Three years into the RosBREED project and the deliverables are really starting to show up. Now halfway through the project’s timeline, it is thrilling to share those research successes, but equally important to notice where adjustments, both large and small, need to be considered. As always, we enjoy the opportunity to share progress towards milestones with our Advisory Panel (AP) members and International Partners at our annual meetings in January. That meeting always sets us up for our Project Participant meeting in March, at which we strategically address an array of technical and biological issues, adjust activity plans, and then get back to work! At that meeting, which this year included 57 participants, we often piggyback a workshop targeted at specific technical issues, like the FlexQTL™ session in East Lansing this year.

Annual AP Member Meeting. RosBREED’s Project Director (PD), Co-PD, and Team Leaders (or their representatives) met with 17 of 31 AP members in San Diego, CA, 12 January 2017. In the morning session, PD Amy Iezzoni and each team leader shared updates on project objectives - major activities, outcomes, impacts - and planned next steps. Afternoon breakout sessions were devoted to crop-based small group discussions of stakeholder opportunities, needs, and bottlenecks. Fortunately, that feedback has usually reinforced that we are on track; however we are have been alerted to areas where adjustments are needed. Of course, we take advantage of some AP members throughout the year. For example, David Karp and Dave Eddy copy edit RosBREED project newsletters, while other AP members critique industry one-pagers and DNA test materials.

Annual International Partner Meeting. The next day, 12 international collaborating partners from seven institutions in six countries joined 15 RosBREED participants to coordinate genotyping needs, focusing on SNP genome scan capabilities and trait-targeted DNA test development. This function is both scientific and personal. Working across disciplines and many rosaceous crops, this collaborative group

(continued next page)
leverages vast resources and strengthens community among worldwide labs and institutions.

**Annual Project Participant Meeting.** Our Year 3 Participant Meeting was held in East Lansing, MI, 8-10 March 2017. The 57 participants (up from 42 in Year 2) included 23 faculty, 15 graduate students, and six post-doctoral associates, with two co-PIs joining via teleconference. PD Iezzoni presented a project overview and Team Leaders delivered in-depth reviews of their Year 2 accomplishments and Year 3 needs. Participants were busy - the meeting had 35 breakout group sessions (!), many of which were concurrent, followed by a wrap-up plenary session.

Next, a “Meet, Greet, and Eat” gathering featured a range of delicious rosaceous products and showcased project breeders, who discussed their progress to combine targeted disease resistance with superior fruit quality. Invited attendees included Adam DeLay, representing U.S. Senator Debbie Stabenow (D-MI), local AP members, Michigan State University Agriculture & Natural Resources College administration, and Michigan tree fruit industry leaders and growers. At the end of a productive day, all left with a shared understanding of objectives accomplished, work planned for the field and lab, and a delighted palate!

**Software Training Workshop.** In another meeting with international flavor, project consultant Eric van de Weg conducted a one-day software training workshop on 7 March 2017, immediately prior to the Participant Meeting. Eric is a co-developer of FlexQTL™, the pedigree-based analysis software used in RosBREED to locate and characterize genomic regions and their variants influencing valuable traits, as well as to sort out the parentage and ancestry of our diverse germplasm. Such information is critical to planning crosses and developing and applying DNA tests to breeding material. The 20 attendees included six faculty, five post-doctoral associates, and six graduate students, who left with an improved ability to convert the output of FlexQTL™ analyses into practical breeding tools and knowledge.

**Putting it all together.** Planning the logistics, agenda, and goals for the four annual meetings consumed a significant amount of time and effort from the project management team. However, having this time with our stakeholders, partners, and project members continues to be key to keeping the project focused and responsive to stakeholder and team input. We are especially thankful to all our AP members, international partners, and internal teams that bring many students and allied scientists to our meetings and workshop. The result is a collaborative, ever-expanding Rosaceae scientific community with strong industry and allied scientist partnerships.
RosBREED's Berry Breeding Team is at the forefront of better berries. The Team comprises a diverse group of scientists working on an array of crops in production areas across the entire U.S. Despite that diversity, each is dedicated to a common goal: Developing and delivering new strawberry, blackberry and raspberry cultivars that meet consumer expectations for superior appearance, aroma, flavor, shelf life, and texture AND that meet industry needs for durable disease resistances and productivity. Thus, this team exemplifies the unifying principle of RosBREED to apply DNA-informed breeding in addressing crop- and region-specific consumer and stakeholder needs.

Not that all this is easy! Breeding for disease resistance and improved fruit quality is no small task in these crops, each of which has large and complex genomes. In fact, some have sets of chromosome 5-6 times greater than the normal two sets. These genomes are usually highly heterozygous, with lots of internal genetic diversity. Furthermore, berry breeders are actively searching for useful alleles in wild relatives, which also have complicated genetic systems. To meet these challenges, the Berry Breeding Team is leveraging knowledge, skill sets, and tools generated by RosBREED 2 with their own and that of other allied projects.

The Team is led by Chad Finn, USDA-ARS Research Geneticist and plant breeder in Corvallis, Oregon. Strawberry breeders are Tom Davis and Lise Mahoney (University of New Hampshire), Jim Hancock (Michigan State University), Vance Whitaker (University of Florida), Pat Moore (Washington State University), and Chad. Blackberry breeders include Chad, along with Margaret Worthington, who has recently assumed leadership of John Clark's blackberry breeding program at the University of Arkansas.

Like the other RosBREED Breeding Teams, collaboration with the Pathology, DNA-Informed Breeding, DNA Testing, and Data Management Teams is a dynamic and exciting process. U.S. production of strawberry, the nation's most widely consumed berry crop, has increased significantly in recent years, but is plagued by numerous diseases. Root and crown rots caused by a range of soilborne pathogens acutely threaten the California strawberry industry, while bacterial angular leaf spot (Xanthomonas fragariae) and crown diseases are of critical concern for the Florida strawberry industry. These diseases result in high management costs for growers and reduce plant productivity.

Building on rigorous screening protocols developed under the leadership of RosBREED plant pathologist, Kelly Ivors (California Polytechnic State University), the Team is developing germplasm that will provide the basis for disease-resistant cultivars. Along the way, some exciting discoveries with implications for improved fruit quality have resulted, including DNA tests predictive for remontancy (perpetual flowering), mesifurane (sherry aroma), and γ-decalactone (fruity and peachy aroma). In blackberry, RosBREED seeks to assist breeders to develop berries with sweeter taste. Sugar profiles for the fruit of hundreds of blackberry selections were evaluated in the University of Arkansas and USDA programs, providing
data to identify genomic regions with major influences on the content of the various sugars. Such discoveries are necessary for development of the predictive DNA tests that enhance breeding effectiveness.

Nahla Bassil (USDA-ARS Corvallis) is another key RosBREED participant in the Berry Team, as she leads the group, including crop post-doc Jason Zurn, in QTL discovery, genome-wide scanning, and DNA testing. Even though not officially a co-PI in RosBREED, UC Davis strawberry breeder Steve Knapp has pitched right in, collaborating to facilitate root and crown rot resistance screening and genome scanning. Members of the Socio-Economics Team – Vicki McCracken (Washington State University), Chengyan Yue (University of Minnesota), and Karina Gallardo (Washington State University) – have evaluated consumer and producer preferences in strawberry production and eating quality, along with a case study of new cultivar adoption in the Florida industry. Lastly, the Extension Team assists with dissemination of information important for berry breeders and growers. This is a winning team contributing to improved berries for all to enjoy.

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WA. His program focuses on breeding and genetics of strawberries and raspberries, emphasizing cultivars suitable for fresh and processing markets. Washington leads the nation in red raspberry production for the processing market, so Pat also focuses on breeding for machine harvestability.

Diseases plague strawberry and raspberry growers in the PNW. Pat uses his studies on genetics and plant breeding to better understand key diseases and leverages his knowledge in developing parents or cultivars that have improved disease tolerance or resistance. Raspberry bushy dwarf virus is one disease he has spent considerable time studying and he is in developing cultivars resistant to this virus, which causes reduced plant size and, most importantly, crumbly fruit. Pat also breeds for improved tolerance or resistance to Phytophthora root rot, a serious soilborne disease caused by a fungus-like organism that can result in reduced yields and plant death.

Well before RosBREED came along, Pat was breeding for disease resistance and improved fruit quality in strawberry and raspberry. Some of Pat's releases include 'Cascade Harvest', 'Cascade Gold', 'Cascade Bounty', 'Cascade Dawn', and 'Cascade Delight' raspberries and 'Puget Crimson' and 'Puget Reliance' strawberries. In 2015, Pat's contributions to fruit breeding were recognized with the award of the Wilder Medal from the American Pomological Society. Pat is a relative newcomer to RosBREED; the project should offer Pat some powerful new tools to make his program even more effective.
Rosaceae Nemesis
Pear Fire Blight

Richard Bell, Research Horticulturist (retired)
USDA-ARS, Kearneysville, WV

Fire blight in pears is a devastating bacterial disease that can greatly decrease fruit production by killing bearing branches and can even kill trees of susceptible cultivars (Fig. 1). All major cultivars grown in the U.S. are susceptible: ‘Bartlett’, ‘Anjou’, ‘Bosc’, their red-skinned sports, ‘Comice’, and ‘Starkrimson’. The causal organism is the same as apple fire blight, *Erwinia amylovora*, and can spread rapidly through an orchard. Few effective control measures exist, making fire blight a major disease nemesis for U.S. pear growers.

RosBREED is discovering and verifying DNA tests that detect genetic factors (alleles) conferring fire blight resistance in pear. Multiple sources of resistance are targeted. A DNA test has been developed for the source of fire blight resistance originating from the old European cultivar Roi Charles de Würtemburg, an ancestor in the USDA pear breeding program (Fig. 2). The presence of genetic factors for resistance from this cultivar’s progeny will be verified in our existing seedling family. The second source of resistance is from the American cultivar Old Home. The third source of resistance is a selection of the Chinese pear species *Pyrus ussuriensis*, Illinois 76.

Seedling families segregating for fire blight resistance will be evaluated and DNA markers analyzed to discover and verify those associated with resistance. Efficient and reliable DNA tests will be developed to routinely identify resistant seedlings, thereby increasing efficiency of the breeding process. A second objective is to develop parents that will serve as efficient donors in subsequent breeding by having genetic resistances combined from multiple sources.

The deliverable to industry? Superior new cultivars with durable fire blight resistance and excellent horticultural quality – a win for both producers and consumers!

Jewels in the Genome
Bloom time in *Prunus*

Amy Iezzoni, Project Director, Michigan State University

Yields of stone fruit and almonds can be greatly reduced when flowers are killed by freezing temperatures. For example, in tart cherry, yields in Michigan are typically reduced one out of every three years due to freeze injury. In the last 12 years, two crop losses have exceeded 80% of tart cherry production, resulting in financial stress on producers and local communities. This year, peach production in the southeast U.S. has unfortunately also been significantly reduced due to spring freeze events. A strategy to decrease the probability of crop loss from spring freeze damage is to grow cultivars that have a delay in the onset of floral bud development. This strategy is based on the knowledge that flower buds become increasingly susceptible to freeze injury as they develop. For example, temperatures of 15°F cause damage to floral bud primordia of ‘Montmorency’ tart cherry when the buds are beginning to swell, but open flowers can be killed by temperatures just below freezing.

Developing *Prunus* cultivars with delayed bloom timing is a high priority for many breeding programs. Fortunately, there is genetic variation for bloom time within elite germplasm, including types that bloom weeks later than commercial cultivar (Fig. 3). A genetic study in almond identified the presence of a Mendelian trait locus named *Lb* (Late bloom) located on chromosome 4 (Ballester et al. 2001). This locus explained about 80% of the phenotypic variation. Plants with the late-blooming allele bloom 15 days later than plants without this genetic factor. Subsequently the *Lb*-containing genomic region was found to contribute to bloom time variation in cherry and apricot also, and to a lesser extent in peach (Dirlewanger et al., 2012).

A DNA test to determine which *Lb* alleles are present was developed (Ballester et al., 2001). The development of a DNA test...
for this region is currently underway in cherry. With these tests, breeders can plan effective crosses and select for late-blooming seedlings in the greenhouse without spending additional time and resources on field-planting genetically undesirable individuals. Therefore, because knowledge of this genetic region will lead to the more effective breeding of Prunus cultivars, it is featured as a RosBREED “Jewel in the Genome”.

References

Student Poster Award

Congratulations to Melinda Yin, RosBREED M.S. student at the University of Arkansas, for winning first place in an oral presentation at the Southern Regional American Society for Horticultural Science meeting in Mobile, AL, February 4, 2017.

Melinda was awarded the Norman Childers M.S. Graduate Student Paper Award for her presentation: “Development of a Standardized Phenotyping Protocol for Blackberry and Evaluation of its Effectiveness in Characterizing Seedling Populations.”
As a breeder, you need DNA information to be valuable – differentiating trait levels important to your breeding program or revealing problematic errors of identity. You need DNA information to be reliable – accurately predicting genetic potential or revealing true identity or origin. And you need DNA information to be accessible – coming from your service provider(s) of choice. By converting trait-predictive DNA tests and identity/relatedness-ascertaining fingerprinting sets to a range of genotyping platforms, RosBREED helps to enhance your access to DNA information.

New DNA tests to predict trait performance have rolled out since the beginning of RosBREED 2. Targeted trait examples include remontancy and sherry aroma for strawberry, blue mold and powdery mildew resistance for apple, and maturity timing and fruit cracking incidence for sweet cherry. Such tests add to the suite that was previously developed, refined, or otherwise rolled out in RosBREED 1. Most of these trait-predictive DNA tests were developed as simple PCR assays (SSRs and SCARs/InDel markers) for the genotyping platforms of RosBREED’s DNA Testing Portals. These tests can be converted to other marker types suitable for the numerous genotyping platforms used by service providers available to U.S. breeders (Fig. 4).

Converting many of the existing DNA tests to increase versatility to multiple platforms will help lower costs and decrease turnaround times to breeders. Cheap and fast are particularly necessary for programs aiming to screen thousands of plants in short seedling-raising windows.Internships have been effective for marker conversion. Plant and Food Research, Ltd (PFR) in New Zealand has expertise in DNA test development for high-resolution melting platforms and for single-SNP assays, and much experience in their deployment at high throughputs. Two members of the DNA-Informed Breeding Team, Stijn Vanderzande (Apple & Cherry Postdoc) and Daniel Edge-Garza (manager of the Apple & Cherry DNA Testing Portal) conducted an internship with PFR last year. Their goal was to convert as many simple PCR tests as possible to SNP-based platforms and gain skills to continue such conversions. Under the guidance of David Chagné of PFR, Stijn and Daniel successfully turned three apple DNA tests into SNP-based assays. These new DNA tests, targeting fruit acidity, firmness, and storability, were showcased at the 8th International Rosaceae Genomics Conference in Angers, France in June 2016. The interns also connected Jim Luby’s University of Minnesota apple breeding program with a commercial DNA service provider (Slipstream Automation, New Zealand) to trial deployment of these three tests for thousands of seedlings. Jack Tillman, Researcher with the Minnesota program, reported that within the year the DNA-based seedling selection was performed, a net $6.5k was immediately saved (re-allocated to better breeding purposes) in stage-1 establishment and maintenance costs! Jack calculates further projected savings over the next nine years to be another $86.5k, all by early elimination of seedlings carrying undesirable alleles.

The internship’s successful DNA test conversion followed a tried and true procedure, building on previous advances. For traits and germplasm of breeding interest, trait locus discoveries identify the genomic locations and allelic diversity, effects, and germplasm distributions of valuable genetic factors. From this foundation, DNA tests can be developed and deployed (Fig. 5).
To develop a DNA test, germplasm subsets are screened with multiple candidate DNA marker assays targeting the locus to find those new assays that reveal the allelic contrasts of the original discovery. The most effective of these candidates are confirmed as a new DNA test. Various marker types and genotyping platforms can be considered at this stage. However, most DNA tests have been developed thus far for marker types and genotyping platforms readily accessible to U.S. breeders. Now, we are anticipating versatile DNA testing access for breeding programs by converting DNA tests to multiple markers and platforms as early as possible in the DNA test development process. RosBREED’s Crop Postdocs, participating graduate students, allied scientist partners across the globe, and commercial service providers are all key players in this tool-widening effort.

The more flexible the DNA tests, the wider the choices of service providers and the more competitive the costs for U.S. Rosaceae crop breeders. These improvements are imperative as we transition this year from federal funding support of DNA testing to commercial services. While scientists conducting the upstream lab-based research get a kick out of creating new genetic assays, they are particularly motivated by knowing there is practical use of those tools, leading to breeding impact. Demand creates supply. Feel free to let us know how else we can help.

Community Events

**National Association of Plant Breeders annual conference**
UC Davis, Davis, CA
August 7-10, 2017
http://napb2017.ucdavis.edu/

**American Society for Horticultural Science annual conference**
Hilton Waikoloa Resort, Waikoloa, HI
September 19-22, 2017
www.ashs.org/?page=GeneralConference

**Ninth International Rosaceae Genomics Conference**
Nanjing Agricultural University, Nanjing, Jiangsu, China
June 26-30, 2018
https://www.rosaceae.org/node/5906179

Meet our Advisory Panel Members

Our Advisory Panel members are a critical part of RosBREED, as they validate from their experience the impact of our research and guide our priorities. They dedicate not only a day (plus two for travel) to our Advisory Panel meetings in January, but they also are called upon during the year to provide feedback to questions our project members have.

Thanks so much to all our Advisory Panel members – see you next year in San Diego!

Robin Buell
Scientific Advisory Panel

What is your job description? How do you help the Rosaceae community?
I am a professor of plant genomics at Michigan State University, where I collaborate with breeders and geneticists to understand genetic diversity in a range of crop species. I am pleased to serve on the RosBREED Scientific Advisory Panel to provide advice and feedback on genomics approaches.

Why are you interested in RosBREED?
It is a highly collaborative project that can enable breeders to use new technologies and advance breeding practices in very important crops. And of course, these are some of my favorite fruits!

How do you feel you can contribute to RosBREED?
In addition to providing advice, I can serve as an advocate for the consortium with other scientists who don’t understand the challenges of perennial tree breeding and help funding agencies to see the power of highly collaborative multi-institutional projects.

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Cultivar Corner

‘WHITE COUNTY’ PEACH

INVENTORS
John R. Clark
James N. Moore

What year was ‘White County’ released?
2004

What makes ‘White County’ special?
It is a unique white, freestone peach with slow melting flesh, large size, and sub-acid flavor, with good resistance to bacterial spot.

When was the cross made?
March, 1993

What is the pedigree?
Ark-392 x Ark-433N

What is the size of the family from which ‘White County’ was selected and are there other siblings from this cross that have commercial potential?
Don’t have the family size records. This was the only selection with commercial potential; two selections total were made from this population, made in 1997.

Has this cultivar been used in RosBREED?
This cultivar has been a parent in one or more populations used in RosBREED research. One population, a cross with a peento, melting peach (A-672) yielded unusual variation for a range of qualitative (shape, flesh color, acidity, pubescence) and quantitative traits (fruit maturity date, fruit size, bacterial spot resistance).

Any other interesting notes about ‘White County’?
This cultivar continues to be a favorite peach from the Arkansas program. This is one JR Clark takes home, without fail.

Why should growers plant ‘White County’?
Extends product line beyond standard yellow peaches, bacterial spot resistant, large and attractive fruit.

John Clark displays some of the virtues of ‘White County.’
RosBREED by the Numbers

25
Number of distinct practical breeding applications of DNA information

16,083
SNPs on the new 9k + 9k peach Illumina array, each positioned on the peach whole genome v2.0

100%
Proportion of Honeycrisp’s parents and grandparents now known


In the next issue

- Meet more members of our Advisory Panel
- Progress and updates from key teams from the second third of the year
- Our next “Jewel in the Genome” focuses on disease resistance
- What will be the next Rosaceae Nemesis? It might be one important for your crop!

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