# Michigan State University Report to NCCC-212, November 2021

### ANNOUNCEMENTS:

**1.** New collaborators needed! If you have access to galls from the blueberry stem gall wasp and interested in collaborating with us, please contact Pat Edger (<u>edgerpat@msu.edu</u>). Additional information regarding this insect pest can be found here: <u>https://www.canr.msu.edu/news/blueberry\_gall\_wasp\_management\_guidelines</u>

# **OBJECTIVE I – DEVELOP IMPROVED FRUIT GERMPLASM THROUGH COOPERATIVE BREEDING**

<u>Blueberry</u>: Molecular breeding efforts in blueberry have been slow due in large part to the lack of genetic information and genomic resources. Previous studies have developed genetic and genomic resources for a wild diploid species (2n=2x=24) of blueberry (Gupta et al., 2015). However, this draft genome has a large number of scaffolds (13,757 total; N50 of ~145kb), high percentage of gaps (~27.35%) in ~393.16Mb assembly, and most importantly, does not reflect the genome complexity of the economically important and tetraploid (2n=4x=48) highbush blueberry. Last year, we published a chromosome-scale genome for the tetraploid northern highbush blueberry (Colle et al., 2019). The haplotype-phased assembly consists of 48 chromosome-length pseudomolecules with 1,679,081,592 bases of assembled sequence, ~1.29% gaps, and an average of 32,140 protein coding genes per haplotype (128,559 total). Benchmarking Universal Single-Copy Orthologs (BUSCO) analysis was performed to assess the completeness of the assembly and quality of the genome annotation. The annotated gene set contains 1,413 out of 1,440 (98%) BUSCO genes. The availability of this draft genome will facilitate the discovery and analysis of genes encoding economically important traits and accelerate genome-guided breeding efforts. We leveraged this genome, combined with gene expression and metabolite data measured across fruit development, to identify candidate genes involved in the biosynthesis of important phytonutrients among other metabolites associated with superior fruit quality.

Genomic resources for blueberry are publicly available: Dataset and Genome Browser.

Current work: As part of a USDA SCRI project (VacCAP; https://www.vacciniumcap.org/), we recently completed assembling and annotating 36 genomes for Northern Highbush (NHB), Southern Highbush (SHB), and cranberry (CB) – goal to construct a pangenome. The vast majority of genomes have a high BUSCO completeness score (>95%). 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.

Pangenome analyses will be conducted to identify the variable gene content that contributes to fruit quality differences between various cultivars. A Vaccinium genotyping platform will be developed as. Part of VacCAP that will target both the core (80%) and dispensable (20%) portions of the pangenome for NHB, SHB and CB.

<u>Strawberry</u>: A high-quality reference genome for the octoploid to serve as a platform for identifying agriculturally important genes and applying genomic-enabled breeding approaches. The assembly of the octoploid strawberry genome, with an estimated genome size of 813.4 Mb, has been particularly challenging due to its high heterozygosity and ploidy level. We recently published a chromosome-scale genome for the cultivated octoploid strawberry (*Fragaria x ananassa*) (Edger et al., 2019). The total length of the final assembly is 805.5Mb distributed across 28 chromosome-level pseudomolecules, plus 408.2Mb of haplotype variants. We annotated 108,087 protein-coding genes along with 30,703 long non-coding RNA (IncRNA) genes. Gene annotation and genome assembly quality were evaluated using the Benchmarking Universal Single-Copy Orthologs (BUSCO) method. Most (99.17%) of the 1,440 core genes were identified in the annotation, supporting a high-quality genome assembly and annotation. Furthermore, we identified genes that encode various important traits, including metabolites associated with fruit quality and disease-resistance.

Genomic resources for octoploid strawberry are publicly available: Dataset and Genome Browser.

We previously also published a near-complete genome of diploid woodland strawberry (*Fragaria vesca*) (Edger et al., 2018). This genome assembly (220.8 Mb total) has a contig N50 length of ~7.9 million base pairs (Mb), representing a ~300-fold improvement of the previous version. The vast majority (>99.8%) of the assembly was anchored to 7 pseudomolecules. We obtained ~24.96 Mb of sequence not present in the previous version of the *Fragaria vesca* genome and produced an improved annotation that includes 1496 new genes. Genomic analyses uncovered numerous, large-scale scaffolding errors present in each chromosome in the previously published version of the *F. vesca* genome. This new version of the genome has already been leveraged to identify resistance locus to Fusarium wilt in octoploid strawberry (Pincot et al., 2018).

Genomic resources for diploid F. vesca are publicly available: Dataset and Genome Browser.

A new reference genome for F. vesca (completely runnerless and red fruited) was published earlier this year (Alger et al., 2021). Genome resources are available here: Dataset

Resources for additional wild species:

A chromosome-scale genome for another diploid progenitor (*Fragaria iinumae*) of the cultivated strawberry is now abaailble (Edger et al., 2020). Genomic resources for diploid *F. iinumae* are publicly available: Dataset and Genome Browser.

In addition, new genomic resources for five species (*Fragaria mandschurica, Fragaria daltoniana, Fragaria pentaphylla, Fragaria nilgerrensis*, and *Fragaria viridis*) were recently published, including new estimates of species relationships, the first estimate of the pangenome for strawberry (~43-45% is core genes), and uncovered evidence for a new diploid strawberry species (Qiao et al., 2021). Furthermore, multiple independent mutations in MYB10 was shown to cause fruit color variation in wild strawberries. Lastly, results from gene family analyses will serve to guide future breeding efforts.

## **Collaborative Projects with NCCC-212 Members**

"VacciniumCAP: Leveraging genetic and genomic resources to enable development of blueberry and cranberry cultivars with improved fruit quality attributes" was funded by NIFA-SCRI from 2019-2024. The project involves multiple NCCC-212 committee members.

#### **References:**

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