## Genetic dissection of Natural Dry-On-Vine trait in grapevine

#### Jonathan Fresnedo Ramírez

Department of Horticulture and Crop Science
July 2018

XII International Conference On Grapevine Breeding And Genetics



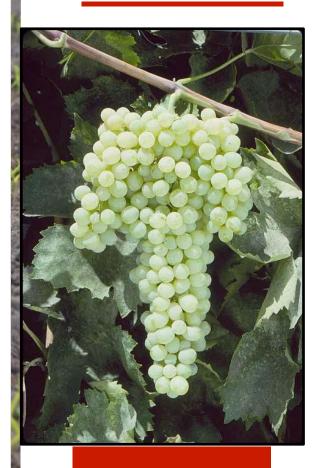


#### Raisins, a little of context

- Dried grapes
- Industry of more than 100 years in California
  - Multiple uses: raw, cooking, baking, brewing
- Trend to mechanization
  - Labor up to 35% of production costs
- Specialized germplasm



#### **Germplasm: traditional**



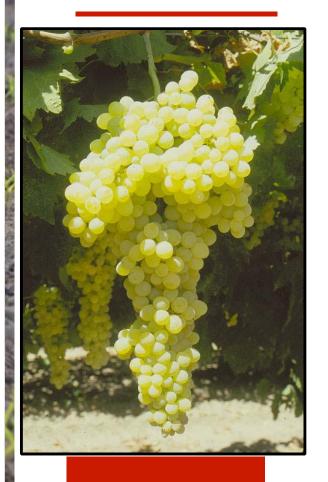




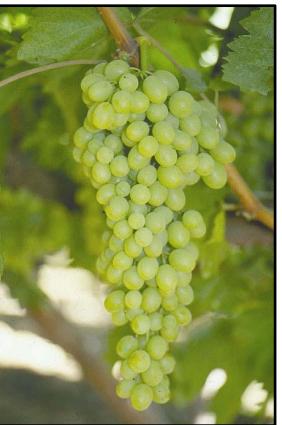
Fiesta

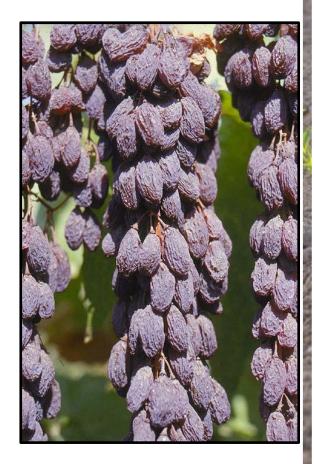
**CFAES** 

#### **Germplasm: DOV**









CFAES

**DOVine** 

Selma Pete

#### Germplasm: Natural Dry-On-Vine (NDOV)





*Sunpreme* (B82-43)

#### **Traditional vs NDOV**





**CFAES** 

#### **How to breed for NDOV raisins?**

- Sunpreme solely source at the moment
- VitisGen
- Mapping of the trait
  - C87-41 × Sunpreme, using presence/absence = No hits!
  - A95-27 x Sunpreme, using % dry at three date = No hits!



#### Using a different approach: candidate gene

- Did we have a well constructed linkage map?
- Are we phenotyping the correct trait?
- Fruit drying: a reason or a consequence?
- Thinking in Vavilov: trait parallel series
  - Which other fruits dry on a vine? → wild tomatoes
- Cuticular Water Permeability 1 (CWP1) gene (Hovav, 2007)
  - cuticle microcracking in fruits of primitive green-fruited wild tomato species, Solanum habrochaites, S. cheesmaniae and S. pimpinellifolium



#### Is Cwp1 present in grapevine?

tblastn query of S. habrochaites Cpw1 on PN40024

**BLAST Results** 





### Collaboration with the USDA ARS San Joaquin Valley Agricultural Sciences Center

- Craig Ledbetter
  - Research Geneticist
  - Segregating populations (Tray-dry × NDOV) and phenotypic evaluation



- Steven Lee
  - Molecular Biologist
  - Gene cloning and transgenic preparation/evaluation



### Gene cloning and (simple) functional evaluation

- Generate primer sequences based on PN40024
- Sunpreme, Valley Perl and Y308-344 (NDOV + REN4)
- Transferring into tomato (ongoing)
  - Phenotypic evaluation on microfissure density



#### What about Cpw1 in other genotypes?

cov pid <b>1201</b>	
1 Cpw1_PN40024 100.0% 100.0%	GCAACAGCTGTAGAGCCGTGGCTGCTTTTACACTGAATTCTTTATTTCTTTTCT
2 Cpw1_CavSavi 100.0% 99.8%	GCACAGCTGTAGAGCCGTGGCTGC TTTTTACACTGAATTCTTTATTTCTTTCAGGTCA TTTCACTGATATCACTGAATATAACTAAACAAA GCACAGCTGTAGAGCCGTGGCTGC TTTTTTACACTGAATTCTTTATTTCTTTCAGGTCA TTCACTTCAC
3 Cpw1_VcineB9 90.8% 96.9%	GCAACAGCTGTAGAGCCGTGGCTGCTTTTACACTGAATTCTTTATTTCTTTCTTTCAGGTCAACCTTCAC
4 Cpw1_FlamSdl 100.0% 100.0%	GCAACAGC TGTAGAGCCGTGGC TGCTTTTACAC TGAATTCTTTATTTCTTTCAGGTCAACCTTCACTGGTATATGAATATAACTAAACAAA
5 Cpw1_Sultana 67.0% 28.2%	TCCACGCCMAAAAGGCCCGAAGCCCGGGACGAAGCTCTAACACAMMGTGAGAAAGGCCAACCCMCCCCMTGCCTMGAACACGCCAACCATGTCCC
6 Cpwl_ValPerl 84.0% 31.3%	GGGCGGCGAGATGG-GCCCGCTGGGACATGGTTGGCTTGGC
7 Cpw1_Sunprem 82.5% 30.3%	GGGCGGGCGAGATGG-GCIICGCCGGTGGGACATGGTTGGCIIIGIAGCAGAGAGGGGGGIIIGGGGAGATGTGCGAAATGTGCGAGAAGCCCCGAAGACCAAGAGAGAG
cov pid <b>1321</b>	
1 Cpw1 PN40024 100.0% 100.0%	
2 Cpw1_CavSavi 100.0% 99.8%	GTATTAAATTGCCTCTTCCCTTGTAGGTC
3 Cpw1_VcineB9 90.8% 96.9%	GTATTAAATT GCCTTCTTCTAGGTC
4 Cpw1_FlamSdl 100.0% 100.0%	GTATTAAATTGCCTCTTCTCCCTTGTAGGTC
5 Cpw1_Sultana 67.0% 28.2%	CA <mark>ACTTCATCCC</mark> T-CCCTCCCAATATATCACA-GCCTTCCCACCATGCTAATGGCTTTGTGGGCCTCAAGAACACAACATGAAAACACACTTGAAAATCATTAAAACCCA
6 Cpw1_ValPerl 84.0% 31.3%	AG <mark>A</mark> CCT <mark>AATTE</mark> TT- <mark>C</mark> GGTT <mark>C</mark> TT <b>G</b> G <mark>AG</mark> AGCAAGAAGAATCCCATGGAATTTGCAGAGGAAGTTATGAAGGAGGCAGATAAGTATAATGGGTTTAACTTGATAATAGCTGATCTTTGTTCCA
7 Cpw1_Sunprem 82.5% 30.3%	$ ext{AGA}$ CCT $ ext{AATTG}$ TT- $ ext{C}$ GGTT $ ext{C}$ TT $ ext{G}$ CAAGAAGAATCCCATGGAATTTGCAGAGGAAGTTATGAAGGAGGCAGATAAGTATAATGGGTTTAACTTGATAATAGCTGATCTTTGTTCCA
cov pid <b>1441</b>	
1 Cpw1 PN40024 100.0% 100.0%	. :
2 Cpw1 CavSavi 100.0% 99.8%	CAGA
3 Cpw1 VcineB9 90.8% 96.9%	CAGA
4 Cpw1 FlamSdl 100.0% 100.0%	CAGA
5 Cpw1 Sultana 67.0% 28.2%	AAAGAACAATACAAAACAAAAACTAAGGGGCATGGCTAACAATGAAAAATCAGACTTACCTGTTGTGATACTCATCTCTGTTGAGCAAGAGAAGAAGAGAGAGAGAGAGAGAGAGAGAG
6 Cpwl ValPerl 84.0% 31.3%	AAACTATGATCTATATAACCAACAGACCAAGAGGAAGCTAATGTTTCTGTTGTAGAGGGTTTCACCTGGTATTCATGTGTCTAAATGCAAGTTTGGACTCACCTTGGCCTAAGGCA <mark>C</mark> GAA
7 Cpw1 Sunprem 82.5% 30.3%	AAACTATGATCTATATAACCAACAGACCAAGAGAAGCTAATGTTTCTGTTGTAGAGGTTTCACCTGGTATTCATGTGCTGTCAAATGCAAGTTTGGACTCACCTTGGCCTAAGGCACCAACCA
cov pid <b>1561</b>	6
1 Cpw1_PN40024 100.0% 100.0%	GATTGAGAGGAAAATT <mark>C</mark> AAGGAAC <mark>TTC</mark> TAGGCAAATATGGTGAAAGTGA <mark>CATTCC</mark> TA <mark>—TAAAAGAGATGATTGAGAAAC</mark> TAATGAGAGATAAAG <mark>TTAAAGC</mark> TG——ACGAGAGTATGTTGC
2 Cpwl_CavSavi 100.0% 99.8%	GATTGAGAGGAAAATTCAAGGAACTTCTAGGCAAATATGGTGAAAGTGA <mark>CATTCC</mark> TA-TAAAAGAGATGATTGAGAAAC <mark>TAATGAGAGATAAAGTTAAAGC</mark> TGACGAGAGTATGTTGC
3 Cpw1_VcineB9 90.8% 96.9%	GA <mark>TT</mark> GAGAGGAAAATT <mark>C</mark> AAGGAAC <mark>TTC</mark> TAGGCAAATATGGTGAAAGTGACATT <mark>CC</mark> TA—TAAAAGAGATGATTGAGAAAC <mark>TAATGAGAGATACAGTTAAAGC</mark> TG—A <mark>C</mark> GAGAGTATGTTGC
4 Cpw1_FlamSdl 100.0% 100.0%	GATTGAGAGGAAAATTCAAGGAACTTCTAGGCAAATATGGTGAAAGTGACATTCCTA-TAAAAGAGATGATTGAGAAAC <mark>TAATGAGAGATAAAG</mark> TTAAAG <mark>CTGACGAGAGTATGTTGC</mark>
5 Cpw1_Sultana 67.0% 28.2%	GAGTGGGTGAGATTGCCCAAACAAAGGCCTGCTATGCCACATT
6 Cpwl_ValPerl 84.0% 31.3%	GACTAGGTCATAAT <mark>TTCAAAGAGCTCTT</mark> GGATAAATATGGTGAAGGTGAGATC <mark>CC</mark> CCACAGAGGGAGATGG <mark>TTGAGAAATTAATGA</mark> AGACACAA <mark>T</mark> CCAAAGACCGATGAAATCGTGCTGC GACTAGGTCATAATTTCAAAGAGCTCTTGGATAAATATGGTGAAGGTGAGATCCCCA-CAGAGGAGATGGTTGAGAAATTAATGAAGAACACAATCAAAGACGATGAAATCGTGCTGC
7 Cpw1_Sunprem 82.5% 30.3%	GACINAGETCATAATITICAAAGAGCIICTIIGGATAAATATGGTGAAGGTGAGATCCCCA-CAGAGGAGATGGTTGAGAATTAATGAAGACACAATCAAAGACGATGAAATCGIIGCIIGC
cov pid <b>1681</b>	7
1 Cpw1 PN40024 100.0% 100.0%	CTCACATTTGTGCTCTTGACTGGGAGTGTGATCAAAGTTCCATTTTCACTGACAGAGACAAAAATGGTATATCTTAGTGGTTTTGCAGTCTCTCTTCAAAAGAACTCACAAACATTG- CTCACATTTGTGCTCTTGACTGGGAGTGTGATCAAAGTTCCATTTTCACTGACACAGAGACAAAAATGGTATATCTTAGTGGTTTTTGCAGTCTCTTCAAAAGAACTCACAAACATTG-
2 Cpwl CavSavi 100.0% 99.8%	CECACATTTGTGCECTCTTGACTGGGAGTGTGATCAAAGTECCATTTCACTGACAGAGACAAAAATGGTATATCTTAGTGGTTTTGCAGTCTCTCTC
3 Cpw1_VcineB9 90.8% 96.9%	CTCACATTTGTGCTCTTGACTGGGAGTGTGATCAAAGTTCCATTTTCACTGACACAGAGACAAAAATGGTATATCTTAGTGGTTTTGCAGTCTCTCTTCAAAAGAACTCACAAACATTGA CTCACATTTGTGCTCTTGACTGGGAGTGTGATCAAAGTTCCATTTTCACTGACACAGAGACAAAAATGGTATATCTTAGTGGTTTTGCAGTCTCTTCAAAAGAACTCACAAACATTGA
4 Cpw1_FlamSdl 100.0% 100.0%	CTCACATTTGTGCTCTTGACTGGGAGTGTGATCAAAGTTCCATTTTCACTGACACAGAGACAAAAATGGTATATCTTAGTGGTTTTGCAGTCTCTCTTCAAAAGAACTCACAAACATTG—
5 Cpw1_Sultana 67.0% 28.2%	
6 Cpw1_ValPerl 84.0% 31.3%	CCTCCGCATCCTATCCTATCCCTCCCAGAGAGGGGAACACCCAGTTA
7 Cpw1_Sunprem 82.5% 30.3%	CECGCATCEARCE

#### **Expectations**

- Test the hypothesis whether or not VviCpw1 is responsible for NDOV trait
  - Provide a tool for the early identification of NDOV genotypes
  - Link NDOV with studies of cuticle characteristics (epidermis) in grape fruits (drying)
  - Collaborate in the selection of NDOV genotypes in a more comprehensive view
- Genotyping of available pedigree and derived germplasm from Sunpreme
- Design of transcriptomic and phenotypic evaluations with additional functional tests



Taken from Hovav *et al.* 2007, The Plant Journal, 52(4) 627-639



#### NDOV is a minute part but relevant



Agronomic

Sensory

**CFAES** 

# Thanks for your attention!



THE OHIO STATE UNIVERSITY

COLLEGE OF FOOD, AGRICULTURAL, AND ENVIRONMENTAL SCIENCES