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IMPACT OF RED BLOTCH DISEASE ON GRAPE AND WINE COMPOSITION AND QUALITY

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Foothills Grape Day 2016: Healthy Vines, Fine Wines
Amador County Fairgrounds, Spur Emporium Building
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Introduction

- Grapevine red blotch-associated virus (GRBaV)
  - Red Blotch disease was first described in Cab Sauv, Zin and Cab Franc in New York and California (1)
  - A DNA virus (GRBaV) was shown to be the causal agent of red blotch diseases (2)
  - Widespread in vineyards in USA and Canada

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(1) Al Rwahnih et al., (2013) Phytopath. 103: 1069-1076
(2) Fuchs (2013) http://lecture.ucanr.org/Mediasite/Play/7e6250539e5e4676ad4cd888051164c1d

Introduction

- **Red Blotch disease symptoms**
  - **RB disease shows symptoms similar to leafroll disease**
    - **Unlike leafroll – RB show red veins on leaf undersides and no rolling**

Introduction

- **Red Blotch disease spread**
  - **Widespread occurrence of Red Blotch disease indicate primary spread through propagation (1)**
  - **Increase incidence in young healthy vines adjacent to infected vineyards suggest vector (3)**
  - **3-cornered alfalfa treehopper (*Spissistilus festinus)* have recently be shown to be able to spread the disease (Bahder and Zalom)**

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(1) Al Rwahnih et al., (2013) Phytopath. 103: 1069-1076
(3) Poojaric et al. (2013) PLosONE 8: e64194

Perceived impact of RB disease on grape composition

- ↓ Sugar accumulation
  - As much 4-5 °Brix less
  - Delay in ripening
- ↓ Color development
- ↑ TA
  - Current research - show not always true
  - ↑ Malic acid
    - True for CH and CS, not Zin
  - For CH, ↓yield

Practices to negate impact of RB disease?

- Dropping 50% of crop
  - Seems to have no impact (CH, CS)
- Other practices? (none formally investigated so far)
  - Pruning?
  - Nutrients?

Study objectives

- To determine the impact of GRBaV on the composition of grapes at harvest and the resulting wines
- To investigate potential sensory and quality differences between wines made from GRBaV positive and negative grapes

Experimental layout

- Virus testing (GRBaV and GRLaV) of subset vines to determine GRBaV (+) and (-) sample plots
- Sample grapes at harvest
  - Chemical panels
  - Metabolomics analysis (primary and secondary metabolite profile)
  - Phenolic profile (AH-assay, RP-HPLC)
  - Tannin composition (SPE isolation, phloroglucinolysis)

Experimental layout

- Winemaking from GRBaV (+) and (-) grapes
  - Chemical analyses similar to grapes (previous slide)
  - Descriptive sensory analysis
    - Correlate wine composition with sensory attributes
    - Impact of GRBaV on wine style/quality

Experimental layout

Variety (site #)	Source County	Grape Sampling	Winemaking
Chardonnay 1a	Sonoma	Yes	Yes
Chardonnay 1b	Sonoma	Yes	No
Chardonnay 2	Sonoma	Yes	No
Merlot 1	Napa	Yes	No
Merlot 2	Napa	Yes	Yes
Cab Sauv 1	Napa	Yes	Yes
Cab Sauv 2	Napa	Yes	Yes

Red Blotch symptoms – Chardonnay Site 1a

Red Blotch symptoms – Site 1 Cab Sauv

Results: Grape chemical composition

Sample	GRBaV Status	Harvest Date	°Brix	pH	TA (g/L)
Chardonnay 1a	-	12-Sep-14	24.4	3.4	6.0
	+	12-Sep-14	23.0	3.5	6.7
Chardonnay 1b	-	11-Sep-14	23.0	3.4	6.6
	+	11-Sep-14	22.5	3.6	6.9
Chardonnay 2	-	16-Sep-14	24.1	3.3	7.8
	+	16-Sep-14	24.2	3.5	8.9

↓6%

↓2%

0%

- ↓°Brix 0-6% GRBaV(+) CH grapes
- Small differences in pH
- ↑ TA in GRBaV(+) grapes

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Results: CH 1a chemical composition

CH 1a	GRBaV Status	Harvest Date	°Brix	pH	TA (g/L)
2014	-	12-Sep-14	24.4	3.4	6.0
	+	12-Sep-14	23.0	[illegible]	6.7
2015	-	9-Sep-15	25.7	3.5	5.3
	+	9-Sep-15	23.6	[illegible]	6.3

↓6%

↓8%

- For both years a ↓°Brix 6-8% GRBaV(+) CH grapes
- Small differences in pH
- ↑ TA in GRBaV(+) grapes

Results: Grape chemical composition

Sample	GRBaV Status	Harvest Date	°Brix	pH	TA (g/L)
Merlot 1	-	29-Aug-14	25.0	3.6	3.2
	+	29-Aug-14	21.1	[illegible]	3.6
Merlot 2	-	26-Sep-14	24.9		4.2
	+	26-Sep-14	23.5	3.5	4.7
Cab Sauv 1	-	18-Sep-14	25.7	[illegible]	7.8
	+	18-Sep-14	20.6	[illegible]	8.6
Cab Sauv 2	-	7-Oct-14	26.3	[illegible]	4.8
	+	7-Oct-14	25.2	3.6	4.9

↓16%

↓6%

↓20%

↓4%

- ↓°Brix 6-16% GRBaV +) ME and 4-20% in CS grapes
- Small differences in pH
- ↑ TA in GRBaV(+) grapes

Results: Grape chemical composition

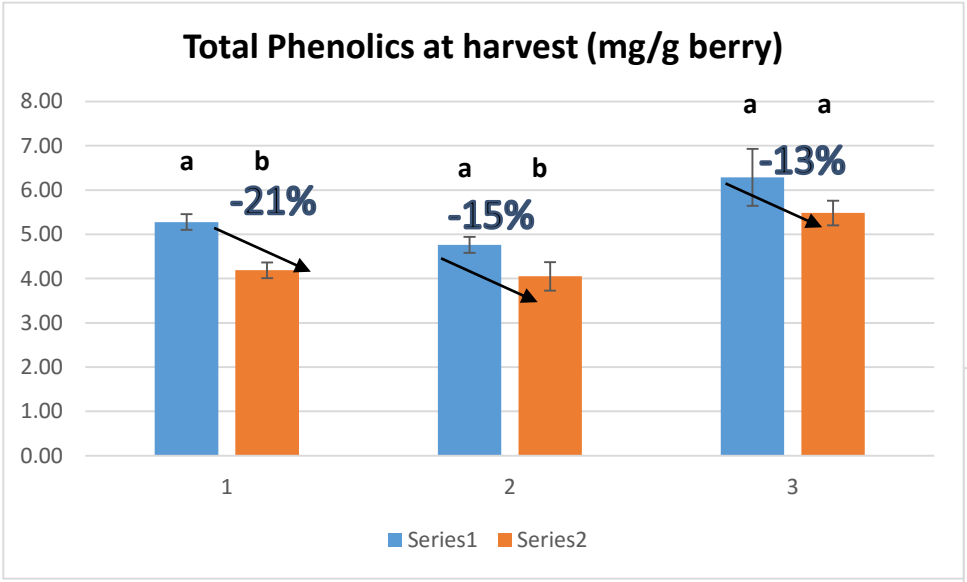
CS 2	GRBaV Status	Harvest Date	°Brix	pH	TA (g/L)
2014	-	7-Oct-14	26.3	3.6	4.8
	+	7-Oct-14	25.2	[illegible]	4.9
2015	-	21-Sep-15	26.0	[illegible]	4.3
	+	21-Sep-15	22.4	3.7	4.4

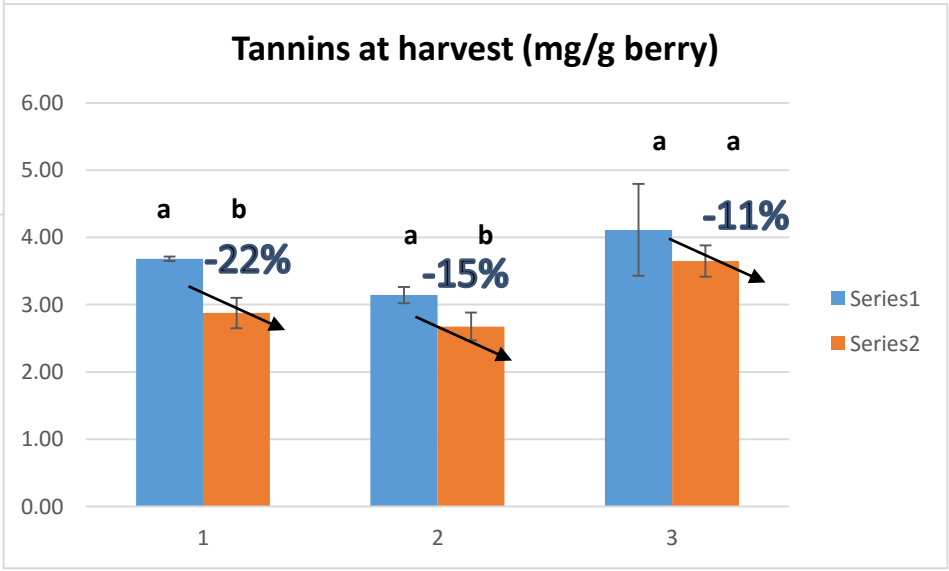
↓4%

↓14%

- Both years ↓°Brix 4-14% GRBaV (+)
- Small differences in pH
- ↑ TA in GRBaV(+) grapes

Results: CH grape composition - AH assay





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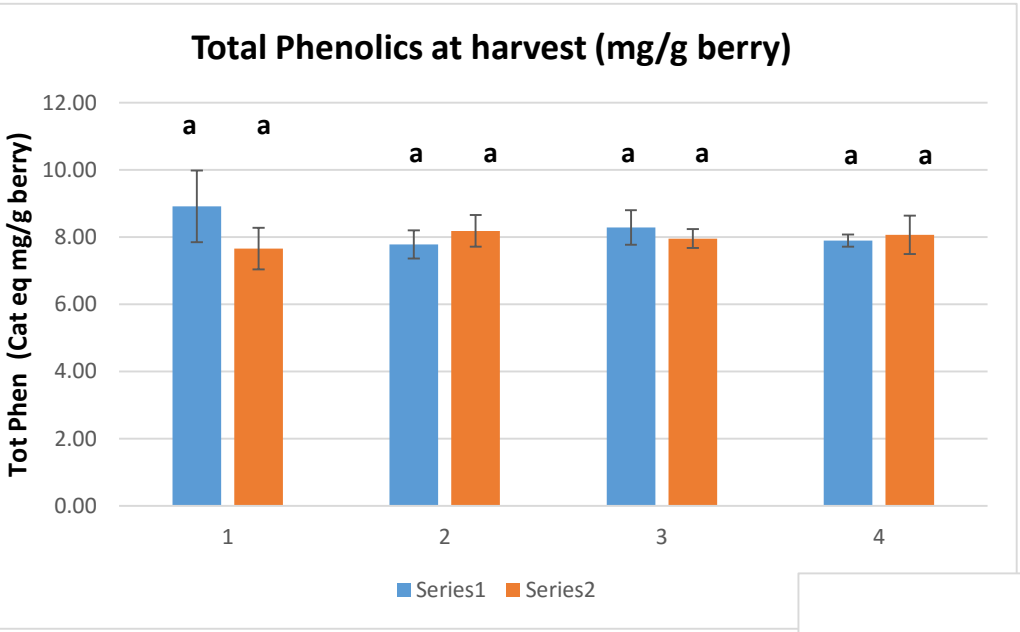
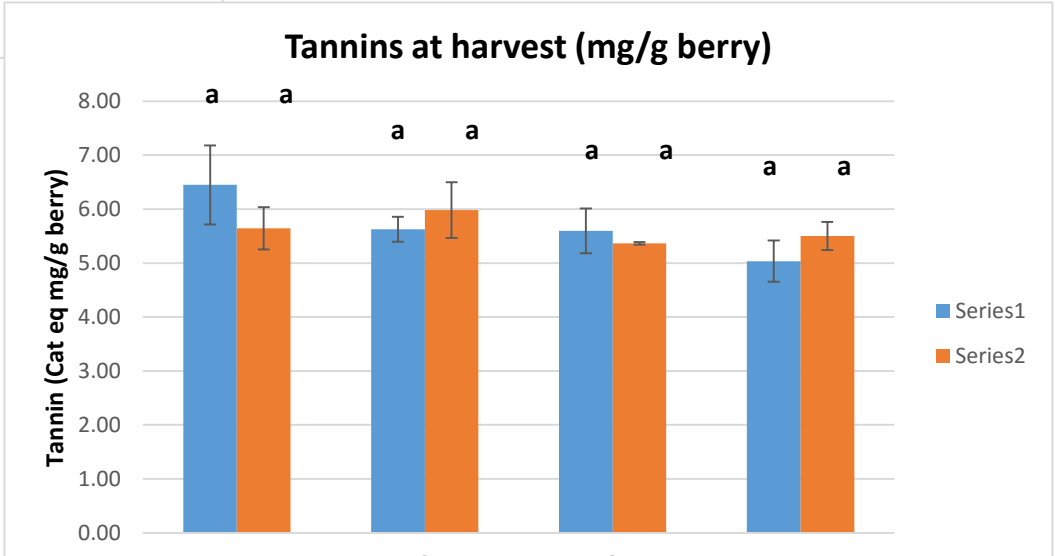
- Bars with the same letter indicate no significant difference within a site

Results: Red grape composition - AH assay





- Bars with the same letter indicate no significant difference within a site

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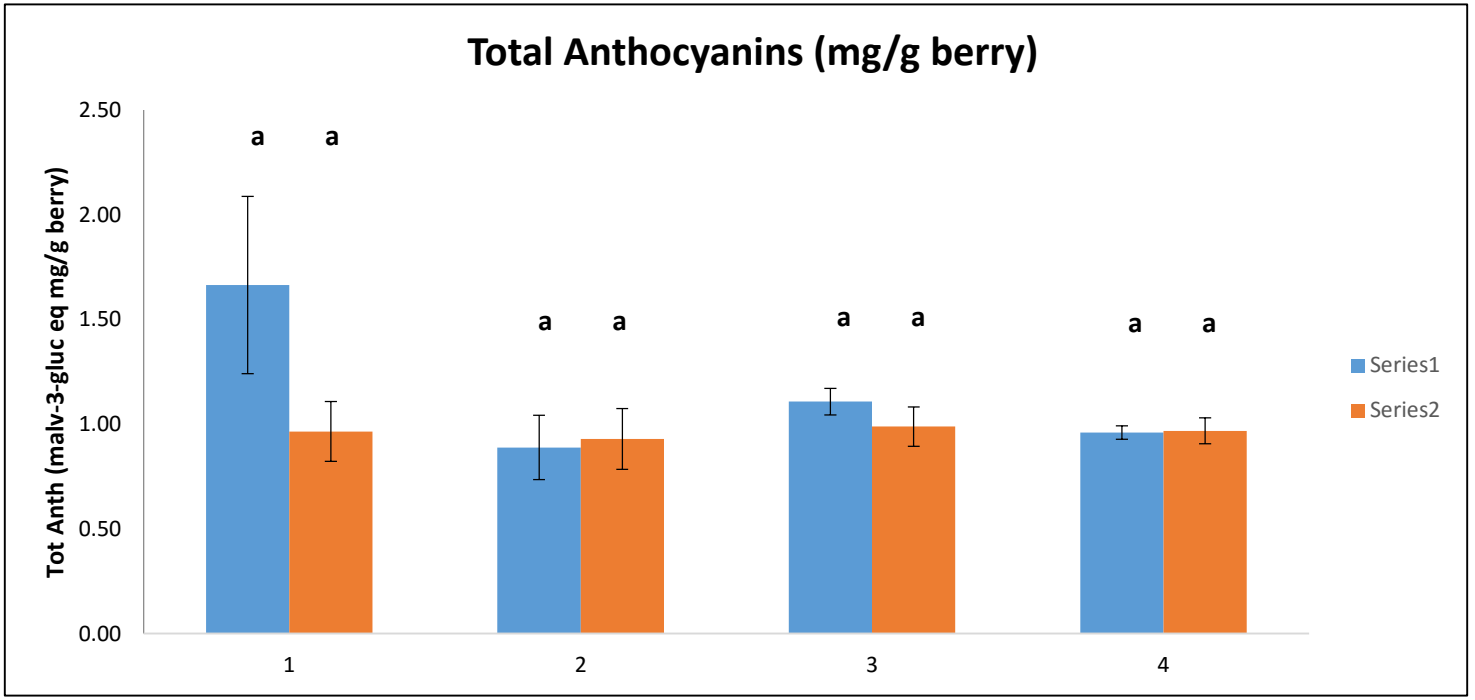
Results: Red grape composition - AH assay



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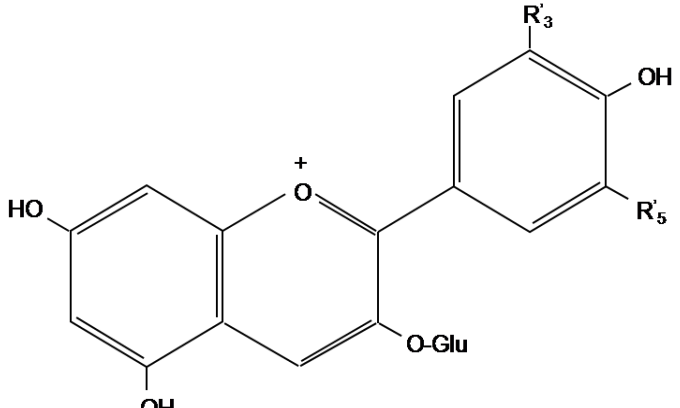
- Bars with the same letter indicate no significant difference within a site

Results: Grape composition RP-HPLC

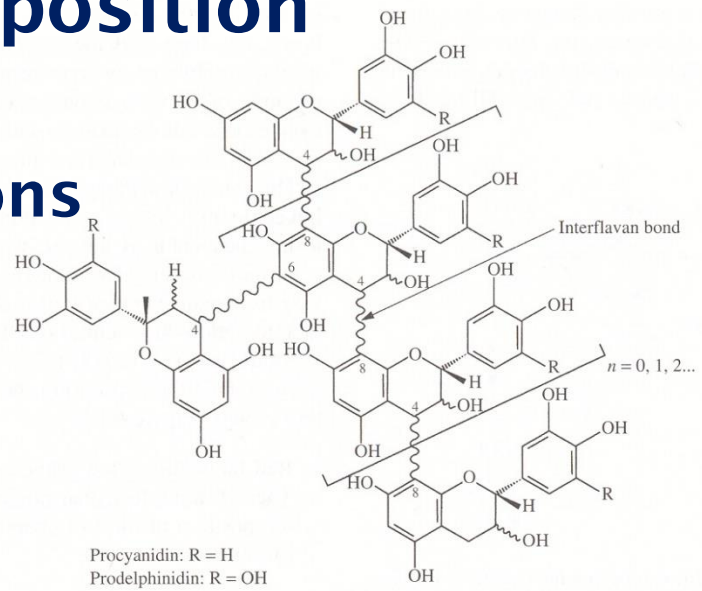
- RP-HPLC results of individual phenols support AH-assay
  - Mostly small differences
    - When significant RB(-) > RB(+)
      - CH 1b and 2 flavan-3-ols RB(-) > RB(+)
      - CS 1 flavan-3-ols, Tot anth, pol pigm RB(-) > RB(+)
- Variable response to RB disease within a variety



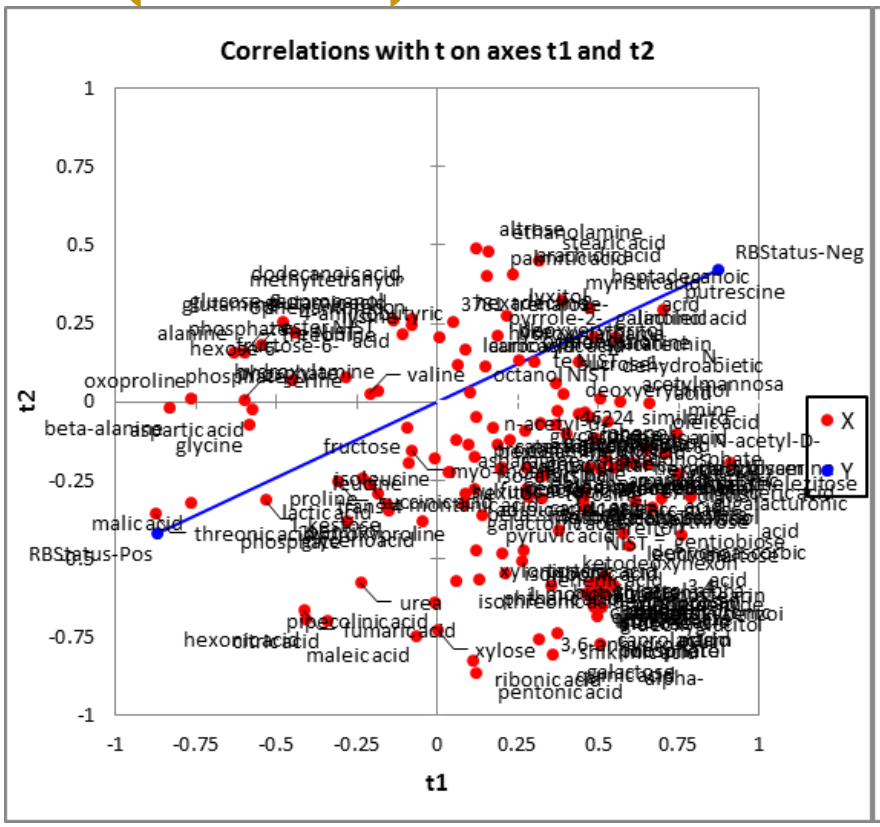
Anthocyanin

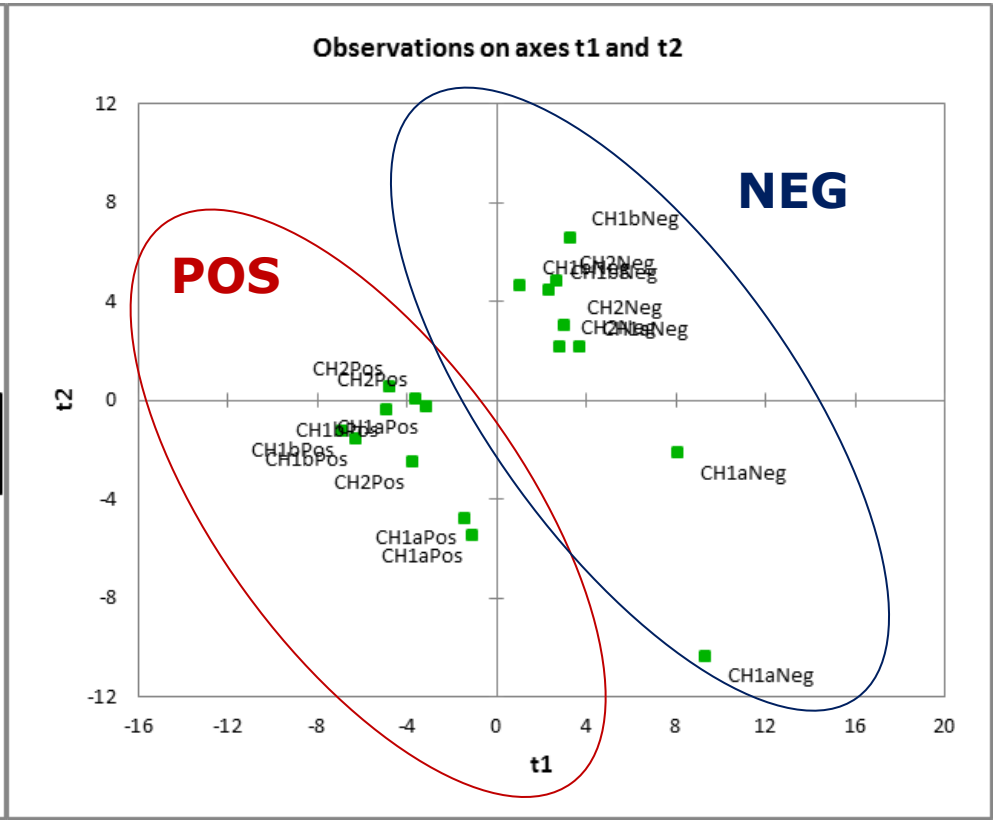
Results: Grape composition phloroglucinolysis

- Tannin analysis showed signf differences among diffr varieties
  - No diffr due to disease status of grapes (mDP, % gallo units, % galloylation)
- It looks as if tannin composition similar
  - However method limitations

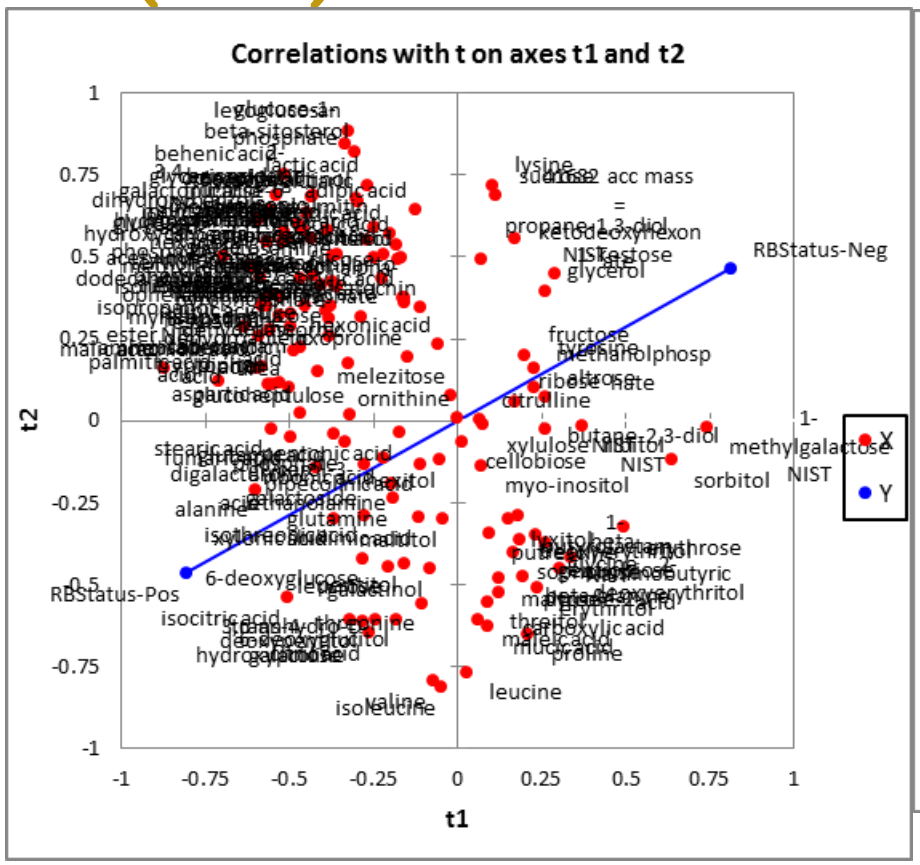


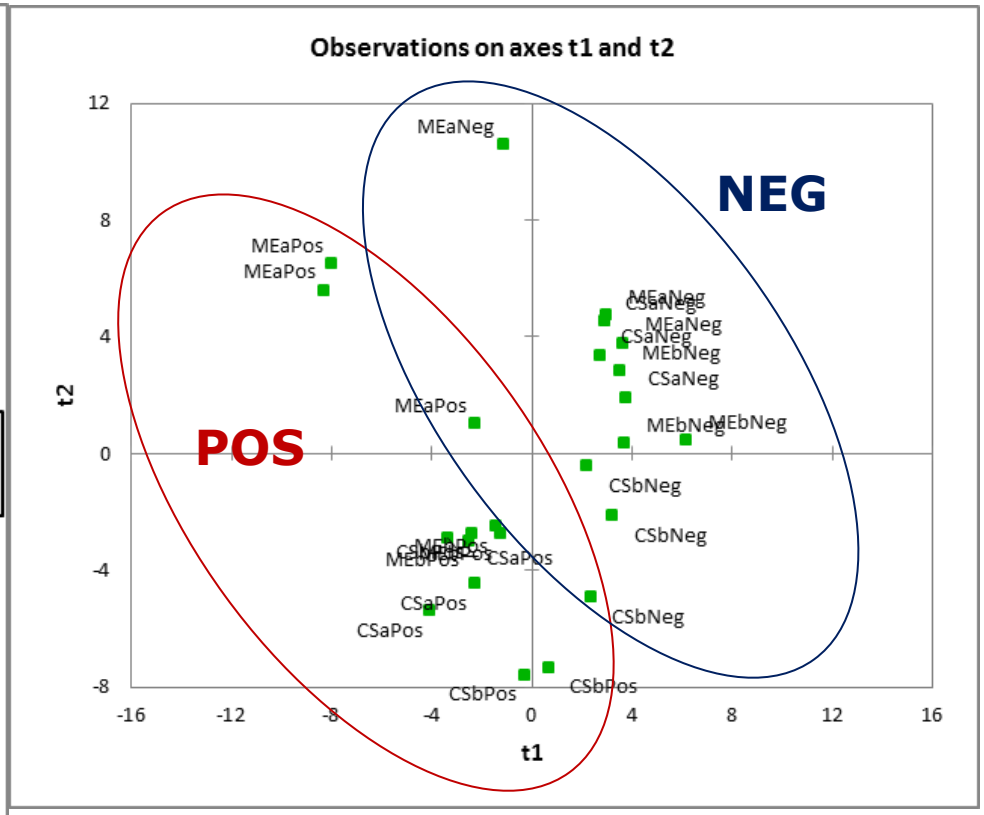
PLS-DA of metabolomics grape data (white)





PLS-DA of metabolomics grape data (red)





Results: Wine chemical composition

Wine	GRBaV Status	EtOH% (v/v)	pH	TA (g/L)	RS (g/L)	AA (g/L)
CH 1a	-	16.1 ± 0.2*	3.6 ± 0.2*	5.2 ± 0.1	1.9 ± 0.2*	0.1 ± 0.0*
	+	15.4 ± 0.0*	3.8 ± 0.2*	5.6 ± 0.0	1.1 ± 0.2*	0.1 ± 0.0*
ME 2 (b)	-	15.3 ± 0.1*	3.7 ± 0.2	5.2 ± 0.1	0.2 ± 0.0	0.0 ± 0.0
	+	14.1 ± 0.1*	3.7 ± 0.2	5.3 ± 0.0	0.1 ± 0.0	0.0 ± 0.0
CS 1 (a)	-	14.6 ± 0.3*	3.2 ± 0.2*	7.4 ± 0.0	0.1 ± 0.0	0.1 ± 0.0*
	+	13.0 ± 0.1*	3.2 ± 0.2*	7.1 ± 0.4	0.1 ± 0.0	0.1 ± 0.0*
CS 2 (b)	-	15.8 ± 0.1*	3.9 ± 0.2*	4.8 ± 0.0*	0.3 ± 0.0	0.1 ± 0.0*
	+	14.9 ± 0.0*	3.7 ± 0.2*	5.5 ± 0.5*	0.2 ± 0.0	0.1 ± 0.0*

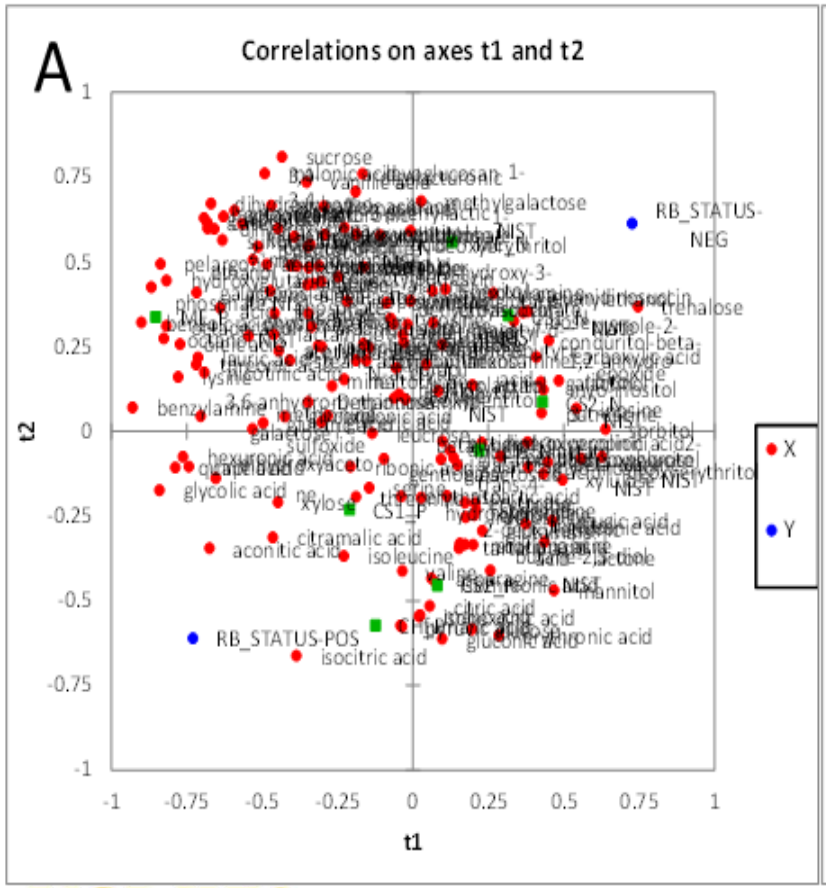
CH = Chardonnay; CS = Cabernet Sauvignon; ME = Merlot

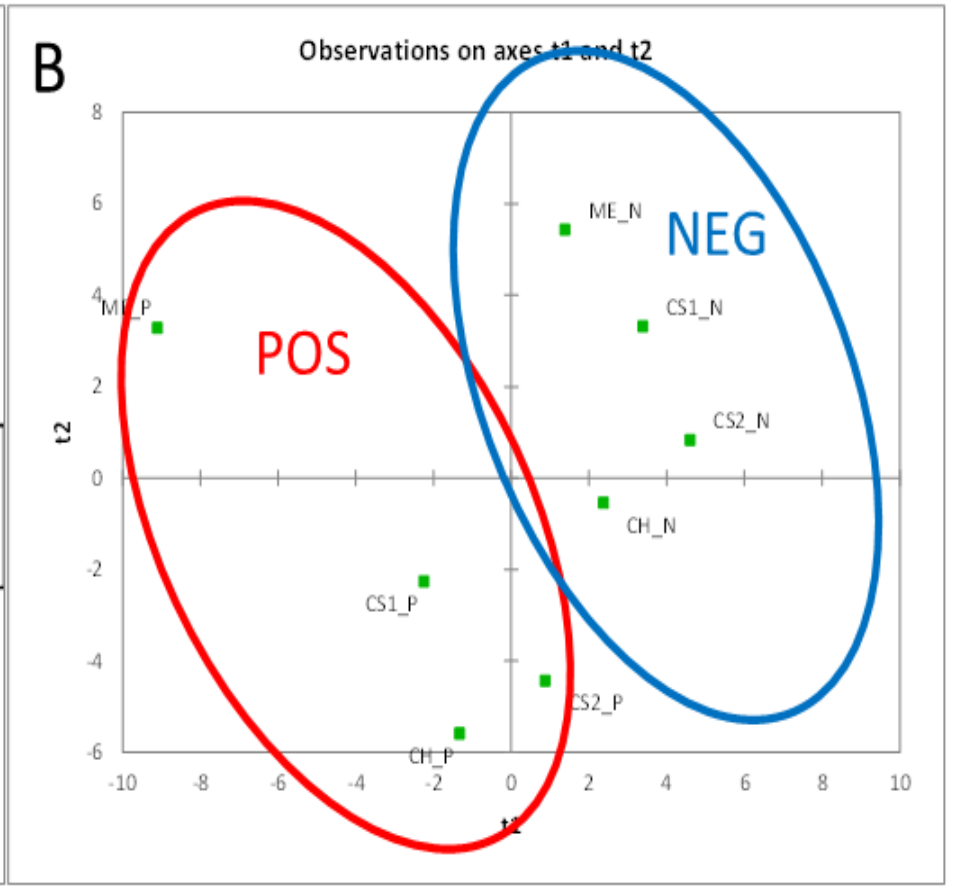
*Indicate significance at $p < 0.05$ within a site

Results: Phenol analysis of wines

- CH only exhibited small differences (RP-HPLC) due to white winemaking protocols
- For both CS sites RB(+) wines signf < pol pigm + phenols
  - Not supported by AH-results
- CS 2 RB(+) signf < anth > quer-glyc

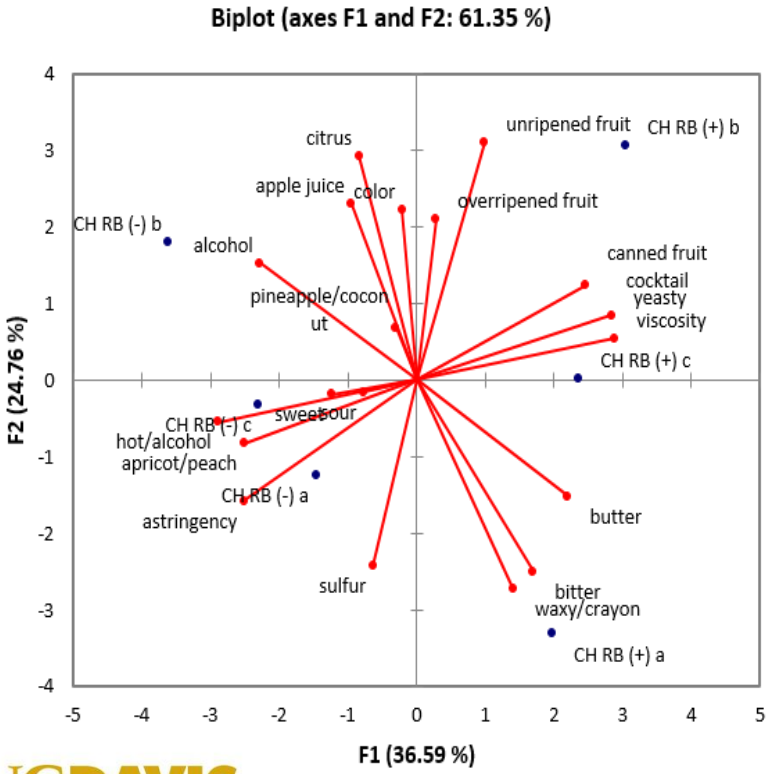
PLS-DA of wine metabolomics data

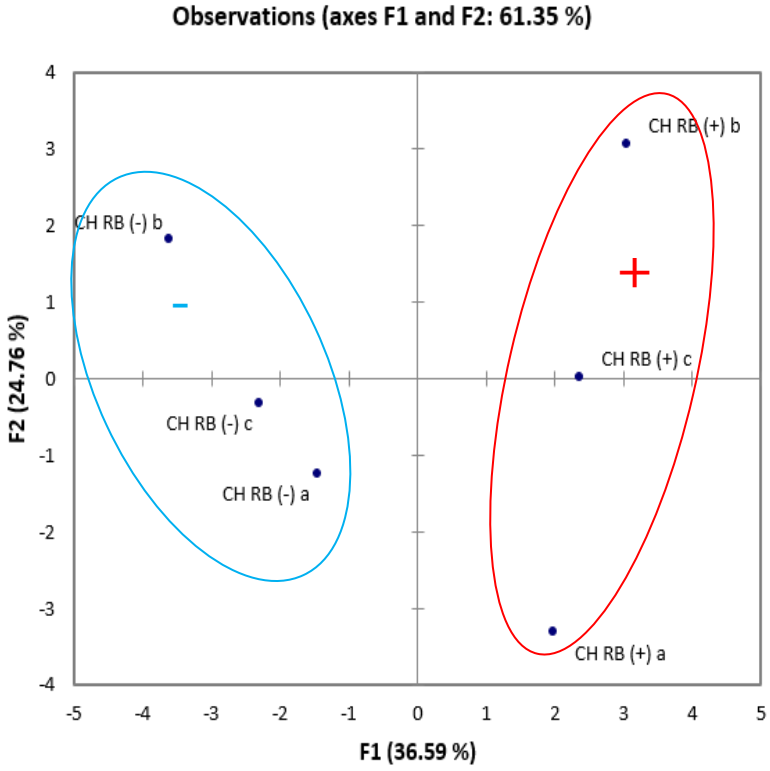




White wine sensory data

PCA scores and loading plot





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PCA separation of the wines although very little diffr

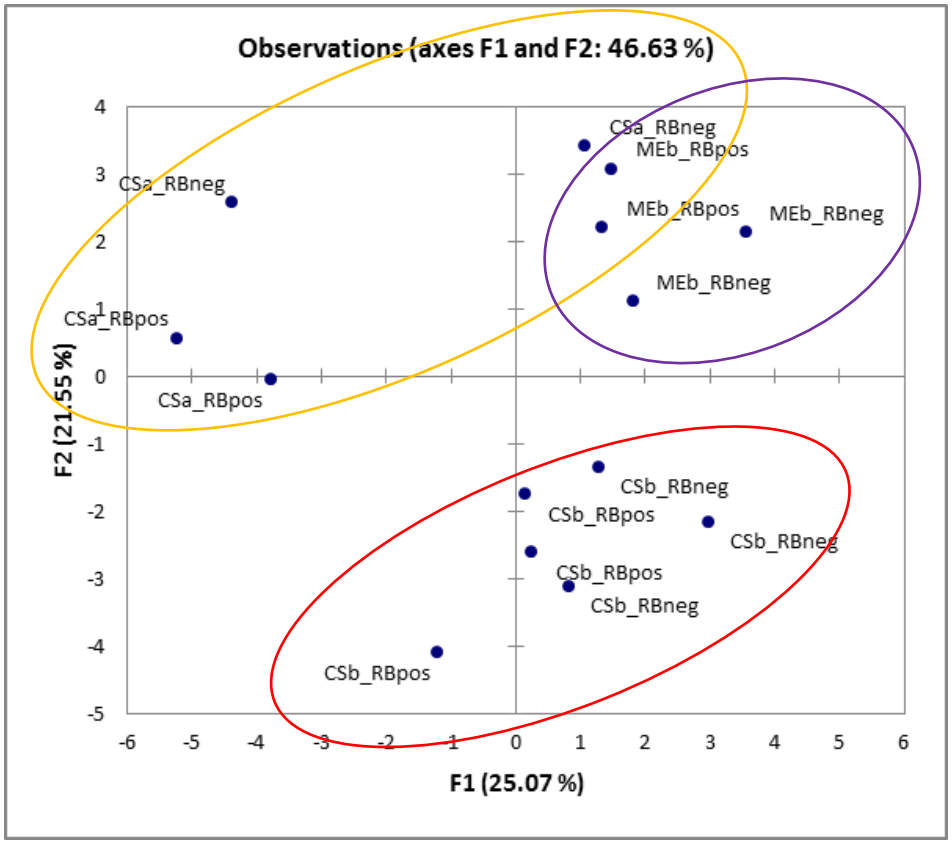
- **Only 1 out of 18 attributes sigf diffr**

Corrected F values for red DA attributes

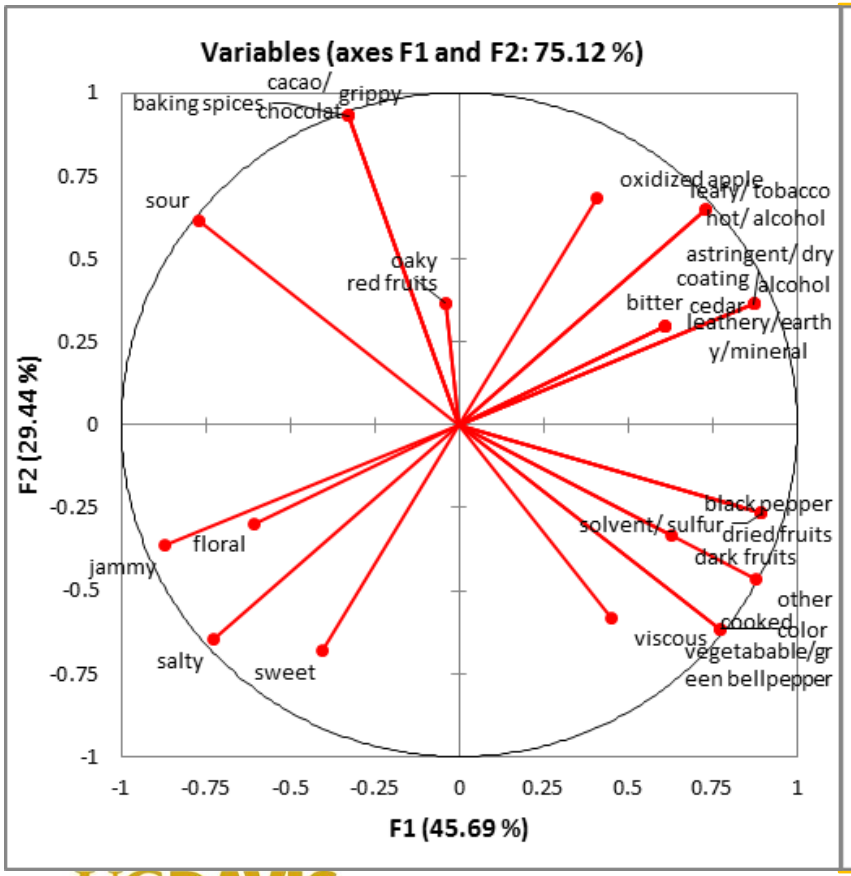
Attributes	F value wine	Significant
red fruits	1.184	no
dark fruits	1.393	no
dried fruits	2.744	yes**
oxidized apple	0.484	no
jammy	0.654	no
cooked vegetables/green bellpepper	1.551	no
leafy/tobacco	2.382	no
ceder	1.085	no
leathery/earthy/mineral	0.874	no
okay	0.970	no
alcohol	3.405	yes***
solvent/sulfur	0.520	no
baking spices	0.586	no
black pepper	0.805	no
cacao/chocolate	1.666	no
floral	1.135	no
sweet	1.994	yes
sour	3.798	yes
salty	1.418	no
bitter	1.753	no
coating	2.205	yes*
viscous	0.579	no
astringent/dry	6.484	yes***
grippy	2.205	yes*
hot/alcohol	2.587	yes**
color	1.630	no

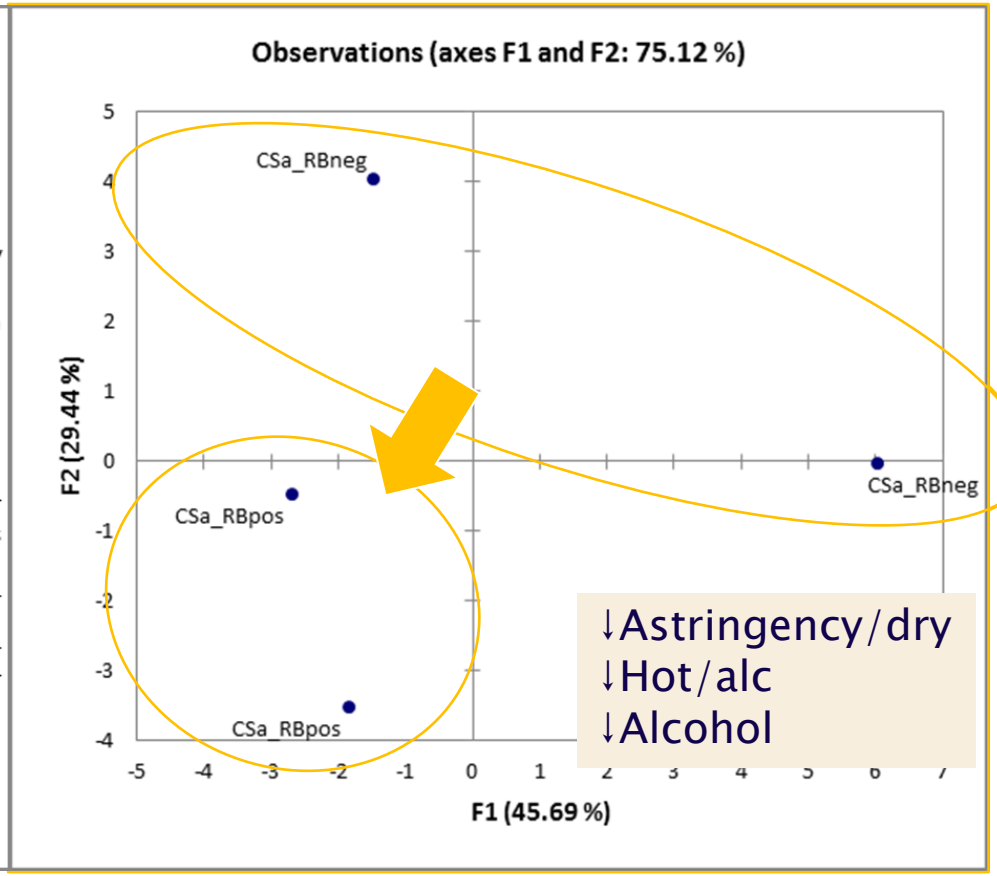
*, ** and *** indicate significance at respectively: $p < 0.05$, $p < 0.01$, $p < 0.0001$

PCA score plot



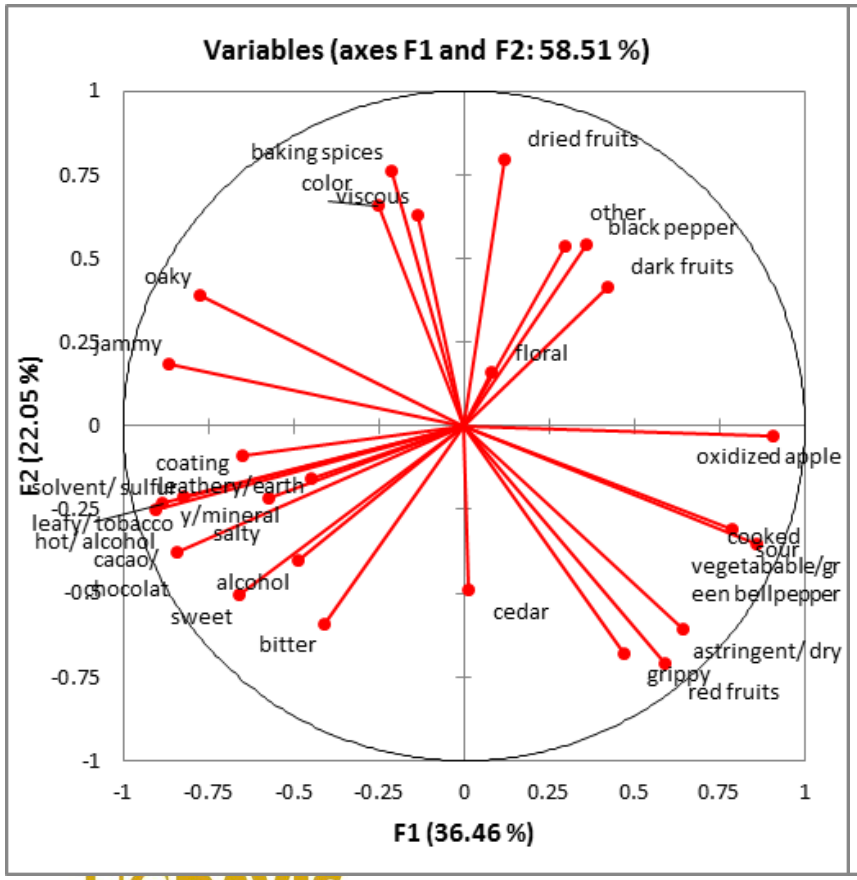
PCA: Descriptive analysis of CS (1)a

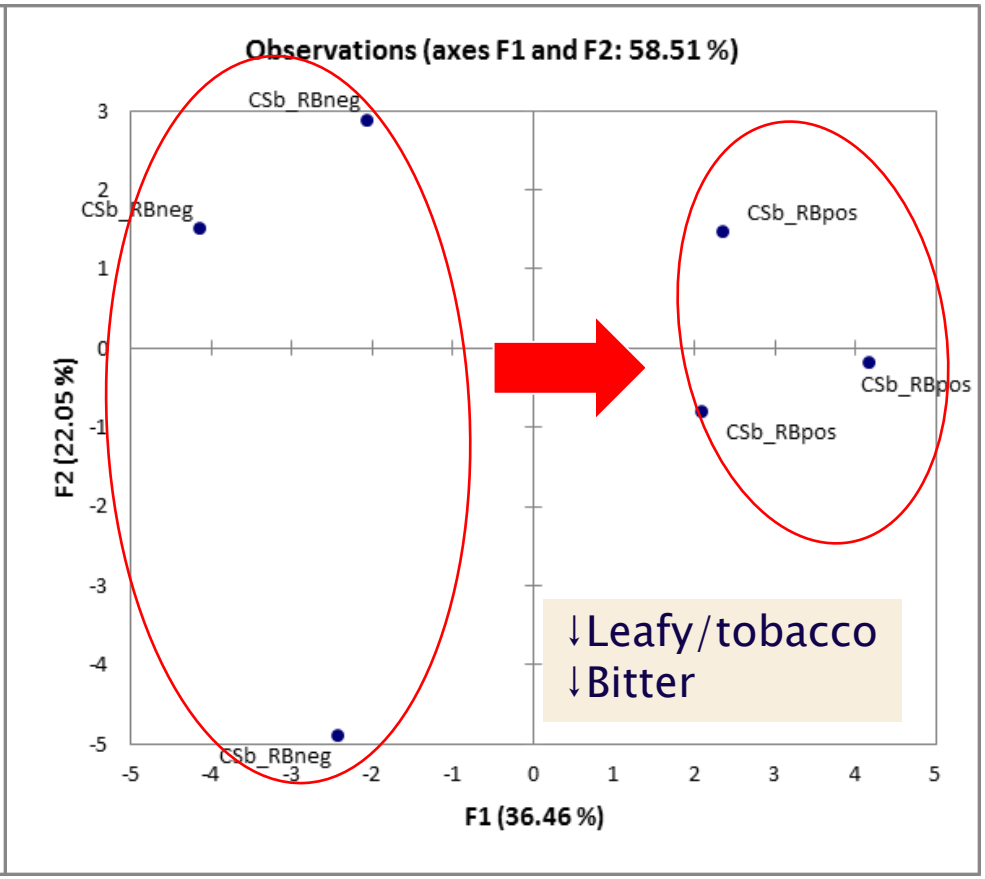




- Phenolic analyses: RB ↓ [tannin], [pol pigments] and % Alc

PCA: Descriptive analysis of CS (2)b





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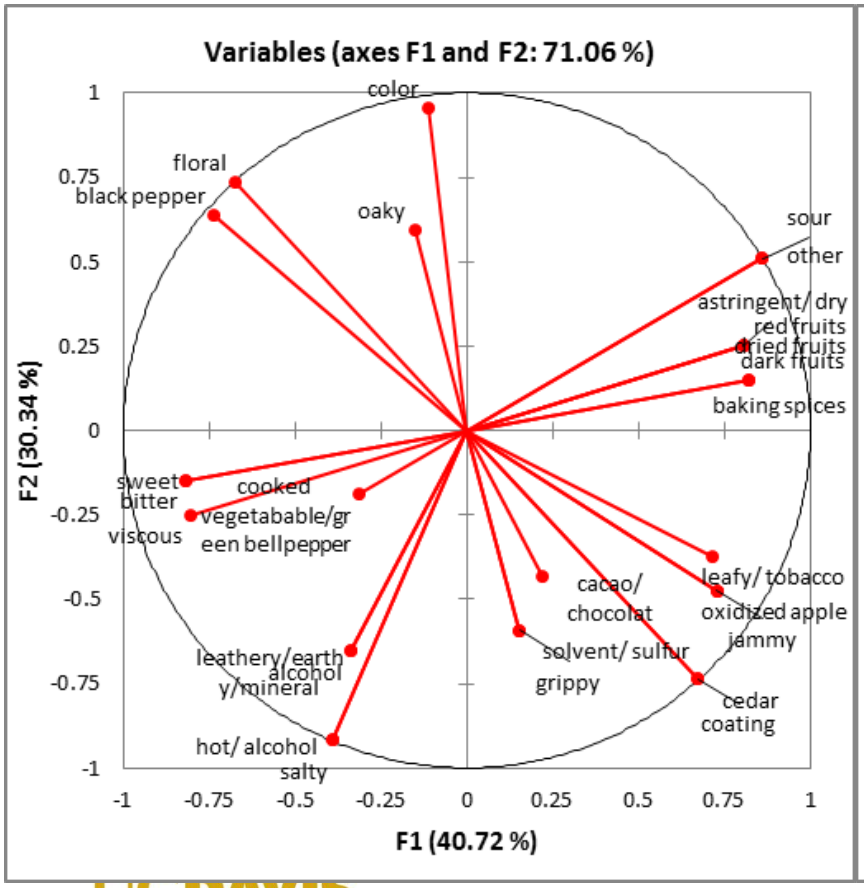
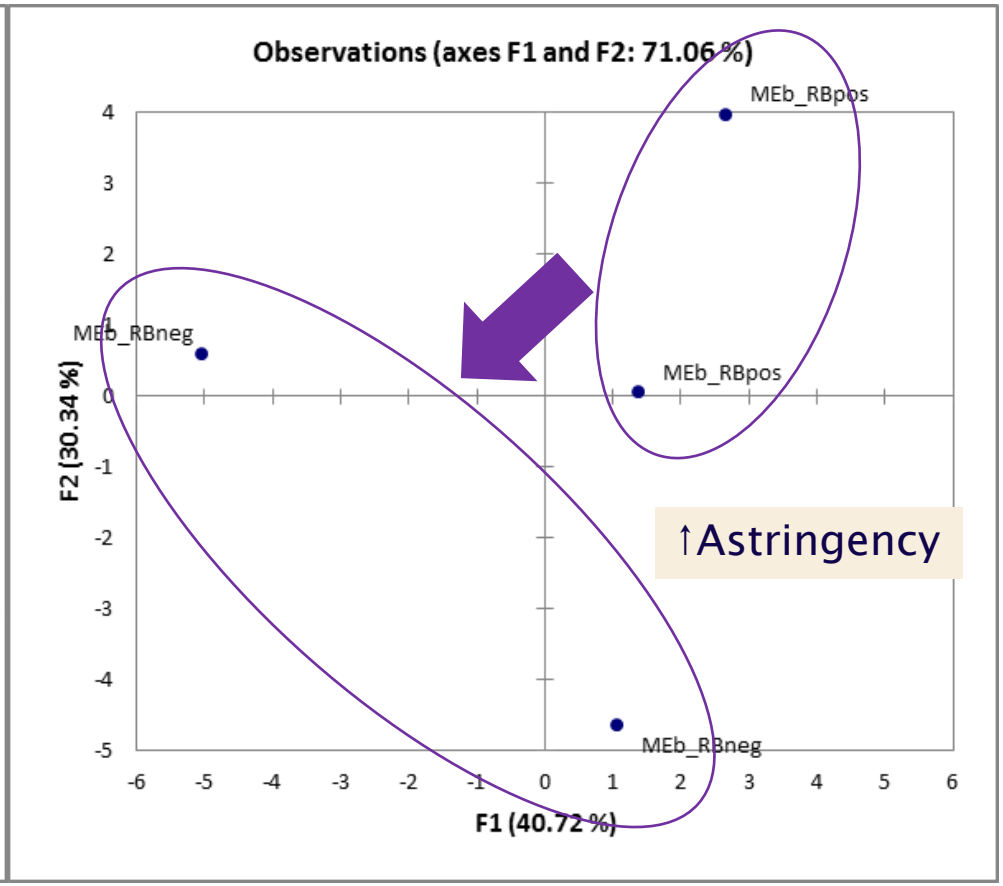
- Phenolic analyses: RB only small differences
- ↓ [anth], [pol pigments], [flavanols]
- ↑ [tannin], [flavonols], % Alc

PCA: Descriptive analysis of ME (2)b





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- Phenolic analyses: RB(+) ↑ in most phenols including [tannin], [anth]

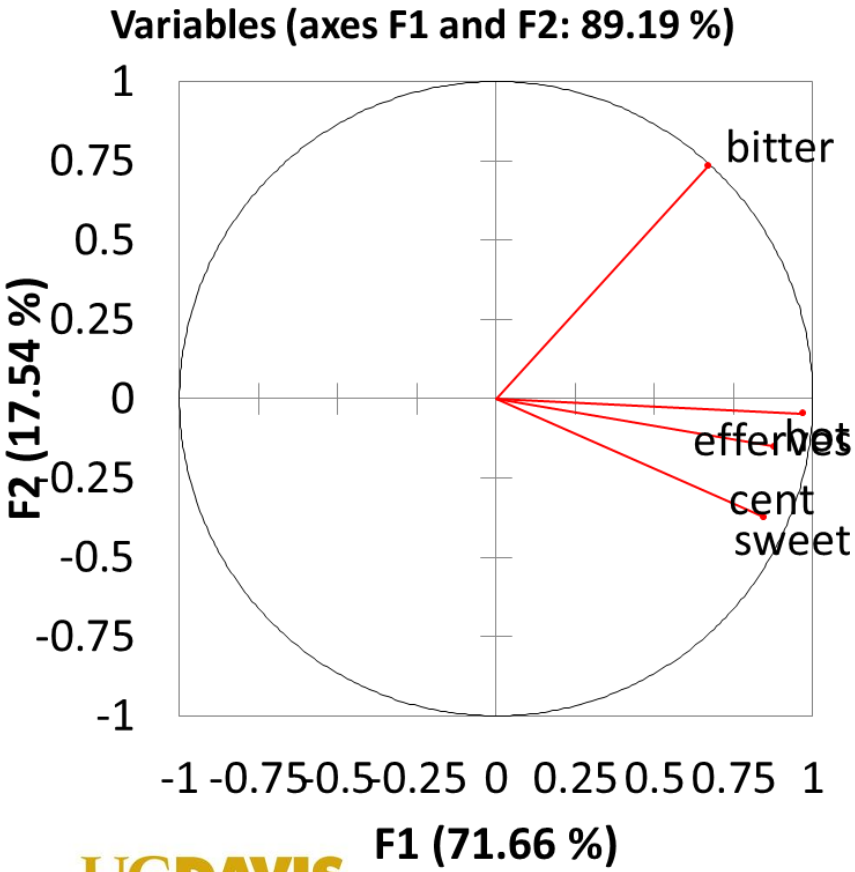
In Summary

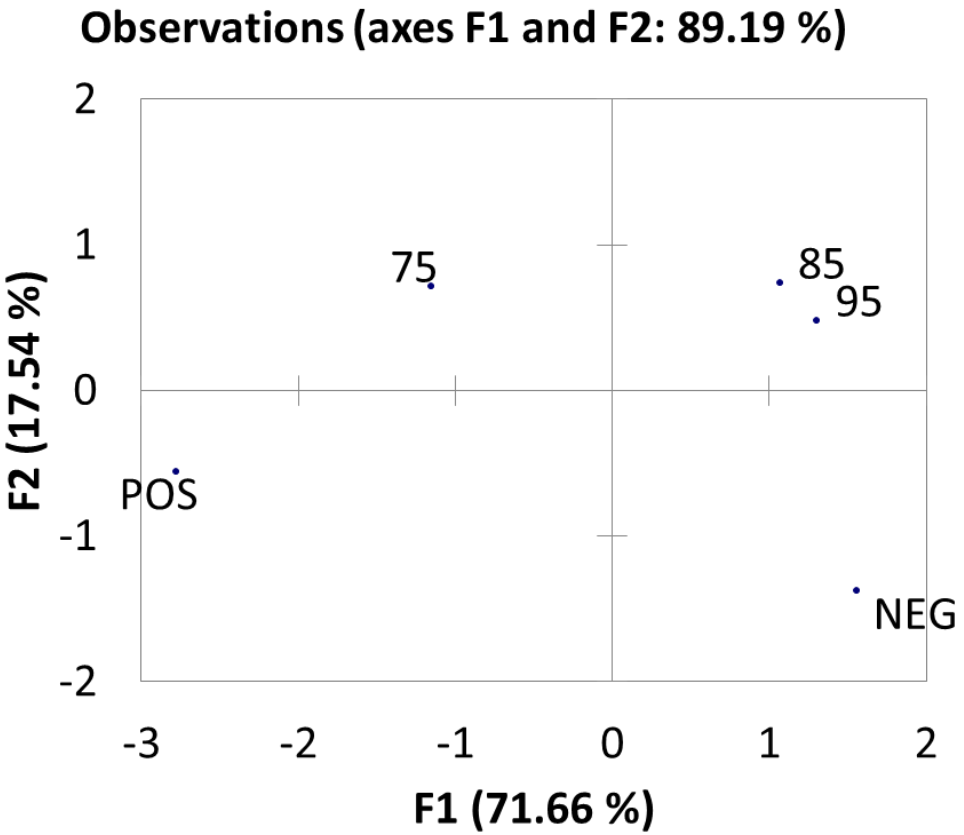
- Relation between RB disease expression (phenology) and compositional and quality impact
  - qPCR indicated similar levels of GRBaV
- Results indicate RB impact is not variety but site specific
- Untargeted metabolomics indicated large impact on primary metabolites
  - Organic acids
  - Sugars
  - Amino acids
  - Polysaccharides
- Some volatile and non-volatile secondary metabolites (phenols, aroma precursors)

Next Steps

- Determine seasonal impact
- Relation between phenological expression and altered grape and wine composition and quality
- How to deal with RB infection in the vineyard
  - Selective harvesting?
  - Making wine with 0, 5, 15 and 25% RB(+) fruit included
    - Chemical (volatile and non-volatile) and sensory profiling

Averaged fermentation reps – signf attributes





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Next Steps

- Make wines from RB (+) and (-) grapes with the same sugar content
- Continue to explore impact of site on variety impact
  - Find correlation with soil, nutrient…..
- Targeted analysis combined with transcriptomics to identify metabolic pathways altered by RB disease resulting in changes in biochemical composition
- Use impact on gene expression to develop potential counter measures

Acknowledgements

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- Napa Valley Grapegrowers
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THANK YOU