Current advances in understanding grapevine virus diseases

Cool Climate Oenology & Viticulture Institute

Brock University

Outline

- How many viruses and which ones are important and why?
- Does symptoms of grape viruses indicate anything?
- How do grapevine viruses spread, what we know so far.
- What type of management options available.

List of CFIA Regulated Grapevine Infecting Viruses in Canada

Virus Name	Family	Genus	Directive*
Grapevine asteroid mosaic virus	*Tymoviridae*	*Marafivirus*	D-97-06, D-94-34
Grapevine Bulgarian latent virus (GBLV)	*Secoviridae*	*Nepovirus*	D-97-06, D-94-34
Grapevine chasselas latent agent	-	-	D-97-06, D-94-34
Grapevine (Hungarian) chrome mosaic virus (GCMV)	*Secoviridae*	*Nepovirus*	D-97-06, D-94-34
Grapevine corky bark virus	-	-	D-97-06, D-94-34
Grapevine enation agent	-	-	D-97-06, D-94-34
Grapevine vein mosaic or necrosis virus	-	-	D-97-06, D-94-34
Cherry leaf roll nepovirus (CLRV)	*Secoviridae*	*Nepovirus*	
Peach rosette mosaic nepovirus (PRMV)	*Secoviridae*	*Nepovirus*	D-97-06, D-94-34
Raspberry ringspot nepovirus (RRSV)	*Secoviridae*	*Nepovirus*	D-94-34
Strawberry latent ringspot nepovirus (SLRV)	*Secoviridae*	NA	D-94-34
Tomato black ring nepovirus (TBRV)	*Secoviridae*	*Nepovirus*	D-98-01, D-97-06, D-96-05, D-94-34
Tobacco ringspot nepovirus (TRSV)	*Secoviridae*	*Nepovirus*	D-97-06, D-94-34
Tomato ringspot nepovirus	*Secoviridae*	*Nepovirus*	D-97-06, D-94-34

(http://inspection.gc.ca/plants/plant-pests-invasive-species/pests/regulated-pests/eng/1363317115207/1363317187811)

Grapevine Viruses

More than 70 viruses world wide

- Viruses cause serious production problems affecting rooting ability, graft take, vine vigour and fruit quality.

Major viral diseases

Grapevine leafroll disease (GLRD)

Grapevine leafroll-associated virus-1,-3,-4 and -7: *Ampelovirus*

Grapevine leafroll-associatedvirus-2: *Closterovirus*

Grapevine red blotch virus (GRBD)

Grapevine red blotch virus: *Grablovirus*

Rugose wood (RW) complex

Grapevine rupestris stem pitting-associated virus (GRSPaV),
Grapevine virus A (GVA), *Grapevine virus B* (GVB),
Grapevine virus D (GVD).

Grapevine fanleaf degeneration complex

Grapevine fanleaf virus (GFLV)

Other viruses:

Grapevine Pinot Gris Virus (GPGV)

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Major Grapevine Viruses: Economic Importance

Leafroll Disease

GLRD in California

$29,902 - 226,405 USD / ha

(Ricketts et al. 2015. AJEV 66:138-147)

GLRD in New York (Finger Lakes)

$25,000 - $40,000 USD / ha

(Atallah et al. 2012. AJEV 63:73-79)

GLRD in Washington

Up to 77% revenue decline / ac

30% yield + 1.0 °Brix reduction

2% price reduction for each 0.1 °Brix below min

(T. Ball. 2014 YVCC, Rayapati's Lab, IAREC, Prosser)

Red-Blotch Disease

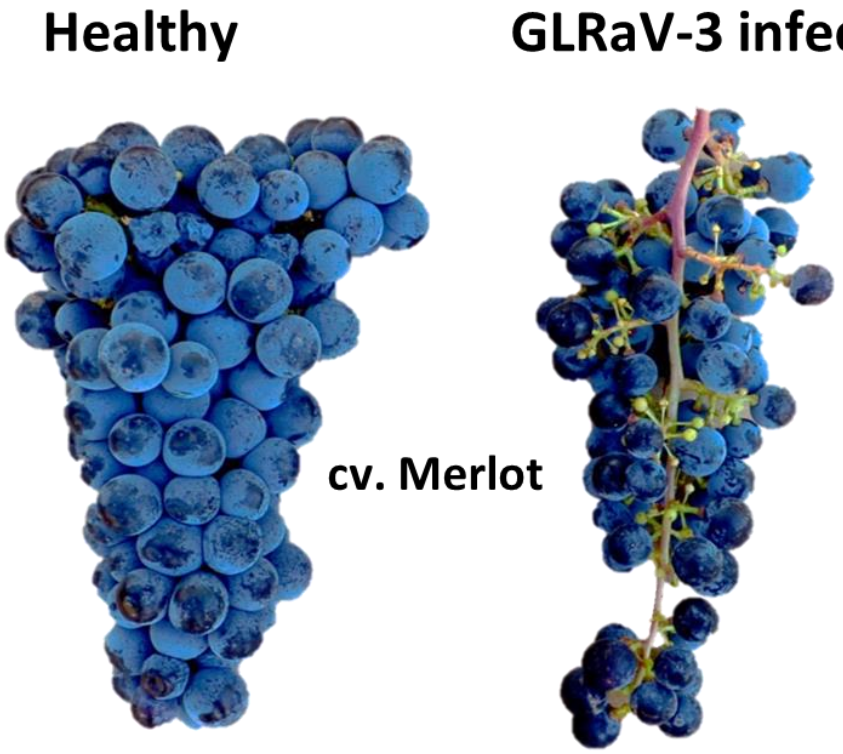
GRBD in California and WA

$2,213 to $68,548 USD / ha

(Ricketts et al. 2016. AJEV
doi: 10.5344/ajev.2016.16009)

No economic impact studies under cool-climate conditions

Leafroll Disease: Why it Matters



Vine

- Graft incompatibility
- Performance depend on variety, clone, rootstock and virus species/strain

Fruit

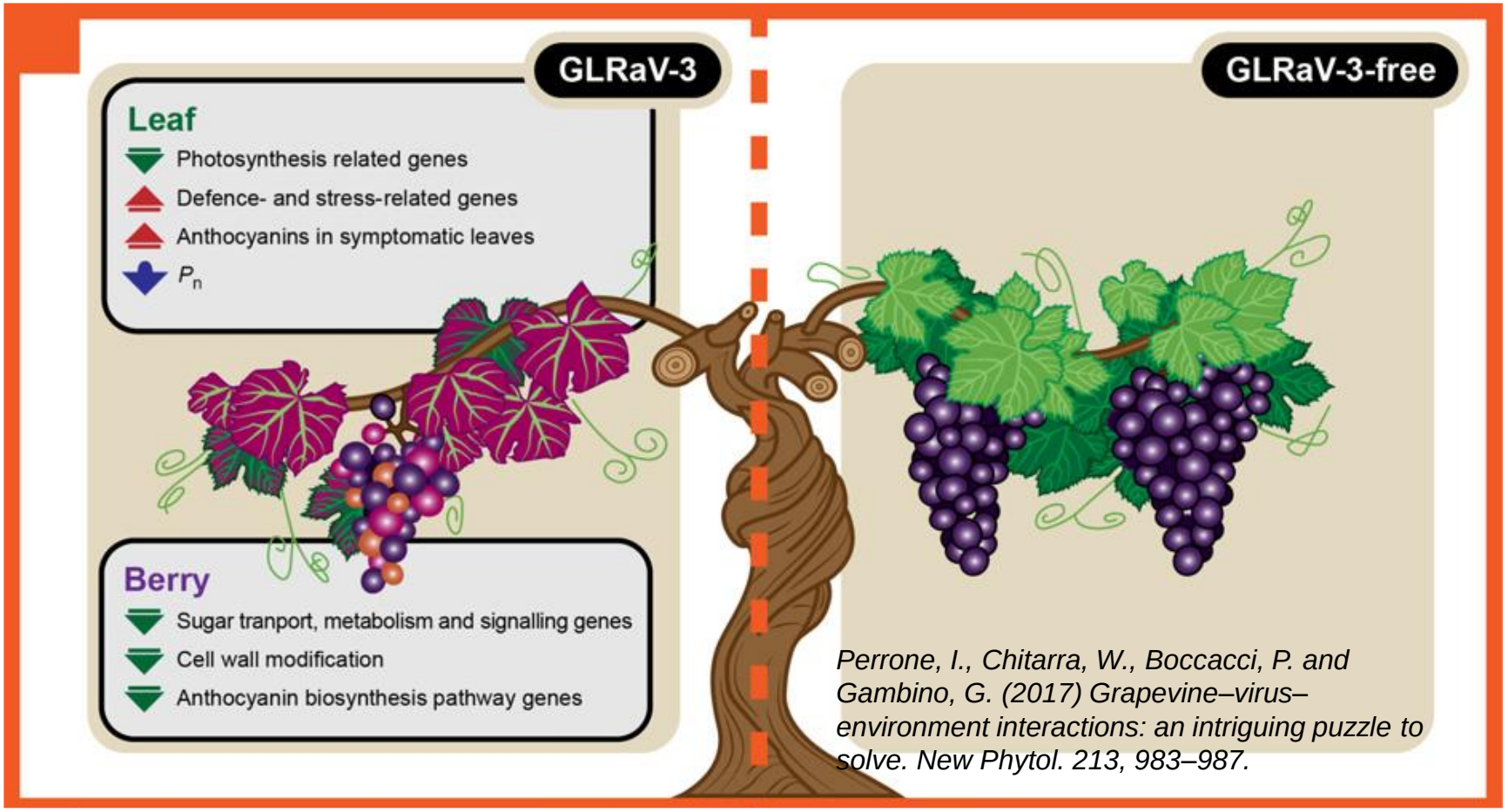
- Lower total soluble solids (TSS)
  - Significantly lower Brix
- Higher titratable acidities (TA)
- Lower anthocyanin concentrations
- Lower pH

Wine

- Less intensely colored
- Less Fruit aroma earthier
- More astringent

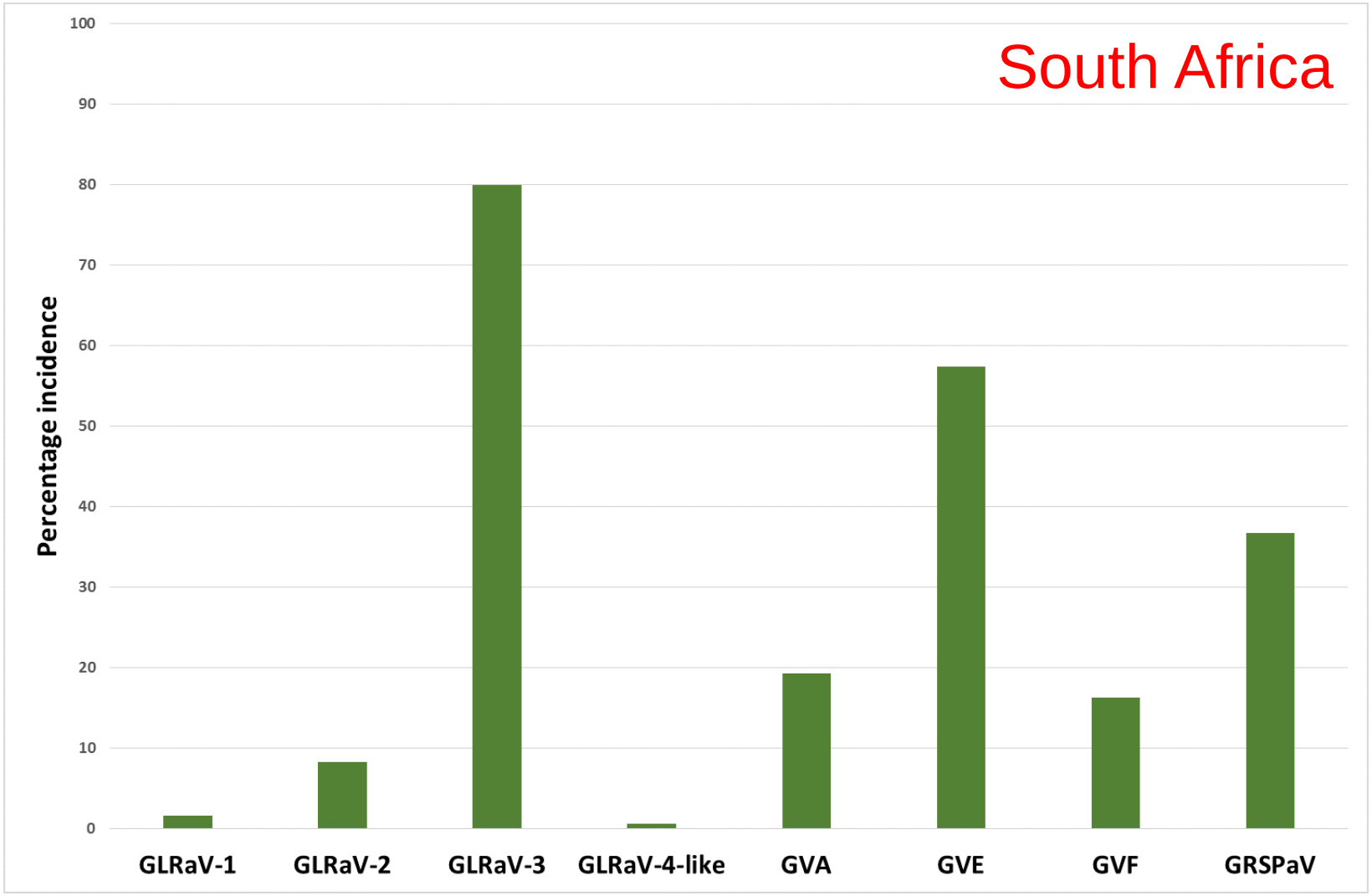
"GLRaV-3 infection effects on wine quality may depend on fruit maturity at harvest" – Pat Bowen et al. 2018, ASEV.

Leafroll Disease: Why it Matters



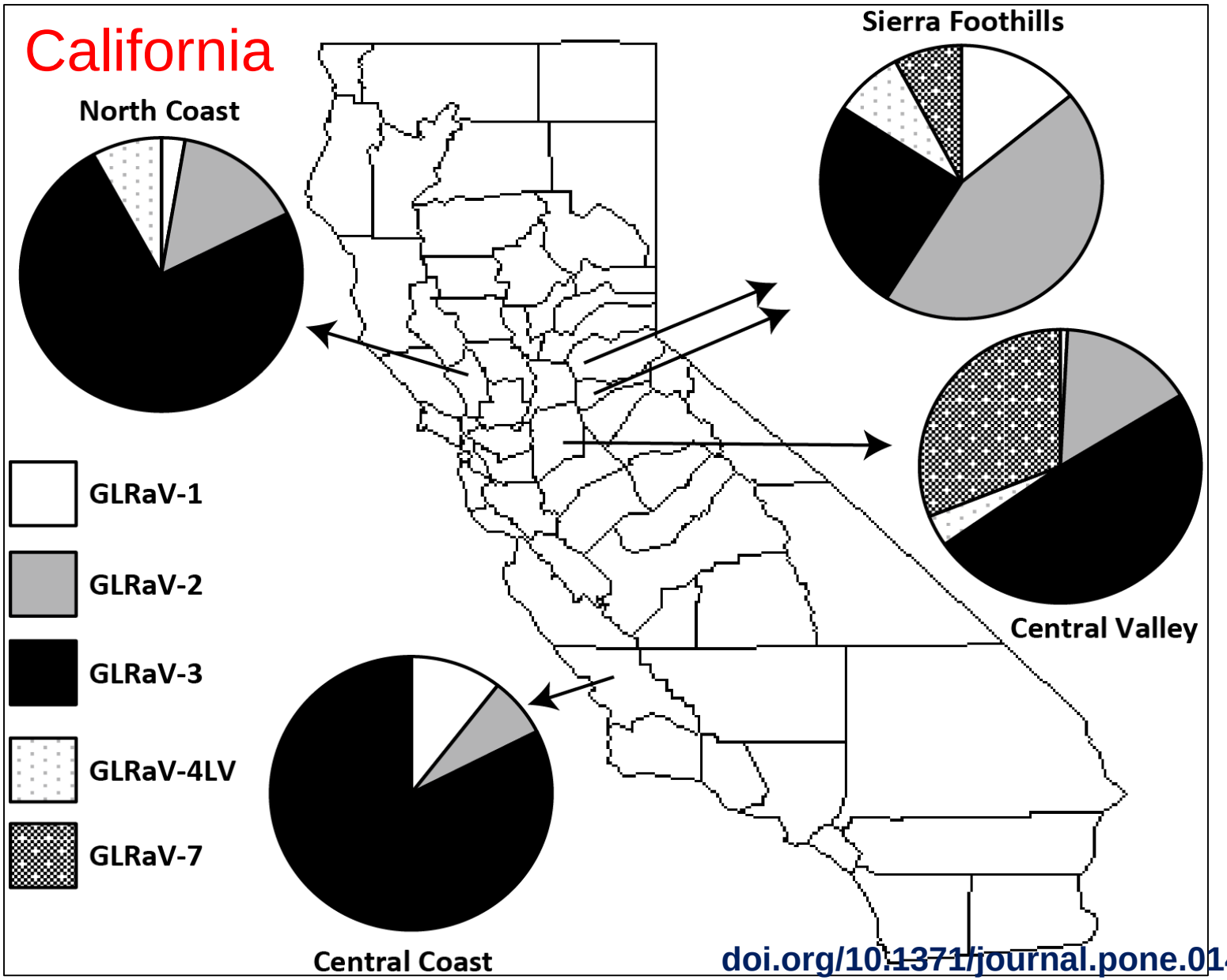
Up-regulation of Anthocyanin gene in GLRaV-3-infected leaves, contributes to the reddish-purple colour of symptomatic grapevine leaves of red/dark berried cultivars (Gutha et al., 2010)

Leafroll Disease: Which virus is the most wide spread



Elize Jooste, Hano Maree & Johan Burger | 1 Mar, 2017 | Viticulture research, Winetech Technical. Multiple viruses identified in grapevine leafroll diseased vineyards

Leafroll Disease: Which virus is the most wide spread



doi.org/10.1371/journal.pone.0142120

Leafroll Disease: Which virus is the most wide spread

New York

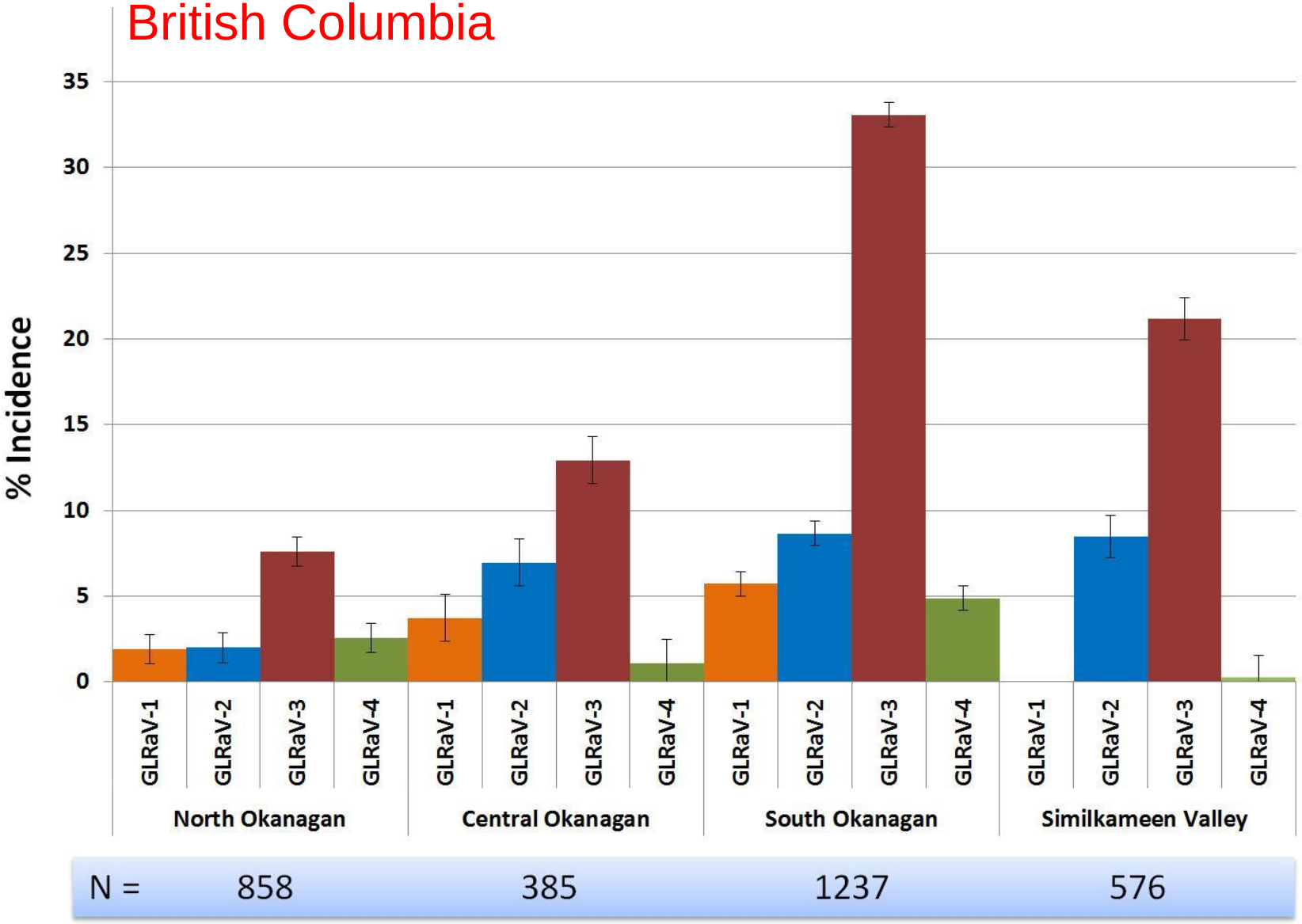
Table 3. Rates of single and mixed infections by *Grapevine leafroll-associated virus 1* (GLRaV-1), *Grapevine leafroll-associated virus 2* (GLRaV-2), and/or *Grapevine leafroll-associated virus 3* (GLRaV-3) in grapevine leaf samples (pooled 15 leaves from a five-vine quadrat) from Finger Lakes vineyards

	Virus-infected samples	
Virus combination	**Infected/tested[a]**	**Percent**
No infection	762/1,124	68
Single infection		
GLRaV-1	113/1,124	10
GLRaV-2	36/1,124	3
GLRaV-3	173/1,124	15
Subtotal	322/1,124	29
Multiple infection		
GLRaV-1 + GLRaV-2	8/1,124	0.7
GLRaV-1 + GLRaV-3	28/1,124	2.5
GLRaV-2 + GLRaV-3	3/1,124	0.3
GLRaV-1 + GLRaV-2 + GLRaV-3	1/1,124	0.1
Subtotal	40/1,124	3.6

[a] Infected samples were detected by double antibody sandwich–enzyme-linked immunosorbent assay.

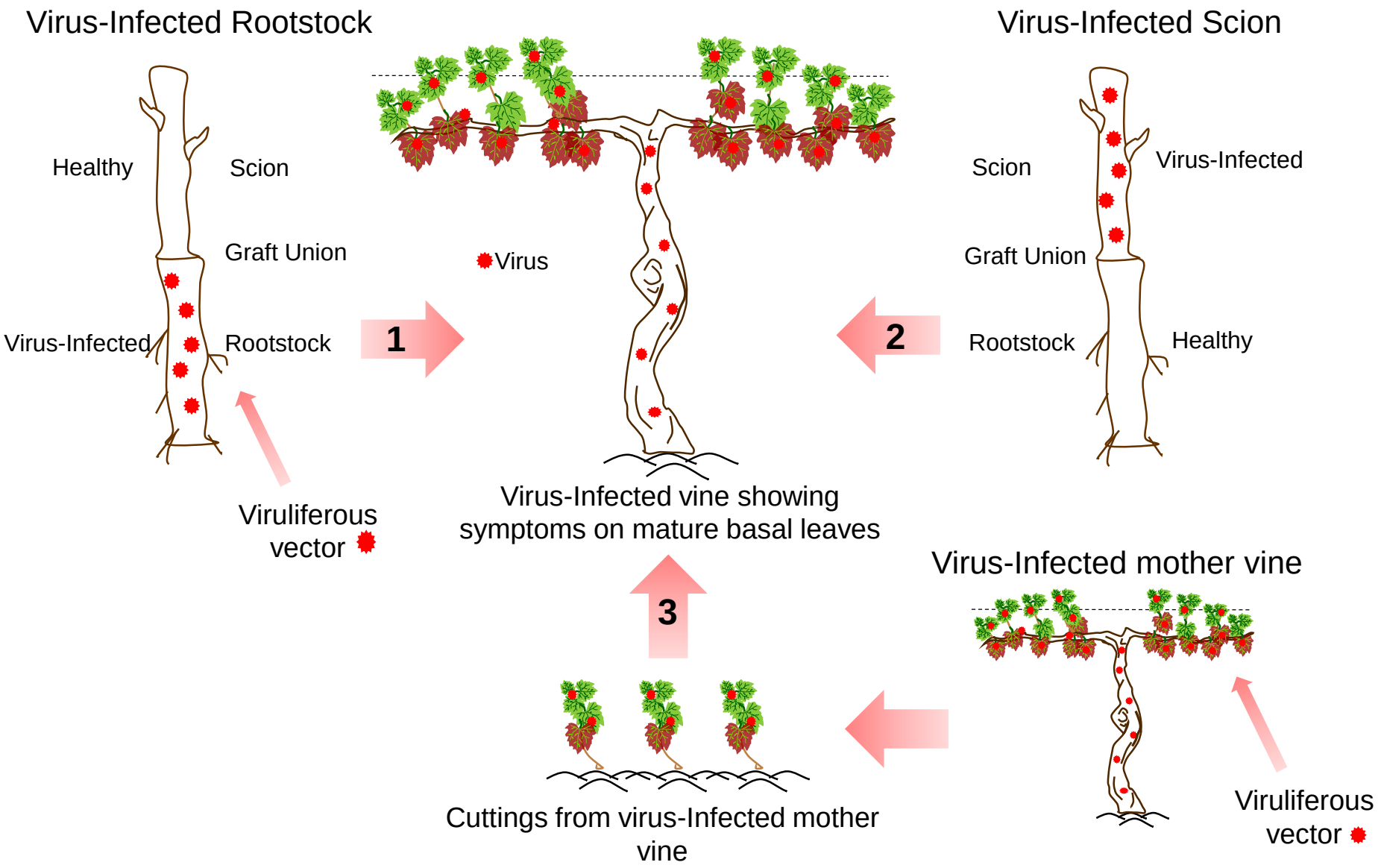
Fuchs, M. et al. Plant Dis. 93:395-401

Leafroll Disease: Which virus is the most wide spread



Poojari et al. 2017. Plant Disease. (https://doi.org/10.1094/PDIS-04-17-0497)

Primary transmission of grapevine viruses

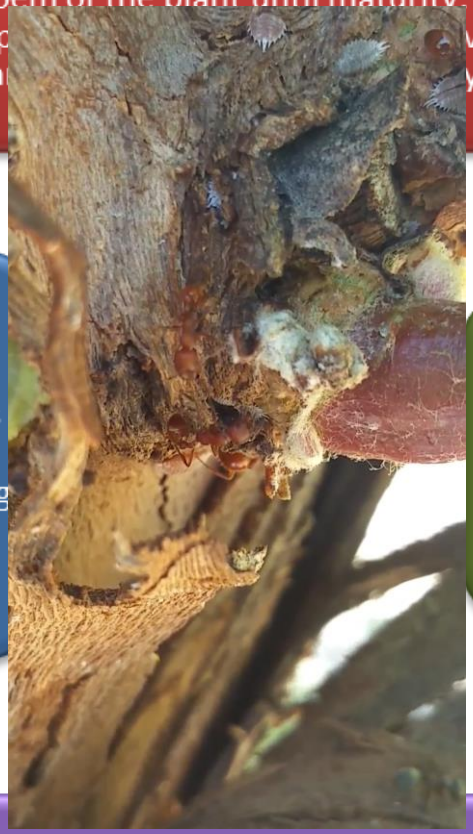


Secondary transmission of GLRD - Reported vectors for Grapevine leafroll viruses

Virus	Mealybugs	Scale insects
GLRaV-1	Bohemian mealybug (*Heliococcus bohemicus*)	*Pulvinaria vitis*
	Apple mealybug (*Phenacoccus aceris*)	***Parthenolecanium corni***
	Obscure mealybug (*Pseudococcus viburni)*	***Neopulvinaria innumerabilis***
	Citrophilous mealybug (*Pseudococcus calceolariae*)	
	Grape mealybug (*Pseudococcus maritimus*)	
	Comstock mealybug (*Pseudococcus comstocki)*	
GLRaV-3	Bohemian mealybug (*Heliococcus bohemicus*)	*Pulvinaria vitis*
	Vine mealybug (*Planococcus ficus*)	***Neopulvinaria innumerabilis***
	Citrus mealybug (*Planococcus citri*)	***Parthenolecanium corni***
	Longtailed mealybug (*Pseudococcus longispinus*)	*Coccus hesperidium*
	Citrophilous mealybug (*Pseudococcus calceolariae*)	*Coccus longulus, Saissetia* sp.
	Grape mealybug (*Pseudococcus maritimus*)	*Parasaissetia nigra*
	Obscure mealybug (*Pseudococcus viburni*)	*Ceroplastes rusci*
	Comstock mealybug (*Pseudococcus comstocki*)	
	Apple mealybug (*Phenacoccus aceris*)	
GLRaV-4 and its strains -5, -6, and -9		
	Vine mealybug (*Planococcus ficus*)	*Ceroplastes rusci*
	Longtailed mealybug (*Pseudococcus longispinus*)	
	Apple mealybug (*Phenacoccus aceris*)	

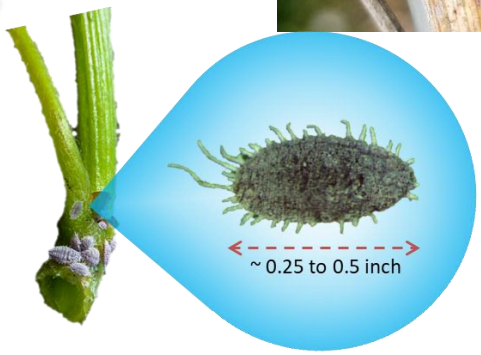
Table: Updated from Rayapati et al., 2014. Plant Disease / Vol. 98 No. 9

March - April: Overwintering eggs hatch around the same time as buds appear. Early crawlers conglomerate around the bud until shoots emerge where they will crawl to the green growth and feed on the phloem of the plant until maturity. Early nymp viruses tha ybug



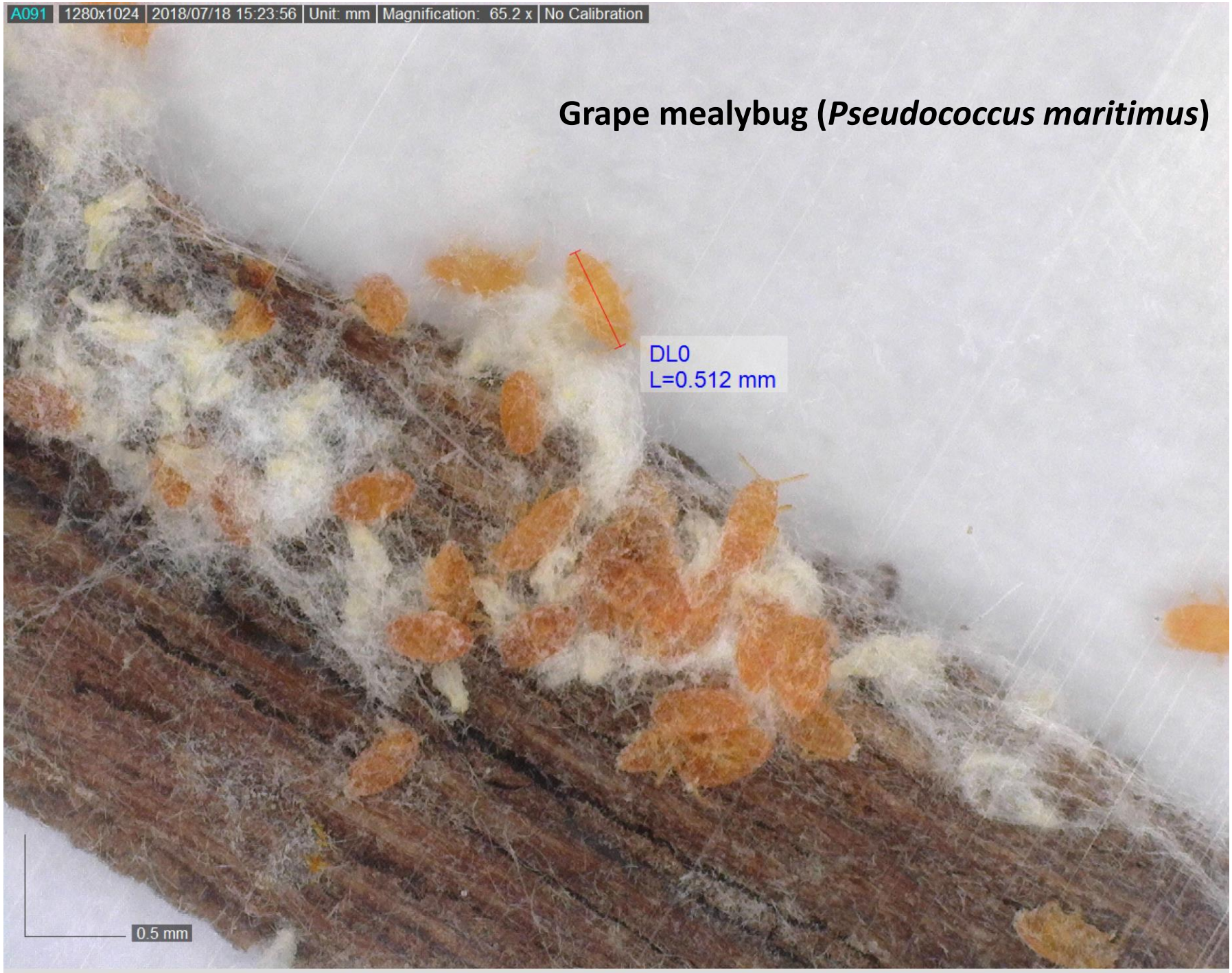
May - June: First generation nymphs mature, mate and then return to the woody regions of the vine to lay eggs and die.

October - February: The grape mealybug population overwinters as eggs. The eggs are the most cold tolerant stage of the grape mealybug lifecycle.



July - September: Second generation crawlers emerge and make their way to the shoots and grape clusters. They will mature quickly before returning to the woody portions of the vine to lay eggs for the winter

Grape mealybug


A091 1280x1024 2018/07/18 15:23:56 Unit: mm Magnification: 65.2 x No Calibration
Grape mealybug (*Pseudococcus maritimus*)
DL0
L=0.512 mm
0.5 mm

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Grapevine fanleaf degeneration complex

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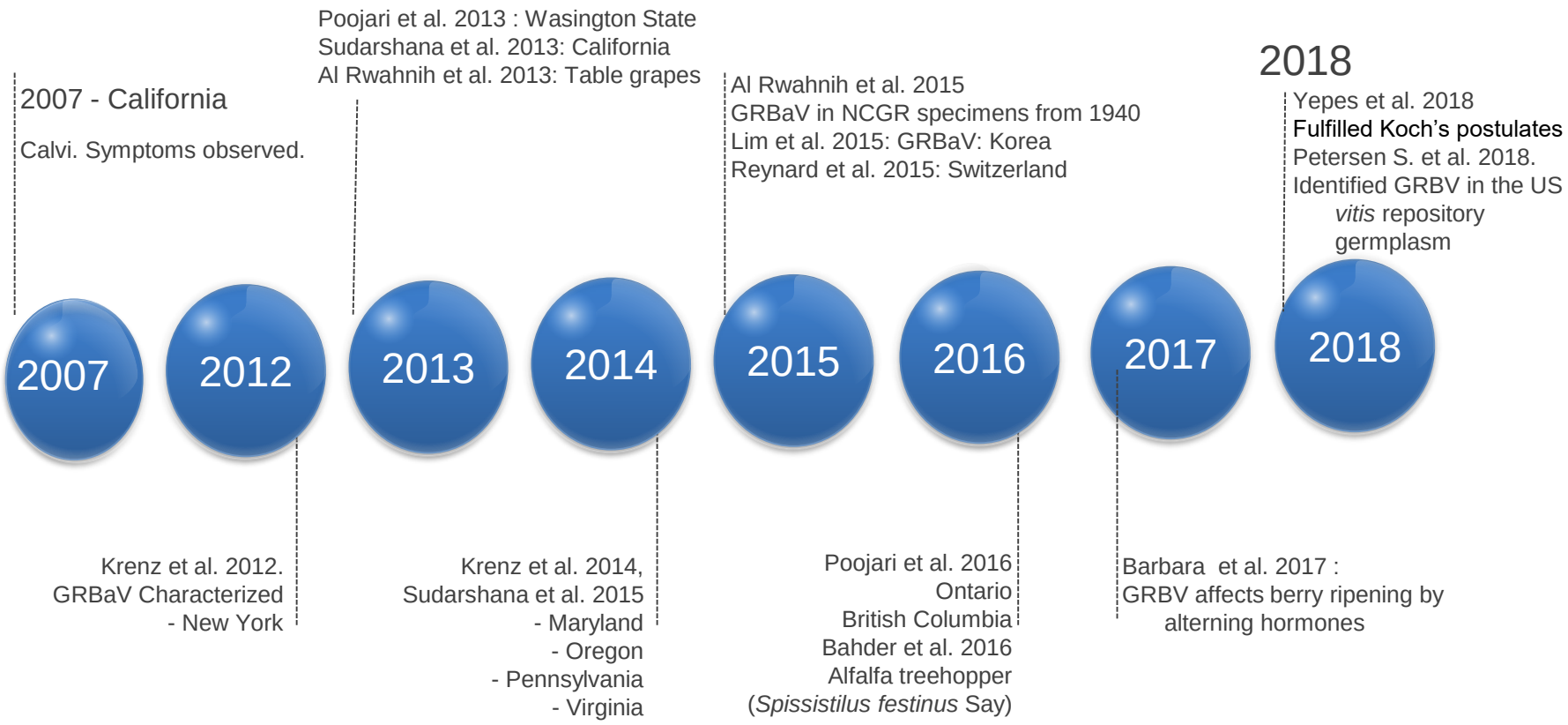
***Grapevine Pinot Gris Virus* (GPGV)**

Grapevine Red Blotch Virus

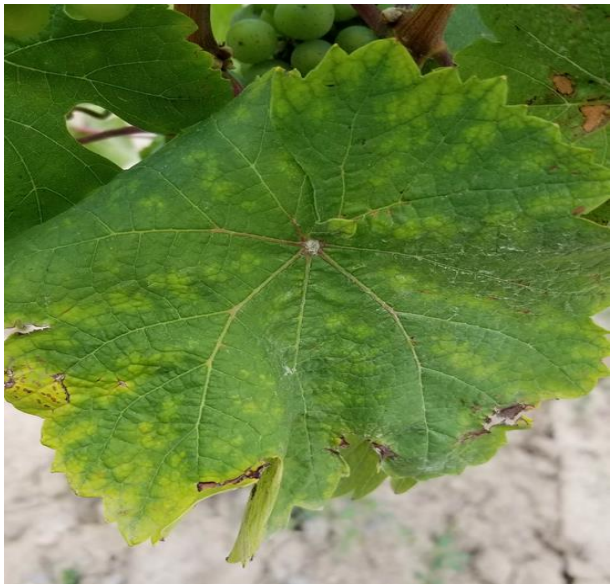


Grapevine Red Blotch Virus: In The Ideal World

GRBV on white cultivars

cv. Chardonnay

Image curtesy @ Jessica Oppenlaender



Grapevine red blotch virus - Update

- Secondary spread of GRBV was recorded in California and Oregon
- Alfalfa treehopper (*Spissistilus festinus* Say) (Bahder et al. 2016) – under controlled laboratory conditions
- GRBV found in field collected insect species belonging to the *Membracidae* (*Ceresa festina*), *Cicadellidae* (*Colladonus reductus* and *Osbornellus* sp.) and *Cixiidae* (unknown species) (Fuchs et al. 2016)

References: http://www.grapesociety.org/uploads/5/5/0/6/55065965/fri_am_3_wgs_bahder_2014.pdf
https://static.cdfa.ca.gov/PiercesDisease/reports%2F2016%2F2016%20CDFA%20Report%20Fuchs.pdf.

Buffalo treehopper (*Stictocephala bisonia*)

- Bright green color and triangular shape
- Size: 6 to 8 millimeters (0.24 to 0.31 in)
- Long and transparent wings.
- It is an occasional pest of grapevines


Treehopper damage on grapevine

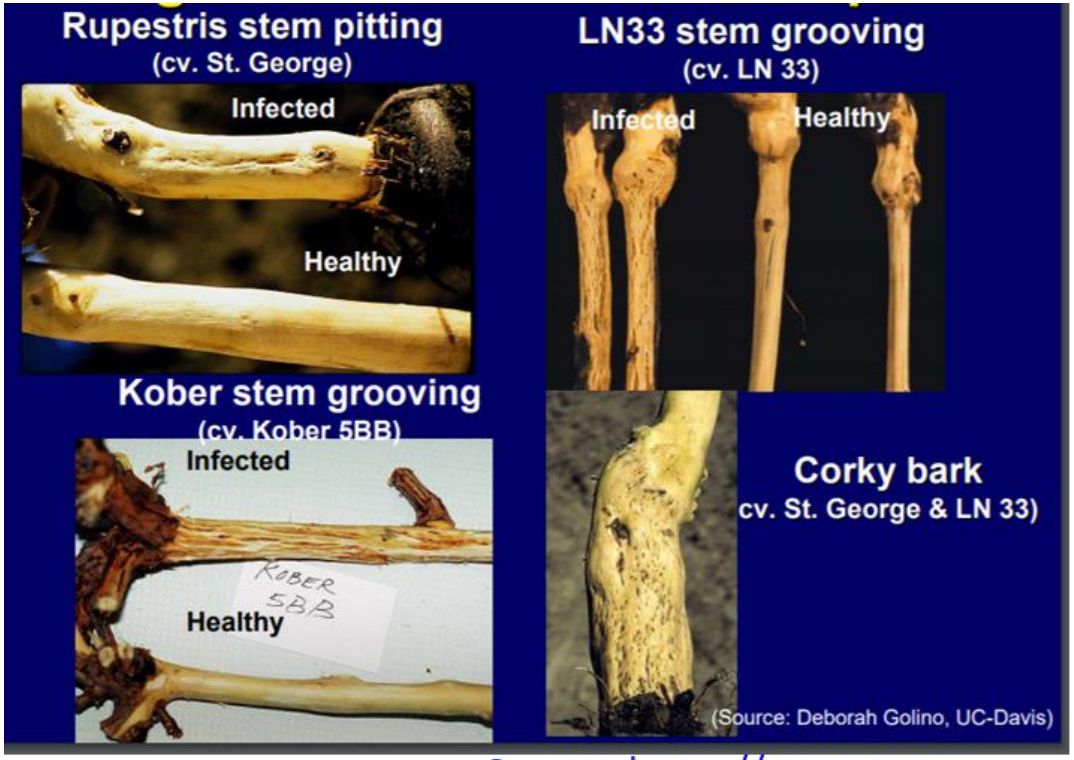
Rugose wood disease complex

- Rugose = wrinkled; corrugated
- Known for typical modifications of ridges and grooves of the woody cylinder: Scion, rootstock or both
- These modifications vary on different cultivars and root stock combinations
- Known to cause synergistic interactions with other viruses and viroids
- RSP is latent (symptomless) in most of European grapevine cultivars and American rootstock species and hybrids but do show veinal necrosis in *Vitis rupestris*

Rugose wood disease complex

Disorders

Rupestris stem pitting	RSP
Kober stem grooving	KSG
LN33 stem grooving	LNSG
Corky bark	CB



Source: https://wawgg.org

Viruses

Grapevine Rupestris stem pitting-associated virus(GRSPaV)

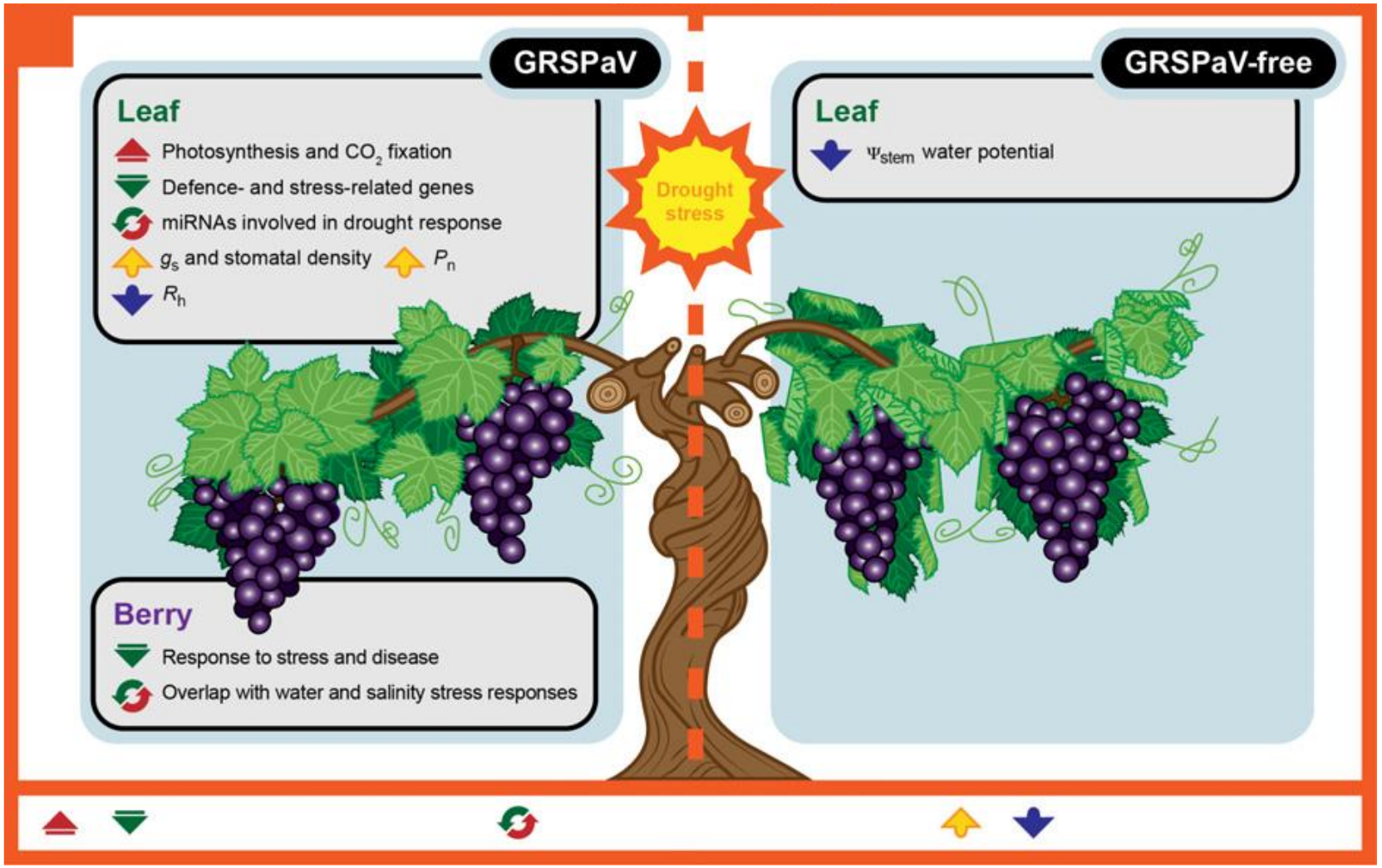
Grapevine virus A (GVA)

Grapevine virus B (GVB)

Grapevine virus D (GVD)

Grapevine virus E (GVE)

Grapevine Rupestris stem pitting-associated virus (GRSPaV)



Perrone, I., Chitarra, W., Boccacci, P. and Gambino, G. (2017) Grapevine–virus–environment interactions: an intriguing puzzle to solve. New Phytol. 213, 983–987.

Pantaleo et al., 2016

Grapevine fanleaf disease

- GFLV is worldwide in distribution
- GFLV is a soil-borne disease - spread by Dagger nematode *Xiphinema index*
- *X. index has not reported in Canada*
- Generally displays a patchy patter in the vineyard
- Infected vines show less growth and irregular fruit clusters
- Symptoms can be confused with herbicide damage

Xiphinema index

***Grapevine fanleaf virus* (GFLV)**

Grapevine Fanleaf Virus

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Other viruses:

***Grapevine Pinot Gris Virus* (GPGV)**

Grapevine Pinot Gris Virus

- GPGV has been reported from major grape growing regions of the world - USA, China, Canada, Europe, Australia
- Belongs to the genus *Trichovirus* in the family *Betaflexiviridae*
- GPGV is known to occur in both red and white wine-grape cultivars as well as in table grapes as latent infections.

  - Two strains: Symptomatic and Asymptomatic
- The virus is possibly transmitted by **Erineum mite** *Colomerus vitis* (Need to be confirmed)

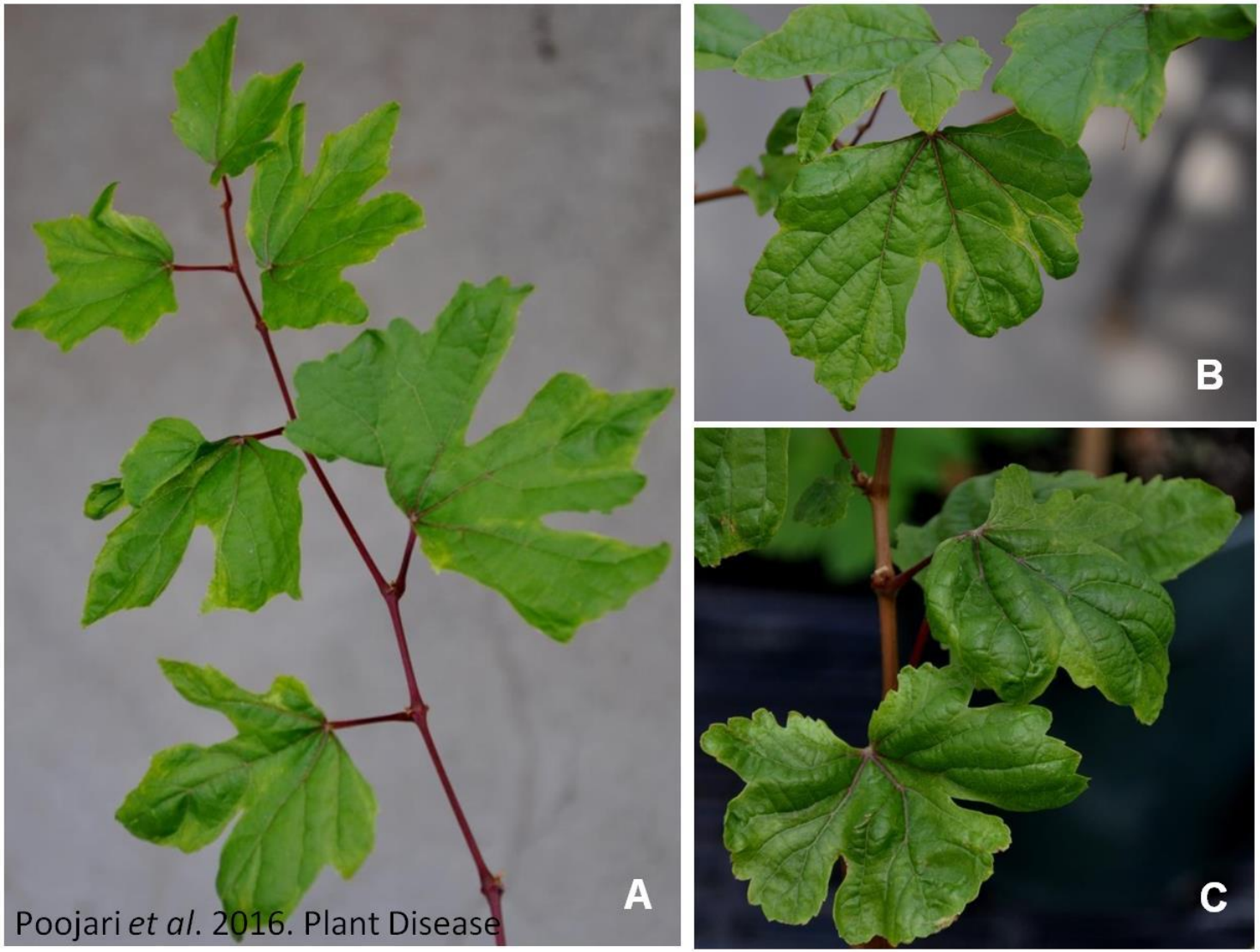
Reported Alternative Hosts

Lamb's quarters
(*Chenopodium album* L.)

White Campion
(*Silene latifolia* subsp. *Alba* (Mill.)

Image source: nature gate and cabi

Grapevine Pinot Gris Virus (GPGV)

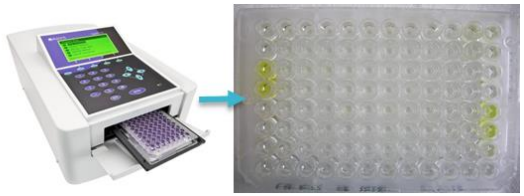


Foliar symptoms of GPGV on cv. *Pinot gris*: Chlorotic mottling and severe leaf deformation

Grapevine Virus Diagnostics

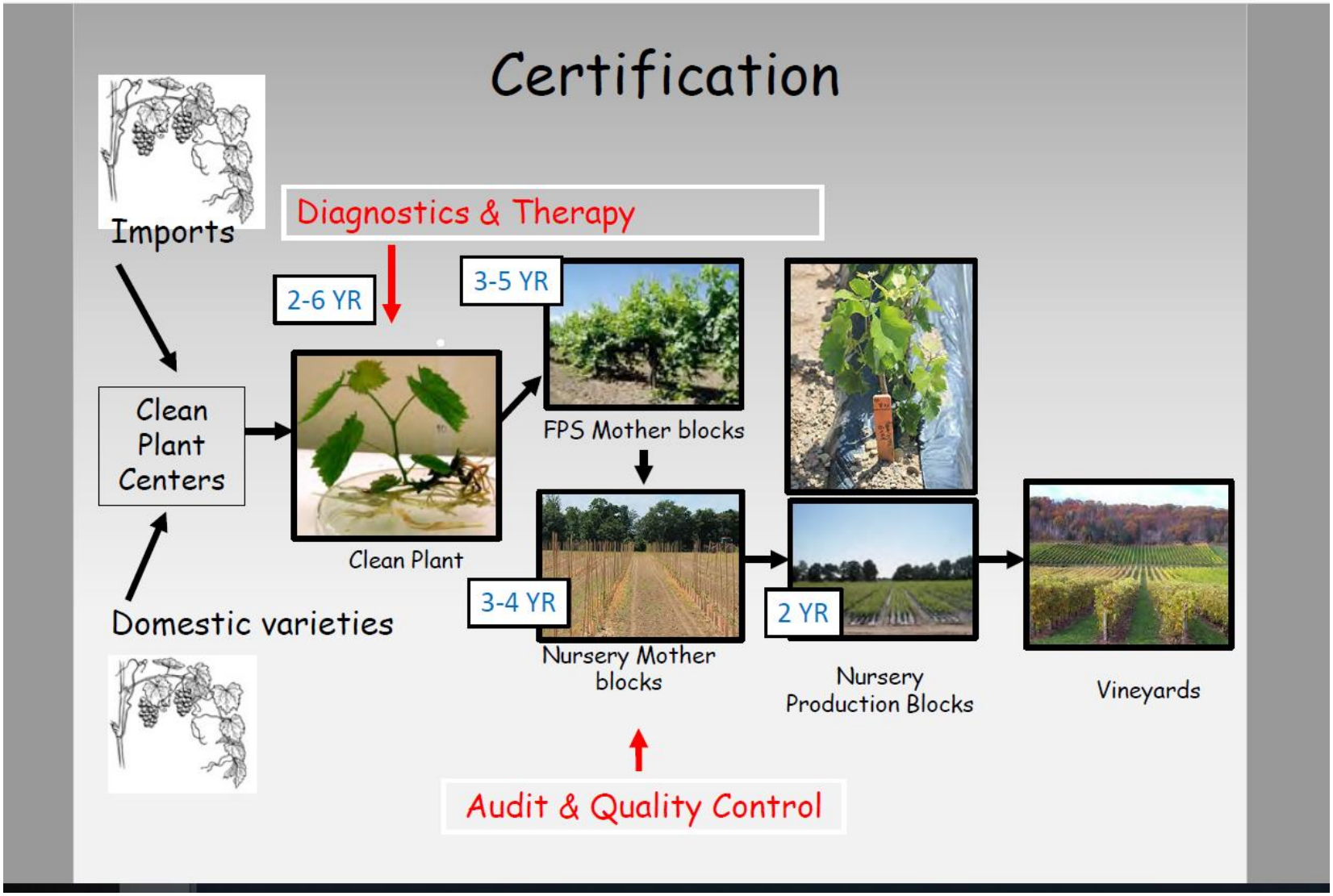
Symptom Based	Biological Indexing	Serological (ELISA)	Molecular (PCR, qPCR, ddPCR & NGS)
Non-specific Inaccurate	Symptom based Labor intensive Time consuming	Specific No Abs for all viruses Less sensitive than PCR	Highly specific Post-PCR process Multiplex End-Point Quantitative

Poojari et al. 2013. PloS ONE 8(6): e64194



Speed, sensitivity, specificity, robustness and cost effectiveness

Clean Plant Program



Source:
https://grapesandwine.cals.cornell.edu/sites/grapesandwine.cals.cornell.edu/files/shared/March%2010%20NCPN%20webinar%20The%20pipeline%20From%20tissue%20culture%20to%20your%20vineyard%20reduced%20size.pdf

Clean Plant Program: Simplified

Classical

Health Status Unknown

NGS based

Diagnostic tests for 30+ pathogens (ELISA, PCR)

NGS testing (days to weeks)

Positive

Virus elimination (meristem culture; 2 yrs.)

Woody + Herbaceous Indexing (2 yrs.)

Retesting (30+ pathogens) (days to weeks)

Negative

Positive

Virus elimination (meristem culture 2 yr)

Retesting with NGS

NGS is a high-throughput analysis tool
– Unambiguous results
– No reflexive testing

CLEAN PLANTS

- Budwood/cuttings distributed
- Mist-propagated vines in greenhouse (1 Yr)
- 3-4 years to full production - Visual Inspection and testing

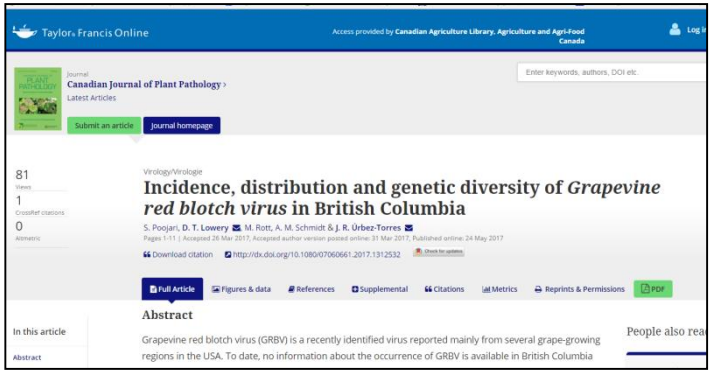
Options for management of virus diseases

- Use virus-free plant materials
- Use reliable laboratory tests:
  - for diagnosing problems in existing vineyards
  - to test disease status of dormant wood before using for propagation of new vines
  - do not compromise with the quality of the planting material
- Removing/Roughing infected vines: 5-20% infection - roughing is optimal; Beyond 20% vineyard replacement (Ricketts et al., 2015)
- Prevent secondary spread by vectors like mealybugs/soft-scales:
  - closely monitoring the vineyards for mealybug/scale infestations
  - applying appropriate pesticides: Timing of pesticides are important

We need clean plants, but its only the first step

We need “start clean stay clean” programs

For more information



Summerland RDC Newsletter
http://bcwgc.org/sites/default/files/project/Kate/flies/SRDC%20Newsletter_GLD_Nov%202016.pdf

CCOVI lecture series @
https://brocku.ca/ccovi/outreach/lecture/

- Improvements in Grapevine virus diagnostics.
  https://brocku.ca/ccovi/wp-content/uploads/sites/125/sud-2017-virus.pdf
- Epidemiology and management of grapevine virus diseases.
  https://brocku.ca/ccovi/wp-content/uploads/sites/125/lowery-2017-epidemiology.pdf
- Demystifying the status of grapevine viruses in British Columbia.
  https://brocku.ca/ccovi/wp-content/uploads/sites/125/2016-03-02.-CCOVI-Lecture-Series.-Urbez-Torres-Grapevine-viruses-in-BC.pdf