Journal of the American Pomological Society 74(2): 104-110 2020

Fruit and Nut Germplasm Collections: Treasuries of Genetic Diversity:

Guardians of the germplasm: hazelnuts, berries, pears, hops, and mint

Kim E. Hummer[1] and Joseph Postman[1]

Additional index words: germplasm, genetic resources, genebanks, conservation,

Abstract

The NCGR-Corvallis is one of about 20 federal facilities and worksites in the U.S. National Plant Germplasm System dedicated to preserving economically important crops and their wild relatives. This genebank opened in May 1981, through a congressional funding mandate of the U.S. Department of Agriculture to conserve hazelnuts, strawberries, hops, mint, pears, currants/gooseberries, raspberries/blackberries, blueberries/cranberries, and their crop wild relatives. The unit collects, maintains, distributes, and evaluates genetic resources for these crops. The collection now includes more than 12,000 accessions. About half are living plants and the rest are seedlots. Clonal collections are conserved in orchards and as containerized plants under screen or in greenhouses. Seeds are stored at -20° C. These living plant collections are some of the most extensive in the world for each genus, and represent heritage cultivars as well as diverse wild species. Since 1981, the NCGR has annually distributed between 6,000 and 10,000 samples (cuttings, plants, plant tissue or seeds) to plant breeders and researchers around the world. Over the past decade, operational funds have been limited and administrative costs have increased. New diseases and pests have expanded their ranges. The objective of this manuscript is to provide examples of significant plant introductions (PIs) that are conserved at NCGR-Corvallis, that have qualities to overcome the pests and diseases of the future.

The USDA-ARS National Clonal Germplasm Repository (NCGR-Corvallis), dedicated in 1981, is one of nine clonal genebanks of the U.S. National Plant Germplasm System (NPGS). Over the past 38 years, the Corvallis facility has been mandated by the US Congress to conserve genetic resources of cultivars and crop wild relatives of hazelnuts (*Corylus avellana* L.), strawberries (*Fragaria* L.), hops (*Humulus* L.), mint (*Mentha* L.), pears (*Pyrus* L.), currants and gooseberries (*Ribes* L.), blackberries and raspberries (*Rubus* L.), and blueberries and cranberries (*Vaccinium* L). These genera were assigned to Corvallis primarily because of their climatic adaptation to this temperate vicinity. Germplasm is the foundation of plant breeding and crop innovation.

This genebank collects, maintains, distributes, and evaluates genetic resources for these crops, as well as documents the related information in an on-line public database, the Germplasm Resources Information Network (GRIN-Global). The plant materials at the NCGR-Corvallis are chosen for inclusion by the curator and crop technical committees because they 1) represent a unique, clonal genotype with economically significant characteristics or desirable evaluation traits, or 2) are a wild species representative of the cultivated crop.

Many consumers who shop at the grocery store may only see a final food product that they have purchased. They may not realize the critical contribution of plant germplasm in establishing that food as a saleable item, much less the broad germplasm and breeding efforts to maintain its constant and consistent supply. Stories behind crop development demonstrate part of the economic value of the intrinsic germplasm.

Each of the more than 12,000 accessions

[1] US Department of Agriculture, Agricultural Research Service, National Clonal Germplasm Repository, 33447 Peoria Road, Corvallis, Oregan 97333, USA

at the NCGR-Corvallis has an origin-story regarding pedigree or heritage, background, or locality information if wild collected. The objective of this manuscript is to provide examples of significant germplasm conserved within the collections at the NCGR-Corvallis. Examples of particular Plant Introduction (PI) accessions are cited with their significant contribution to crop development.

Materials and Methods

Plant conservation

The NCGR-Corvallis is located on the Lewis Brown Horticultural Farm of the Oregon State University. The facilities include about 1000 m^2 greenhouses, 1660 m^2 screenhouses, and about 121 ha of field plantings. The staff includes a research leader, curator, geneticist, 4 crop managers, 2 technicians, a program support assistant plus student and seasonal helpers.

First, the primary germplasm collections of the berry crops, mint, and hop are conserved as containerized living plants under protective cultivation with insect-exclusion screening. The plants receive their natural chilling during fall and winter. In the summer, flowers and fruits are removed and seedlings rogued to preserve clonal identity of the foundation plant. Primary collections of pears, hazelnuts and other tree crops are maintained as orchards.

Pathogen testing, for phytoplasmas, viroids, and viruses, is performed every 3-5 years using enzyme-linked immunosorbent assays (ELISA) and molecular tests. Known infected plants are removed from pathogen-negative areas and are preserved in separate houses. As resources are available, pathogen elimination techniques including thermotherapy and micro-propagation are applied to produce pathogen-negative germplasm.

Cultivars are tested for trueness-to-type using DNA fingerprinting sets of microsatellite or simple sequence repeat (SSR) markers (Akin et al., 2016; Bassil et al., 2012, 2015a, 2015b 2019; Freixas-Coutin et al., 2019; Gilmore et al., 2011, Zurn et al., 2018) and morphological evaluation from original cultivar releases or plant descriptions are compared to confirm horticultural and botanical identity.

Information management

Curators enter the information on newly received accessions into the Germplasm Resources Information Network (GRIN-Global, 2019) upon receipt of the plant material. These crop accessions in the Corvallis collection are assigned a 6-digit plant information, i.e., "PI" number, and a Corvallis local inventory number. The local number consists of a four-letter code and a number to recognize and individualize inventory records. The code contains "C" (for Corvallis) plus a three-letter genus code such as "CCOR" for the genus *Corylus* or "CFRA" for *Fragaria*. Integer numbers are sequentially attached after the four-letter code in the order of receipt by acquisition. A 3-number decimal suffix is attached to represent either subclones for pathogen testing purposes or seedlings from a seedlot. Germplasm can be found and requested through the on-line ordering system by placing the PI number, the Corvallis local number, or the plant name in the accession search box of the GRIN-Global System (GRIN-Global, 2019).

Results and Discussion

Significant Corylus

Oregon produces about 98% of the US hazelnuts (*Corylus avellana*). The US is the third largest hazelnut producing country, behind Turkey and Italy. The demand for these nuts by the hazelnut industry (particularly from candy manufacturers) greatly exceeds the supply, so there is a global economic need for additional nuts.

Eastern filbert blight (EFB), caused by *Anisogramma anomola*, is a disease native to North America east of the Rocky Mountains. It kills susceptible hazelnut trees and is one of the reasons the Eastern United States has limited commercial hazelnut production. The main European hazelnut cultivars are suscep-

Table 1. PI numbers from the USDA ARS National Clonal Germplasm Repository (NCGR-Corvallis) that have significantly contributed to the production and development of their crop.

Crop	Plant name	PI number	Corvallis local number	Origin	Significant contribution	Estimated value
Hazelnuts	'Gasaway'	557042	CCOR 54.001	Selection of *Corylus avellana* from Battle Ground, Washington, by Richard Gasaway. Introduced in 1926 as a pollinizer nut for 'DuChilly' production by Carroll D. Bush Nursery, Barton, Oregon.	Gene for resistance to the Oregon race of Eastern filbert blight caused by *Anisogramma anomola.*	Saved Oregon hazelnut production valued at $91.8 million annually.
Strawberries	*Fragaria virginiana* subsp. *glauca*	551718	CFRA 322.002	Selections of this subspecies that have perpetual bloom (or day-neutral blooming); male parent of 'Brighton' and 'Hecker'; original species collected from Hecker Pass in the Wasatch Mountains, Utah	Everbearing strawberry.	Expanded strawberry production to ripen every day of the year in California. University of California cultivar releases predominately have this gene.
Blackberries	'Hillquist'	553951	CRUB 723.001	Selection of *Rubus argutus* from Virginia.	Primocane fruiting blackberry.	Extended harvest time for blackberries in Midwest, expanded production throughout the world in warmer climates.
Blueberries	'Florida 4B'	554904	CVAC 1790.001	Natural complex hybrid of *V. fuscatum* and *V. darrowii* from Florida	Low chilling genes enabled the production of southern highbush blueberries to grow in warm climates without any chilling.	Blueberry production previously a North American product, this innovation expanded blueberry production throughout the world in warm climates.
Pears	'Old Home'	541456	CPYR 429.001	*Pyrus communis* selection	Vigorous, fireblight resistant rootstock	Mother for the majority of clonal rootstocks sold in the US. Also used as an interstem between quince and pear cultivars in Europe.
Hops	'Brewer's Gold'	302781	CHUM 130.001	Manitoba BB1 × open pollinated *H. lupulus* var. *lupulus*	Genes for verticillium wilt resistance and high alpha acid production.	Important in world production of hops (including Europe, UK, Australia, New Zealand, and the United States).
Mint	*M. longifolia* subsp. *capensis*	557767	CMEN 585.001	*M. longifolia* subsp. *capensis*	Genes for verticillium wilt resistance and a good essential oil profile.	Important in developing a diploid genome map for metabolic engineering and molecular breeding.

tible to this disease. An Oregon State quarantine on movement of hazelnut trees kept EFB out of the main growing areas for about 20 years. However, during the past 8 years, one race of the disease has spread throughout the cultivation acreage of Oregon.

Dr. Maxine Thompson, Geneticist and Plant Breeder from Oregon State University (OSU) and now Emeritus Professor, began a hazelnut-breeding program in 1969. She first recognized that 'Gasaway' (PI 557042, CCOR 54) a pollinizer hazelnut with a small nut, was resistant to EFB and began making crosses.

Since then, Dr. Shawn Mehlenbacher, Geneticist and Plant Breeder from OSU, has combined the resistance of 'Gasaway' with improved nut quality for many new cultivars and pollinizers. These new releases have energized farmers so that more than 50,000 new acres of hazelnuts were planted in Oregon during the past decade (Rodakowski, 2019). Each of these resistant trees have the 'Gasaway' gene in their parentage. That gene and the hard work of the OSU breeders and staff not only saved the Oregon hazelnut industry but have almost tripled the production

acreage during the past decade by providing new cultivars (Gökirmak et al., 2009, Sathuvalli et al., 2017). Dr. Mehlenbacher has not stopped there – the job is not yet done! He has now screened the NCGR-Corvallis *Corylus* collection with EFB isolates and molecular markers and found 170 additional PI accessions that represent other sources of resistance to this disease (Rodakowski, 2019). His crosses will pyramid numerous sources of resistance into improved cultivars to resist multiple strains of EFB and hazelnuts will continue to thrive and produce in Oregon's temperate climate.

Significant Fragaria

Since the turn of the 1900s, strawberry breeders were interested in the everbearing or perpetual flowering trait. Dr. Royce Bringhurst, Geneticist from the University of California (UC) at Davis, made a monumental find at Hecker Pass in the Wasatch Mountains, Utah, in the 1950s where he collected *Fragaria virginiana* subsp. *glauca* (S. Watson) Staudt that bloomed in late summer. Bringhurst was visionary to see "that only time and breeding was needed before the fruit [of crosses with this subspecies] is truly equal to the best of the short-day types." Indeed, his breeding program paved the way for the production of ripe strawberries somewhere in California every day of the year by utilizing this everbearing (or day-neutral) trait!

The NCGR-Corvallis strawberry collection includes the significant accession donated by Dr. R. Bringhurst, selection CA 65.65-601 (PI 551718, CFRA 322.002) which is a day-neutral *F.* ×*ananassa* strawberry derived from the original *F. virginiana* subsp. *glauca* from the Wasatch Mountains, Utah. The genetic and economic importance of the wild Utah strawberry and its derivatives to the California strawberry industry cannot be understated.

The recent molecular study (Hardigan et al., 2018) confirmed the broad distribution of the Wasatch flowering gene throughout the UC strawberry cultivar releases. The NCGR-Corvallis also has 17 other accessions of wild strawberries collected from that region in Utah, advanced backcrossed selections of those wild collections, and early cultivar releases from the UC, Davis program. The cultivars developed with the everbearing genes from that Utah subspecies built the modern strawberry industry.

In 2017, the US produced 725,748 MT of strawberries, valued at $3.5 billion (NASS, 2017). The U.S. strawberry industry is primarily located in the southern and coastal areas of California (Geisseler and Horwath, 2019). In 2017, the US harvested strawberries from 52,700 acres located in California, Florida, Oregon, North Carolina, Washington, New York, Michigan, Pennsylvania, Wisconsin, and Ohio (NASS, 2017). https://www.agmrc.org/commodities-products/fruits/strawberries.

Significant Rubus

One of the most exciting events in blackberry (*Rubus* subg. *Rubus* hybrid) production in the past two decades has been the development of the primocane-fruiting blackberry. Drs. Jim Moore and John Clark, Geneticists and Plant Breeders at the University of Arkansas (UA), recognized that 'Hillquist' (PI 553951, CRUB 723), a public domain blackberry, originally collected from the wild in Virginia, had a propensity to bloom on first year canes, unlike other blackberries. This previously unknown genotype provided the source of the primocane-fruiting for the new cultivar releases of the UA program (Clark, 2008). Dr. John Clark has been breeding and improving these "primocane" blooming plants by combining this trait with improved flavor, fruit size, shipping quality, and other characters to develop a series of blackberry cultivars. The 'Hillquist' source of primocane fruiting has resulted in the release of five public cultivars from the University of Arkansas with several million plants established by both commercial and garden growers. Production is in the US, Mexico, and other countries, indicating an interna-

tional impact of this germplasm. This has led to the commercial domestic production of blackberries in the late summer and fall, where there was no prior substantial production. The germplasm will further revolutionize blackberry production to broader climates across the world.

In 2017, U.S. blackberry production was valued at $31.1 million, up from the previous year (NASS, 2017).

Significant Humulus

Professor Earnest Stanley Salmon was appointed as a mycologist at the South Eastern Agricultural College, in Kent, England, in 1906. His initial research on powdery mildew of hop, caused by *Podosphaera macularis* (formerly *Sphaerotheca humuli*), encouraged him to start a breeding program on hops. He collected cones from a wild hop (*Humulus lupulus* var. *lupuloides*) BB1 from Morden, Manitoba, Canada, which was open pollinated from male European hops (Salmon, 1934). The offspring produced 'Brewer's Gold' (PI 302781, CHUM 130) and 'Brewer's Favourite'. He also crossed a female 'Canterbury Golding' with a male OB21. The OB21 was the result of 'Brewer's Gold' x American Male OY1 to produce 'Northern Brewer' (PI 558710, CHUM 146). These two significant hop cultivars, 'Brewer's Gold' and 'Northern Brewer' changed the hop industry by being powdery mildew resistant types with high-alpha acid content. Virtually all of today's high-alpha varieties were derived from these two cultivars. In addition, the USDA ARS NCGR-Corvallis Hop collection contains wild hops collected by R. Hampton, in 1989 (Hampton, 2001).

In 2018, production for Idaho, Oregon, and Washington totaled a record high 107 million pounds, up one percent from the 2017 crop of 106 million pounds. The value of production for the US totaled $583 million, down one percent from the previous year. (NASS, 2018)

Significant Mentha

One of the most significant economic challenges in 'Black Mitcham' peppermint (*Mentha ×piperita*) or Scotch spearmint (*Mentha spicata*) production is verticillium wilt damage caused by the fungus, *Verticllium dahlia*. Vining et al. (2005) examined *Mentha longifolia*, one of the diploid crop wild relatives from which these mints were derived on an evolutionary scale. Upon screening and evaluation of the *Mentha longifolia* in the NCGR collection, one South African accession particularly stood out (PI 557767, CMEN 585.001). This accession had resistance to verticillium wilt, with an essential oil composition of 32.8% pulegone, 24.3% menthone, 11.3% 1,8-cineole. This accession has now provided draft *de novo* genome and plastome assemblies (Vining et al., 2017). Vining et al. (2017) state that "the 353 Mb genome contains 35,597 predicted protein-coding genes, including 292 disease resistance gene homologs, and nine genes determining essential oil characteristics. A genetic linkage map ordered 1,397 genome scaffolds on 12 pseudochromosomes. More than two million simple sequence repeats were identified." This accession has facilitated molecular marker development and is a valuable resource for metabolic engineering and molecular breeding.

Significant Pyrus

The Corvallis *Pyrus* genebank maintains more than 1,300 cultivars and about 900 crop relatives as trees in a living collection at Corvallis. Pear orchards are grown on rootstocks and one of the most economically important rootstocks and progenitor of rootstocks is the cultivar 'Old Home' (PI 541456, CPYR 429). This rootstock originated as a chance seedling, apparently a pure form of *P. communis*, selected by B.O. Curtiss in Paris, Illinois, before 1957. This selection forms a vigorous and well-formed trunk that is fireblight (caused by *Erwinia amylovora*) resistant with a great framework. Seedlings of 'Old Home' are vigorous, grow uniformly, and tend to be blight resistant. Lyle Brooks (1984), Melvin Westwood, and Porter Lombard developed a

series of "Brooks®" rootstocks. OH x F 97 and 87 remain as the most offered by major wholesale nurseries in the US and OH x F 333 is sold to homeowners. European pear growers widely use 'Old Home' as an interstem between quince rootstock and pear cultivars (Mielke et al., 2008).

Significant Vaccinium

The development of low chilling blueberries, also known as southern highbush blueberries, has been significant in the expansion of this originally North American-only crop. Now blueberries are grown in Central and South America, as well as Europe and Asia. The original northern highbush blueberry cultivars were initially selected or bred about 100 years ago by Dr. Fredrick Coville and Ms. Elizabeth White. They started with elite hand-selected plant germplasm native to New Hampshire and New Jersey. The buds of these blueberries required about six weeks of chilling temperature in the winter when the plants were dormant before the buds would begin growing in the spring. A collaboration of breeders, including Dr. Arlen Draper, Geneticist and Plant Breeder at the USDA, Dr. Ralph Sharp, Geneticist from the University of Florida, and others began making wide crosses within the section *Cyanococcus*, the true blueberries. They began using a natural highbush × lowbush hybrid, *Vaccinium fuscatum* × *V. darrowii* 'Florida 4B' (PI 554904, CVAC 1790). This selection is in the pedigree of many successful southern highbush blueberries. Using this selection and others from lower chilling regions of Florida and through many backcrosses, the breeders reduced the chilling requirement of new cultivars first to 500 hours, then to 400 hours, and finally to 0 hours, over a decade. These low or no chilling blueberries, are being widely planted in warm areas under protected cultivation in high tunnels. A novel horticultural production scheme called "evergreening" was developed to be used with these no chilling, southern highbush blueberries. This technique allows for berry production in warm climate every 8 months rotated through the year. Now consumers going to markets in the Northern and Southern Hemispheres can enjoy eating fresh blueberries anytime in the year, even in winter. In 2016, fresh and processed cultivated blueberries from the US were valued at $720.2 million.

Conclusions

These germplasm stories of "Hall of Fame" PI numbers are only a few examples of recent genetic innovations and unusual ancient genotypes that produce high value agricultural crops and resolve economic challenges for growers. These stories exemplify that the security and success of our food supply depends on long-term conservation of diverse plant germplasm and an excellent system of applying agricultural crop innovation and research.

Acknowledgements

This research is supported by the U.S. Department of Agriculture, Agricultural Research Service CRIS 2072-21000-049-00D. The USDA is an equal opportunity provider and employer.

Literature cited

Akin, M., A.M. Nyberg, J. Postman, and N.V. Bassil. 2016. A multiplexed microsatellite fingerprinting set for hazelnut cultivar identification. Eur. J. Hortic. Sci. 81 (6): 327-338.

Bassil, N.V., A.M. Nyberg, K.E. Hummer, J. Graham, M. Dossett, and C. E. Finn. 2012. A universal fingerprinting set for red raspberry. Acta Hortic. 946: 83-87.

Bassil, N., A. Nyberg, C. Finn, J. Clark, C. Peace, and A. Iezzoni. 2015a. Development of a multiplexed fingerprinting set in blackberry. Acta Hortic. 1133: 89-96.

Bassil, N. and J.D. Postman, 2010. Identification of European and Asian pears using EST-SSRs from *Pyrus*. Genet. Res. Crop Evol. 57**:** 357-370.

Bassil, N.V., A.M. Nyberg, Y.-K. Kim, and J. Postman. 2015b. Improved microsatellite markers for quince (*Cydonia oblonga*) genetic analysis. Acta Hortic. 1094: 57 - 65.

Bassil, N.V., A. Bidani, A. Nyberg, K. Hummer, and L.J. Rowland. 2019. Microsatellite markers confirm identity of blueberry plants in the USDA-ARS National Clonal Germplasm Repository collection.

Genetic Resources and Crop Evolution. In Press.
Brooks, L.A. 1984. History of the OH x F pear rootstocks. Fruit Var. J. 38:126–128.
Clark, J.R. 2008. Primocane-fruiting blackberry breeding. HortScience 43: 1637–1639.
Freixas-Coutin, J.A., S. An, J. Postman, N.V. Bassil, B. Yates, M. Shukla, and P.K. Saxena. 2019. Development of a reliable *Corylus* sp. reference database through the implementation of a DNA fingerprinting set. Planta 249(6):1863-1874. https://doi.org/10.1007/s00425-019-03131-4
Gilmore, B.S., N.V. Bassil, and K.E. Hummer, 2011. DNA extraction protocols from dormant buds of twelve woody plant genera. J. Amer. Pomol. Soc. 65:201-207.
Gökirmak, T., S.A. Mehlenbacher, and N.V. Bassil. 2009. Characterization of European hazelnut (*Corylus avellana*) cultivars using SSR markers. Genet. Res. Crop Evol. 56**:** 147-172.
GRIN-Global. 2019. https://npgsweb.ars-grin.gov/gringlobal/search.aspx accessed 4 August 2019.
Geisseler, D. and W.R. Horwath. 2019. Strawberry Production in California http://apps.cdfa.ca.gov/frep/docs/Strawberry_Production_CA.pdf
Hampton, R., E. Small, and A. Haunold. 2001. Habitat and variability of *Humulus lupulus* var. *lupuloides* in Upper Midwestern North America. J. Torrey Bot. Soc. 128(1): 35-46.
Hardigan, M. A., T. J. Poorten, C. B. Acharya, G.S. Cole, K.E. Hummer, N.V. Bassil, P.P. Edger, and S.J. Knapp. 2018. Domestication of temperate and coastal hybrids with distinct ancestral gene selection in octoploid strawberry. Plant Gen. 11:180049 doi:10.3835/plantgenome2018.07.0049
Mielke, E.A., J. Turner, and D. Sugar. 2008. Pear production on 'Old Home × Farmindale' (OHXF) interstem-rootstock combinations. Acta Hortic. 800, 645-652 DOI:10.17660/ActaHortic.2008.800.86 https://doi.org/10.17660/ActaHortic.2008.800.86
NASS, 2017. USDA-National Agricultural Statistics Service, Noncitrus Fruits and Nuts 2013 Summary, Nov. 2017.
NASS, 2018, USDA-National Agricultural Statistics Service, National Hop Report. Dec. 19, 2018.
Rodakowski, G. 2019. Hazelnut Control Order. In: 2019 Growers Handbook, 104th Annual Meeting, Corvallis, Oregon. January 17, 2019. pp 144-146.
Sathuvalli, V.R., S.A. Mehlenbacher, and D.C. Smith. 2017. High-resolution genetic and physical mapping of the Eastern Filbert Blight resistance region in 'Jefferson' hazelnut (*Corylus avellana* L.). Plant Gen. 10(2) doi:10.3835/plantgenome2016.12.0123
Salmon, E.S. 1934. Two new hops: Brewer's Favourite and Brewer's Gold. J. South-East Agricultural College, Wye, Kent, England 34:93-105.
Vining, K., Q Zhang, A.O. Tucker, and T.M. Davis. 2005. *Mentha longifolia* (L.) L.: A model species for mint genetic research. HortScience 40(5): 1225-1229.
Vining K.J., S.R. Johnson, A. Ahkami, I. Lange, A.N. Parrish, S.C. Trapp, R.B. Croteau, S.C.K. Straub, I. Pandelova, and B.M. Lange. 2017. Draft genome sequence of *Mentha longifolia* and development of resources for mint cultivar improvement. Mol. Plant.10, 323–339.
Zurn, J.D., K.A. Carter, M.H. Yin, M.W. Worthington, J.R. Clark, C.E. Finn, and N.V. Bassil**.** 2018. Validating blackberry seedling pedigrees and developing an improved multiplexed microsatellite fingerprinting set. J. Amer. Soc. Hort. Sci. 143: 381-390 doi.org/10.21273/JASHS04474-18.
Zurn, J.D., K.A. Carter, M.H. Yin, M.W. Worthington, J.R. Clark, C.E. Finn, and N.V. Bassil**.** 2018. Validating blackberry seedling pedigrees and developing an improved multiplexed microsatellite fingerprinting set. J. Amer. Soc. Hort. Sci. 143: 381-390 doi.org/10.21273/JASHS04474-18.