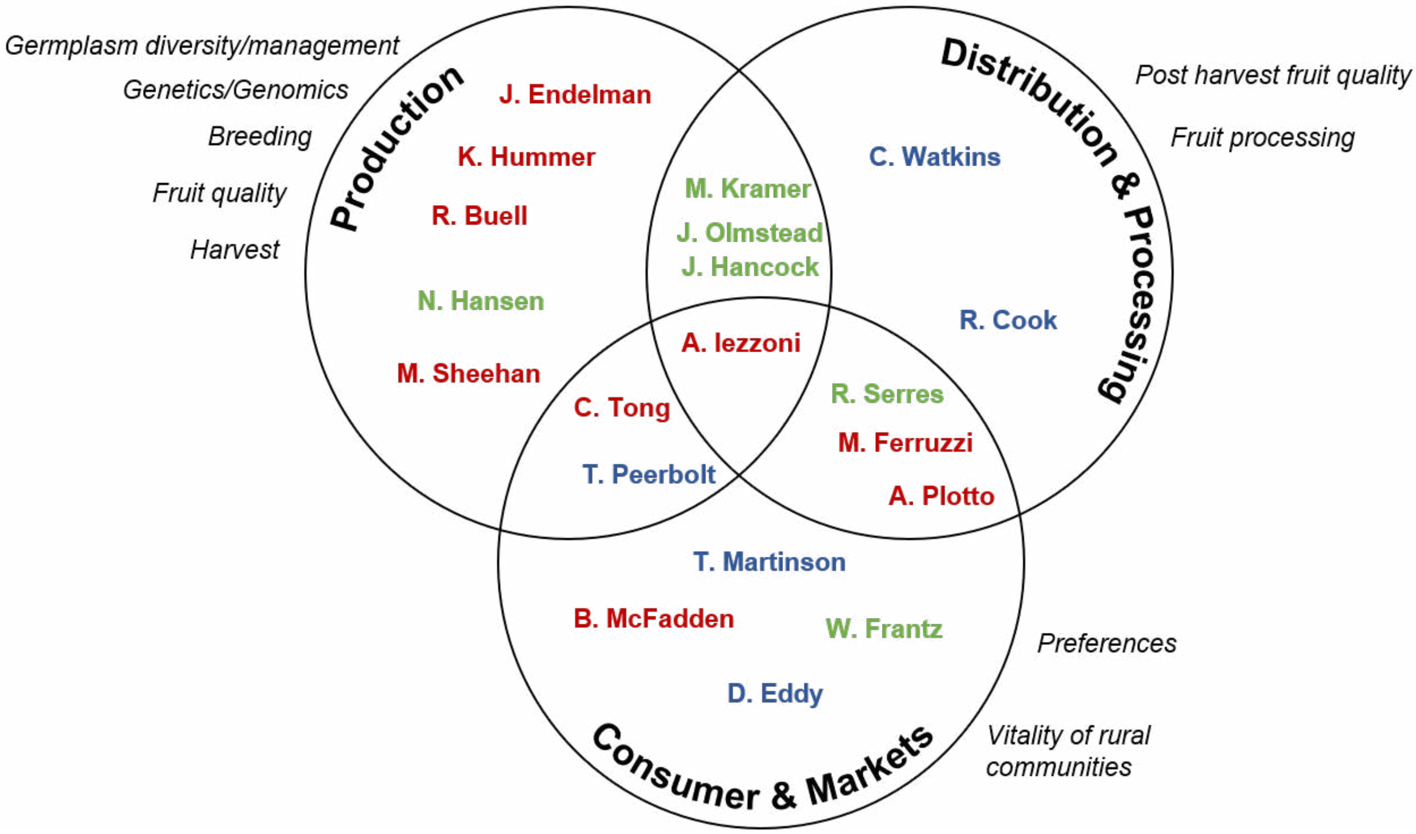
- James Hancock, Berry Blue LLC
- Nicole Hansen, Cranberry Grower
- Matt Kramer, Fall Creek Farm and Nursery
- James Olmstead, Driscoll's, Inc.
- Jessica Gilbert, Driscoll's, Inc.
- Mandie Driskill, Fall Creek Farm and Nursery
- Marivi Colle, Berry Blue LLC
- Rod Serres, Ocean Spray Cranberries, Inc.
- 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)
- Michael Dossett, BC Berry Cultivar Development Inc.
- Amy Iezzoni, Michigan State University
- Brandon McFadden, University of Delaware
- Anne Plotto, USDA-ARS (FL)
- Moira Sheehan, Cornell University
- Cindy Tong, University of Minnesota
- Jeannine Lisa Rowland, USDA-ARS



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

Timeline

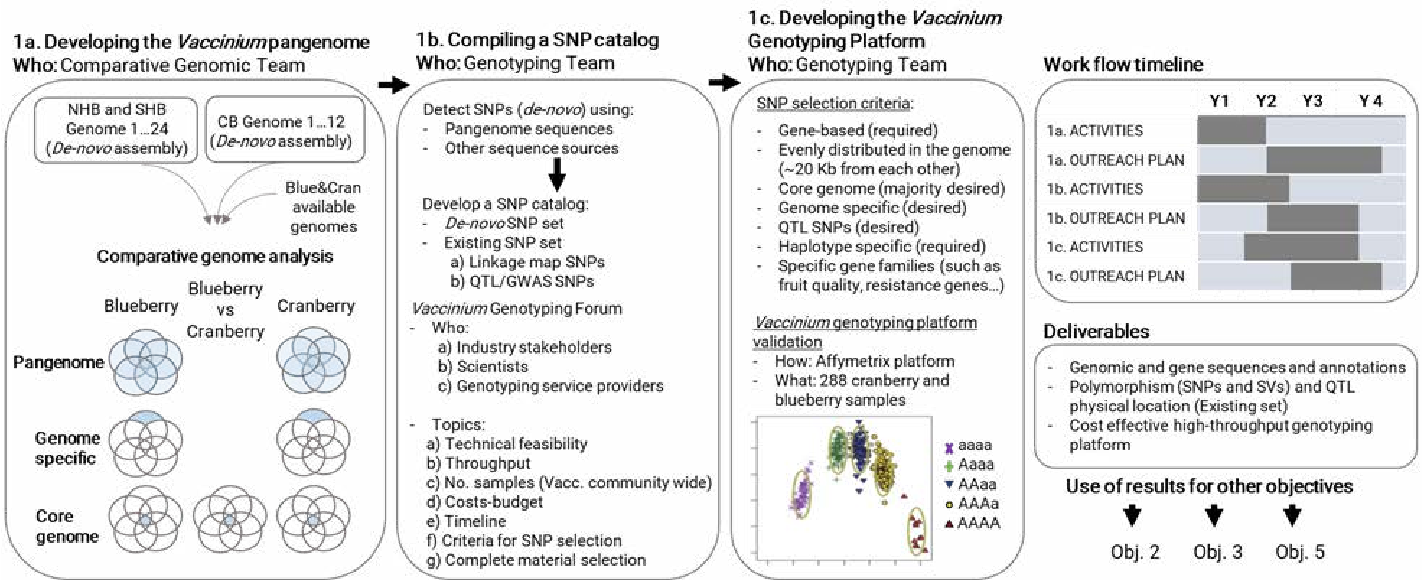
Cranberry Harvest Season   Blueberry Harvest Season

VacciniumCAP: Leveraging genetic and genomic resources to enable development of blueberry and cranberry cultivars with improved fruit quality attributes	Fall	Winter	Spring	Summer	Fall	Winter	Spring	Summer	Fall	Winter	Spring	Summer	Fall	Winter	Spring	Summer
Objective 1: Establish genomic resources to enable effective association mapping studies	**YEAR 1 2019-20**				**YEAR 2 2020-21**				**YEAR 3 2021-22**				**YEAR 4 2022-23**			
1a. Developing the *Vaccinium* pangenome (Comp. Genom. Team)	■	■	■	■	■	■										
1b. Compiling a SNP catalog (Genot. Team)	■	■	■	■	■	■										
1c. Developing the *Vaccinium* genotyping platform (Genot. Team)			■	■	■	■	■	■	■							
Outreach plan for results of Objective 1							■	■	■	■	■					
Objective 2: Discover DNA markers and fruit characteristics that match consumer preferences and maximize industry profitability in blueberry and cranberry																
2a. Phenotyping Fruit Characteristics (Pheno. Teams)	■	■	■	■	■	■	■	■	■	■	■	■	■			
2b. Discover DNA markers associated with FCs (Stat. Gen. Team)												■	■	■		
2c. Identifying FCs that contribute to essential fruit quality traits (Fr. Qual. Team)					■	■	■	■	■	■	■	■				
Outreach plan for results of Objective 2					■	■	■	■	■	■	■	■				
Objective 3: Deliver molecular and genetic resources to improve blueberry and cranberry fruit quality traits that maximize industry profitability and consumers preferences																
3a.Phenotyping, FC QTLs validation and fine mapping (Pheno. & Stat. Gen. Teams)				■	■	■	■	■	■	■	■	■				
3b. Develop and validate a high-throughput affordable assay for *Vaccinium* FCs (Stat. Gen. Team)												■	■	■	■	■
3c.Use validated SNPs to pyramid key traits (Breed. and Stat. Gen. Teams)														■	■	■
Outreach plan for results of Objective 3											■	■	■	■	■	■
Objective 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 FCs (Soc. Econ. Team)					■	■	■	■	■	■	■	■	■			
4b. Evaluate consumer behavior response to fruit tasting using biometric parameters (Soc. Econ. Team)								■	■	■	■	■	■	■		
4c. Estimate consumers' reactions to "added-sugar" labelling and labelling information in cranberry products (Soc. Econ. Team)				■	■	■	■	■								
Outreach plan for results of Objective 4								■	■	■	■	■	■	■	■	■
Objective 5: Engage U.S. *Vaccinium* stakeholder groups to transfer advanced phenomic and genomic tools to build a more efficient cultivar development system																
5a. Update and expand online platforms (Data Man. and Ext. Teams)	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■
5b. Develop newsletters (Ext. Team)	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■
5c. Develop webinars (Co-PI and Ext. Team)		■	■	■	■	■	■	■	■	■	■	■	■	■	■	■
5d. Develop workshops and forums (Co-PI and Ext. Team)			■		■			■				■				■
5e. Participate in commodity group meetings (Co-PI and Ext. Team)	■	■			■	■			■	■			■	■		
5f. Annual meeting			■					■				■				■
5g. Engage the public (Outreach, Co-PI and Ext. Team)	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■

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 3 Progress Summary

Obj. 1. Establish genomic resources to enable effective association mapping studies in blueberry and cranberry	
Contributed by: Edger, Bassil, Iorizzo, Zalapa, Vorsa/Cicalese, Polashock, Chagne', Munoz, Luby	
Presented by: Edger, Yocca, Bassil	
1a. Developing the Vaccinium pangenome [Comparative Genomic Team]	**To whom***
Compelted construction of the pan-genome. Fully annotated (genes and repetitive features) genomes of blueberry and cranberry cultivars that were used to construct the pangenome. Using the pangenome graph, ancestral states estimated and genomic variants identified unique to blueberry, cranberry and individual genomes. The blueberry and cranberry core and dispensable genes were used by the Genotyping Team for selecting target SNPs for probe design.	Statistical Genetic Team, Breeder and Geneticist Partners
Development of new genomes and genetic resources for genetic and comparative analysis. Released a new blueberry genome (W85_v2) and five linkage maps. Uncovered a chromosome translocation in the blueberry cultivar 'Draper' that affects recombination behavior and demonstrated that tetraploid blueberry behaves as an autotetraploid. Completed annotation of the blueberry and cranberry pan-genomes, and conserved and genome specific genes were identified and used to design the genotyping platform. Mining of the pangenome for specific gene families is ongoing.	
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 *Ericaceae* and across other plant families.	
1b. Compiling SNP catalog [Genotyping Team]	**To whom**
Compiled SNP catablog (cranberry). Completed compiling and annotation of the cranberry SNP catalog. The catalog includes about 30M SNP/indels and 2.4 M mapped to a single location on the reference genome (Stevens). These variants were used to select 60,000 SNPs that were submitted to Rapid genomics for probe design/optimization which initaited the testing phase.	Statistical Genetic Team, Breeder and Geneticist Partners
1c. Developing genotyping platform [Genotyping Team]	**To whom**
Completed testing phase and initiatied genoyping phase (blueberry). A total of 50,000 probes were selected, synthetized and tested in 192 blueberry samples (valdiation set). After screening and SNP calls, a set of 21,935 highly performing probes (containing SNPs and re-producible across samples) were selected for use in the genotyping phase. About 73% of these probes span highly conserved blubeerry genes (hard and soft core genes) an only about 9% span disposable genes. The remaining probes largely span intergenic space that were targetted to reach uniform distribution acorss chromosomes. Average distance between probe is 57kb, and 65 probes are cover SNPs previously associated with QTLs. Preliminary analyses of genotype data generated with the 22K platform in 192 blueberry samples indicated that this tool is not only informative in the tetraploid cultivated highbush blueberries that were targeted (4.2% missing value, on average) but that it will work well in the hexaploid rabbiteye blueberry (identical 4.2% missing value, on average) as well. This high throughput platform generated genotype data for > 150,000 SNPs in the cultivated blueberry (NHB, SHB, & RE) in addition to diploid species even in non-Cyanococcus sections. For each loci (probe) haplotype inforamtion were also extracted. 205 loci were assoiated with QTLs previously mapped for 16 traits (Table 2). In total, 6,696 blueberry samples were submitted by members of the Vaccinium Genotyping consortium for genotyping. In total 8 members from private and public breeding programs from US, France, New Zeland, Canada and Italy have submitted samples for using the platform. New partners expressed interest to use the genotypig platform and joined the consortium. Parameters and formatting needs for SNPs and haplotype call were identified. SNPs and haplotype data are being mined for population genetics, and marker traits association analysis.	Breeding, Statistical Genetic Teams, Breeder and Geneticist Partners
Completed testing phase (cranberry). Leaf samples were collected from 192 cranberry accessions that included representative samples from four industry partners and academic institutions in the US. Throuhg this process 17,502 probes were selected based on data uniformity and consistency of recovery among replicates sequenced. About 80% of the probes are located in core genes, 18% in dispensable genes and 2% in intergenic region. Also, 8 SNPs were associated with known QTLs (Table 3). These probes were used to evaluated the testing set and SNPs data being mined. In total between blueberry and cranberry, a total of 7,934 samples were submitted by the Vaccinium Genotyping consortium for genotyping.	

Expected impact 1b-c. A cost effective genotyping platform that is useful to the *Vaccinium* scientific community worldwide that will enable genetic studies to identify loci and markers associated with fruit qualiy and other important traits.
Challenges & changes. The delay of funding release from the USDA-NIFA during Year 1 and COVID-19 requirements during 2020-2022, delayed the development of genotyping platforms (Obj. 1). As a result, delivery of the genotypic data for genetic studies expected in Y3 was delayed and will be completed in Year 4.
Addressing challenges and recommendations. A recommendation from the advisory panel and imputs from the pangenome webinar was to plan the release of the data used for the pangenome construction. Raw genomic data as well as assembled genomes for all samples included in the pangenome construction were already released on NCBI adn GDV (see Table 1). This will permit various organizations to utilize that data for sequence alignment, etc. The pangenome, and 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 4. 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 1-2-3.

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.	Completed**	Mengist et al. 2022	Genome assembly, annotation and raw data released in GDV
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 released in GDV
Blueberry pangenome	Edger P.	Work in progress		Raw data released on NCBI, raw and assembled data released on GDV
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.	Completed**	Mengist et al. 2022	SNPs sequences released in GDV
Draper Selection 44392 x Jewel	Iorizzo M.	Completed**	Mengist et al. 2022	SNPs sequences released in GDV
Draper x Biloxi	Giongo L.	Completed**	Mengist et al. 2022	SNPs sequences released in GDV
Hortblue Petite x Nui	Chagne D.	Completed**	Montanari et al., 2022	SNPs sequences released in GDV
Nui x Hortblue Petite	Chagne D.	Completed**	Montanari et al., 2022	SNPs sequences released in GDV

* completed during the previous reporting cycle (Y1-Y2)
** completed during this reporting cycle (Y3)

Table 2. List of traits and # of SNPs represented in the 22K blueberry genotyping probes

Trait	# SNPs*
BlueFDF	2
Scar	1
Size	21
Weight	5
Firmness	40
Flavor	3
SizeRetention	6
Color	23
Totalsolublesolids	1
Fullbloom(FB)	1
Earlygreen(EG)	14
Coldhardiness(ColdHard)	9
DeltaFDF	11
DeltaFDF/FirmRetention	4
Diameter(Dia)	58
Chillingrequirement(ChillReq)	6

* these SNPs span QTL regions but were not validate

Table 3. List of traits and # of SNPs represented in the 17K cranberry genotyping probes

Trait	# SNPs*
Wax	1
FFR	4
CA	1
MA	2

* these SNPs span QTL regions but were not validate

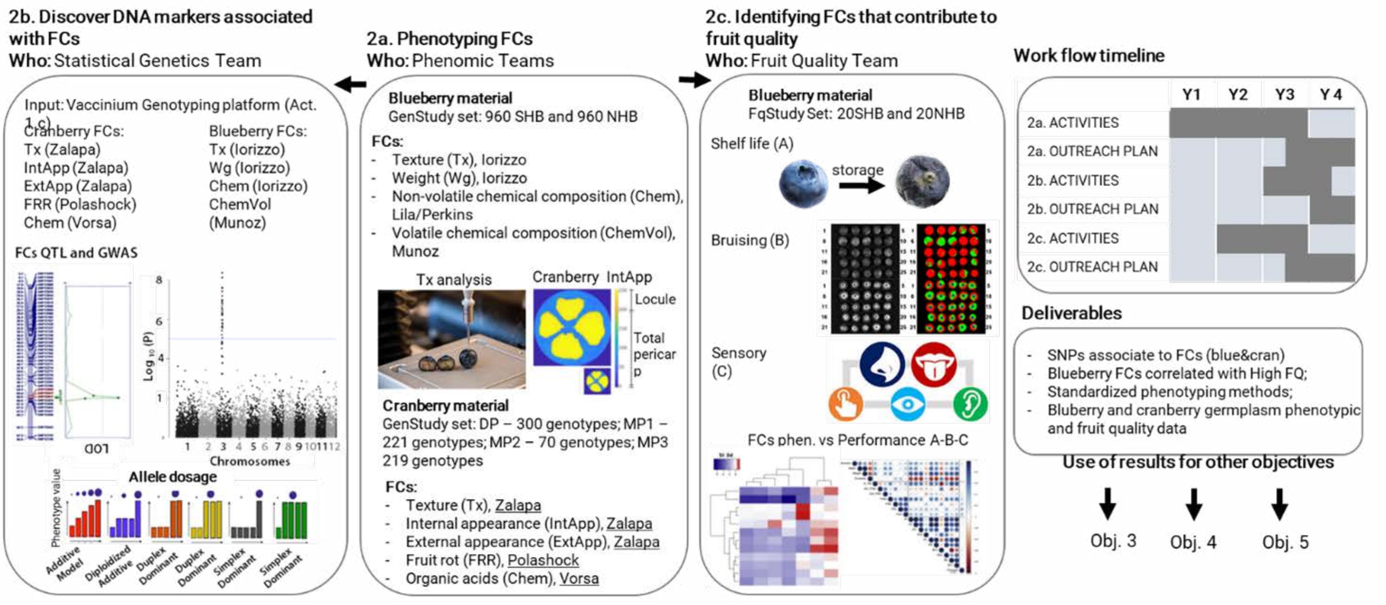
Obj. 1 - VacCAP Plan for Year 4

Obj. 1. Establish genomic resources to enable effective association mapping studies in blueberry and cranberry
1a. Developing the *Vaccinium* pangenome [Comparative Genomic Team]
Finalize analysis of the pangenome. Identify genes (Dispensable/Core) that contrbute to fruit quality traits. Understand the temporal expression patterns of these genes during fruit development & identify putative genes functions and pathways
Develop machine learning models to more easily identify dispensable genes, particularly those associated with fruit quality traits, in future Vaccinium Genomes.
Continue to collaborate with the VacCAP Co-PIs and partners to generate new and revised genome assemblies and annotations for various Vaccinium species
Communicate outcomes through publications and presentations
1b-c. Developing genotyping platform [Genotyping Team]
Complete genotyping phase for cranberry
Complete data analysis for testing phase blueberry and cranberry
Communicate outcomes through publications and presentations
Expected challenges. None at this time.

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 3 Progress Summary

Obj. 2. Discover DNA markers and fruit characteristics that improve fruit quality in blueberry.	
Contrbuted by: Perkins-Veazie, Lila, Giongo, Iorizzo, Munoz, Luby, Bassil, Li, Espley	
Presented by: Iorizzo, Ferrao, Giongo, Perkins-Veazie, Oh, Mengist-Fentie	
2a. Phenotyping fruit characteristics (FCs) [Phenomic Teams]	**To whom***
Finalize phenotyping protocols, equipment needs and settings. **Blueberry:** 1) tested an acoustic system to evaluate fruit texture; 2) Improved a user-friendly learning-based web application to evaluate blueberry bruise from mechanical harvesters; 3) To increase throughtput for organic acid analysis in the GenStudy and ValStudy sets (SHB and NHB) a LC-MS method that use a 96 well plate system was implemented.	Fruit Quality, Statistical Genetic and Breeding Teams and Partners
Phenotype FCs. Blueberry: Completed evaluation of FCs (except organic acids in NHB and SHB GenStudy set) and shelf life indicators on the NHB GenStudy set (3 years data), SHB GenStudy set (2 year data), a diversity panel representing NHB from the germplasm repository (OR, Corvallis) and two mapping populations RxA (2 years data) and DSxJ (2 years data) (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). Evaluation of Organic acids in SHB adn NHB GenStudy sets is ongoing. Preliminary analysis of the data indicated: pH and TA were highly correlated; TSS and total or individual sugars were not highly correlated; the relative ratio of the different sugars (fruictose, glucose, sucrse) was relatively stable across samples; Size and TSS constributed the most to separate SHB and NHB, with SHB having larger berries and lower TSS; FCs had from low to high genetic inheritance; chemistry characteristics have no or very limied impact on texture characteristisc; genetic gains for all traits was observed. Phenotipic data are being analyzed and were used for breeding purposes and genetic analysis (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 is allowing us to better understand which texture parameters and/or other FCs contribute to fruit firmness and shelflife, and if it is genetically inherited. For instance the low or no correaltion between sugars and TSS pose questions about use of TSS measurments, a parameter commonly used by research and breeding community, as a proxy for sugar content in blueberry. Phenotypic data provided information to Oregon and Florida Breeding Programs for advancing selection and select 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 and genotype GenStudy set: Completed DNA extraction and genotyping.	Breeding and Genotyping Teams, and geneticist partners
Perform Genetic analyses. Blueberry: Completed two QTL mapping study for fruit quality traits (pH, TA, TSS, Wg, anthocyanin content and composition) in two biparental populations DraperSelection x Jewel (DSxJ) and Nui x HardyBlue (NxHB)(Table 5). In total, 353 QTLs were identified, with eleven QTL regions spanning several QTLs that were stables across years. Stable QTLs across three years for TA and pH overlapped in the same regions and coincide with the same pH and TA QTL regions detected in a previous QTL study in RxA, that was completed in year 2. Similarly stable QTLs for anthocyanin acylation was indetified across multiple studies. Preliminary GWAS and QTL mapping in the GenStudyes sets (NHB, SHB, RxA, DSxJ) identified several QTLs for most of the traits, and suggests that citric acid regulate TA and pH. QTLs are being used for validation, candidate gene analysis and evaluated for use in DNA marker assay development.	
Expected impact. FCs-DNA marker associations are being identified and preliminary results indicated that we expect to detect more significant associations. This outcome provides 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
Perform FQ analysis - shelf life: A set of 60 cultivars (named here NC-FqStudy set) were harvested in North Carolina and held up to 6 weeks at 3°C to follow changes in texture, fruit quality, and fruit chemistry (Table 6). Completed evaluation of the following FCs include 4 Tx parameters using needle probe, 14 Tx paramenters using a flat probe, 24 Tx parameters using the double compression probe, Weight (Wg), Scar Tear (ScT), Scar Diameter (ScD) and the shelf life indicators: Wrinkle, Leakage, Mold; chemistry (non-volatile) parameters: pH, TA, TSS, organic acids, sugars. All FC's were measured at harvest (T0), and two (T2), four (T4), and six (T6) weeks in post-storage. Tx with needle and double compression probes were evaluted for 1 year and all other FCs were evalauted for two years. Preliminary analysis indicates: chemistry profile did not change much during the storage; Wg has a very limited crontribution for change in texture during storage; three Tx parameters (flat probe) had a large contribution on changes in texture during storage; models to predict the performance in post-harvest of these three paramters, wrinkle and weight loss using measurements colelcted at harvest were developed. The predictive value of the three texture parameters ranged from >0.1 - 0.6 (adjusted R2), predictive value for wrinkle ranged between 0.35 - 0.48 (adjusted R2), predictive value for Weight loss was 0.85 (adjusted R2). Similar analysis are being performed using the GenStudy and ValStudy sets evalauted in Obj. 2a, at Oregon and Florida locations.	Breeding, Fruit Quality and Other industry partners
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 the harvest. A protocol for cell wall compisition analysis was established. Data analysis indicated: monosaccharide composition differed among cultivars and tissue types; several polysaccharide linkages were identified and many differed among cultivars; firm and crisp fruits had higher amount of hemicellulosic heteromannan. Overall differences in the cell wall structure and composition in soft, firm, and crisp types may be mirroring their difference texture.	Fruit Quality team and partners
Perform FQ analysis bruising: A machine learning model based web application software that can automatically determine the blueberry bruise developed during Y1-Y2 was used t evaluate the brusing rate of the NC-FqStudy set (Table 6). Preliminary analysis indicated significant variation among cultivars and ranking of cultivars with the lowest and highest bruising rate was consistent across the two years. Bruising data in this set are being used for correlation with all traits measured at harvest time.	Breeding, Fruit Quality 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 for evaluate texture and chem composition will contribute to increasing the number of the potential users of the technique used to phenotype these FCs in blueberry.	
Challenges & Changes. Due to a problem in the software used to extract the texture data from the texture analyzer data for the GenStudy and ValStudy sets had to be re-extracted and re-analyzed causing some delay with the data analysis and mining. Data from the sensory analysis that were originally planned as part of Obj. 2 were integrated into Obj. 4 as the two experiments were evalauted in paralelle.	

* 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 (N=960)			GenStudy set SHB (N=960)		Draper x Jewel (DxJ)(N=196)	Reveille x Arlen (RxA)(N=364)	Diversity Panel OR NHB	New methods reference
				2020	2021	2022	2021	2022	2020-21-22	2021-22	2019-2020	
FC Traits												
Texture and Appearance FC												
Texture (TxNp) Needle probe		4 mechanical parameters	TA.XTPlus***									
Texture (TxPp) Penetration probe		24 mechanical parameters	TA.XTPlus***	Completed	Completed		Completed		Completed			
Texture (TxPp) Double compression probe	T0* and T1*	23 mechanical parameters	TA.XTPlus***							Completed	NP	Giongo et al. 2022, *PBT 183, 111696*; Iorizzo et al. 2021, ISHS presentation
Weight (Wg) (Size**)		Avg. individual berry weight and avg. 10-12 berry weight	High Precision Mettler Scale***	Completed	Completed	Completed	Completed	Completed	Completed			
Scar (ScD)		Diameter (ScD)	Digital caliper***									
Chemical composition												
Non-Volatile chemical composition (Chem)		Titratable acidity (TA), pH, Soluble Solid Content (SSC)	Digital refractometers and pH meter	Completed	Completed	Completed	Completed	Completed	NP	Completed	Completed	Perkins et al. 2021, Acta Hort.
	T0	Sugars (SSg): fructose, glucose, sucrose	NIR & HPLC						NP		NP	
		Organic acids (Ac): malic, citric, quinic, shikimic	HPLC and LC-MS	NP	Ongoing	NP	Ongoing	NP	NP		NP	
Volatile chemical composition (ChemVol)		Volatile organic composition (>60 volatile compounds)	GC-MS	NP	Completed	NP	Completed	NP	NP	NP	NP	
Shelf life indicators												
Wrinkle/Shrivel (Shr)		Scale 0-3, and % yes/no	Visual***									
Leakage (Lk)		% yes/no	Visual***									
Mold	T0 and T1	% yes/no	Visual***									
Mold color		Visual	Visual***									
Stem scar Tear (ScTr)		% yes/no	Visual***									
Wet stem scar (WSc)		% yes/no	Visual***	Completed	Completed	Completed	Completed	Completed	Completed	Completed	NP	Giongo et al. 2022, *PBT 183, 111696*; Iorizzo et al. 2021, Proceeding of XII ISHS Int Vac Symp
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		NxHB* (N=128)	Anthocyanin types	165	3/2years	Montanari et al., 2022, F 13:965397 doi: 10.3389/fpls.20
	Chem	DSxJ* (N=190)	Anthocyanin types	172	4/3 years	Mengist et al. 2022, Front. Pla doi: 10.3389/fpls.202
			Total Anthocyanin content	5	1/3 years	
			Chlorogenic acid	3	1/3 years	
			pH	2	1/2 years	
			TA	4	1/3 years	
			SSC	2		
	Chem	RxA** (N=287)	pH	5	1/3 Yrs	Mengist et al. 2021, Hort F (2021). https://doi.org/10.1(00605-z
			TA	3	2/3 Yrs	
			SSC	3	2/3 Yrs	
	Appearance		Wg/Size	7	1/3 Yrs; 2/2 Yrs	
				371	19	

* completed during this reporting cycle (Y3)

Table 6. Summary of blueberry phenotyping activities completed or ongoing for the NC-FqStudy set.

Traits	Phenotyping time	Type and # parameters	Methods	NC-FqStudy set (N=62)		New methods reference
				2021	2022	
FC Traits						
Texture and Appearance FC						
Texture (TxNp) penetration needle probe	T0 and T4*	4 mechanical parameters	TA.XTPlus***	Completed		
Texture (TxPp) penetration flat probe	T0 and T2, T4, T6*	14 mechanical parameters	TA.XTPlus***			
Texture (TxPp) Double compression probe	T0 and T2, T4, T6*	23 mechanical parameters	TA.XTPlus***		Completed	Giongo et al. 2021, *PBT 183, 111696*; Iorizzo et al. 2021, ISHS presentation
Weight (Wg) (Size**)	T0 and T2, T4, T6*	Avg. individual berry weight and avg. 10-12 berry weight	High Precision Mettler Scale***	Completed		
Scar (ScD)		Diameter (ScD)	Digital caliper***			
Chemical composition						
Non-Volatile chemical composition (Chem)	T0 and T2, T4, T6*	Titratable acidity (TA), pH, Soluble Solid Content (SSC)	Digital refractometers and pH meter	Completed	Completed	Perkins et al. 2021, Acta Hort.
		Sugars (SSg): fructose, glucose, sucrose	NIR & HPLC	Completed	Completed	
		Organic acids (Ac): malic, citric, quinic, shikimic	HPLC and LC-MS	Completed	Completed	
Shelf life indicators						
Wrinkle/Shrivel (Shr)	T0 and T2, T4, T6*	Scale 0-3, and % yes/no	Visual***	Completed	Completed	Giongo 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			
Bruising	T0	Bruising rate	Image analysis	Completed	Completed	Ni et al. 2022, CEA 201, 107200.

* T0= 12-24 hours post-harvest; T2= 2 weeks post harvest, T4= four weeks post-harvest; T6= six weeks post harvest. Material was stored at the following conditions: 2°C and 80% RH.
** 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

Obj. 2 Blueberry – VacCAP: Plan for Year 4

Obj. 2. Discover DNA markers and fruit characteristics that improve fruit quality in blueberry.
2a. Phenotyping fruit characteristics [Phenomics Team]
Complete analysis of year 1-3 data
Continue testing new protocols in GenStudy set: testing the acoustic methods for blueberry texture will continue. The team is also considering working on identifying a common set of blueberry sensory descriptors.
2b. Discover DNA markers associated with FCs [Statistical Genetic Team]
Complete QTL analysis for multiple FCs in all GenStudy sets.
Prepare manuscripts for dissemination of results.
2c. Identifying FCs that contribute to essential fruit quality traits [Fruit Quality Team, Penelope Perkins Lead]
Complete data analysis for the following studies:
1. Variation and predictability of FCs changes during storage using Double compression probe.
2. Variation of bruising rate across cvs, stability across years and texture parameters that contribute to low bruising rate
3. Evaluation of texture profile by sensory analysis and mechanical texture.
Prepare manuscripts for dissemination of results.
Excepted challenges. Expect some delay in completing the GWAS studies.

Obj. 2 Cranberry – VacCAP: Year 3 Progress Summary

<table>
<tr><td colspan="2">Obj. 2. Discover DNA markers and fruit characteristics that improve fruit quality in cranberry.</td></tr>
<tr><td colspan="2">Contributed by: Zalapa, Polashock, Vorsa, Cicalese, Bassil</td></tr>
<tr><td colspan="2">Presented by: Zalapa, Lopez Moreno, Loarca, Knowles, Polashock</td></tr>
<tr><td>2a. Phenotyping fruit characteristics (FCs) [Phenomic Teams]</td><td>To whom*</td></tr>
<tr><td>Finalize phenotyping protocols, equipment needs and settings. Finalized data analysis for texture methods. Results indicated that 30 fruits are enough to evaluate texture in cranberry. Kramer method has the highest reproducibility measured, followed by the blade. Temperature and fruit orientation have a significant effect on texture measurements. Tested a new fruit harvester to collect samples that has increased speed of collection, allowing phenotyping of more accessions. Also tested a hyperspectral imaging system for higher throughput phenotyping to evaluate fruit rot, color, shape, size, waxiness.Methods were developed for UHPLC of organic acids.</td><td>Fruit Quality, Breeding, Statistical Genetic and Breeding Teams and Partners</td></tr>
<tr><td>Phenotype FCs. Evaluated fruit weight, yield, % fruit rot, total anthocyanin, brix and titratable acidity, organic acids, epicuticular wax on berries, leaf disease and multiple morphological and phenological traits (e.g. percent bloom, #flowers, #fruit per upright) on MP3 (N=219). I Initiated evaluation for resistance to fruit scald (Table 7). Cranberry Y1-3 data are being integrated and analyzed; this mapping population is segregating for all traits analyzed. Samples were collected from the Diversity Panel (N=300) in 2022 for analysis of texture, fruit chemistry and other FC traits.</td><td>Statistical Genetic and Breeding Teams</td></tr>
<tr><td colspan="2">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 rot resistance and other 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.</td></tr>
<tr><td>2b. Discover DNA markers associated with FCs [Statistical Genetics Team]</td><td>To whom</td></tr>
<tr><td>Collect material, extract DNA from GenStudy set. Genotyping of the cranberry GenStudy sets was initiated. A new genetic study set, that includes a wide array of accessions and breeding populations (N=847), was sent for genomic analysis.</td><td rowspan="2">Breeding and Genotyping Teams, Breeders and geneticist partners</td></tr>
<tr><td>Perform Genetic analysis. Completed two genetic studies for upright traits, berry shape/size, yield, fruit rot resistance, and other fruit characteristics (TAcy, brix, titratable acidity, PAC, epicuticular wax) (Table 8). Highlights from these studies were: yield traits positively correlated with plot and upright traits and negative correlate with PAC; QTLs were identified for all the the traits, some QTLs were stables across the years. Three QTL for yield, one for TAcy and one for fruit rot resistance overlap with those identified in previous studies.</td></tr>
<tr><td colspan="2">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. If reliable markers for fruit rot resistance are identified, this could expediate breeding and release of resistant cultivars, significantly impacting the sustainability of cranberry production.</td></tr>
<tr><td colspan="2">Challenges & Changes. Delay in the delivery of genotypic data is in turn delaying some of the genetic studies.</td></tr>
<tr><td colspan="2">Addressing challenges and recommendations. To ensure success of the genetic studies, additional mapping populations were evaluated for FCs. Evaluation of the Diversity panel, that was delayed due to COVID-19, was completed this year. Based on preliminary data analysis a second year of evaluation will be considered.</td></tr>
</table>

* To whom results are transferred during the project

Table 7. Summary of cranberry 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)			DP (N=293)	New methods reference
			2019	2020	2021	2019	2020	2021	2019	2020	2021	2022	2019	2020	2021	2019	2020	2021	2022	
FC Traits																				
Texture and Appearance FC																				
Texture (TxCp) compression probe	31 mechanical paramenters	TA.XTPlus***																		Lopez-Moreno et al., 2023. Horticulturae publication in review
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	Done	Done	NP*	NP*	NP*	NP*	NP*	NP*	NP*		Done	
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.
Berry Weight (Wg)	Avg. Wg	Precision scale									Done									
Fruit wax		Visual rating	NP*	NP*	NP*	NP*	NP*	NP	NP*	NP*	NP	Done	Done	Done		Done	Done		NP	
Fruit Shape		Visual rating	NP	NP	NP	NP	NP	NP	NP	NP	NP	Ongoing		Ongoing		Done	Done		NP	
Chemical composition																				
Non-Volatile chemical composition (Chem)	Total Anthocaynin	Spectrophotometric	NP**	NP**		NP**	NP**	NP	Done	Done	Done	NP							NP**	
	Organic acids: quinic, citric, and malic	HPLC	NP	NP	NP	Ongoing		NP		Done	Ongoing	NP		Ongoing	Ongoing		Ongoing	Ongoing	Ongoing	
	Brix	Refractometer	NP**	NP**	NP	Done	Done	NP	Done	Done	Done	NP							NP**	
	TA	Titration	NP**	NP**	NP	Done	Done	NP	Done	Done	Done	NP		Done	Done		Done	Done	NP**	
Fruit Rot Resistance (FRR)	% of rotted fruit/sq ft	Precision scale	Done	Done	NP	Done	Done	NP	Done	Done	Done	NP								
Yield (total Wg)	Total Wg/sq. ft.	Precision scale	NP**	NP**	NP	Done	Done	NP	Done	Done	Done	NP								
Leaf disease		Visual rating							Done		Done	Done	NP	NP	NP	NP	NP		NP**	
Bloom time	% of open & past flw/total flw in 10 uprights	Count	NP	NP	NP	NP	NP		NP		NP	Done								
fruit set	#fruit per 10 uprights	Count										Done								

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

Table 8. Summary of FC QTL studies completed in cranberry.

Crop	FC	Material	FC sub-component	# QTL	# Stable QTL/Years	Reference
Cranberry	Fruit Weight Fruit rot resistance Yield	DP (N=293)*	Avg Fruit Weight Fruit rot Total Yield	6 5 1	NA NA NA	Diaz-Garcia et al., 2021. Front Plant Sci.;11:607770. https://doi.org/10.3389/fpls.2020.607770
	Chem - organic acids	CM151* (N=49)	Low Malic acid (mala) < 2.5 mg/g	1	1 / 2 Yrs	Fong et al. 2021, Tree Genetics & Genomes 17, 4. https://doi.org/10.1007/s11295-020-01482-8
		CM100* (N=33)	Low Malic acid (mala) < 2.5 mg/g	1	1 / 2 Yrs	
	Fruit rot Resistance TA TACY brix	MP3** N=200	Fruit rot Titratable Acidity total anthocyanins soluble solids	1 1 3 1	2 2 2 2	
	Epicuticular Wax	MP4** (N=141)	Epicuticular Wax	1	1	Erndwein et al. 2023 Submitted to BMC Plant Biology
				21	6	

* completed during the previous reporting cycle (Y1-Y2)
* completed during this reporting cycle (Y3)

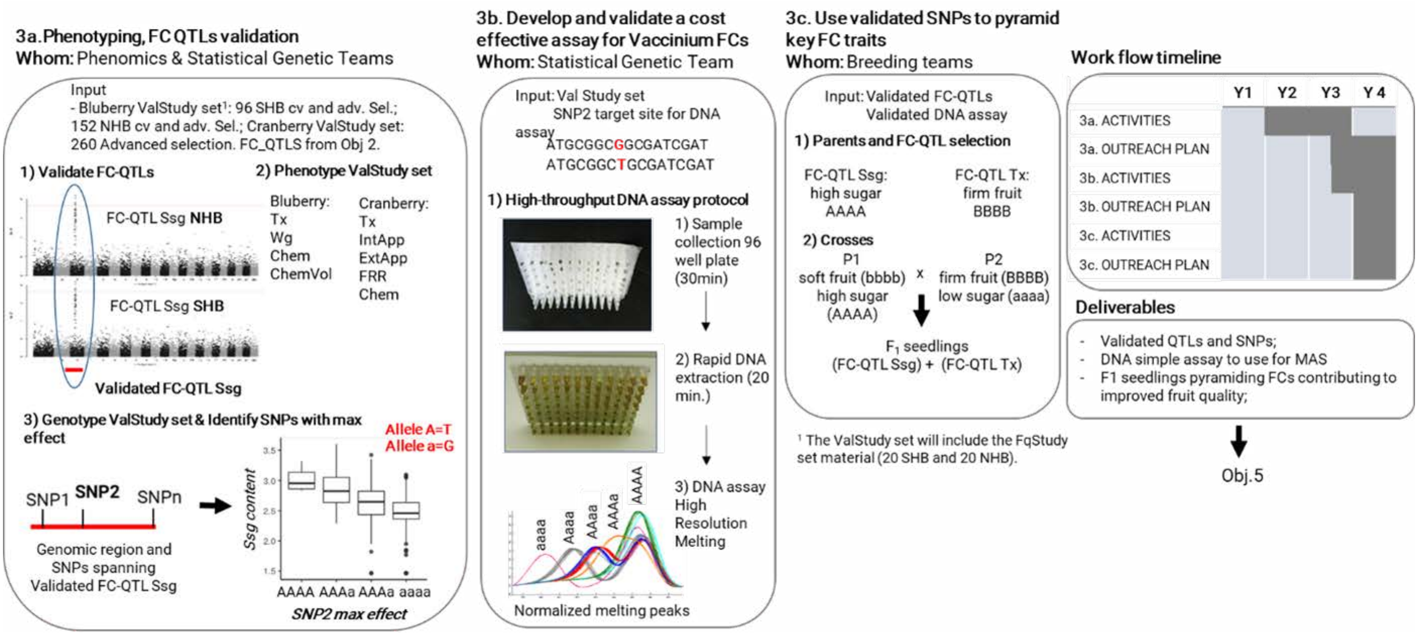
Obj. 2 Cranberry – VacCAP: Plan for Year 4

Obj. 2. Discover DNA markers and fruit characteristics that improve fruit quality in cranberry.
2a. Phenotyping fruit characteristics [Phenomics Team]
Continue testing and developing new methods for phenotyping FC: continue testing phenotyping methods for harvest and image analysis.
Continue phenotyping FCs. Consider phenotyping DP for one additional year.
Prepare manuscripts for dissemination of results.
Complete analysis of year 1-2-3 data
2b. Discover DNA markers associated with FCs [Statistical Genetic Team]
Complete genotyping GenStudy sets
Perform QTL mapping analysis for data collected in year 1-2-3.
Prepare manuscripts for dissemination of results.
Expected challenges. Delays in delivery of genotypic data

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 3 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	
Contributed 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***
Phenotype ValStudy Set. Blueberry. Completed evaluation of FCs (except organic acids in NHB and SHB ValStudy set) and shelf life indicators on the NHB ValStudy set (3 years data), SHB GenStudy set (2 year data) (Table 9). Except for chemical composition all FCs were evaluated at the harvest time and after storage (six or seven weeks under controlled conditions) (Table 9). Evaluation of Organic acids in SHB adn NHB ValStudy sets is ongoing. **Cranberry**: Phenotyping carried out on several mapping populations and a diversity panel for multiple cranberry traits, including yield, fruit rot, fruit weight, fruit shape and texture, organic acids, fruit wax, Brix, total anthocyanins, titratable acidity, leaf disease, bloom time, and upright traits.	Genotyping and Breeding teams, Vaccinium Breeders, and Geneticist Partners.
Collect material, extract DNA from ValStudy set. Completed genotyping the validation study set both for blueberry and cranberry.	
Develop list of target existing FC-QTL. Blueberry: Anchored stable QTL identified across three mapping populations to the blueberry genome. Three QTL regions associated with pH/TA, anthocyanin acylation and anthocyanin glycosylation are being considered as validated, as they were detected across multiple genetic backgrounds. Also QTL for volatiles that overlapped across multiple studies were identified. Some were considered as validated QTL and used to design DNA assays to test their prediction ability, and thus, their potential for Marker Assisted Selection. **Cranberry**: Two regions harboring major QTL for total anthocyanin and color variation, that were mapped in two populations, overlap on the same physical map region and are being considered as validated. Three QTL for yield, one for TAcy, and one for fruit rot resistance overlap with those identified in previous studies and will be evaluated further.	
3b. Develop and validate a high-throughput affordable assay for Vaccinium FCs [Statistical Genetic Team]	**To whom**
Finalize selection and protocol for simple DNA assay. Blueberry: continued testing of a rapid DNA extraction method and two genotyping high-throughput assays including High Resolution Melting (HRM) and Kompetitive allele-Specific PCR (KASP). Cost analysis estimates indicated that outsourcing DNA assay is more cost effective. Two genes associated with the control of Linalool and Eucalyptol volatiles were targeted for these assays. Preliminary analysis indicated that low to high level of allele vs phenotype (high vs low Linalool or Eucalyptol) predictability were obtained. The results highlighted that additional work is needed to optimize primer design, especially in understanding variation at the genomic level in regions surrounding genes where primers are placed, which affect their amplification. Other challenges with these methods is represented by the dosage estimates. Also ideal target genes will be those that are functionally characterized. **Cranberry**: validated a DNA assay using PACE technology targeting SNPs associated with epicuticular wax within a mapping population. The marker is highly sensitive for predicting waxy fruit (Table 10).	Genotyping and Breeding teams, Vaccinium Breeders, and Geneticist Partners.

3c. Pyramid key traits using validated SNPs and phenotypic data [Breeding Teams]	To whom
Leverage FC and FQ phenotypic data for advancing breeding selections. Blueberry and cranberry: continued making selections for genotypes with good performance for FC and propagated selections made in Y2 for trialing. Crosses between superior individuals were also performed and will be germinated in early 2023.	Genotyping and Breeding teams, Vaccinium Breeders, and Geneticist Partners.
Expected impact 3a-c. Validated QTL will provide opportunities to evaluate these markers for MAS and also to perform functional characterization of candidate genes. With these validated QTL 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.	
Challenges & changes. Due to the delay in completing genetic studies, validation of QTL was also delayed. To facilitate integration of the novel QTL that are likely to be detected from these genetic studies, team members initiated efforts to anchor QTLs identified during Y1-Y3 in mapping populations, and from previous studies into the blueberry and cranberry physical maps. As new QTL studies are being completed, this framework will facilitate identification of QTL that overlap across multiple studies and will be considered as validated across multiple genetic stocks.	

* To whom results are transferred during the project

Table 9. Summary of blueberry phenotyping activities completed or ongoing for ValStudy sets.

FC Traits	Phenotyping time	Type and # parameters	Methods	ValStudy set NHB (134)			ValStudy set SHB (N=109)	
				2020	2021	2022	2021	2022
FC Traits								
Texture and Appearance FC								
Texture (TxPp) Penetration probe	T0	24 mechanical parameters	TA.XTPlus***	Completed	Completed	Completed	Completed	Completed
Texture (TxPp) Double compression probe	T0	23 mechanical parameters	TA.XTPlus***	NP	NP	Completed	NP	Completed
Weight (Wg) (Size**)	T0	Avg. individual berry weight and avg. 10-12 berry weight	High Precision Mettler Scale***	Completed	Completed	Completed	Completed	Completed
Scar (ScD)	T0	Diameter (ScD)	Digital caliper***	Completed	Completed	Completed	Completed	Completed
Chemical composition								
Non-Volatile chemical composition (Chem)	T0	Titratable acidity (TA), pH, Soluble Solid Content (SSC)	Digital refractometers and pH meter	Completed	Completed	Completed	Completed	Completed
	T0	Sugars (SSg): fructose, glucose, sucrose	NIR & HPLC	Completed	Completed	Completed	Completed	Completed
	T0	Organic acids (Ac): malic, citric, quinic, shikimic	HPLC	NP	Ongoing	NP	Ongoing	NP
Volatile chemical composition (ChemVol)	T0	Volatile organic composition (>60 volatile compounds)	GC-MS	NP****	Completed	NP	Completed	NP
Shelf life indicators								
Wrinkle/Shrivel (Shr)	T0 and T1	Scale 0-3, and % yes/no	Visual***	Completed	Completed	Completed	Completed	Completed
Leakage (Lk)	T0 and T1	% yes/no	Visual***	Completed	Completed	Completed	Completed	Completed
Mold	T0 and T1	% yes/no	Visual***	Completed	Completed	Completed	Completed	Completed
Mold color	T0 and T1	Visual	Visual***	Completed	Completed	Completed	Completed	Completed
Stem scar Tear (ScTr)	T0 and T1	% yes/no	Visual***	Completed	Completed	Completed	Completed	Completed
Wet stem scar (WSc)	T0 and T1	% yes/no	Visual***	Completed	Completed	Completed	Completed	Completed
Weight loss (WgLo)	T0 and T1	Avg. 10-12 berry weight (T0-T1)	High Precision Mettler Scale***	Completed	Completed	Completed	Completed	Completed
Storage Index (SI), (estimated for all Tx and shelf life indicators)	T0 and T1	SI = log2(TiPH/TiH) TiH= Tx at harvest TiPH= Tx post storage	TA.XTPlus	Completed	Completed	Completed	Completed	Completed

* 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 10. 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 Low CA < 2.5mmg/g; Moderate CA 3-6 mg/g; High CA >6 mg/g	scf258d	SSR	Biparental populations	0.94	0.99	Fong et al. 2020. Tree Genetics & Genomes 16: 42 https://doi.org/10.1007/s11295-020-01432-4
			SNP CA_609	KASPs	Biparental populations	0.91	0.98	
		Low vs Moderate vs High Malic Acid Low MA < 2.5mmg/g; Moderate MA 3.5-5 mg/g; High CA >5 mg/g	SNP MA_271	KASPs	Biparental populations	0.94	0.81	Fong et al., 2021. Tree Genetics & Genomes 17, 4. https://doi.org/10.1007/s11295-020-01482-8
			SNP MA_476	KASPs	Biparental populations	0.94	1	
	Epicuticular Wax	Epicuticular Wax	SNP	PACE	Biparental populations	_	_	Erndwein et al. 2023 Submitted to BMC Plant Biology

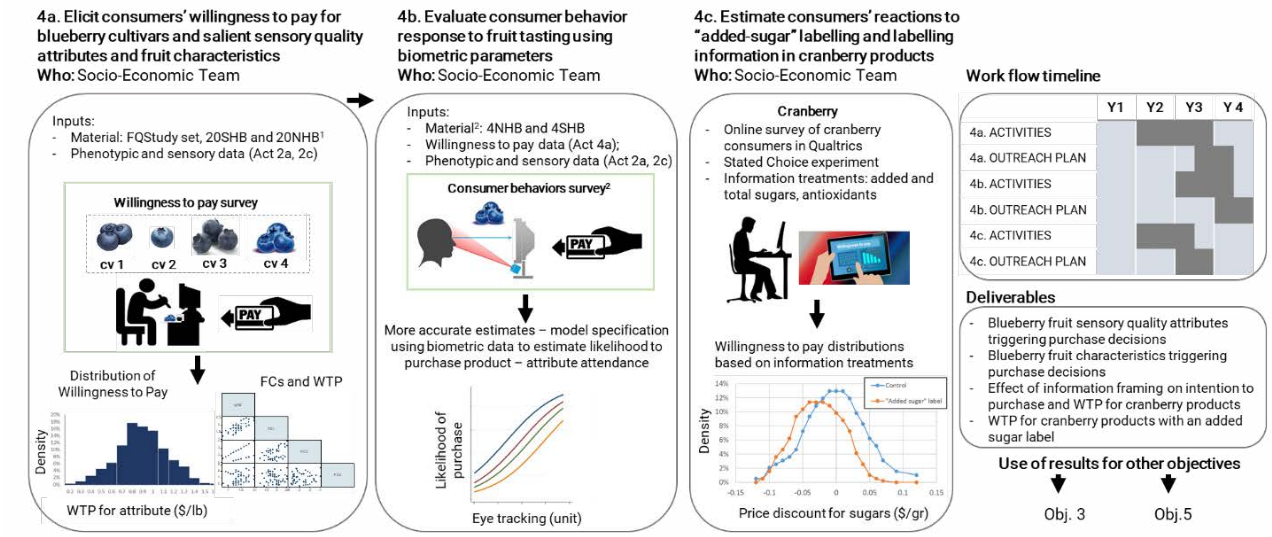
Obj. 3 – VacCAP: Plan for Year 4

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]
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 and cranberry)
Test DNA assays for organic acids, wax, fruit rot resistance, and fruit shape that were developed for cranberry in more diverse populations to evaluate sensitivity and specificity.
3c. Pyramid key traits using validated SNPs and phenotypic data [Breeding Teams]
Validate performance of genotypes selected during year 1-3 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. None at this time

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.



¹ The willingness to pay survey will be carried in conjunction with sensory panel survey.
² For the consumer behaviors study 4 SHB and 4 NHB were selected form the set of cultivars used for the FQStudy set (used in Act 2c and 4a).

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 3 Progress Summary

Obj. 4 Assess the potential socio-economic impact of blueberry and cranberry fruit quality improvements on market demand.	
Contributed by: Gallardo, Canales, Sims, Munoz, Iorizzo, Lila, Perkins-Veazie, Bassil, Colonna, Luby, Paul Sandefur	
Presented by: Gallardo, Ma, Canales, Sims	
4a. Elicit consumers' willingness to pay for blueberry cultivars and salient sensory quality attributes and fruit characteristics [Socio Economic Team]	**To whom***
Perform WTP and consumer preference questionnaire. A second year of WTP and consumer preference surveys were performed for 22 NHB and 20 SHB varieties, which total 42 NHB and 40 SHB samples across the two years, with 27 varieties assessed for both years. Data is being used to identify salient sensory attributes and fruit characteristics.	Breeding, Fruit Quality and Statistical Genetic teams, Blueberry industry partners
Evaluated fruit characteristics. Fruit characteristics (FC) including pH, SSC, TA, texture, sugars, organic acids, volatiles content and profile are measured for integration with sensory and WTP data. Texture was evalauted using the penetration flat probe and double compression probe as described in Obj. 2. Measurements of texture, pH, SSC, TA. organic acids and sugars for both sets FL and OR, both years were completed. Volatile analsis for the 2022 sets 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 fir blueberry breeding programs about fruit traits that maximize consumer value.	
4b. Evaluate grower and consumer behavior response to fruit quality attibutes [Socio Economics Team]	**To whom**
Finalized blueberry industry and consumer preference survey questionaires. The grower survey was first disseminated with the support of the USHBC on FEB 6. On FEB 14 there were 23 responses. We plan to request USHBC if they can send reminders to increase our response rate. The grower survey goal is to estimate growers' willingness to-pay considering blueberry quality traits and information that growers in other countries are adopting new blueberry varieties. Also this first study will re-asses industry breeding priorities. The consumer blueberry survey was first implemented FEB 8. By FEB 14, the survey collection was finalized and data collected was being check for quality control. The goal of the consumer survey is to measure consumers' propensity to purchase blueberries considering different labels that inform buyers about blueberry quality traits (e.g. "Sweety").	Breeders, growers, distributors,
Expected Impact. Information about blueberry quality traits preferred by GROWERS and information on what taste-related word that would trigger consumers' purchase, will be useful for breeders and growers in identifying quality traits with higher consumer acceptance and potential market performance, and distributors to develop marketing strategy for preferred quality traits.	

<table>
<tr><th>4c. Estimate consumers' reactions to "added-sugar" labelling and labelling information in cranberry products [Socio Economics Team]</th><th>To whom</th></tr>
<tr><td>Finalized a cranberry study assessing WTP discount for added sugars. 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). The study indicated that consumers discount for increases in added sugars in cranberry products (dried and cranberry juice), and such discount was not offset by health claims about consumption of cranberries. Also, consumers preferred other fruit juices as a sweetener compared to the addition of regular sugar. A second study assessing WTP for cranberry products from fruit developed using CRISPR technology with the result in a reduction in total sugar content is being completed. Results show that consumers will discount more for regular sugar content in relation to reduced sugar content, compared to the discount for CRISPR in relation to conventional breeding.</td><td>Breeding and Statistical Genetics teams, cranberry industry partners (nurseries, processors, growers, commodity group organizations)</td></tr>
<tr><td colspan="2">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.</td></tr>
<tr><td colspan="2">Challenges & Changes: Due to logistic difficulties in accessing large amounts of fruit from selected blueberry cultivars, as well as feedback from stakeholders on research questions with higher priorities, the proposed research on consumer behavioral response to fruit tasting using biometric parameters was re-designed into a two-part study described in 4b.</td></tr>
</table>

* To whom results are transferred during the project

Table 11. Summary of blueberry phenotyping activities completed or ongoing for SensoryStudy sets.

FC Traits	Phenotyping time	Type and # parameters	Methods	SensoryStudy set NHB		SensoryStudy set SHB	
				2021 (N=20)	2022 (N=22)	2021 (N=20)	2022 (N=20)
FC Traits							
Texture and Appearance FC							
Texture (TxPp) Penetration probe	T0	14 mechanical parameters	TA.XTPlus***	Completed	Completed	Completed	Completed
Texture (TxPp) Double compression probe		23 mechanical parameters	TA.XTPlus***	NP****		NP****	
Weight (Wg) (Size**)		Avg. individual berry weight and avg. 10-12 berry weight	High Precision Mettler Scale***	Completed		Completed	
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	Completed	Completed	Completed	Completed
		Sugars (SSg): fructose, glucose, sucrose	NIR & HPLC		Ongoing	Ongoing	Ongoing
		Organic acids (Ac): malic, citric, quinic, shikimic	HPLC	Completed	Completed	Completed	Completed
Volatile chemical composition (ChemVol)		Volatile organic composition (>60 volatile compounds)	GC-MS	Completed	Completed	Completed	Ongoing

* T0= 12-24 hours post-harvest;
** 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

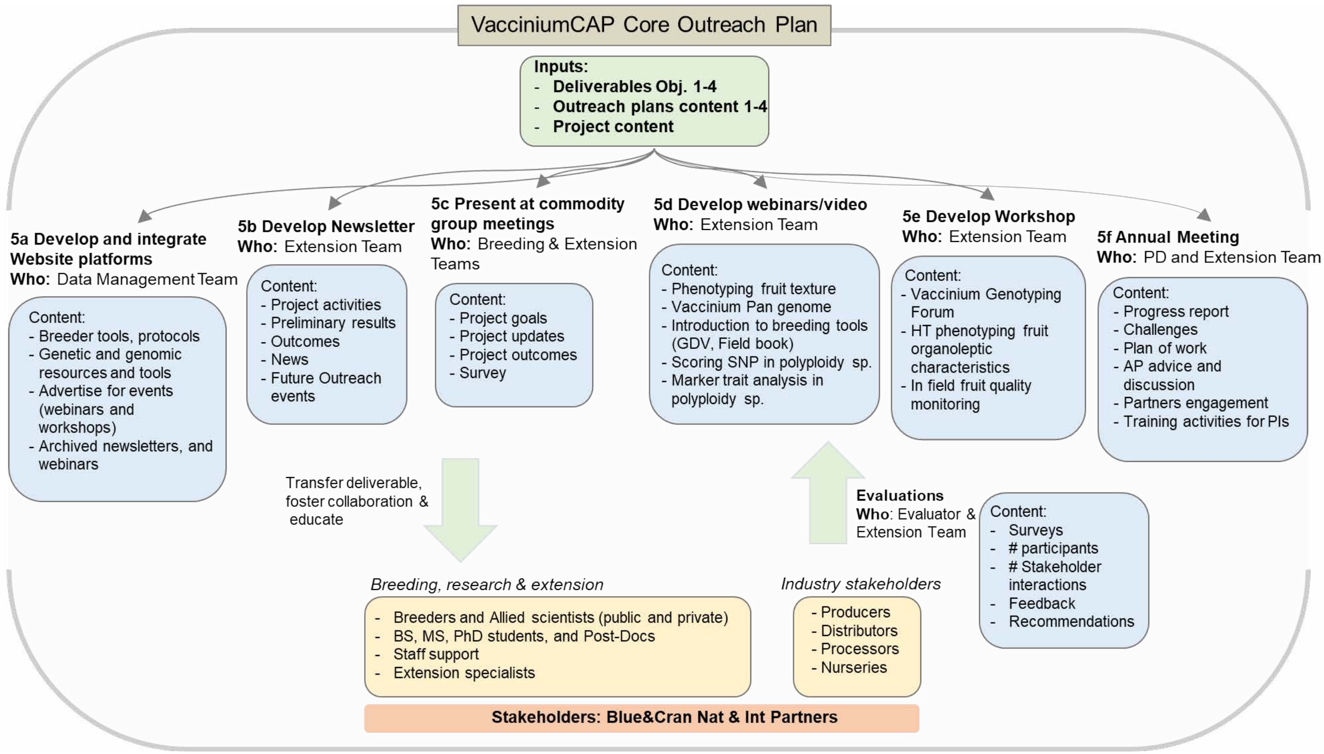
Obj. 4 – VacCAP: Plan for Year 4

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]
Analyze the data an preapare manuscripts for dissemination of results.
4b. 4b. Evaluate grower and consumer behavior response to fruit quality attibutes [Socio Economics Team]
Complete analyze the data from the two studies, preapare manuscripts for dissemination of results
4c. Estimate consumers’ reactions to “added-sugar” labelling and labelling information in cranberry products [Socio Economic Team]
Communicate outcomes through publications and presentations. One manuscript (added sugar) is in revised and resubmit. The other is being submitted soon.
Excepted challenges: Current challenge is the delay for the dissemination, as researchers prefer to have the manuscripts accepted for publication prior to dissemination.

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 3 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	
Presented by: Atucha, Main, Coe	
Reported by: Atucha, DeVetter, Main and all other PIs	
5.a. Update and expand online platforms [Data Management team]	**To whom**
Update VacCAP project website: Updates that were done to the VacCAP website in YR3: - created specific categorized content for user groups such as breeders and stakeholders and using icons to organize information on these pages. - Development of VacTrait and VacCAP for all deliverables - Updated webinar videos and newsletters - Website stats summary (September 1, 2021–August 31, 2022): 5,719 pageviews; 2,614 users; 3,211 sessions (1.23 sessions per users).	VacCAP core labs and the Vaccinium genomics, genetics and breeding research community
Update Genome Database for Vaccinium (GDV): During year 3 of the project, new GDV data included: 33 genomes, 2,685,225 annotated genes, 2,717,231 annotated transcripts, 5 genetic maps, 126,348 markers, and 203 QTLs. GDV was accessed by 6,037 users from 99 countries, with 11,054 visits and 100,439 pages served (Google Analytics).	
5.b. Develop material for newsletters and other social media [Extension Team]	**To whom**
Develop and release newsletters: Two VacCAP newsletters and three GDV newsletters were issued during year 3. Newsletters are archieved on the VacCAP ad GDV websites and are delivered by email to our stakeholders (breeders, researchers, extension specialists, and industry stakeholders, as well as project participants). Newsletter stats: 325 subscribers get the newsletter directly by email, with an opening rate of >50% (compared to an average of 45% among similar email campaigns).	VacCAP core labs, partners, stakeholders, general public
Maintained VacCAP accounts on social media: A Twitter account (@VacciniumCAP) was mantained 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 2021–August 2022): 347 followers; 7,118 (average 593/month) profile visits. Youtube stats: 774 video views, 65.9 hours of watch-time.	
5.c. Develop webinars [Extension Team]	**To whom**
Deliver webinars: delivered three webinars (see VacCAP pub and presentations for titles and link), attended by 170 participants.	VacCAP core labs, breeding and genetic partners
5d. Protocols and methodology training videos [Extension Team]	**To whom**
Develop video training. Eight instructional videos were produced during Year 3 and posted on the project's YouTube channel (see VacCAP pub and presentations for title and link). (https://www.youtube.com/channel/UCpAdtvTEebzZjvJ4SJcoXwg). These videos feature the use of new methodology to evaluate fruit quality of blueberries and how to use tools and resources avaiable on GDV .	VacCAP core labs, partners, and stakeholders
5e. Participate in commodity group meetings or field days [Extension Team]	**To whom**
Attend commodity group meetings [meetings in collaboration with industry extension network team]: Delivered 10 presentations at grower association meetings and field days across US states. Developed handouts flyer that summarized the the objectives of the VacCAP project.	Blueberry and cranberry growers, processors, distributors, nurseries and breeding partners

5f. Annual meeting [Extension Team]	To whom
Facilitate annual meetings targeting project participants, AP members, and national and international partners: An annual report that summarized Year 3 activities and plans for Year 4 was prepared and shared with members of the advisory panel prior to the meeting. Information collected during the annual meeting will be used to integrate reccomendations from the AP and plan Year 4 activities. The annual report was posted on the VacCAP web site at https://www.vacciniumcap.org/annualreport.	Team members, AP members, USDA program officers
External Evaluation [Extension Evaluator]	**To whom**
Coordinate the design and implementation of surveys for webinar, workshop and meeting participants and assist with annual report forms for project partipants: Participant surveys for the webinars noted in Obj 5c were developed with input from team members. Online surveys were used to collect feedback from webinar participants with a combination of standardized questions across webinars and customized questions related to each webinar topic. Resulting data was aggregated and summarized across webinars. Assisted with the updated design and implementation of the annual project report form for project Co-PIs and Co-PDs.	Team members, AP members, USDA program officers, extension audiences
Manage, analyze, interpret and report project evaluation feedback, recommendations, and related data: survey data, feedback and recommendations were collected from 48% of webinar participants, including breeders and scientists from public and private organizations, technical staff members, post-docs and students. Two thirds or more reported that each webinar “greatly” or “moderately” improved their understanding of the topics (66% to 97%) and almost all would recommend the webinars to others (83% to 100%). Over 11,700 users/viewers and 325 newsletter subscribers from >99 countries accessed these updated products/information in VacCAP and GDV. Overall, evaluation matrix indicates that the project is reaching a very broad audience and the resources/information generated in the project have a positive impact on this community. Feedback and recommendations for the project from webinar participants, Advisory Panel and Working Group participants were reported back to the team and are being used to inform future project activities.	Team members, AP members, extension audiences, stakeholders, USDA program officers

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 over the long term will result in a 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 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.

Challenges & changes. Engagement with the public and commodity groups was still limited due to limited in person events organized at each location due to COVID-19.

Addressing challenges and recommendations. 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.

Obj. 5 – VacCAP: Plan for Year 4

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 and GDV: continue updating VacCAP and GDV content. Updates will include: new genetic and genomic data, links to peer-reviewed publications, newsletter articles, short publications on fruit quality traits, video recordings of webinars and trainings, and social media updates.

5.b. Develop newsletters and other social media [Extension Team]

Release additional VacCAP newsletter editions: During Year 4 we will issue 3 editions of our VacCAP Newsletter that will be distributed to our stakeholders and partners. Also new issues of the GDV newsletter will be released to update national and international partners on new tools and resources available on GDV.

Prepare article for trade magazines: We plan to release one article in a trade magazine during Year 4.

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]

Five webinars are planned for Year 4: genotyping platform (Dr. Bassil); W85 genome and polyploid genetics (Dr. Iorizzo); Cranberrry socio economic studies (Dr. Gallardo and Canales); cranberry fruit quality traits genetics (TBD); Blueberry fruit quality genetics (TBD).

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

Two instructional videos will be produced during Year 4, posted on the VacCAP 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. Also new training videos on how to use GDV will be posted on our webiste by the team monthly.

5e. 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. This meetings will also serve as an event to re-assess industry breeding priorities.

5g. Annual meeting [Extension Team]

The extension team will facilitate annual meetings targeting project participants, AP members, and national and international partners.

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

The team expects that activities to engage and educate the general public about the project outcomes and impacts will re-start during year 4.

External Evaluation [Extension Evaluator]

Coordinate the design and implementation of surveys for webinar, workshop and meeting participants and assist with annual report forms for project partipants. Continue to develop and update surveys, feedback forms, report forms and related instruments and systems as needed during Year 4.

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 4.

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.

VacCAP Team Publications and Presentations

Journal Article

- Montanari S, Thomson S, Cordiner S, Günther CS, Miller P, Deng CH, McGhie T, Knäbel M, Foster T, Turner J, Chagné D and Espley R. (2022) High-density linkage map construction in an autotetraploid blueberry population and detection of quantitative trait loci for anthocyanin content. Frontiers in Plant Science. 13:965397. https://doi.org/10.3389/fpls.2022.965397
- Dare AP, Günther CS, Grey AC, Guo G, Demarais NJ, Cordiner S, McGhie TK, Boldingh H, Hunt M, Deng C, Karppinen K, Jaakola L and Espley RV. (2022). Resolving the developmental distribution patterns of polyphenols and related primary metabolites in bilberry (Vaccinium myrtillus) fruit. Food Chemistry 374, 131703. https://doi.org/10.1016/j.foodchem.2021.131703
- Lafferty DJ, Espley RV, Deng CH, Günther CS, Plunkett B, Turner JL, Jaakola L, Karppinen K, Allan AC and Albert NW. (2022). Hierarchical regulation of MYBPA1 by anthocyanin- and proanthocyanidin-related MYB proteins is conserved in *Vaccinium* species. Journal of Experimental Botany. 73, 1344-1356. https://doi.org/10.1093/jxb/erab460
- Lafferty DJ, Espley RV, Deng CH, Dare AP, Günther CS, Jaakola L, Karppinen K, Boase MR, Wang L, Luo H, Allan AC and Albert NW. 2022. The coordinated action of MYB activators and repressors controls proanthocyanidin and anthocyanin biosynthesis in Vaccinium. Frontiers in Plant Science. 13:910155. https://doi.org/10.3389/fpls.2022.910155
- Samkumar A, Karppinen K, McGhie TK, Espley RV, Martinussen I, Jaakola L. 2022. Flavonoid biosynthesis is differentially altered in detached and attached ripening bilberries in response to spectral light quality. Frontiers in Plant Science. 2529. https://doi.org/10.3389/fpls.2022.969934
- Günther CS, Plunkett BJ, Cooney J, Jensen D, Trower T, Elborough C, Nguyen HM, Deng C, Lafferty DJ, Albert NW, Dare AP and Espley RV. 2022. Biotic stress-induced and ripening-related anthocyanin biosynthesis are regulated by alternate phytohormone signals in blueberries. Environmental and Experimental Botany. https://doi.org/10.1016/j.envexpbot.2022.105065
- Edger PP, Iorizzo M, Bassil NV, Benevenuto J, Ferrão LFV, Giongo L, Hummer K, Lawas LMF, Leisner CP, Li C, Munoz PR, Ashrafi H, Atucha A, Babiker EM, Canales E, Chagné D, DeVetter L, Ehlenfeldt M, Espley RV, Gallardo K, Günther CS, Hardigan M, Hulse-Kemp AM, Jacobs M, Lila MA, Luby C, Main D, Mengist MF, Owens GL, Perkins-Veazie P, Polashock J, Pottorff M, Rowland LJ, Sims CA, Song G, Spencer J, Vorsa N, Yocca AE and Zalapa J. 2022. There and back again; historical perspective and future directions for *Vaccinium* breeding and research studies. Horticulture Research 9, 10.1093/hr/uhac083. https://doi.org/10.1093/hr/uhac083
- Ni X, Takeda F, Jiang H, Yang WQ, Saito S and Li C. 2022. A deep learning-based web application for segmentation and quantification of blueberry internal bruising. Computers and Electronics in Agriculture. 201:107200. https://doi.org/10.1016/j.compag.2022.107200
- Alan E, Yocca, Patrick P. Edger PP. 2022. Current status and future perspectives on the evolution of cis-regulatory elements in plants. Current Opinion in Plant Biology. 65: 102139. https://doi.org/10.1016/j.pbi.2021.102139
- Teresi SJ, Teresi MB and Edger PP. 2022. TE Density: a tool to investigate the biology of transposable elements. Mobile DNA. 13, 11 https://doi.org/10.1186/s13100-022-00264-4
- Mengist MF, Bostan H, De Paola D, Teresi SJ, Platts AE, Cremona G,Xinpeng Q, Mackey T, Bassil NV, Ashrafi H, Giongo L, Jibran R, Chagné D, Bianco L, Lila MA, Rowland LJ, Iovene M, Edger PP and Iorizzo M. 2022. Autopolyploid inheritance and a heterozygous reciprocal translocation shape chromosome genetic behavior in tetraploid blueberry (*Vaccinium corymbosum*). New Phytologist, in press. https://doi.org/10.1111/nph.18428
- Fong SK, Kawash J, Wang Y, Johnson-Cicalese J, Polashock J, Vorsa N. 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
- Ma X, Gallardo RK, Canales E, Atucha A, Zalapa J and Iorizzo M. 2022. "Consumers' discount for added sugars: An application to cranberry products under different nutrition-related information treatments. Applied Economic Perspectives and Policy
- Trandel M, H Oh, Johanningsmeier S, Iorizzo M and Perkins-Veazie P. Cell wall polysaccharide linkage assembly in blueberry peel and pulp reveals hemicelluloses specifically associated with the 'Indigocrisp' cultivar. Frontier in Plant Science. https://doi.org/10.3390/plants10030553

Conference Papers and Presentations

- Ma X, Gallardo RK and Canales E. 2022. "Consumer's willingness to pay for gene editing breeding: Application to cranberry products." Annual Meetings of the Agricultural and Applied Economics Association. Anaheim, CA. August 1–2.

VacCAP Team Publications and Presentations

- Trandel-Hayse M, Oh H, Johanningsmeier S, Iorizzo M, Perkins-Veazie P. 2022. Cell wall polysaccharide composition in the peel and pulp of blueberry cultivars differing in fruit texture quality. 135th Florida State Horticulture Conference, June 5-7, Sarasota, FL.
- Ma X, Canales E, Gallardo RK. "Consumers discount for added sugars: An exploratory analysis of two cranberry products under different nutrition-related information treatments." Online Agricultural and Resource Economics Seminars (OARES). December 1, 2021.
- Boase MR, McGhie T, Wang L, Zhang H, Luo H, King I, Lafferty DJ, Albert NW, Allan AC and Espley RV. 2022. Overexpression of VcMYBA1, a key anthocyanin transcription factor in highbush blueberry elevated anthocyanins, turning leaves dark red. Society of In Vitro Biology Meeting, 4-7 June 2022 in San Diego, California
- Munoz P. 2022. Genome wide association of blueberry fruit quality traits. International Horticultural Congress. August 14-19, 2022.
- Xueping N, Takeda F, Jiang H, Yang WQ, Saito S and Li C. 2022. A deep learning-based web Application to quantify blueberry internal bruising. International Horticultural Congress. August 14-19, 2022.
- Iorizzo M, Lila MA, Perkins-Veazie P, Luby 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, De Vetter L, Coe M, Chagné D, Colonna A and Espley R. 2022. VacciniumCAP, a community-based project to develop advanced genetic tools to improve fruit quality in blueberry and cranberry. Acta IHC ISHS 2022
- Knowles S, Kawash J, Johnson-Cicalese J, Polashock J, Vorsa N. 2021. Relationships between fruit rot resistance and horticultural traits in American cranberry (*Vaccinium macrocarpon* Ait.). 12th International Vaccinium Symposium, International Society for Horticultural Sciences. Remote. August 30 – September 1, 2021.
- Mengist MF, Grace M, Mackey T, Bassil N, Luby C, Ferruzzi M, Lila MA and Iorizzo M. Dissecting the genetic basis of anthocyanins accumulation and diversity in blueberries (*Vaccinium corymbosum* L). ASHS 2022 Annual Conference, July 29-August 3, 2022, Chicago, IL, USA.
- Yalcin O, Finn C, Mackey T, Pottorff M, Giongo L, Iorizzo M, Mengist MF, Coe K, Hardigan M, Luby C, King R and Bassil N. 2022. QTL Analysis of phenological and fruit quality traits in a tetraploid highbush blueberry population. ASHS 2022 Annual Conference, July 29-August 3, 2022, Chicago, IL, USA.
- Iorizzo M, Molla MF, Bostan H, De Paola D, Teresi S, Teresi A, Cremona G, Qi X, Mackey T, Bassil N, Ashrafi H, Giongo L, Jibran R, Chagne D, Bianco L, Lila MA, Rowland LJ, Iovene M and Edger P. 2022. Comparative genome analysis in blueberry revealed autopolyploid recombination behavior and a heterozygous reciprocal translocation. ASHS 2022 Annual Conference, July 29-August 3, 2022, Chicago, IL, USA.
- Iorizzo M, Lila MA, Perkins-Veazie P, Pottorff M, Mengist MF, Colonna A, Vorsa N, Edger P, Bassil N, Luby C, Mackey T, Munoz P, Zalapa J, Gallardo KR, Atucha A, Main D, Giongo L, Li C, Polashock J, Sims C, Canales E, DeVetter L, Chagne D, Espley R and Coe M. 2022. *Vaccinium* CAP status and updates: advancing genetic and genomic tools to improve fruit quality in blueberry and cranberry. NABREW conference, June 27-29, 2022, virtual.
- Mengist MF, Bostan H, De Paola D, Teresi SJ, Platts A, Cremona G, Qi S, Mackey T, Bassil NV, Ashrafi H, Giongo L, Jibran R, Chagné D, Bianco L, Finn C, Lila MA, Rowland LJ, Iovene M, Edger PP and Iorizzo M. 2022. Autopolyploid inheritance and a heterozygous reciprocal translocation shapes chromosome genetic behavior in tetraploid blueberries. XXIX Plant & Animal Genome, January 8-12, 2022, San Diego, California, USA.
- Iorizzo M, Lila MA, Perkins-Veazie P, Pottorff M, Mengist MF, Colonna A, Vorsa N, Edger P, Bassil N, Luby C, Mackey T, Munoz P, Zalapa J, Gallardo KR, Atucha A, Main D, Giongo L, Li C, Polashock J, Sims C, Canales E, DeVetter L, Chagne D, Espley R and Coe M. 2022. *Vaccinium* CAP: A community-based project to develop advanced genetic and genomic tools to improve fruit quality in blueberry and cranberry. Acta Horticulturae, Proceeding of the International Symposium on Breeding and Effective Use of Biotechnology and Molecular Tools in Horticultural Crops, August 14-20, 2022, Angers, France.
- 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.
- Iorizzo M. 2022. *Vaccinium*CAP, a community-based project to develop advanced genetic tools to improve fruit quality in blueberry and cranberry. Southeast Regional Fruit & Vegetable Conference, January 69, 2022, Savannah, GA.
- Perkins-Veazie P, Ma G, Oh H, Trandel-Hayse MA, Bassil N, Luby C, Munoz PR and Iorizzo M. Development of a high-throughput method to evaluate soluble sugar content of large sets of blueberry fruit. ASHS 2022 Annual Conference, July 29-August 3, 2022, Chicago, IL, USA.

VacCAP Team Publications and Presentations

Conference Posters

- Oh H, Pottorff M, Giongo L, Iorizzo M, Perkins-Veazie P. 2022. Texture characteristics associated with fruit shelf-life in blueberry. Southern Region Meeting, American Society for Horticultural Science, February 11, 2022, New Orleans, LA. Poster
- Bassil et al., 2022 VacCAP. Produces a targeted capture genotyping platform for blueberry. International Horticultural Congress. August 14-19, 2022. ePoster
- Oh H, Pottorff M, Mengist MF, Giongo L, Iorizzo M and Perkins-Veazie P. Examination of texture characteristics at harvest and postharvest and identification of QTLs in blueberry. The 31st International Horticultural Congress (IHC). August 14-20, 2022, Angers, France. Poster
- Trandel-Hayse MA, Oh H, Johanningsmeier S, Iorizzo M and Perkins-Veazie P. 2022. Peel and pulp texture parameters are negatively correlated to pectin and cellulose content in ten highbush blueberry cultivars. ASHS 2022 Annual Conference, July 29-August 3, 2022, Chicago, IL, USA. Poster
- Oh H, Pottorff M, Giongo L, Iorizzo M and Perkins-Veazie P. 2022. Dissecting the texture characteristics determining the phenotypic variation and postharvest change of fruit texture in blueberry. ASHS 2022 Annual Conference, July 29-August 3, 2022, Chicago, IL, USA. Poster
- Perkins-Veazie P, Oh H, Trandel M, Ma G, Mainland CM, Lila MA and Iorizzo M. 2022. Changes in blueberry fruit chemistry after six weeks cold storage. Acta Horticulturae, Proceeding of the International Symposium on Advances in Berry Crops, August 14-20, 2022, Angers, France. Poster
- Pottorff M, Coe K, Giongo L, Bassil N, Luby C, Munoz PR, Ferrao F, Perkins-Veazie P and Iorizzo M. 2022. Analysis of northern highbush and southern highbush blueberry populations using an integrated phenotyping system to capture fruit texture characteristics and other traits at harvest and post-harvest. ASHS 2022 Annual Conference, July 29-August 3, 2022, Chicago, IL, USA. Poster
- Oh H, Pottorff M, Mengist MF, Giongo L, Iorizzo M and Perkins-Veazie P. Examination of texture characteristics at harvest and postharvest and identification of QTLs in blueberry. Acta Horticulturae, Proceeding of the International Symposium on Advances in Berry Crops, August 14-20, 2022, Angers, France. Poster

Webinars

- Perkins-Veazie P, Giongo L and Zalapa J. 2022. Exploring firmness and texture in blueberry and cranberry. February 3, 2022. 76 participants. https://www.youtube.com/watch?v=Xnmvefb_X9c&t=1s
- Yocca A. 2022. Generating a *Vaccinium* pangenome to unlock previously hidden genetic variation. August 22, 2022. 41 participants. https://www.youtube.com/watch?v=dUnFxrzZAY0
- Munoz P. Making sense of volatiles and their effect on blueberry flavor. May 26, 2022. 53 participants. https://www.youtube.com/watch?v=JReCVw2cJk0&t=131s

Presentations for Growers or Other Industry Stakeholders

- Iorizzo M. VacCAP project updates. North Carolina Blueberry Council – 56th Annual Open House and Trade Show, January 11, 2022, Harrells, NC, USA. Presentations for Growers or Other Industry Stakeholders
- Knowles S, Kawash J, Johnson-Cicalese J, Polashock J, Vorsa N. 2022. Phenotyping and genotyping CNJ14-31: A unique population segregating for fruit rot resistance, fruit chemistry and other traits (Bog 18). Annual Summer Meeting of the American Cranberry Growers Association. P.E. Marucci Center, Chatsworth, NJ. August 18, 2022. Presentations for Growers or Other Industry Stakeholders.
- Kawash J, Polashock J, Vorsa N, Johnson-Cicalese J. 2022. Identifying genetic markers for defense mechanisms in cranberry fruit. Winter Meeting of the American Cranberry Growers Association. January 20, 2022 (virtual). Presentations for Growers or Other Industry Stakeholders.
- Johnson-Cicalese J, Vorsa N, Polashock J, Oudemans P, Knowles S. 2022. Update on breeding for fruit rot resistance in cranberry. Winter Meeting of the American Cranberry Growers Association. January 20, 2022 (virtual). Presentations for Growers or Other Industry Stakeholders.
- Polashock J, Kawash J. 2022. Visualizing cranberry fruit rot in a different light. Winter Meeting of the American Cranberry Growers Association. January 20, 2022 (virtual). Presentations for Growers or Other Industry Stakeholders.

VacCAP Team Publications and Presentations

- Phillips M, Lopez-Moreno H, Diaz-Garcia L, Grygleskic E, Giongo L, Atucha A, Iorizzo M, and Zalapa J. 2022. Cranberry school presentation. Cranberry fruit quality genetic research and the VacCap project: distinguishing soft and firm berries. Cranberry School Proceedings 30, January 19-20, 2022. Presentations for Growers or Other Industry Stakeholders. https://fruit.wisc.edu/cranberries/cranberry-school-proceedings/
- Blueberry Field Day, Oregon State University, North Willamette Research & Extension Center, July 20, 2022, 2022. Improving Fruit Quality Through VacCAP https://extension.oregonstate.edu/sites/default/files/documents/11671/field-day-agendablueberry2022.pdf Oregon Blueberry Conference, Salem Convention Center, Salem, OR, February 14, 2022. Presentations for Growers or Other Industry Stakeholders.
- Luby C. Innovative blueberry breeding: thoughts, priorities, and insights for the future. Oregon Blueberry Conference. Salem Convention Center, Salem, OR. February 14, 2022. Presentations for Growers or Other Industry Stakeholders.
- DeVetter LW, Luby C and Bassil N. 2022. Improving blueberry fruit quality through VacCAP. Oregon State University Blueberry Field Day. 20 July, 2022. Aurora, OR. Presentations for Growers or Other Industry Stakeholders.
- Atucha A. 2022. Cranberry fruit quality traits. Wisconsin Cranberry State Growers Association Field Day. 10 August, 2022. Black River Falls, WI. Presentations for Growers or Other Industry Stakeholders.

VacCAP newsletters

- Atucha A, Russo J, DeVetter L and Giongo L. Breaking down fruit quality - Texture and its role in blueberry. VacCAP Newsletter Issue 4, April 2022. https://www.vacciniumcap.org/textureinblueberry
- Atucha A, Russo J, DeVetter L and Edger P. Some assembly required: how the VacCAP team is developing the first Vaccinium pangenome. VacCAP Newsletter Issue 4, April 2022. https://www.vacciniumcap.org/firstvacciniumpangenome
- Atucha A, Russo J, DeVetter L and Luby C. Get to know VacCAP team member Dr. Claire Luby. VacCAP Newsletter Issue 4, April 2022. https://www.vacciniumcap.org/clairelubyfeature
- Atucha A, Russo J, DeVetter L and Chagné D. A chromosome-scale assembly of the bilberry genome identifies a complex locus controlling berry anthocyanin composition. VacCAP Newsletter Issue 5, August 2022. https://www.vacciniumcap.org/bilberrygenome
- Atucha A, Russo J, DeVetter L and Iorizzo M. High-density linkage map construction and identification of loci regulating fruit quality traits in blueberry. VacCAP Newsletter Issue 5, August 2022. https://www.vacciniumcap.org/highdensitymap
- Atucha A, Russo J and DeVetter L. There and back again; historical perspective and future directions for *Vaccinium* breeding and research studies. VacCAP Newsletter Issue 5, August 2022. https://www.vacciniumcap.org/thereandbackagain
- Humann JL and Main D. Genome database for *Vaccinium* newsletters, Issues 2, (January 2022). https://www.vaccinium.org/sites/default/files/files/GDV%20Newsletter-January2022.pdf
- Humann JL and Main D. Genome database for *Vaccinium* newsletters, Issues 3, (April 2022). https://www.vaccinium.org/sites/default/files/files/GDV%20Newsletter-April2022.pdf
- Humann JL and Main D. Genome database for *Vaccinium* newsletters, Issues 4, (July 2022). https://www.vaccinium.org/sites/default/files/files/GDV%20Newsletter-July2022.pdf

Other Products

- Zalapa J, Russo J, Atucha A. 2022. VacTrait: Color in cranberries. April 5, 2022. Online (https://www.vacciniumcap.org/sites/default/files/inline-files/Vactrait%20Color%20in%20Cranberries.pdf). Handout
- Perkins-Veazie P, Iorizzo M, Russo J, Hancock J. VacTrait: Soluble solids concentration. April 5, 2022. Online: https://www.vacciniumcap.org/sites/default/files/inline-files/VacTrait%20SSC.pdf) Handout.
- Humman JL and Main D. Humman JL and Main D. Learn how to go from BLAST to mRNA details to JBrowse. https://www.youtube.com/watch?v=FBN9s4-a7BU
- Humman JL and Main D. Learn how to use MegaSearch for Genetic Maps. https://www.youtube.com/watch?v=a8iMUo8i1po

VacCAP Team Publications and Presentations

- Humman JL and Main D. Learn how to use MegaSearch for QTL. https://www.youtube.com/watch?v=zihzFcngoqY
- Humman JL and Main D. Learn how search markers on GDV. https://www.youtube.com/watch?v=5O80zt614vA
- Humman JL and Main D. Learn the basics of how to use the MapViewer tool to explore genetic maps. https://www.youtube.com/watch?v=byG8fq62M2E
- Humman JL and Main D. Learn how to use MegaSearch. https://www.youtube.com/watch?v=Ia3v650TZAg
- Humman JL and Main D. Learn how to use the Synteny Viewer Tool. https://www.youtube.com/watch?v=t6Kl2z_dCqs

Join Us on Social Media

VacCAP
@VacciniumCAP

VacCAP
Project

Do you want to stay up to date on all the latest news and resources? Follow us on Twitter @VacciniumCAP. You will be able to:

- 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"

VacCAP
IMPROVING FRUIT QUALITY

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

www.vacciniumcap.org
vaccapsocial@gmail.com | @VacciniumCAP