# June 2021 Final Report California Grape Rootstock Improvement Commission California Grapevine Rootstock Research Foundation American Vineyard Foundation California Table Grape Commission CDFA Improvement Advisory Board

### Project Title: Development of next generation rootstocks for California vineyards.

Principal Investigator: Andrew Walker, Dept. Viticulture and Enology, UC Davis, 530-752-0902 awalker@ucdavis.edu

### Reporting Period: January 2019 to June 2021

**Overall Summary:** Since my last report Nina Romero has made excellent improvements to our rootstock screening and is currently re-vamping our ring nematode resistance screening, and we have replaced three technicians who departed over the past year. There are 444 genotypes in testing for resistance to nematodes, salt or both. Our 2019 crosses again focused on using fertile and tetraploid VR hybrids to get rotundifolia forms of resistance into better rooting backgrounds, and mostly failed (due to the genetic distance between Vitis grape species and rotundifolia. We have been successful with crosses to two VR hybrids both of which resist phylloxera and combined with rootstocks. Most of the seeds from the 2018 crosses are in storage for the next grape breeder, except for the 18113 (GRN3 x V. acerifolia 9018. Summaira Riaz used a limited mapping approach and identified a major QTL for Cl- exclusion on chromosome 8; the same chromosome where the Cl- locus was identified from V. berlandieri. Chris Chen is working on his PhD with this population which brings excellent and broad nematode resistance to our best form of salt tolerance. We have improved our phylloxera screening in the greenhouse and have verified a number of fertile VR hybrids also have strong phylloxera resistance. A new post-doc (Erin Galarneau from the Baumgartner lab) was hired to direct examinations of phenolic compounds responsible for phylloxera and nematode resistance. They will also assist our efforts to determine how O39-16 induces fanleaf degeneration resistance. We are also making good progress on identifying the basis of red leaf virus tolerance. These efforts were being directed by a visiting scholar from China, but he has returned to China. We have rootstock examples of strong tolerance (St. George and AXR1) and very sensitive (Freedom and 101-14) and rapid tissue-culture and greenhouse-based screens that mimic field test results.

**2019 Pollinations:** The 2019 crosses which were designed to use tetraploid and diploid *Vitis Muscadinia* hybrids from crosses of 101-14 x *M. rotundifolia* Trayshed (the 07107 population. We are advancing these as rootstocks but have thus far not been able to find fertile forms that we could introgress the strong resistance from *rotundifolia* into other backgrounds that root well. The few seeds we have produced were not viable. We made many crosses with tetraploid and diploid 101-14 x Trayshed progeny (some of which we had doubled their chromosome numbers. None of these crosses were successful. We made crosses with T6-42 (a fertile VR – *vinifera rotundifolia*) hybrid has repeatedly tested as resistant to phylloxera and ring nematodes. Very few seeds were produced, but T6-38 (also a fertile VR with good phylloxera resistance) was a more successful parent and produced 125 seeds in crosses to five standard rootstock parents. These seeds are in storage for the new grape breeder.

**2018 Pollinations and planting:** None of the 2018 crosses were germinated except 18-113 (GRN-3 x *acerifolia* 9018). The first 78 genotypes have been planted in the field. Thus far this group appears to segregate 1:1 for root-knot HarmAC, salt tolerance appears multigenic, and rooting is skewed towards hard to root.

Nina ran a trial with this set to see if the seedling root architecture could predict the architecture of green and hardwood cuttings. She completed the comparison with greens and there was poor correlation both in respect to root angle ( $R^2$ =0.49) and root thickness ( $R^2$ =0.15). In addition, she tried different containers, both 4" pots and Styrofoam cups as well as two media, perlite and our standard seedling mix which is the UC agronomy mix cut about ¼ with perlite. The correlations were not strong, but observations indicated that perlite gave better results especially for root diameter.

### **2018** Pollinations and planting

The first 78 vines of the 18-113 population (GRN-3 x *acerifolia* 9018) were planted in May of 2019. After testing for salt and nematode resistance and evaluation for horticultural traits, the number retained by this time has been reduced to 19. In late May of this year Nina planted another 150 genotypes to the field for horticultural evaluation. Of these 141 have been tested for salt resistance and 136 tested for HarmAC (the mixture of 2 root-knot strains capable of damaging Harmony and Freedom rootstocks).

### Root knot nematode screening

Nina's focus over this reporting period was to test as many of the remaining populations and individual selections as possible in preparation for Walker's retirement and hiring of a new breeder. This allowed the testing of some larger populations to document the capability of some crosses to produce highly resistant progeny. Over the period to date, 457 tests were conducted on 408 genotypes. Table 2 shows the results for six populations with 07-107 providing a very high percentage of HarmAC resistant progeny. The 16-063 cross with b55-1 (a tetraploid VR) was hypothesized to confer resistance from its *rotundifolia* background but proved unsuccessful. A bit surprising was the relatively high recovery of category 4 resistant progeny in the two crosses to *acerifolia* as neither parent rates a 4 on their own. For the 18-113 cross about a third were resistant. The group of 58 miscellaneous crosses were tested to find any worthwhile individuals in older crosses.

#### **Ring nematode**

Over the period Nina conducted 172 ring tests on 155 different genotypes. The results confirm that the only reliable ring resistance of the many tested so far derives from *Muscadinia* although not all progeny are resistant, as reported previously for 6 of the 34 tested genotypes in the 07-107 population. All 6 genotypes in the most resistant category in this period were VR or VM or were one generation removed from such a cross.

# Xiphinema index (Xi)

No successful dagger nematode trials were completed in this reporting period. The 07-107 and VRs trial was contaminated with RKN. Currently we have five 25 gallon barrels of sandy soil in Greenhouse 19 co-culturing on Colombard and St. George.

# **Field trials**

### UCD grafted rootstock trial

There was significant activity over the period in establishing and expanding field trials with focus on nematode resistance and salt and fanleaf tolerance. In the first two weeks of June our fifth grafted rootstock trial at UCD will be planted. Note that all have better nematode resistance and some better rootability than the 101-14Mgt and 1103P reference selections. From previous testing, we know there is no salt resistance in the 07-107 line so these selections weren't tested for chloride exclusion. Similarly there are no ring results for the 16141-003 and the 18-113 line since they don't have *rotundifolia* in their background, the only reliable source of ring nematode resistance. Hence the gaps in ring results.

# **UCD Fanleaf Trial**

The 07-107 (101-14Mgt x Trayshed VM = Vitis X Muscadinia) population offers an opportunity to combine resistance to 3 problematic nematode species and can provide rootstock induced resistance to

fanleaf degeneration. Table 3 presents an assessment of scion growth on nine 07-107 selections and 4 commercial rootstocks 10 weeks after grafting to clean and infected Cabernet Sauvignon. Figure 1 clearly demonstrates the variable performance between stocks and between clean and infected scion material on a given stock. Table 3 presents the nematode resistance and rooting ability of the stocks chosen for the 2021 UCD fanleaf trial. Stock -091 is tolerant to fanleaf while -012 is moderately tolerant. The other 3 stocks in the 07-107 line will provide the chance to assess their level of fanleaf tolerance.

**Phylloxera** – **Phenolic compounds in grapevine roots:** We are studying phenolic compounds and their relation to phylloxera resistance. Phenolics do play a major role in the hypersensitive response (HR) against insect herbivores and microorganisms. We are also exploring an association between grape color and infestation level of own-rooted vines that suggests that white cultivars might exhibit a higher susceptibility (Arancibia et al. 2018). We extracted phenolic compounds from red, pink and white varieties plus 2 rootstocks to compare their phenolic composition through LCMS-QTOF profiling at the Food Safety & Measurement Facility. Total phenolics and tannins were quantified with the Adams-Harbertson assay. Based on PCA analysis of first round of results using the UC Davis metabolite database, we have selected anthocyanins and 12 additional compounds to continue towards target analysis. We added Pinot noir, Pinot blanc, Pinot gris, Dolcetto, Alicante Bouschet, Pedro Giménez Arg, 140 Ru, SO4 and 5BB to the several other cultivars. Dr. Celeste Arancibia is in my lab for a three month research fellowship and expects results later this summer.

# **Drought tolerance/avoidance**

One of our long-term goals has been to produce stocks with a deep plunging root phenotype that can exploit the entire soil profile to scavenge for water in seasons and times of year when water is limiting. Table 6 shows rooting ability for 23 stocks at 3 different time periods. Stocks that don't have scores for a particular time period failed to propagate beyond the previous level. Figure 2 shows the root behavior of the two most promising stocks at 17 days coming out of callus and at 12 weeks after growth in deep pots. The two most promising stocks are included in the 2021 UCD rootstock trial (Table 2).

Kevin Fort is now working in the Cooperative Extension Office in Solano County but we have a good start on root depth and architectural traits and it will be ready for the next breeder. This work utilizes various gel and agars left the lab to work for an environmental consulting agency in Sacramento. We are continuing his work on root fibrosity/depth and salt tolerance with increasing concentrations of 'agar' (Gelzan, Phytotechnology Labs) to develop a simple system to discriminate deep vs shallow root growth.

We collaborated with the McElrone lab studying root anatomical and morphological differences between 110R and 101-14. Together, we are developing a two-layered system where the lower half has been infused with PEG to modify the water potential of the medium. Plant apices plus one expanded leaf from *in vitro* plants were grown in 50 ml tubes containing 20 ml MS medium supplemented with 5 g/l gelzan (Phytotechnology labs). As soon as roots were visible, plantlets were transferred with original medium on top of 20 ml medium supplement with 0, 200 or 400 PEG 8000 (Sigma). Each treatment was replicated 5 times. Root growth was recorded for 2 months using digital images and noninvasive high-resolution x-ray computed microtomography imaging. Data is currently being analyzed.

Using *CRISPR* technology to study grape aquaporins: PIP proteins (plasma membrane intrinsic proteins) are aquaporins that facilitate the transport of water and small neutral molecules across cell membranes. Cecilia Agüero has collaborated with others to design gRNAs targeting the *V. vinifera PIP2-1* gene to knock it out. Plasmid construction with DNA harboring CRISPR–Cas9 and guides was performed by Dr. M. Ron at Britt's lab. Transformations of embryogenic callus of Thompson Seedless, Chardonnay, and St George via *Agrobacterium* have been initiated, and we are currently acclimating the first group of Chardonnay and Thompson Seedless edited plants to greenhouse conditions. Sequencing of

mutations has shown that high editing efficiency occurred with one the guides tested; with 10 CH and 8 TS lines, out of 11 lines each, containing indels at the target site (1 CH and 3 TS had at least one wild type (WT) allele). Phenotyping of these plants will be performed in collaboration with the McElrone lab.

### Salt Testing

Nina conducted 234 tests of chloride exclusion on 200 different genotypes. The main focus was on the 18-113 GRN-3 x *acerifolia* 9018 population where 143 genotypes were tested and chloride exclusion fell almost uniformly into the 4 exclusion categories with 32, 37, 38 and 36 in categories 1-4 respectively. The only other population tested with category 4 (most excluding) was also based on *acerifolia* 9018. Additionally, tests showed *doaniana* 9024, *doaniana* 9026, *girdiana* -8 and *treleasei* NM11-082 to be high excluders and promising parents (Figure 3). Nina's outdoor salt trial has grown significantly over the last year (Figure 4) and treatment with salty irrigation water started early in May of this year. One of the main goals of this trial is to determine whether chloride excluders prevent salt from getting into the fruit.

**Chloride exclusion, germplasm and mapping population screening:** We are using 75mM (about 12% sea water) salt concentrations to test germplasm previously identified as salt tolerant at 25-50 mM concentrations. We hope this more severe test will identify the most useful parents for crosses. There are currently 494 genotypes in testing; 162 for HarmAC, 157 for ring and 175 for salt. Of these, 71 and 101 genotypes have been inoculated for HarmAC and ring respectively. In salt tested, 149 genotypes have completed testing and await chloride analysis. All other remainders are in various stages of preparation.

Chris Chen is close to finishing his PhD. Recent progress in rootstock chloride tolerance has focused on establishing a mapping population for screening; following methods established throughout the past 11 months as standard protocol for greenhouse studies. This methodology requires use of special medium to prevent dispersion in response to high chloride concentrations in the soil, allow for sufficient aeration and drainage, and retain cations which compete for binding sites with chloride. Applied to the soil is a complete nutrient solution amended with [NaCl] = 75mM; a concentration which invokes chloride toxicity symptoms within a short time frame without causing plant death. To discover a source of natural tolerance to chloride toxicity we are testing wild-type grapevines from the arid regions of the southwestern United States. In total we have tested 60 individuals using our screening protocol for each of two propagation methods, herbaceous propagules and hardwood-dormant propagules, to compare effects of chloride uptake between the two methods of propagation. Both root and shoot chloride concentrations were quantified.

An individual with excellent chloride exclusion potential, *V. acerifolia* 9018 (same as *longii* 9018), was identified as having the lowest chloride accumulation in leaves following the NaCl application period (Fig. 3); consistent with previous reporting. A cross of GRN3 (susceptible) x *V. acerifolia* 9018 was made to determine heritability of salt-tolerant phenotypes in this salt-excluding species. A total of 238 F1 seedling plants have been established in the field. All have been DNA screened and are true to type. Chloride screening was completed for 223 F1 seedling plants including both parents and two control plants. DNA was extracted for the entire population and markers from chromosomes 8 and 11 were tested on a subset of 8 samples for polymorphism. These two chromosomes were selected based on the previous published information that Cl- and Na- exclusion are two independent loci mapped on chr8 and 11, respectively. A total of 20 polymorphic markers that provide coverage for both chromosomes were added to the entire data set. Mapping and QTL analysis was performed with JoinMap and MapQTL software. Interval mapping analysis identified a major QTL on chromosome 8 that explained up to 26% phenotypic variation at LOD 14.54. This population is also expected to yield nematode resistant progeny and initial data suggest segregation within the population.

GRN3 x *V. acerifolia* 9018, has shown that variance in the population's expression of salt-tolerance is continuous and strongly suggests multigenic control of this trait; while root chloride accumulation is not

as closely related to accession as leaf chloride accumulation, there are still significant differences within this single population.

For consolidation, we have also shown (in a separate test) that these uptake rates vary by genotype, only after being exposed to high external NaCl concentrations; with a  $R^2 = 0.54$  in salted accessions of Resseguier 2 and *V. berlandieri* and  $R^2 = 0.11$  in control groups; indicating some change in chloride uptake rates by accession once exposed to external NaCl. Current and future trials include a variable NaCl concentration trial using common rootstocks and our best and worst performers from previous trials. NaCl concentrations will range from 25mM to 100mM; enough to kill young grapevines. This trial will include physiological measurements while the plant is still alive to ascertain the effect of varying degrees of NaCl exposure on photosynthetic capacity and growth rates. Additionally, we will test the best and worst performers from previous trials in a grafted experiment using Cabernet Sauvignon and Chardonnay.

**Developing a consensus DNA fingerprint database of the Walker lab southwestern US germplasm** for diversity and population genetic studies: I have amassed a very large collection of grape germplasm from the southern US – particularly the southwestern States (over 700 accessions). This collection is a very valuable resource for the rootstock breeding program. We have developed SSR fingerprint database with 17 - 20 markers for a set of unique 1047 accessions to carry out population diversity studies that would help us to identify germplasm from different genetic groups. The collection also serves as the foundation for a NSF project to sequence many of these species and selections that is now underway. The sequencing and testing of these individuals for salt tolerance and PD resistance continues.

**Transcriptomic analysis of grapevine infected by red leaf viruses:** Prof. Nihal Buzkan was on a 1.5 yr-long sabbatical with me and returned to Turkey in September. She was working on this virus tolerance project and continues the analysis with colleagues in Italy with a manuscript expected by the end of this year. Experiments were carried out with Cabernet franc infected with red leaf viruses; leafroll (GLRaV-1) and rugose wood viruses (GVA) and two rootstocks Freedom (highly sensitive to red leaf viruses) and St. George (tolerant to red leaf virus disease) in field and *in vitro* conditions. Virus strains were LR131 for GLRaV-1 and LR132 for GVA. She also overlapped with Dr. Zhenhua Cui who is working on various aspects of the same project. Please see the June 2019 progress report of complete details on Nihal's research. Zhenhua has been more focused on the cause of graft union collapse due to Red Leaf and Vitiviruses.

**Inheritance of the GFLV Tolerance Trait in a 101-14 x Trayshed Population:** Dr. Erin Galarneau is working with recent PhD graduate Dr. Andy Viet Nguyen to screen phytohormone biosynthesis genes and phytohormone concentrations of nine genotypes (plus controls) with different flower to fruit set ratios in reaction to fan leaf virus. **Tolerant:** 07107-005, 07107-043, 07107-008, **Moderate:** 07107-125, 07107-007, 07107-135, **Susceptible:** 07107-077, 07107-108, 07107-102, **Controls:** 101-14, St. George, O39-16, Cab Sauv were collected to determine the mechanism of the tolerance trait. Collections of buds were collected before bud break, along with inflorescences (2 collection times), flowers (pre-, 10% capfall, and 90-100% cap fall), and young berries (setting, peppercorn, and pea sized) have occurred March-June 2020 and are almost completed March-May 2021. Analyses of phenolics, sugar concentrations, and phytohormones are ongoing. Currently phenolics and sugars significantly vary only according to phenological stage. Phytohormone expression is currently being processed.

#### Dr. Nguyen's dissertation abstract

One of the most destructive grapevine viruses is grapevine fanleaf virus (GFLV), the causal agent of fanleaf degeneration. This virus is vectored from root-to-root by the dagger nematode (*Xiphinema index*) and can result in crop losses of up to 80% by greatly reducing fruit set and causing formation of 'shot berries,' small, seedless berries that do not mature. Currently, fanleaf degeneration is

controlled by grafting vines onto the rootstock O39-16, which suppresses the expression of fanleaf degeneration symptoms in the scion. However, the *Vitis vinifera* parentage of O39-16 raises concerns about the rootstock's long-term susceptibility to grape phylloxera and other pests and diseases. Additionally, O39-16 is susceptible to root-knot nematodes and induces high vigor to scions grafted on it. Due to these reasons, breeding efforts to produce alternative fanleaf degeneration rootstocks have continued.

In 2007, 101-14 Mgt. was crossed with *Muscadinia rotundifolia* cv. Trayshed and the resulting progeny have been growing at the University of California, Davis. *Muscadinia rotundifolia* is the source of rootstock-induced tolerance observed in O39-16, and 101-14 Mgt. (*V. riparia x V. rupestris*) is a popular commercial rootstock commonly chosen for its ease of propagation, moderate nematode and phylloxera resistance, and the ability to control vigor to scions grafted on it. We quantified GFLV resistance and fanleaf degeneration tolerance in the progeny from this cross and studied the inheritance of these traits. Both traits segregated in the 101-14 x Trayshed population as quantitative traits controlled by multiple genes.

Additionally, we developed a novel method that utilized digital imaging to obtain fruit set ratios for individual grape clusters as a means to quantify fanleaf tolerance in vines. Utilizing this method, we identified six progeny from this hybrid population (07107-012, 07107-043, 07107-091, 07107-112, 07107-135, and 07107-148) that were able to control fanleaf degeneration as well as O39-16 and exhibited desirable viticultural traits. This study is the first to have extensively quantified fruit set as a method to determine the degree of fanleaf degeneration tolerance induced by a rootstock to GFLV-infected vines.

In addition to exploring the 101-14 x Trayshed breeding population, we also studied five recentlyreleased rootstocks with broad and durable nematode resistance. Although these five rootstocks (GRN-1, GRN-2, GRN-3, GRN-4, and GRN-5) possess resistance to *X. index*, their ability to induce fanleaf tolerance (such as the case in O39-16) is unknown and the overall performance of these rootstocks on a GFLV-infested site has not been thoroughly evaluated. We are preparing the first evaluation of these new rootstocks on a fanleaf site in Lodi. After ten years of growth, all five GRN rootstocks performed comparably to O39-16. The low GFLV infection rate in vines grafted on GRN-1 shows the potential for GRN-1 to become a viable alternative to O39-16. Although further studies are needed to evaluate the long-term performance of these rootstocks, these initial results are promising.

**Performance of GRN rootstocks on a Gallo fanleaf site in Acampo (Lodi):** Although the GRN rootstocks possess resistance to *X. index*, their ability to induce fanleaf tolerance (such as the case in O39-16) is unknown and the overall performance of these rootstocks on a GFLV-infested site has not been thoroughly evaluated. After 10 years of growth on a fanleaf site in the San Joaquin Valley, all 5 GRN rootstocks performed similarly to O39-16, the only commercially available fanleaf tolerant rootstock (Table 7 and Table 8). The low GFLV infection rate in vines grafted on GRN-1 shows the potential for GRN-1 to become a viable alternative to O39-16 (Table 9). These results are encouraging, but additional studies to observe the long-term performance of these rootstocks are required to fully assess their suitability on fanleaf sites. Furthermore, these rootstocks should be tested on more sites with varying degrees of pest and disease pressure to gain greater confidence in the rootstocks' ability to act as a control measure against *X. index* and GFLV. This trial has greatly benefited from maintenance and data collection by E&J Gallo winery.

**Propagation of hard to root rootstocks:** James Shoulders (MS student and current Production Manager at FPS) has been working on detailing propagation for hard to root materials such as O39-16, GRN1, and 420A. Needed rootstocks in the future will come from non-traditional species and will be harder to root.

That said, I do not have trouble rooting or grafting O39-16, GRN1 or GRN5. Some key points: The mothervines need to be mature (young vines do not propagate as easily as older vines); don't over grow (excess water and N fertilizer) late in the season. The cuttings root and graft more easily when mature (lignified not green). This can be problematic because there are southern species in their parentage that grow longer into the Fall and shed leaves later. For potted plants, pre-callusing for extended periods works well as does callusing after grafting and longer growth periods in the greenhouse.

Caution: keep moist not wet; prepare callusing /greenhouse areas where you can treat these pre- and postgrafted plants separately from standard rootstocks – more shade, less watering, high humidity, good air circulation, active fungicide program. Ensure that your production crews don't treat these plants in the same way as they treat standard rootstocks.

# Presentations to Industry Groups / Grower Advice

Presentations June 2020 to June 2021

- Walker, M.A. Sonoma Vit Tech Group. Grape breeding. June 18, 2020.
- Walker, M.A. Farm Call to Rodney Strong regarding ring nematode numbers and resistance. June 24
- Walker, M.A. Farm Call with Crop Care PD issues with Cain Cellars and Staglin, also checked on GRN plot with Bob Steinhauer, July 7.
- Walker, M.A. Laffort USA seminar. Grape Breeding talk, July 14.
- Walker, M.A. Interview for Dan Baron Podcast, July17.
- Walker, M.A. Farm call with Ashley Anderson, Spring Mtn PD and new varieties, August 8.
- Walker, M.A. Interview with Agostino Petron, PD breeding, Aug 11.
- Walker, M.A. Farm call with Bob Gallagher (Medlock Ames), winter cold damage to O39-16 latte planting, Aug 14
- Walker, M.A. PD wine tasting with Silverado Winery organized through Crop Care, Aug 21
- Walker, M.A. "Office hours with Dave and Anita" talk on PD PM resistant wine grapes. Aug 25.
- Walker, M.A. Talk for Expo 2020 ... Can viticulture be sustainable without disease resistant varieties, Oct. 14.
- Walker, M.A. Farm call with Tyler Klick, stuck ripening in the Petaluma Gap (cold or red leaf). Oct. 27
- Walker, M.A. Fire damage in vineyards. North Coast Fire meeting, Dec. 15
- Walker, M.A. 2020. New PD resistant winegrapes from UCD interview with Elaine Corn, Ag Press, UC Davis, Jan. 8, 2020
- Walker, M.A. 2020. Breeding drought resistant rootstocks and update on the GRN rootstocks. Daniel Roberts Client, Martinelli Winery, Santa Rosa, CA, Jan. 10, 2010.
- Walker, M.A. 2020. Grapevines on Freedom rootstock: sudden vine decline associated with graft incompatibility. Grape Day UC Davis, Jan. 21, 2020.
- Walker, M.A. Unified PD resistant Wine tasting with virtual tasting, and lecture, Jan. 28
- Walker, M.A. Healthy vineyard soils symposium -- Nematode control. Feb. 10. 20
- Walker, M.A. USDA Parlier talk on "30 yrs. of grape breeding at UC Davis", Virtual talk, Fe. 16
- Walker, M.A. On the Road Seminars from UC Davis, Kern County. Grape rootstocks for the San Joaquin Valley. Mar. 10
- Walker, M.A. On the Road Seminars from UC Davis, Kern County. PD resistant wine grapes are ready to plant. Mar. 16.
- Walker, M.A. Wine Executive Program: presentation on vineyard challenges. Mar 23.
- Walker, M.A. Recent Advances Viticulture and Enology. Presentation, 30+ years of grape breeding and I'm just getting started! Apr. 8
- Walker, M.A. RMI Forum Walker/Waterhouse session. Breeding legacy, May 12.
- Walker, M.A. interview with Agostino Petroni PD breeding article, May 27
- Walker, M.A. Mitigating drought and drought tolerant rootstocks, Mendocino Growers and Water Board, June 3.

Walker, M.A, Phylloxera consult with Sandy Henson, June 7.

- Walker, M.A. Farm call with Walsh Vnyd Mang (Ben Leachman) concerns regarding GRN1 vigor. June 14.
- Walker, M.A. Farm call with Lynn Wunderlich to Zielmsky Ranch, Somerset, training establishment issues, June 17, 2021.

#### Posters/Abstracts at Scientific Meetings

- Huerta-Acosta, K., S. Riaz, O. Franco-Mora and M.A. Walker. 2019. Search for new sources of resistance to Pierce's Disease: characterization of the PD resistant accession b46-43. ASEV National Conference, Napa, CA, June 19, 2019.
- Summaira Riaz, Alan Tenscher, Rong Hu and M Andrew Walker. 2019. Characterization of Pierce's disease resistance in b41-13, an accession collected from Tamaulipas, Mexico. ASEV National Conference, Napa, CA, June 19, 2019.
- Ines Hugalde, Summaira Riaz, Cecilia B. Agüero, Marcos Paolinelli, Nina Romero, Andy V. Nguyen, Hernán Vila, Sebastian Gomez Talquenca, M. Andrew Walker. 2019. Genetic determination of vegetative vigor in a Ramsey x Riparia GM population. ASEV National Conference, Napa, CA, June 19, 2019.
- Cecilia B. Agüero, Marco Rocha-Figueroa, and M. Andrew Walker. 2019. Effect of agar on growth of roots of six grapevine rootstocks. ASEV National Conference, Napa, CA, June 19, 2019.
- Summaira Riaz, Cecilia Agüero, Rong Hu and M Andrew Walker. 2019. Comparative sequence analysis of the PD resistance locus *PdR1* in two resistant accessions –b43-17 and b40-14. ASEV National Conference, Napa, CA, June 19, 2019.
- Christopher Cody Lee Chen, Nina Romero, and M A Walker. 2019. Rapid screening for salt-stress tolerance through chloride-ion accumulation in leaves of wild *Vitis* spp. rootstocks. ASEV National Conference, Napa, CA, June 19, 2019.
- Laila Fayyaz, Alan Tenscher, Huma Qazi, M. Andrew Walker. 2019. Powdery mildew resistance varies in western US *Vitis* accessions. ASEV National Conference, Napa, CA, June 19, 2019.

#### **Publications**

- Cui, Z.-H., C.B. Agüero, Q.C. Wang and M.A. Walker. 2019. Validation of micrografting to identify incompatible interactions of rootstocks with virus-infected scions of Cabernet franc. Australian Journal of Grape and Wine Research 25: 268-275 doi: 10.1111/ajgw.12385
- Riaz, S., D. Pap, J. Uretsky, V. Laucou, J-M. Boursiquot, L. Kocsis and M.A. Walker. 2019. Genetic diversity and parentage analysis of grape rootstocks. Theoretical and Applied Genetics DOI: 10.1007/s00122-019-03320-5
- Heinitz, C. C., Uretsky, J., Peterson, J. C. D., Huerta-Acosta, K. G., and Walker, M. A. 2019. Crop wild relatives of grape (*Vitis vinifera* L.) throughout North America. North American Crop Wild Relatives, Volume 2 (pp. 329-351): Springer.
- Cantu, D. and M.A. (Eds.) 2019. The Grape Genome. Volume within Compendium of Plant Genomes. Springer.
- Dodson-Peterson, J.C., R. Duncan, D. Hirschfelt, C. Ingels, G. McGourty, R. Smith, E. Weber, J. Wolpert, M. Anderson, J. Benz and M.A. Walker. 2019. Grape rootstock breeding and their performance based on the Wolpert trials in California. In, Cantu D and Walker MA (Eds) The Grape Genome. Compendium of Plant Genomes. Springer. pp. 301 318.
- Walker MA, Heinitz CC, Riaz S and Uretsky J. 2019. Grape taxonomy and germplasm. In, Cantu D and Walker MA (Eds). Compendium of Plant Genomes. Springer. pp. 25 38.
- Vondras, A.M., Mino, A., Blanco-Ulate, B., Figuero-Balderas, R., Penn, M.A., Zhou, Y., Seymour, D., Ye, Z., Liang, D., Espinoza, L.K. Anderson, M.M., Walker, M.A., Gaut, B., and Cantu, D. 2019. The genomic diversification of grapevine clones. BMC Genomics 20:972.

- Cuneo, I, F. Barrios-Masias, T. Knipfer, J. Uretsky, C. Reyes, P. Lenain, C. Brodersen, A. Walker and A. McElrone. 2020. Differences in grapevine rootstock sensitivity and recovery from drought are linked to fine root cortical lacunae and root tip function. New Phytologist doi: 10.1111/nph.16542
- Amaral, BD, AP Viana, EA Santos, RM Ribeiro, FA da Silva, M Ambro'sio and MA Walker. 2020. Prospecting for resistance of interspecific hybrids of *Vitis* spp. to *Plasmopara viticola*. Euphytica 216:68
- Heinitz, CC, S Riaz, AC Tenscher, N Romero and MA Walker. 2020. Survey of chloride exclusion in grape germplasm from the southwestern United States and Mexico. Crop Science 60:1946-1956 (doi: 10.2135/cropsci2019.05.0348)
- Riaz, S., CM Menéndez, AC Tenscher, D. Pap and MA Walker. 2020. Genetic mapping and survey of powdery mildew resistance in the wild Central Asian ancestor of cultivated grapevines in Central Asia. Horticultural Research 7:104
- Adam R. Zeilinger, A.R., C. Wallis, D. Beal, A. Sicard, A. Walker, R.P.P. Almeida. 2020. Non-linear dynamics of vector transmission of a plant pathogen: a test of theory and application to disease management. Ecosphere (In Press).
- Merrill NK, I García de Cortázar-Atauri, AK Parker, MA Walker and EM Wolkovich. 2020. Exploring grapevine phenology and high temperatures response under controlled conditions. Frontiers in Environmental Science, section Biogeochemical Dynamics. In Press
- Wallis, C.M., Zeilinger, A.R., Sicard, A., Beal, D.J., Walker, M.A. and Almeida, R.P.P. 2020. Impact of phenolic compounds on progression of *Xylella fastidiosa* infections in susceptible and PdR1-locus containing resistant grapevines. PLoS ONE 15: e0237545.
- Arancibia C, E Malovini,1 CB Agüero, F Buscema, R Alonso, MA Walker and LE Martinez. 2021. Evaluation of two phylloxera genotypes in Argentina on six *Vitis vinifera* cultivars and three rootstocks. American Journal of Enology and Viticulture 72: 94-100.
- Riaz S, AC Tenscher, CC Heinitz, KG Huerta-Acosta, M A Walker. 2021. Genetic analysis reveals an east-west divide within North American *Vitis* species that mirrors their resistance to Pierce's disease. PLOS ONE(In press).
- Inés Hugalde<sup>1,2</sup>, Cecilia B. Agüero<sup>2</sup>, Felipe H. Barrios-Masias<sup>2,3</sup>, Nina Romero<sup>2</sup>, Andy V. Nguyen<sup>2</sup>, Summaira Riaz<sup>2</sup>, Patricia Piccoli<sup>5</sup>, Andrew J. McElrone<sup>2,4</sup>, M. Andrew Walker<sup>2</sup>, Hernán Vila. 2021. Modeling vegetative vigor in grapevine: Unraveling underlying mechanisms <sup>1</sup>Heliyon (In Press)
- Fayyaz, L., A. Tenscher, A. Viet Nguyen, H. Qazi and M.A. Walker. 2021. *Vitis* species from the southwestern United States vary in their susceptibility to powdery mildew. Plant Disease (In Press)

Table 1. 2021 UCD Grafted rootstock trial selections with nematode, salt and rootability scores. Nematode resistance is measured on a 1 to 4 scale with 1 highly susceptible and 4 resistant with no nematode damage. Rootability is reported from typical duration (17 days) for hardwood cuttings coming out of callus and an added 6-7 weeks out of sleeves. Scale is 0 with no usable plants and 4 excellent shoots and roots. The added healing time greatly improves rooting ability.

			Avg	Avg	Avg	Avg	Avg Sleeves
	Female		HarmAC	Ring	Chloride exclusion	HW Deet	Root-
Genotype	parent	Male parent	resist- ance	resist- ance	rating	Root- ability	ability - Typical
07107-012	101-14Mgt	Trayshed	3.0	3.5	- wing	1.3	4.0
07107-091	101-14Mgt	Trayshed	4.0	3.0		1.0	3.3
101-14Mgt			2.8	1.3	2.0	2.3	4.0
1103P			1.0	1.5	1.7	3.0	
16141-003	doaniana 9026	GRN-4	4.0		3.0	1.5	4.0
10111 005	7020	acerifolia	1.0		5.0	1.5	1.0
18113-022	GRN-3	9018	4.0		4.0	3.0	3.5
18113-047	GRN-3	<i>acerifolia</i> 9018	4.0		4.0	3.0	4.0
18113-067	GRN-3	<i>acerifolia</i> 9018	4.0		4.0	3.7	3.5
2011-188-16	T6-42	St. George	3.8	4.0	1.5	1.2	3.8
2011-188-6	T6-42	St. George	3.5	4.0	2.0	2.6	3.6
2012-108-28	101-14Mgt	doaniana 9028	3.4	1.0	3.0	0.9	4.0
2014-137-038	Dog Ridge	<i>berlandieri</i> 9031				3.0	3.0
2014-145-004	Ramsey	girdiana NV11-116				2.5	3.7
acerifolia 9018			1.8	1.0	4.0	3.4	3.8

Table 2. Nematode resistance to combined Harmony A and C strains for 6 populations and a miscellaneous group of 58 different crosses tested in the reporting period. Resistance is measured on a 1 to 4 scale with 1 highly susceptible and 4 resistant with virtually no nematode damage.

		Ha	HarmAC Resistance			
						Cross
						ID
Cross ID	Cross	1	2	3	4	Total
07-107	101-14Mgt x Trayshed		4	2	16	22
16-063	5BB x b55-1	15	2			17
16-191	SC2 x GRN-4	4	3	2	1	10
17-028	101-14Mgt x acerifolia 9018	7	6	4	5	22
17-032	101-14Mgt x acerifolia 9035	1	1	3	6	11
18-113	GRN-3 x acerifolia 9018	40	92	8	60	200
58 Misc crosses		34	18	5	69	126
Resistance Total		101	126	24	157	408

		Scion growth			
Rootstock	Clean	Fanleaf Infected			
07107-012	Good	Good			
07107-043	Good	Fair			
07107-091	Good	Fair-Good			
07107-112	Fair	Poor			
07107-191	Very good	Poor			
07107-204	Good	Good			
07107-208	Good	Poor			
07107-229	Very good	Fair-Good			
07107-241	Good	Poor-Fair			
GRN-1	Very good	Very Good			
GRN-3	Excellent	Fair-Good			
O39-16	Good	Good			
St. George	Very good	Very Good			

Table 3. Scion growth for clean and fanleaf infected Cabernet Sauvignon scions on nine 07-107 selections and 4 commercial stocks about 10 weeks after putting them into callus conditions.

Table 4. Stocks selected for the 2021 UCD Fanleaf trial along with nematode resistance and rooting ability over three time points. Nematode resistance is measured on a 1 to 4 scale with 1 highly susceptible and 4 resistant with no nematode damage. Rootability is reported from typical duration (17 days) for hardwood cuttings coming out of callus; 6-7 weeks in sleeves; and 10 weeks for sleeves-slow. Scale is 0 with no usable plants and 4 excellent shoots and roots.

	Avg HarmAC	Avg Ring	Avg Xi	Avg HW	Avg Sleeves Rootability	Avg Sleeves Rootability
Genotype	resistance	resistance	resistance	Rootability	- Typical	- Slow
07107-012	3.0	3.5	2.0	1.3	4.0	4.0
07107-091	4.0	3.0	4.0	1.0	3.3	4.0
07107-204	4.0	3.0	3.0	1.0	3.0	3.0
07107-229	3.0	3.5	3.0	1.0	3.0	2.0
07107-241	4.0	4.0	3.0	1.0	4.5	3.0
O39-16	3.2	3.9	3.0	0.7	2.7	1.7
St. George	1.4	1.6	1.4	1.8	3.7	4.0
GRN-1	4.0	3.9	3.0	0.8	2.4	1.5
GRN-3	4.0	2.5	3.7	2.0	3.0	

Table 5. Salt tolerant species being used as rootstocks for Chardonnay vines for a field trial in a salty and
high boron vineyard site in Solano County, CA. Salt resistance ratings use a scale with a score of 1
accumulating high levels of leaf chloride while 4 represents very low levels of chloride.

	Mean Leaf Chloride	Average Chloride	
Genotype	(ppm)	exclusion rating	Times Tested
doaniana 9024	43	3.5	4
doaniana 9026	41	4.0	6
doaniana 9042	65	3.0	1
girdiana -8	63	3.5	4
acerifolia 9018	38	4.0	21
acerifolia 9035	71	3.5	6

Table 6. Potentially drought tolerant selections tested for rootability and downward growth of roots. Rootability is reported from typical callusing duration (17 days) for hardwood cuttings coming out of callus, 6-7 weeks for sleeves and 12 weeks for pots. Scale is 0 with no usable plants and 4 excellent shoots and roots.

Test_Genotype	Female_parent	Male_parent	HW Rootability	Sleeves Rootability	Pot Rootability
2014-137-038	Dog Ridge	berlandieri 9031	4	3	4
2014-145-004	Ramsey	NV11-116	3	3	4
2014-150-010	Ramsey	UT12-078	4	2	4
15157-016	Ramsey	ANU77	3	3	3
2014-137-012	Dog Ridge	berlandieri 9031	4	3	3
2014-145-003	Ramsey	NV11-116	4	2	3
2014-145-013	Ramsey	NV11-116	4	2	3
15157-003	Ramsey	ANU77	3	3	
15157-004	Ramsey	ANU77	2		
15157-043	Ramsey	ANU77	4	2	
2014-135-001	Dog Ridge	ANU77	3		
2014-135-007	Dog Ridge	ANU77	3	2	
2014-135-013	Dog Ridge	ANU77	2	0	
2014-135-031	Dog Ridge	ANU77	4	2	
2014-136-013	Dog Ridge	2011-175-15	2		
2014-137-001	Dog Ridge	berlandieri 9031	2		
2014-137-026	Dog Ridge	berlandieri 9031	4	3	
2014-137-027	Dog Ridge	berlandieri 9031	4		
2014-137-037	Dog Ridge	berlandieri 9031	2		
2014-139-002	Dog Ridge	NV12-049	4		
2014-139-009	Dog Ridge	NV12-049	4		
2014-145-012	Ramsey	NV11-116	2		
2014-145-020	Ramsey	NV11-116	4	0	
2014-150-011	Ramsey	UT12-078	4	2	

Table 7. Pruning weight of *V. vinifera* cv. Malbec grown on thirteen rootstocks at Acampo, CA for three selected years. Pruning weight data for a given year were collected in February of the following year. Letters indicate significant differences according to Tukey's test (alpha = 0.05).

	Pruning Weight (kg/vine)					
Rootstock	2014	2018	2020			
101-14	0.855 de	1.141 cd	0.666 bc			
1103P	1.022 bcd	1.343 abc	0.848 ab			
3309C	0.922 cde	1.323 abc	0.858 ab			
GRN-1	1.087 abc	1.327 abc	0.953 ab			
GRN-2	1.239 a	1.563 ab	1.133 a			

GRN-3	1.079 abc	1.303 abc	0.836 ab
GRN-4	1.151 ab	1.673 a	1.147 a
GRN-5	1.077 bcd	1.622 a	0.988 ab
Harmony	0.931 cde	1.191 bcd	0.896 ab
O39-16	0.935 cde	1.683 a	1.097 a
RS-3	0.809 de	1.052 cd	0.748 abc
RS-9	0.765 e	0.855 d	0.366 c
St. George	0.954 bcde	1.167 bcd	0.654 bc

Table 8. Fruit yields of *V. vinifera* cv. Malbec grown on thirteen rootstocks at Acampo, CA for three selected years. Letters indicate significant differences according to Tukey's test (alpha = 0.05).

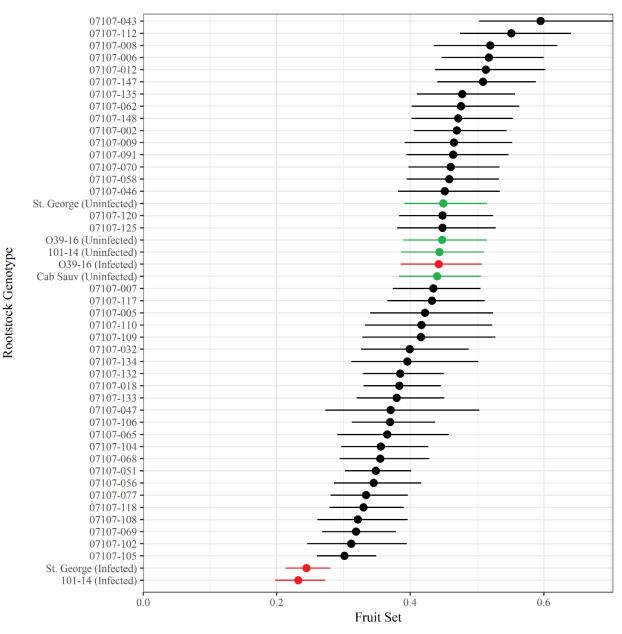
	Fruit Yield (kg/vine)				
Rootstock	2014	2018	2020		
101-14	16.1 a	10.0 bc	12.5 cde		
1103P	16.6 a	11.7 b	14.1 bcd		
3309C	17.2 a	12.9 ab	16.8 ab		
GRN-1	16.0 a	15.6 a	20.4 a		
GRN-2	16.4 a	12.7 ab	17.8 ab		
GRN-3	18.0 a	13.0 ab	15.1 bc		
GRN-4	14.7 ab	11.7 b	15.4 bc		
GRN-5	14.7 ab	11.8 b	14.5 bcd		
Harmony	16.4 a	12.1 b	13.9 bcd		
O39-16	16.3 a	12.4 b	17.3 ab		
RS-3	14.5 ab	8.6 cd	9.5 ef		
RS-9	11.2 b	6.7 d	7.5 f		
St. George	15.5 ab	8.2 cd	10.9 def		

	Proportion of Vines					
		Marginally	Moderately			
Rootstock	Negative	Infected	Infected	Highly Infected		
1103P	0.35	0.13	0.17	0.35		
GRN-1	0.92	0	0	0.08		
GRN-2	0.80	0.12	0	0.08		
GRN-3	0.72	0.08	0.08	0.12		
GRN-4	0.60	0	0.20	0.20		
GRN-5	0.58	0.13	0.17	0.13		
O39-16	0.96	0	0	0.04		
St. George	0.35	0.13	0.16	0.35		

Table 9. ELISA results of *V. vinifera* cv. Malbec grown on GRN-1, GRN-2, GRN-3, GRN-4, GRN-5, 1103P, O39-16, and St. George at Acampo, CA after the 2020 growing season.



Figure 1. Clean (blue strip labels) and fanleaf virus infected (red strip labels) Cabernet Sauvignon grafted on two VM stocks 07107-012 left side and 07107-112 right side showing relative scion performance as detailed in Table 3.



**Figure 2.** Fruit set ratios of *V. vinifera* cv. Cabernet Sauvignon grafted on rootstock genotypes from the 07107 (101-14 x *M. rotundifolia* 'Trayshed') population. Unless otherwise indicated, all vines are infected with fanleaf degeneration. Values represent mean fruit set ratio  $\pm$  95% confidence intervals. Infected controls are represented in red and uninfected controls are represented in green.

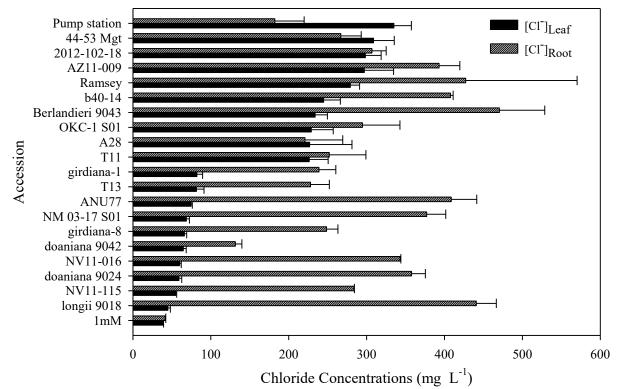


Figure 3. Chloride concentrations compared in leaves and roots of resistant breeding selections



Figure 4. Outdoors, large pot trial at UCD. On the left is 5/26/2020 and right is 5/27/2021 showing plant development. NaCl at 50 uM has been applied since 5/1/2021.