

# **Inception, progression, and compositional consequences of the sugar accumulation disorder (SAD)**

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# Sugar Accumulation Disorder (SAD) a.k.a. 'Berry shrivel'

- Impeded sugar accumulation
- Healthy appearing rachis
- Poor color development (in red varieties)
- Premature berry desiccation

**BSN**

**SAD**

BSN

BS

**BSN**

**SAD**

**Dead rachis,  
sweet (up to  
33.5° Brix),  
raisin-like  
flavor**

**Live rachis,  
sour (rarely  
more than 23°  
Brix), poor  
coloration**

BSN

BS



SAD



BSN



**Cabernet  
Sauvignon and  
Sauvignon blanc  
clusters  
displaying  
symptoms of  
SAD.**

# SAD

The background of the slide is a photograph of a vineyard. It shows several clusters of ripe, dark blue grapes hanging from the vines. The leaves are a mix of green and yellow, suggesting some autumnal change. The overall scene is brightly lit, with a slightly hazy or soft-focus quality.

Grapes displaying SAD have been seen:

- Throughout California
- In Oregon and Eastern Washington
- In Europe (Austria)

Sample	Sample Date	condition	Berry weight (g) ± SD	Brix ± SD	pH ± SD
Round Pond (Rutherford) Cabernet S.	8/14/07	SAD	0.84 ± 0.09	11.2 ± 0.3	3.1 ± 0.07
		Control	1.08 ± 0.04	17.4 ± 0.6	3.3 ± 0.04
Juliana (Napa) Sauv. Blanc	8/23/07	SAD	0.8 ± 0.03	15 ± 1.3	3.1 ± 0.04
		Control	1.2 ± 0.04	26.2 ± 1.2	3.6 ± 0.06
Chimney Rock (Napa) Sauv. Blanc	8/27/07	SAD	1.34 ± 0.15	13.2 ± 2.1	3.3 ± 0.07
		Control	1.7 ± 0.08	25.8 ± 0.9	3.6 ± 0.09
Oakville Exp. V. (Napa) Cabernet S.	9/24/07	SAD	1.00 ± 0.1	19.3 ± 0.9	3.3 ± 0.1
		Control	1.11 ± 0.03	23.2 ± 3.2	3.4 ± 0.1
Jordan (Sonoma) Cabernet S.	10/7/07	SAD	0.73 ± 0.1	15.5 ± 2.0	3.2 ± .06
		Control	1.06 ± 0.01	22.8 ± .8	3.5 ± .03
Chimney Rock (Napa) Cabernet (mature)	10/16/07	SAD	0.82 ± 0.008	13.8 ± 1.4	3.4 ± .1
		Control	1.04 ± 0.05	22.4 ± 1.0	3.7 ± .05
Chimney Rock (Napa) Cabernet (young)	10/23/07	SAD	0.77 ± 0.03	15.6 ± 1.4	3.4 ± .07
		Control	0.89 ± 0.07	22.2 ± .9	3.5 ± .03
<b>SAD Average</b>			<b>0.9 a</b>	<b>14.8 a</b>	<b>3.26 a</b>
<b>Control Average</b>			<b>1.15 b</b>	<b>22.9 b</b>	<b>3.51 b</b>

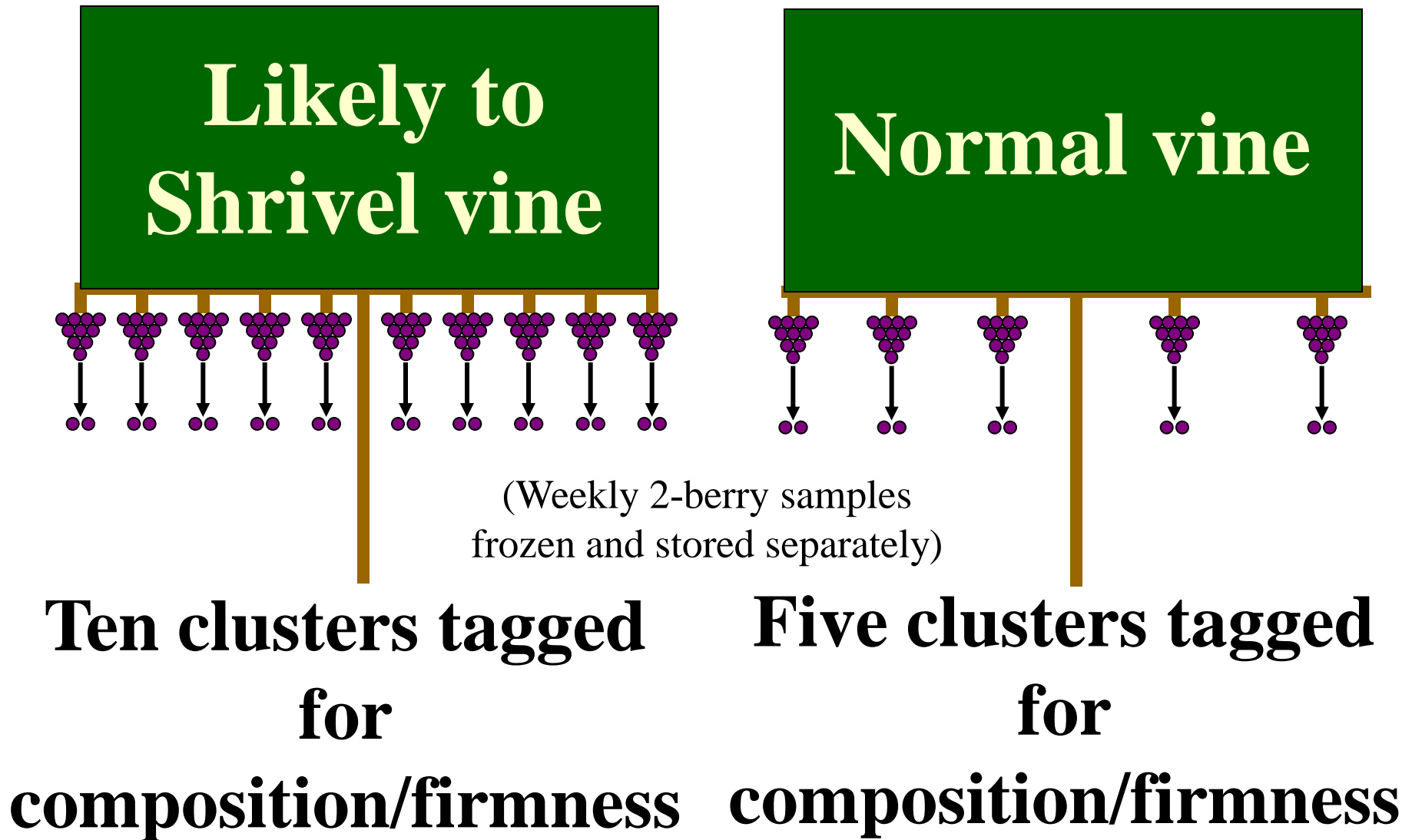


Our work originated at the  
Oakville Experimental Vineyard

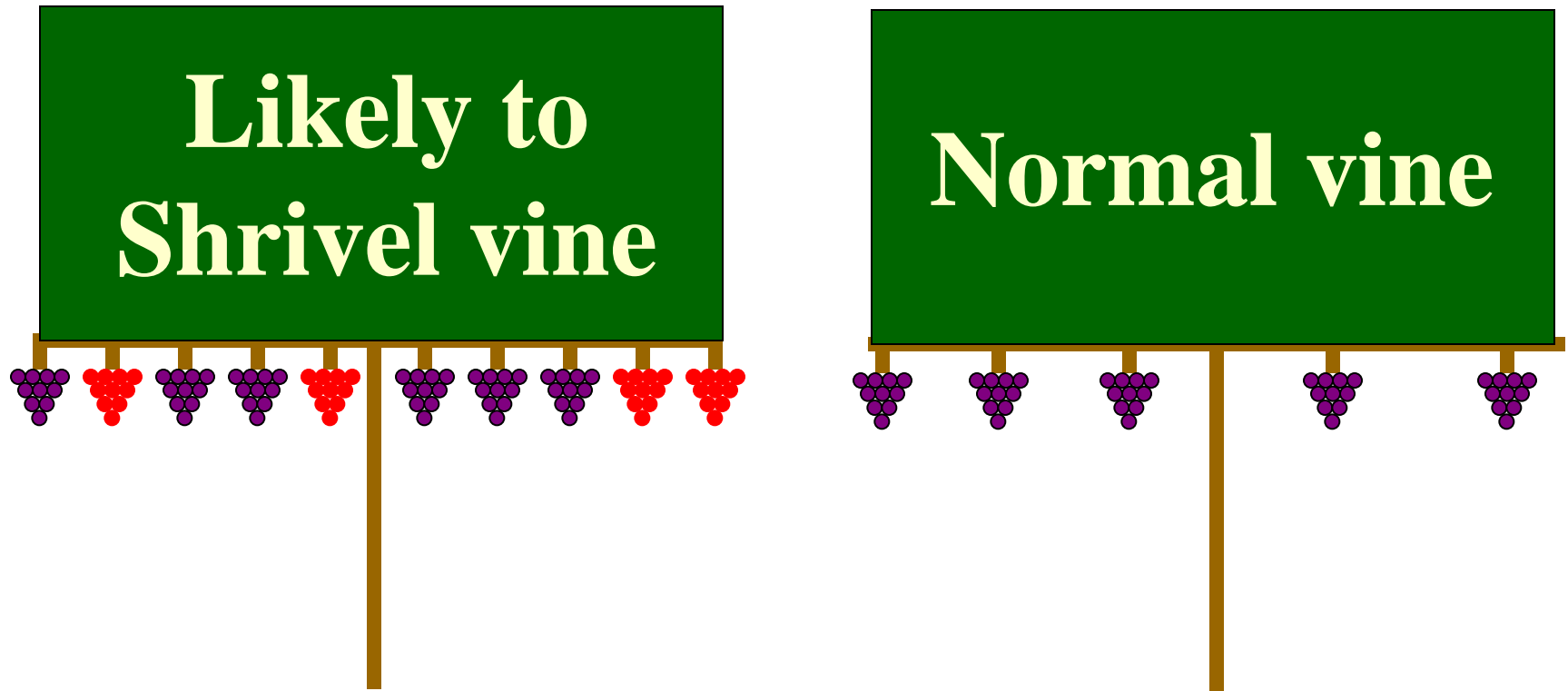
With Cabernet Sauvignon vines that  
had consistently shown berry shrivel  
symptoms over several seasons


Some clusters showed shrivel and  
others did not.

# Sampling protocol: pre-symptom expression



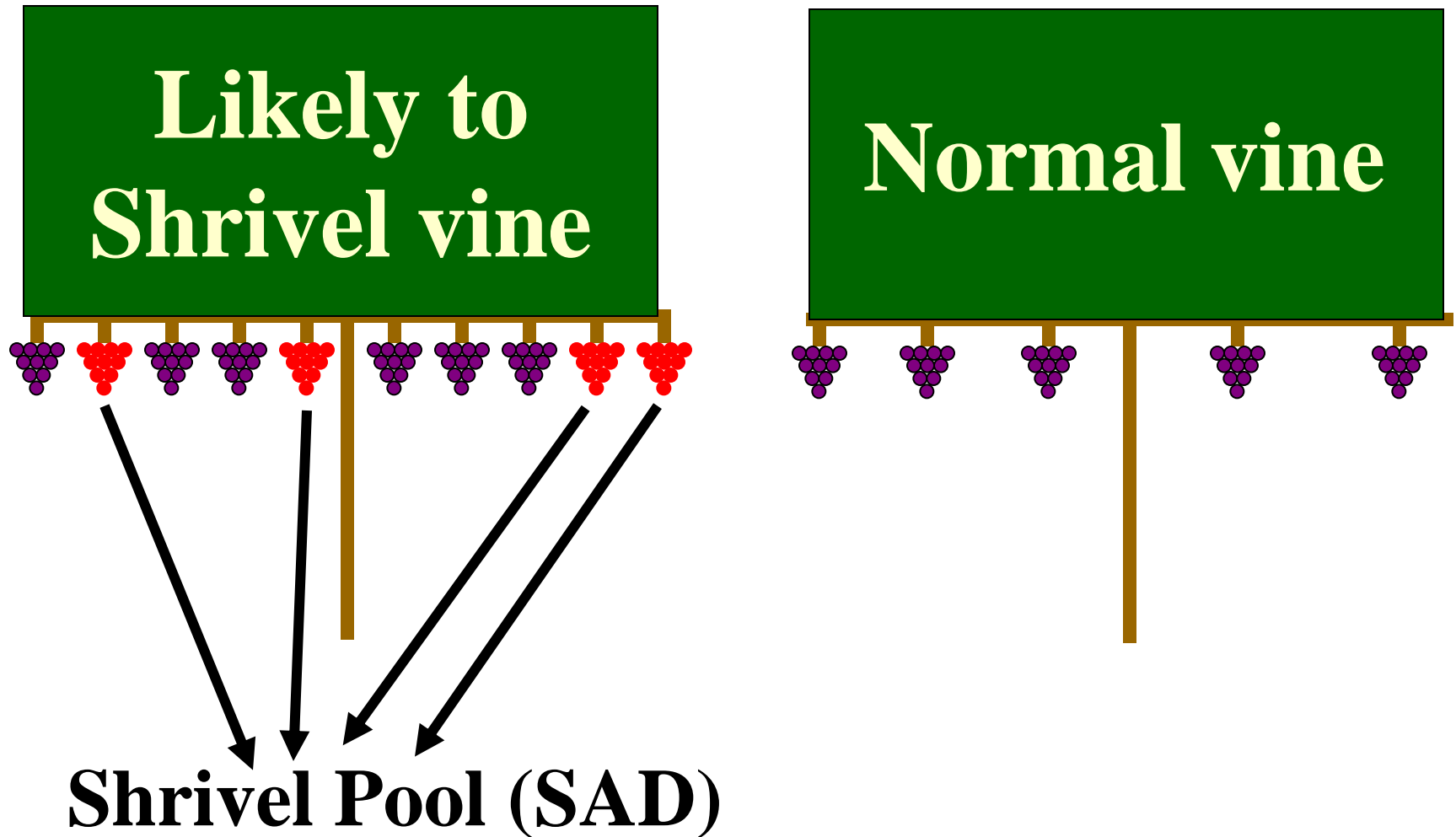
# Symptom expression



“Healthy” clusters 

Shriveled clusters 

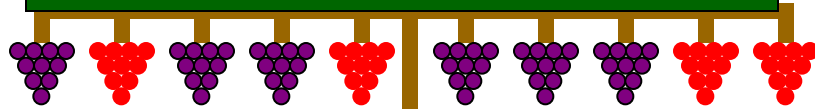
# Pooling 2-berry samples within/between vines



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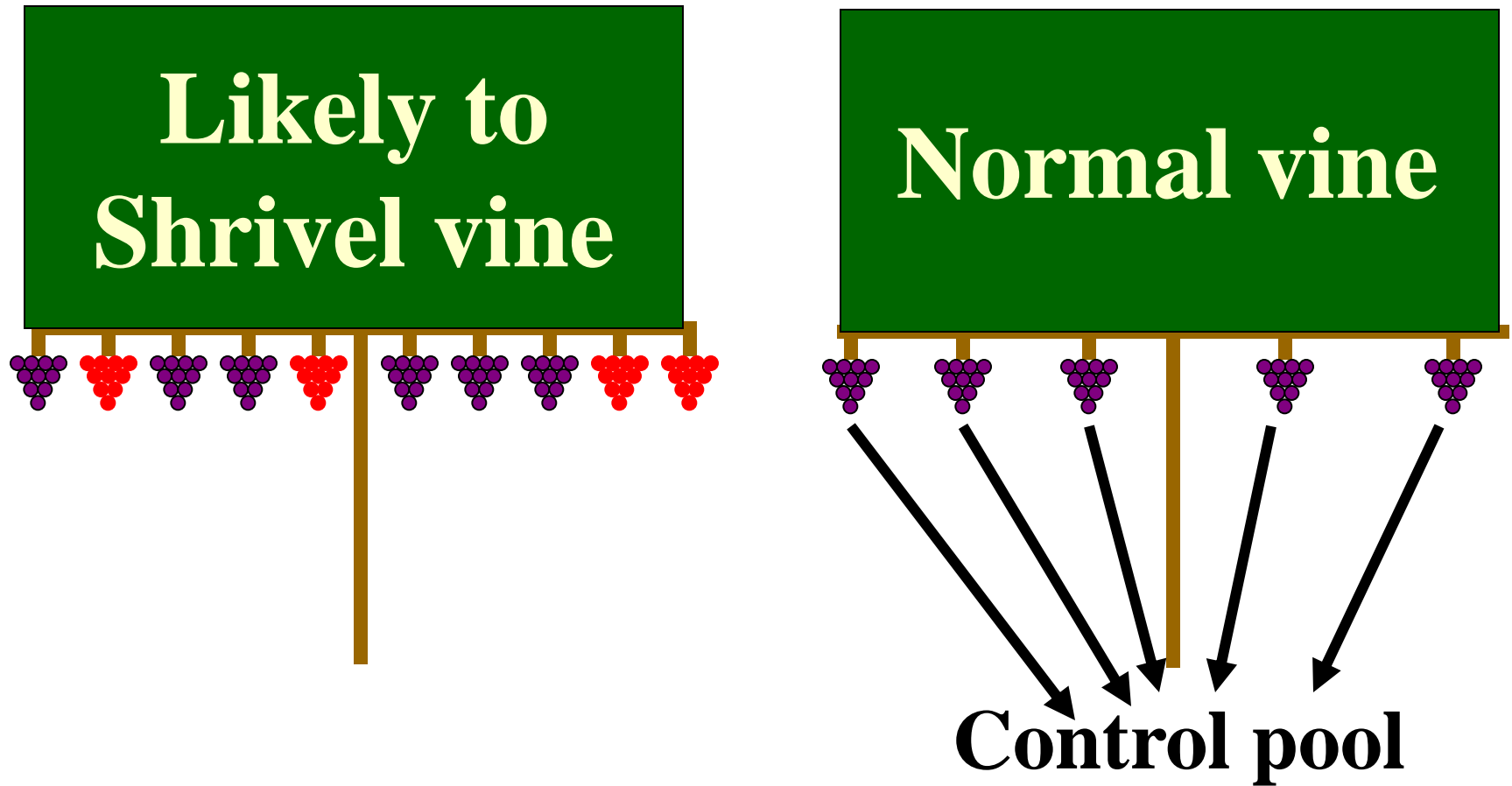
Likely to  
Shrivel vine

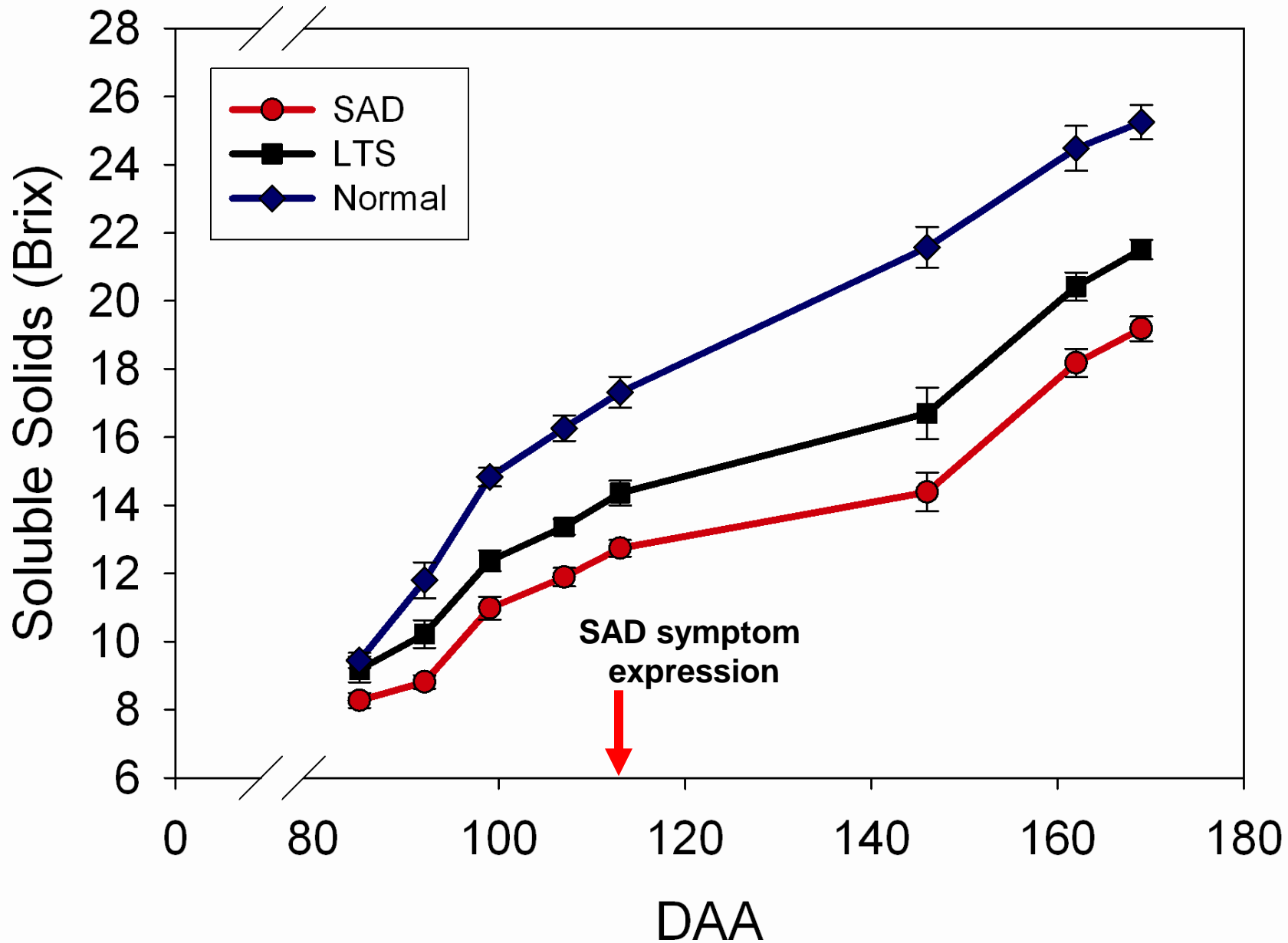
Normal vine

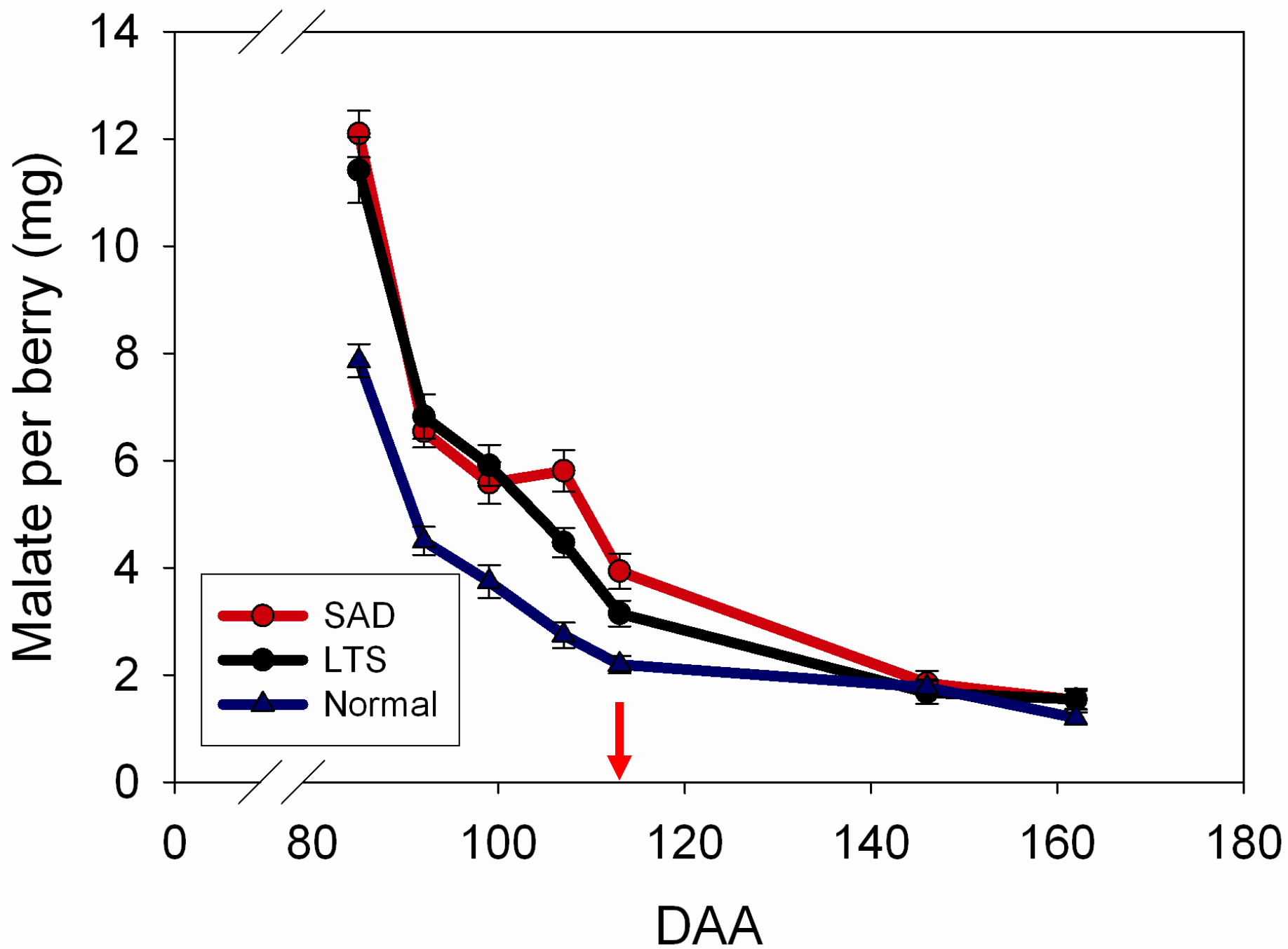


“Likely to shrivel” (LTS)  
pool

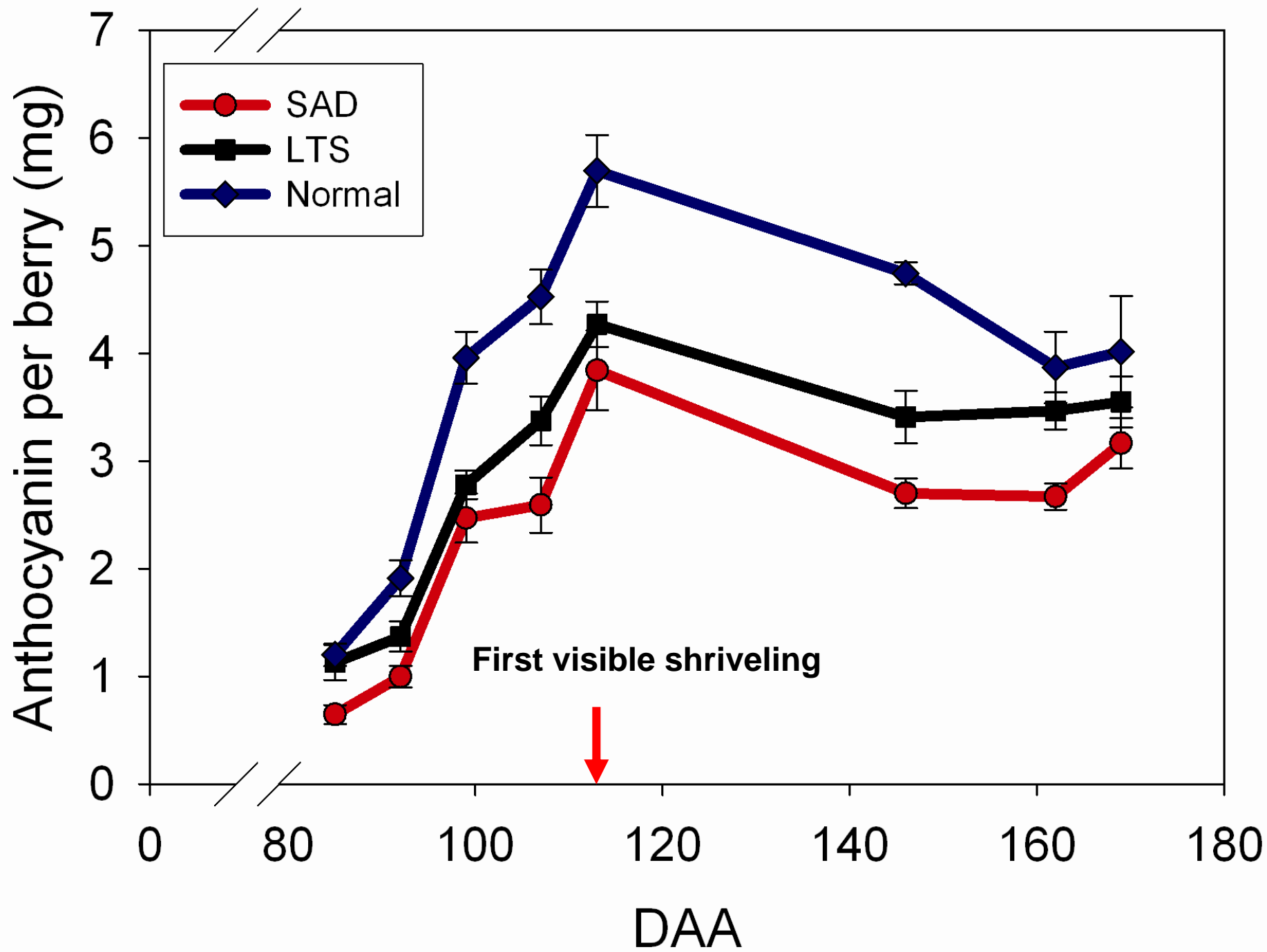
# Pooling 2-berry samples within/between vines

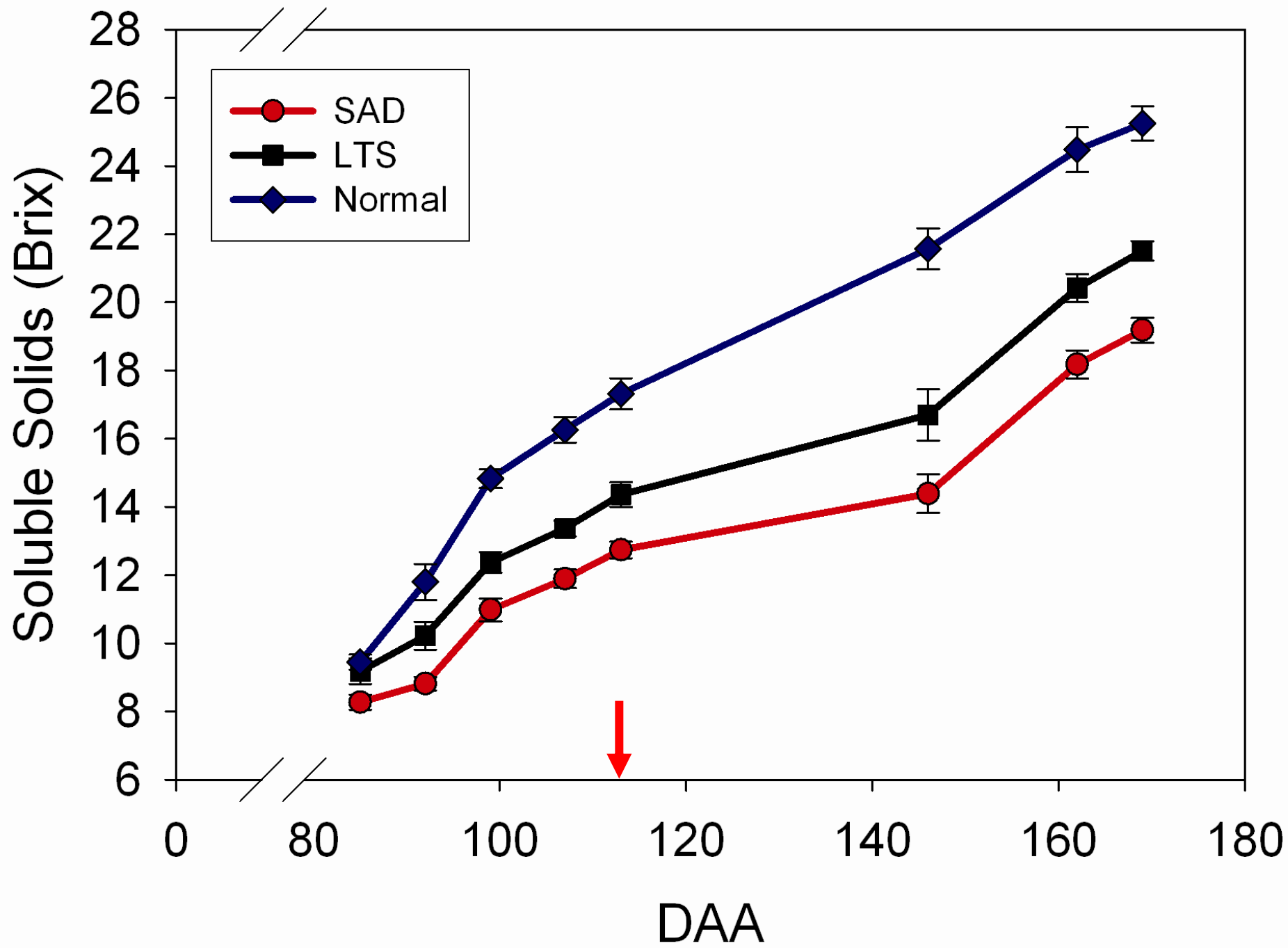


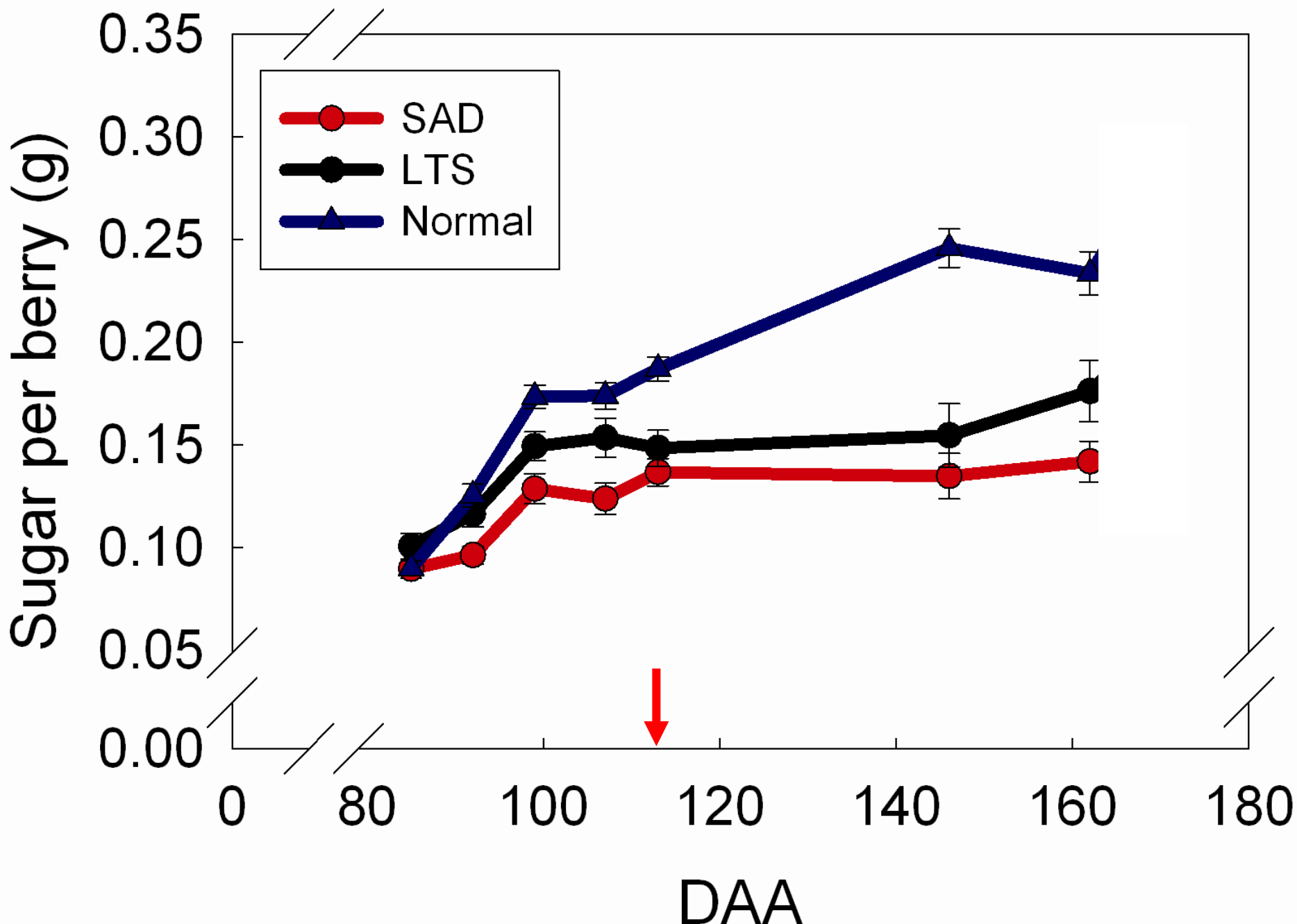










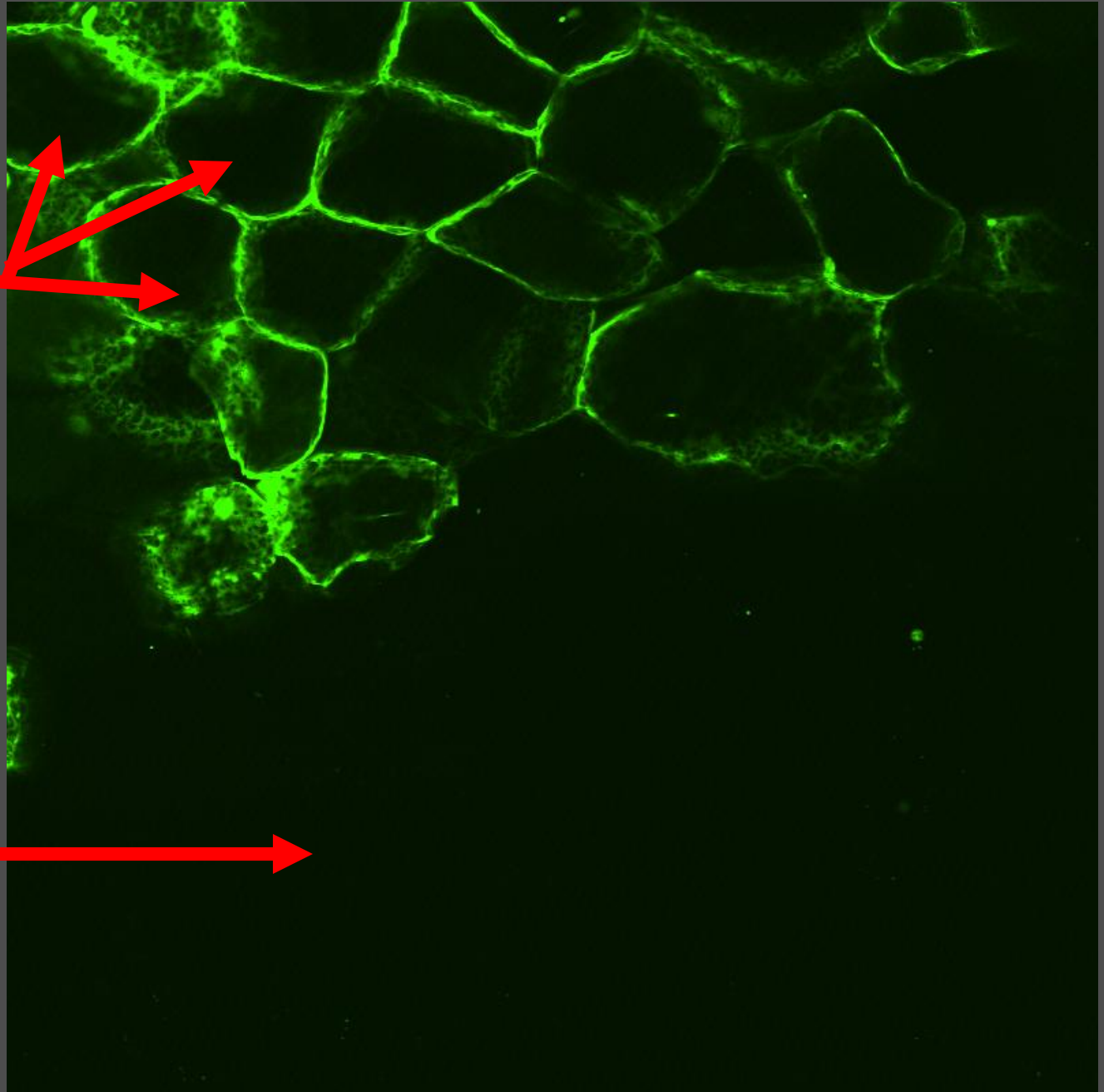


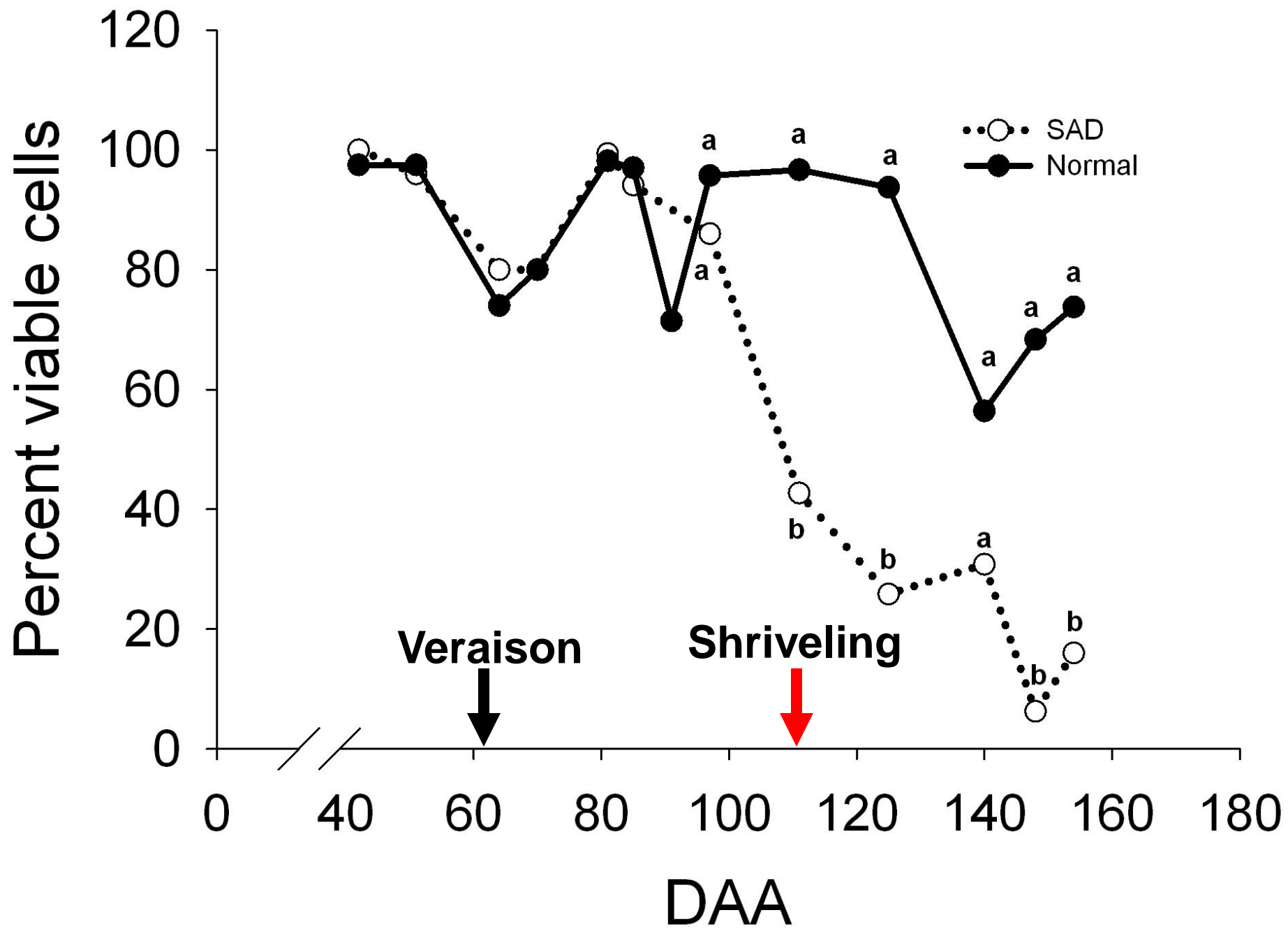
# Fluorescein Diacetate cell viability staining in fruit – Ken Shackel

- Allows differentiation of living versus dead cells

Living cells

Dead cells





# SAD symptom development

- SAD fruit has lower Brix, pH, and anthocyanin concentration than control fruit

SAD clusters stop accumulating sugars weeks before visible symptoms

- Cells die as symptoms become visible
- Vine phenomenon - nonsymptomatic clusters on SAD vines develop similarly to SAD clusters

Is shrivel [SAD] the result of impaired phloem transport?

Stopping sugar import by girdling the cluster should mimic SAD



# Girdling experiments to mimic SAD

Girdling was done with a knife above and below the peduncle to stop sugar transport to the berries.

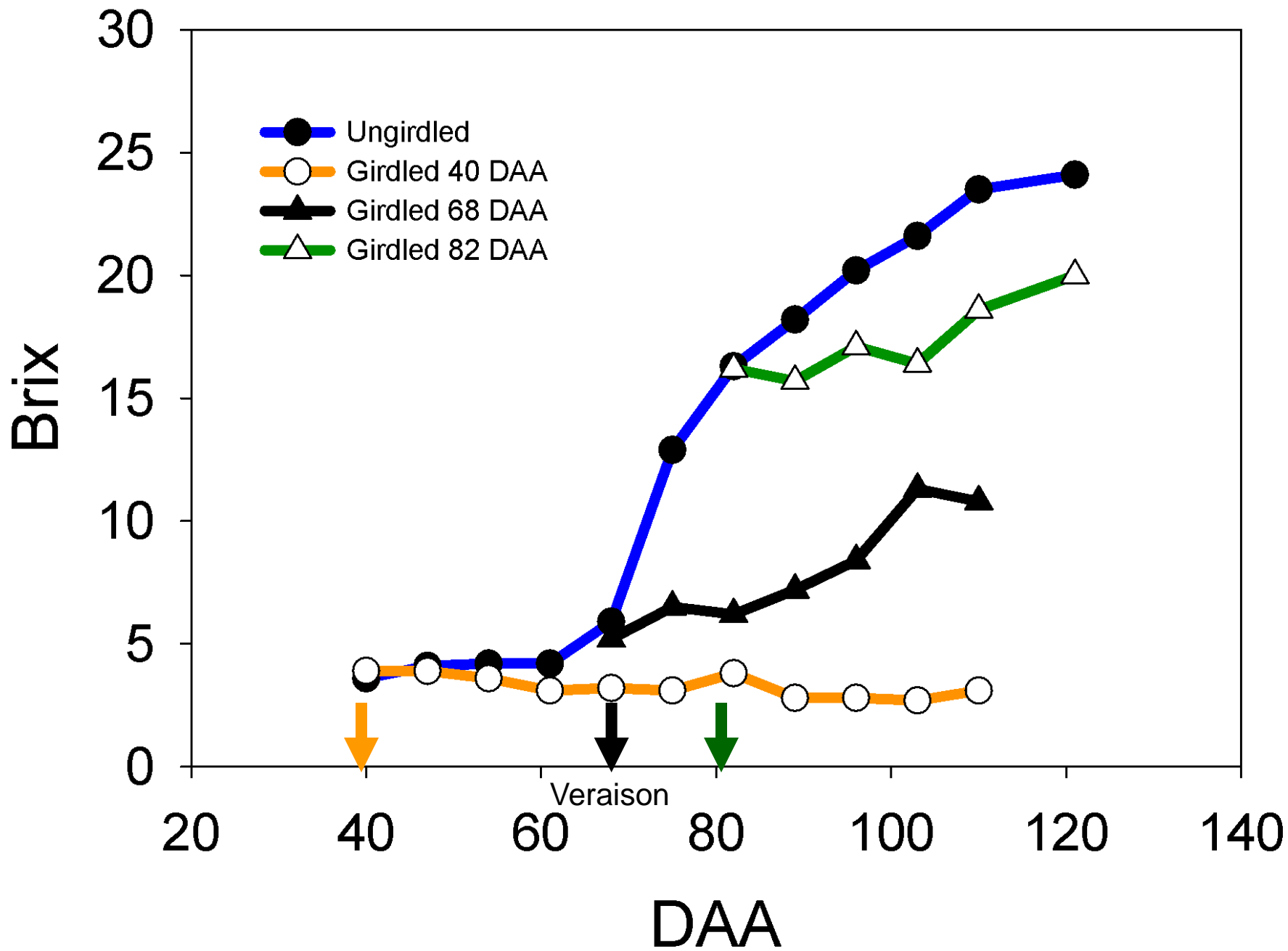


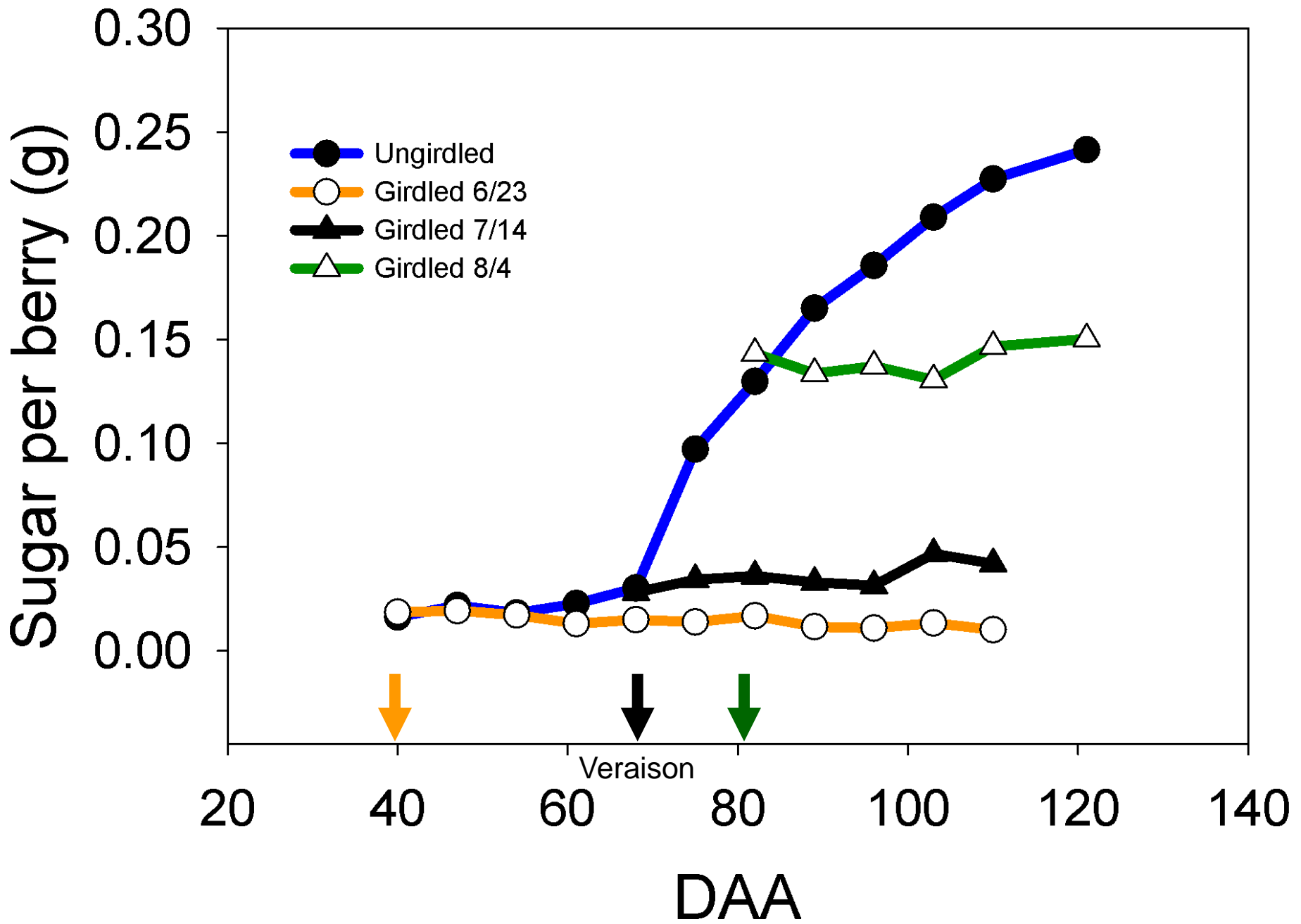
# Cabernet Sauvignon

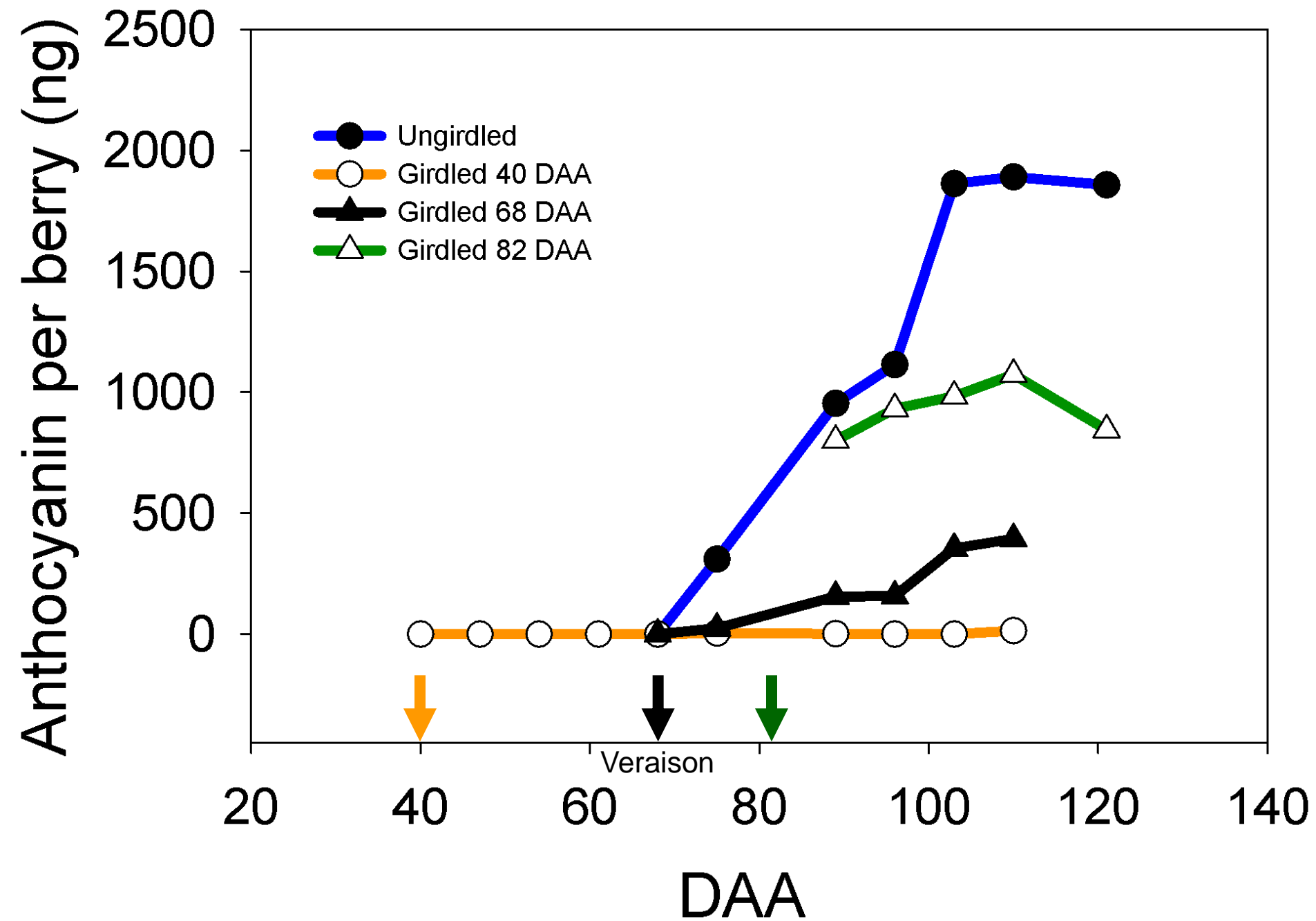
Not  
girdled



Girdled  
before  
veraison.  
Picture  
taken  
103 DAA  
(63 DAG)







Yes, girdling mimics SAD with respect to berry growth, sugar accumulation, and color development.

# Crop Thinning Experiments



**Clusters dropped  
at set**



**Greenest clusters  
dropped at veraison**

**2007  
% SAD**

<b>Site</b>	<b>Unthinned</b>	<b>Set thinned</b>	<b>Veraison thinned</b>
Stag's Leap	1.0 a	0 a	0.3 a
Sonoma	6.3 a	0.9 a	4.7 a
Rutherford River	3.2 a	6.5 a	6.5 a

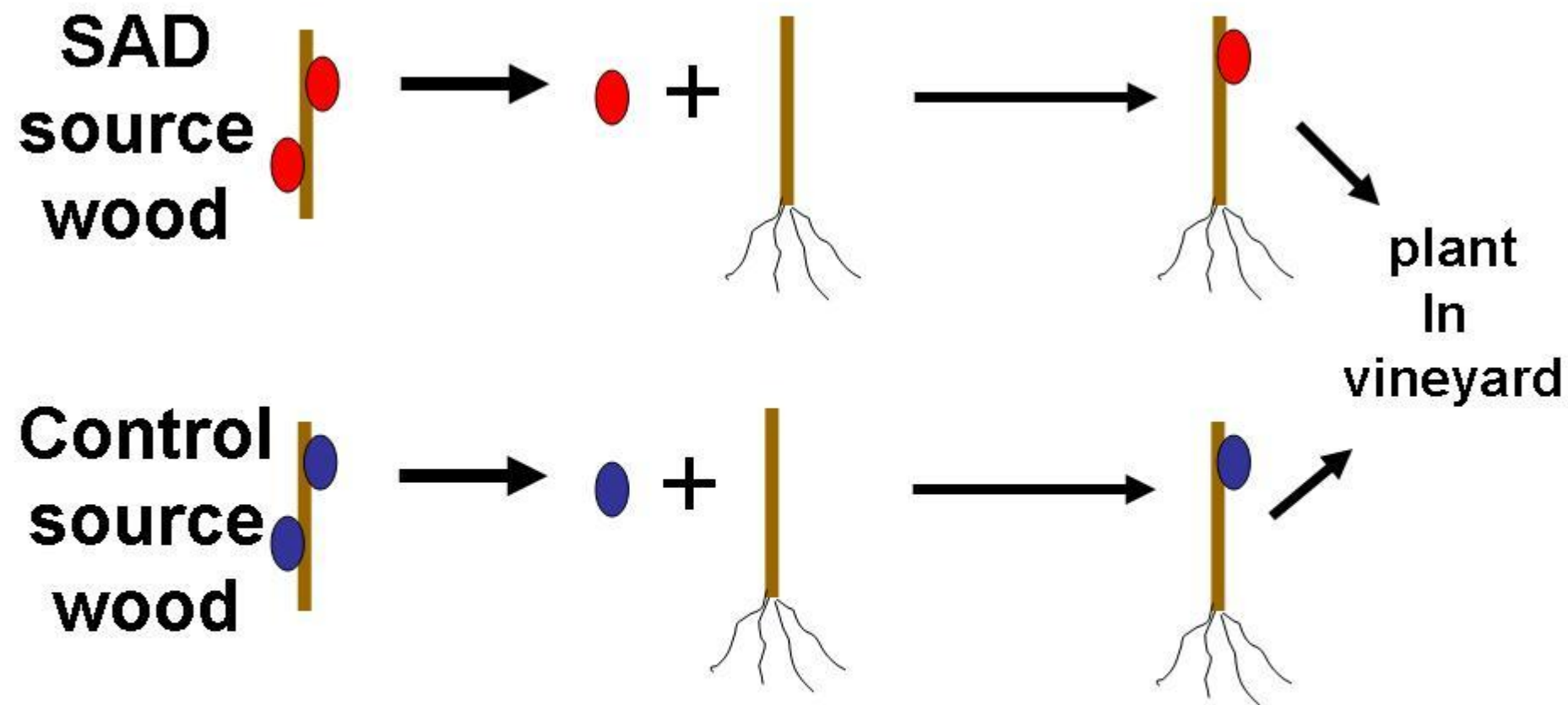


2008

% SAD --- same lack of thinning effect

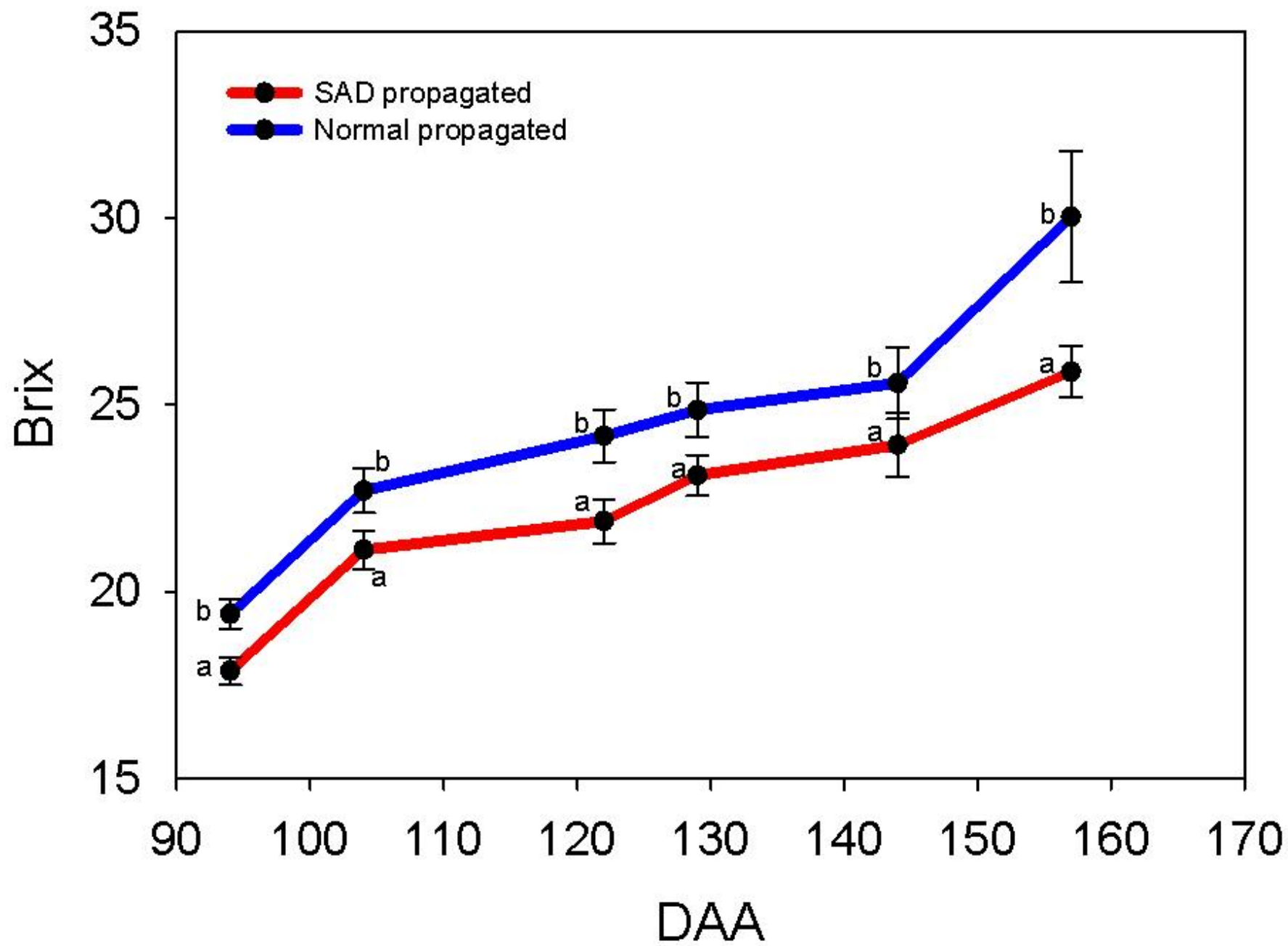
Thinning crop has not diminished incidence of SAD symptoms in remaining fruit.

# Schematic of SAD propagation experiment



Vineyard plot design

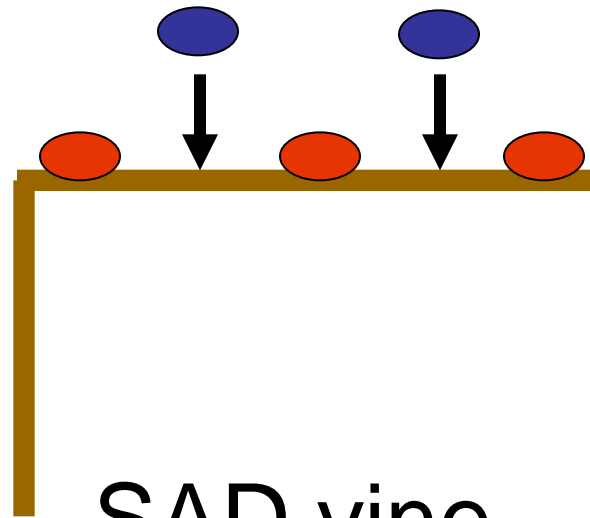
SAD	Cont	SAD	Cont	SAD	Cont	SAD	Cont	SAD	Cont
Cont	SAD	Cont	SAD	Cont	SAD	Cont	SAD	Cont	SAD



# 2007 Budding Experiments

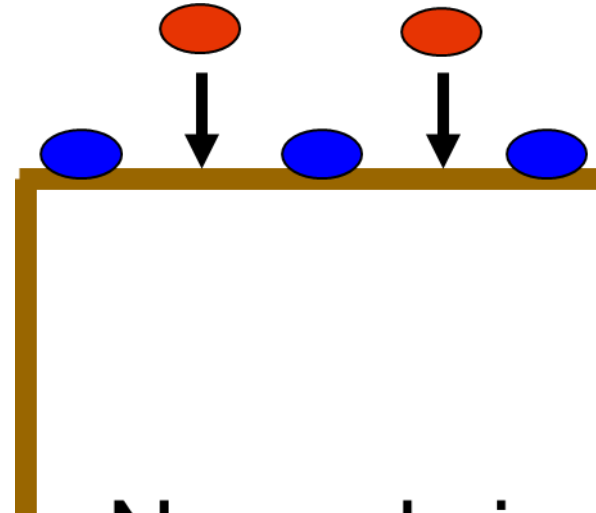
Chip budding from normal to SAD vines and SAD to normal vines

Normal buds



SAD vine

SAD buds



Normal vine



30 vine sets  
(15 SAD, 15 healthy)  
were planted in:

- Oakville (1773 GDD)
- Davis (2069 GDD)
- Hopland (1937 GDD)
- Parlier (2469 GDD)

Normal  
propagated vine



SAD  
propagated vine



Normal source



SAD source



# 2009 Davis vine harvest data

<b>Sample</b>	<b>Harvest weight (g/vine)</b>	<b>Brix</b>
Normal propagated (n=10)	442 <b>a</b>	25.7 <b>a</b>
SAD propagated (n=11)	372 <b>a</b>	17.6 <b>b</b>

Similar results at all locations in 2010.



Controls:  
Source Brix  
3A 22.0a  
4A 21.9a  
2A 20.5a

SAD:  
Source Brix  
1B 18.8a  
3B 17.3 b  
5B 15.7 c  
2B 15.0 c

Clonal propagation: 3 independent wood sources of control and 4 independent wood sources of SAD, all planted at 4 locations (OEV, UCD, KAC, and Hopland). Due to budget limitations, only the OEV and UCD vines now remain.

The 2003 budding experiments  
demonstrated that SAD is  
transmissible

2007 Budding experiments demonstrated that healthy buds grafted onto SAD vines develop SAD fruit

and that SAD is able to spread  
from grafted buds to affect an  
entire vine;

And - there are consistent clonal  
differences

# SAD and pathogens

The SAD vines at OEV have been tested for the following pathogens by FPS

- Leafroll viuruses
- Fanleaf virus
- Phytoplasmas
- Tomato ringspot virus
- Grapevine vitiviruses
- Arabis mosaic virus
- *Xylella fastidiosa*
- Rootstock stem lesion associated virus
- Grapevine fleck virus
- Rupestris stem pitting associated virus

**All tests have yielded negative results,  
[or both SAD and healthy vines have  
given positive results]**

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# Conclusions

- Sugar accumulation stops prematurely
- Anthocyanin accumulation stops about same time as sugar accumulation
- SAD symptoms may arise from impaired phloem flux, like leaf roll virus
- SAD is propagate-able
- SAD at Oakville is a vine phenomenon
- SAD may arise from multiple causes, including a pathogen

# Acknowledgements

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- **Ed Weber and Rhonda Smith**
- **Jason Benz**
- **Daniel Robledo - grafting**
- **Nate Weis**
- **Derek Cronk**
- **Elizabeth Vianna and Doug Fletcher**
- **Dana Grande and Mark Bailey**
- **Adib Rowhani**
- **Valentina Canutti**
- **Marco LiCalzi**
- **Sue Ebeler**
  
- **AVF and NCVRG**



# Different types of SAD?

<b>Location</b>	<b>Year</b>	<b>Condition</b>	<b>Juice pH</b>	<b>Brix</b>	<b>Sugar per berry (g)</b>
Oakville	2005	Normal vines	3.71 a	25.3 a	0.350 a
Oakville	2005	LTS clusters	3.63 a	22.0 b	0.282 b
Oakville	2005	SAD clusters	3.61 a	19.2 c	0.204 c
Sonoma	2008	Normal vines	3.45 a	24.0 a	0.218 a
Sonoma	2008	LTS clusters	3.45 a	23.0 a	0.238 a
Sonoma	2008	SAD clusters	3.27 b	18.2 b	0.127 b

# Aroma compound profiling – Sue Ebeler

## Headspace solid-phase microextraction–gas chromatography–mass spectrometry for profiling free volatile compounds in Cabernet Sauvignon grapes and wines

**Table 5**  
Volatile compounds identified in skins of Cabernet Sauvignon expressed as  $\mu\text{g kg}^{-1}$  of grape unless otherwise noted.

Grape compounds	Retention time (min)	Calculated <i>I</i> (literature) <sup>a</sup>	H1		H2		H3		H4		H5		H6	
			Mean $\pm$ SD	RSD (%)	Mean $\pm$ SD	RSD (%)	Mean $\pm$ SD	RSD (%)	Mean $\pm$ SD	RSD (%)	Mean $\pm$ SD	RSD (%)	Mean $\pm$ SD	RSD (%)
Hexanal	8.33	1087 (1024)	20.99 <sup>b</sup> $\pm$ 2.14	10.2	42.82 <sup>b</sup> $\pm$ 2.84	6.6	32.44 <sup>b</sup> $\pm$ 1.75	5.4	11.87 <sup>b</sup> $\pm$ 1.67	14.1	10.11 <sup>b</sup> $\pm$ 0.72	7.1	6782.56 $\pm$ 880.47	13.0
Isovalerone <sup>c,d</sup>	11.72	1178 (1207)	0.37 $\pm$ 0.07	7.9	0.68 $\pm$ 0.21	30.5	0.55 $\pm$ 0.07	13.0	0.63 $\pm$ 0.07	11.5	0.65 $\pm$ 0.03	3.9	0.49 $\pm$ 0.04	7.3
(Z)-2-Hexenal <sup>c,d</sup>	12.24	1196 (1207)	190.96 $\pm$ 13.46	7.1	227.63 $\pm$ 10.44	4.6	172.37 $\pm$ 9.30	5.4	144.17 $\pm$ 19.70	13.7	129.09 $\pm$ 9.42	7.3	100.00 $\pm$ 17.48	17.5
4-Methyl-2-heptanone <sup>c,d</sup>	13.02	1213 (1206)	0.17 $\pm$ 0.01	5.1	0.39 $\pm$ 0.04	10.5	0.33 $\pm$ 0.01	2.6	0.31 $\pm$ 0.04	14.5	0.30 $\pm$ 0.03	10.2	0.25 $\pm$ 0.03	12.5
(E)-2-Hexenal	13.36	1218 (1212)	140.66 $\pm$ 12.98	9.2	182.34 $\pm$ 8.90	4.9	113.64 $\pm$ 6.82	6.0	60.04 $\pm$ 5.96	9.9	46.10 $\pm$ 3.84	8.3	17.33 $\pm$ 3.36	19.4
3-Octanone <sup>c,d</sup>	14.13	1236 (1251)	<LOD		<LOD		<LOD		<LOD		3.63 $\pm$ 0.24	6.7	0.53 $\pm$ 0.04	7.9
2-Octanone	17.39	1285 (1283)	<LOQ		<LOQ		<LOQ		<LOQ		<LOQ		<LOQ	
1-Hexanol	21.56	1340 (1345)	745.23 $\pm$ 68.91	9.2	1090 $\pm$ 39.03	3.6	1194.73 $\pm$ 45.03	3.8	1065.02 $\pm$ 84.71	7.9	1802.15 $\pm$ 218.11	12.1	1251.41 $\pm$ 265.97	21.2
(E)-3-Hexen-1-ol <sup>d</sup>	21.82	1349 (1367)	0.13 $\pm$ 0.07	50.7	0.26 $\pm$ 0.03	12.6	0.36 $\pm$ 0.02	5.9	0.23 $\pm$ 0.06	24.6	0.17 $\pm$ 0.06	36.8	0.34 $\pm$ 0.06	16.6
Nonanal	22.80	1381 (1392)	3.23 $\pm$ 0.70	21.6	2.54 $\pm$ 0.89	35.1	1.43 $\pm$ 0.60	42.4	0.12 $\pm$ 0.01	11.5	<LOD		<LOD	
3-Octanol	23.71	1399 (1394)	<LOD		<LOD		<LOD		<LOD		2.16 $\pm$ 0.65	30.2	16.27 $\pm$ 3.18	19.6
(Z)-2-Hexen-1-ol <sup>d</sup>	24.41	1441 (1411)	12.33 $\pm$ 1.33	10.7	7.18 $\pm$ 0.21	3.0	8.01 $\pm$ 0.27	3.3	13.04 $\pm$ 0.81	6.2	4.29 $\pm$ 0.33	7.7	0.20 $\pm$ 0.03	16.9
1-Octen-3-ol	26.45	1456 (1451)	0.62 $\pm$ 0.02	0.3	0.64 $\pm$ 0.05	8.3	0.61 $\pm$ 0.01	1.7	1.22 $\pm$ 0.10	8.3	27.82 $\pm$ 1.59	5.7	16.99 $\pm$ 1.79	10.6
2-Ethyl-1-hexanol	26.76	1493 (1494)	<LOQ		<LOQ		<LOQ		<LOQ		<LOQ		<LOQ	
Dihydroedulan <sup>c,d</sup>	27.30	1529 (1506)	0.40 $\pm$ 0.07	16.4	0.38 $\pm$ 0.10	25.61	0.25 $\pm$ 0.02	7.12	0.34 $\pm$ 0.02	5.0	0.31 $\pm$ 0.05	16.8	0.26 $\pm$ 0.07	26.3
(E)-2-Nonenal	27.74	1537 (1532)	<LOQ		<LOQ		<LOQ		<LOQ		<LOQ		<LOQ	
$\beta$ -Linalool	28.22	1554 (1545)	1.25 $\pm$ 0.03	2.7	1.28 $\pm$ 0.07	5.5	1.30 $\pm$ 0.06	4.8	1.38 $\pm$ 0.20	14.1	1.26 $\pm$ 0.07	5.2	1.13 $\pm$ 0.22	19.7
1-Octanol	28.54	1566 (1565)	0.47 $\pm$ 0.02	4.3	0.67 $\pm$ 0.05	7.5	0.57 $\pm$ 0.02	4.1	0.57 $\pm$ 0.12	20.4	0.52 $\pm$ 0.02	3.8	0.51 $\pm$ 0.09	17.1
(E,Z)-2,6-Nonadienal	28.39	1582 (1576)	0.82 $\pm$ 0.03	3.3	0.48 $\pm$ 0.06	11.7	0.51 $\pm$ 0.03	5.1	0.82 $\pm$ 0.12	14.4	0.50 $\pm$ 0.05	10.2	0.25 $\pm$ 0.08	30.8
1-Nonanol	30.59	1685 (1653)	0.06 $\pm$ 0.02	38.8	0.08 $\pm$ 0.01	16.0	0.06 $\pm$ 0.01	21.6	0.07 $\pm$ 0.01	15.6	0.07 $\pm$ 0.02	26.1	0.03 $\pm$ 0.01	41.8
(Z)-3-Nonen-1-ol <sup>c,d</sup>	30.98	1688 (1664)	0.47 $\pm$ 0.05	10.9	0.53 $\pm$ 0.01	1.3	0.44 $\pm$ 0.01	2.9	0.43 $\pm$ 0.08	19.0	0.48 $\pm$ 0.05	9.7	0.35 $\pm$ 0.06	16.0
$\beta$ -Citronellol <sup>c,d</sup>	32.51	1811 (1744)	0.13 $\pm$ 0.01	9.3	0.16 $\pm$ 0.04	23.4	0.12 $\pm$ 0.02	12.7	0.13 $\pm$ 0.02	13.5	0.11 $\pm$ 0.01	7.6	0.09 $\pm$ 0.03	30.9
$\beta$ -Damascenone	33.07	1830 (1841)	0.18 $\pm$ 0.10	57.3	0.22 $\pm$ 0.03	12.5	0.17 $\pm$ 0.08	50.0	0.13 $\pm$ 0.07	54.0	0.30 $\pm$ 0.04	13.2	0.33 $\pm$ 0.22	67.0
Nerol	33.35	1875 (1849)	<LOQ		<LOQ		<LOQ		<LOQ		<LOQ		<LOQ	
Geranyl nerylacetone <sup>c,d</sup>	33.43	1881 (1858)	0.97 $\pm$ 0.59	61.2	0.46 $\pm$ 0.28	61.2	0.38 $\pm$ 0.06	15.2	0.71 $\pm$ 0.34	47.4	0.84 $\pm$ 0.22	25.7	<LOD	
2-Phenylethanol	34.23	1923 (1939)	4.36 $\pm$ 0.33	7.7	5.78 $\pm$ 0.62	10.8	4.95 $\pm$ 0.22	4.4	5.16 $\pm$ 1.19	23.0	4.48 $\pm$ 0.20	4.4	5.90 $\pm$ 0.30	5.1
$\beta$ -Ionone	34.60	1955 (1956)	0.33 $\pm$ 0.02	5.0	0.33 $\pm$ 0.02	6.2	0.33 $\pm$ 0.01	4.4	0.34 $\pm$ 0.05	14.4	0.32 $\pm$ 0.02	5.9	0.27 $\pm$ 0.05	19.2

LOQ, Limit of quantitation; LOD, limit of detection.

<sup>a</sup> Literature sources: <http://www.odour.org.uk>, <http://www.flavornet.org>, Ferhat et al. [44], Jennings and Shibamoto [45].

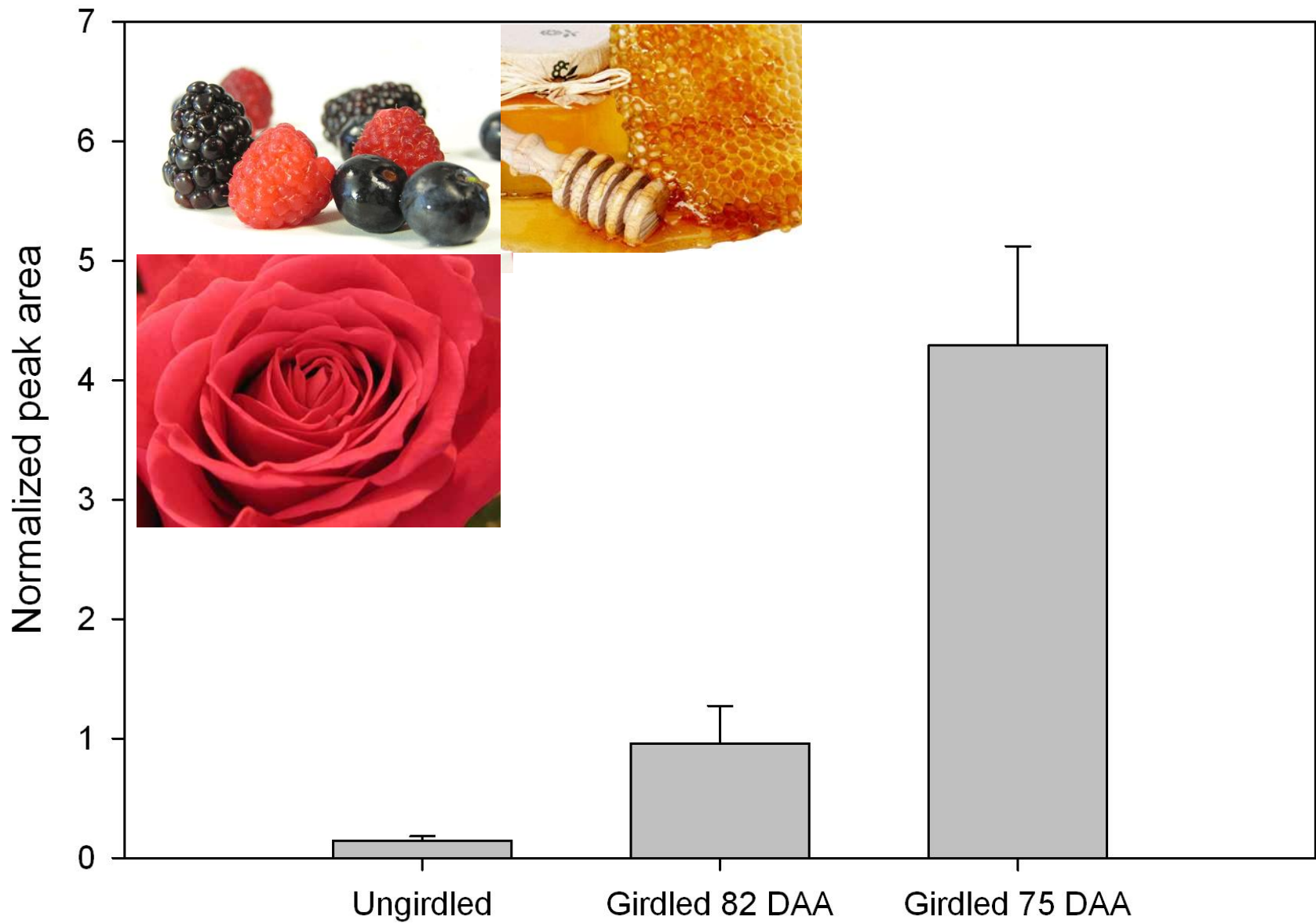
<sup>b</sup>  $\text{mg kg}^{-1}$  of grape.

<sup>c</sup> Compounds tentatively identified by matching to the NIST MS library spectra and comparison of Kovats' retention indices (*I*) to literature values.

<sup>d</sup>  $\mu\text{g}$  equivalents of 2-octanol internal standard per kilogram of grapes.

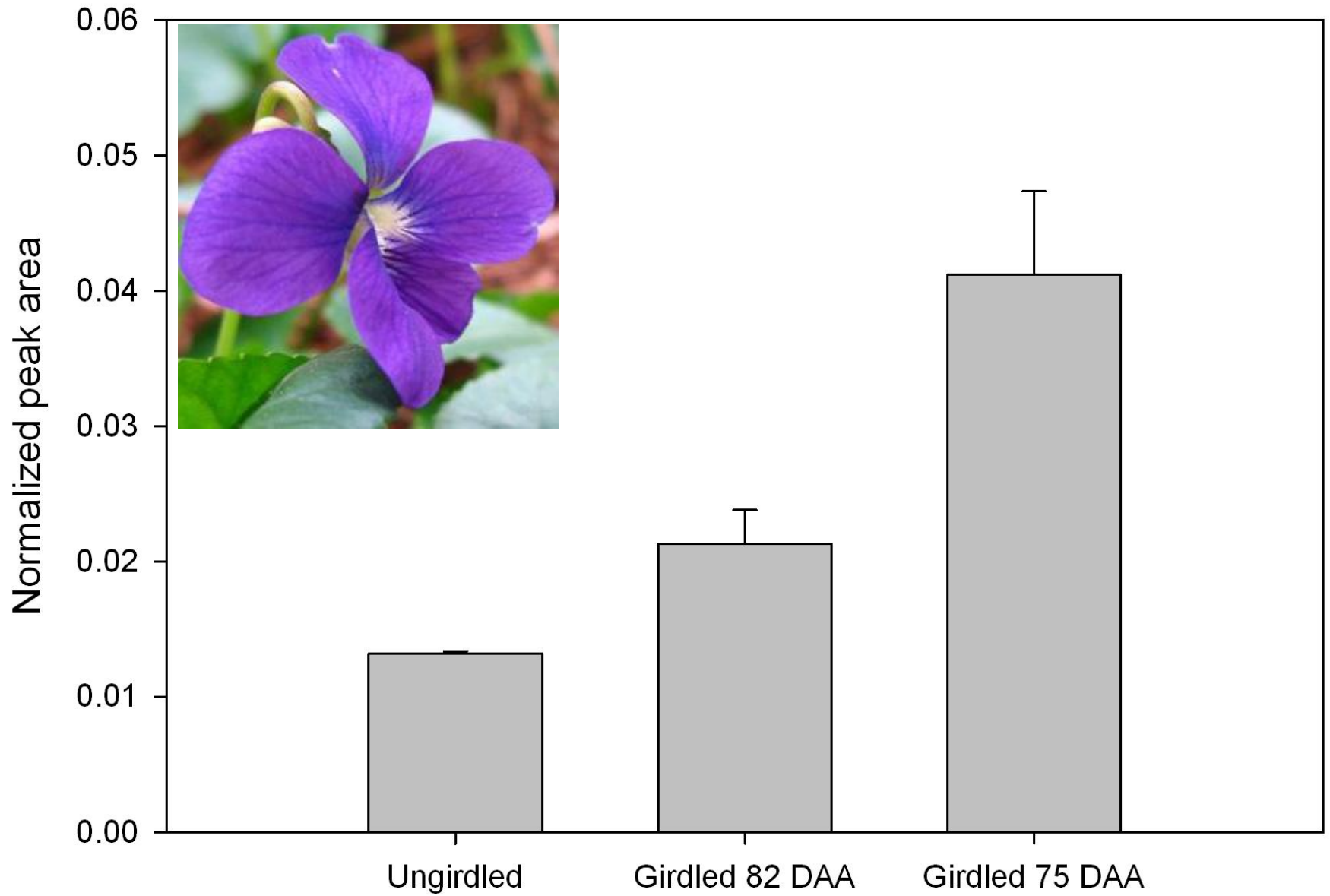
Canutti et al. 2009

# Damascenone content (105 berries) +/- SE

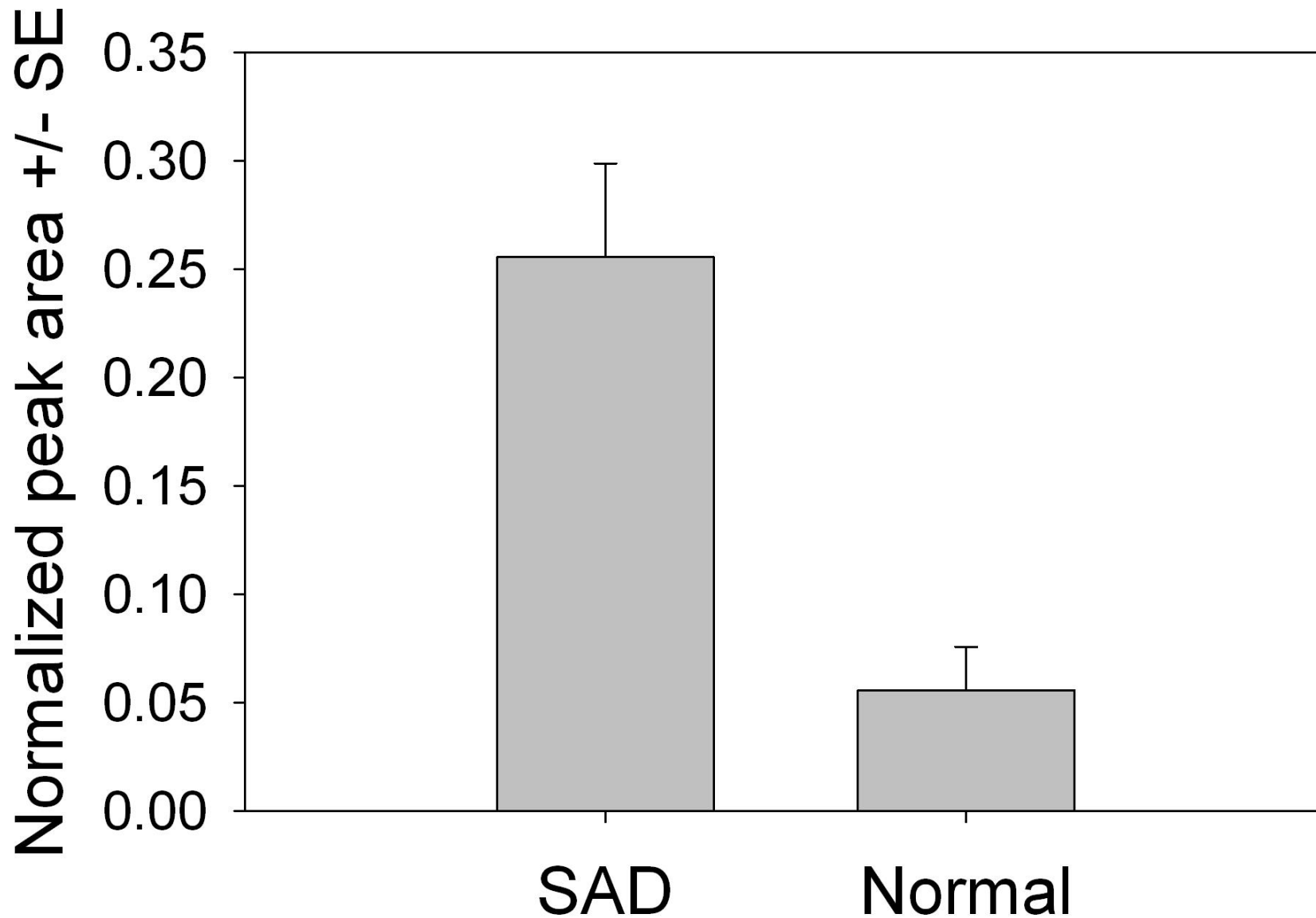




# Ionone content (105 berries) +/- SE

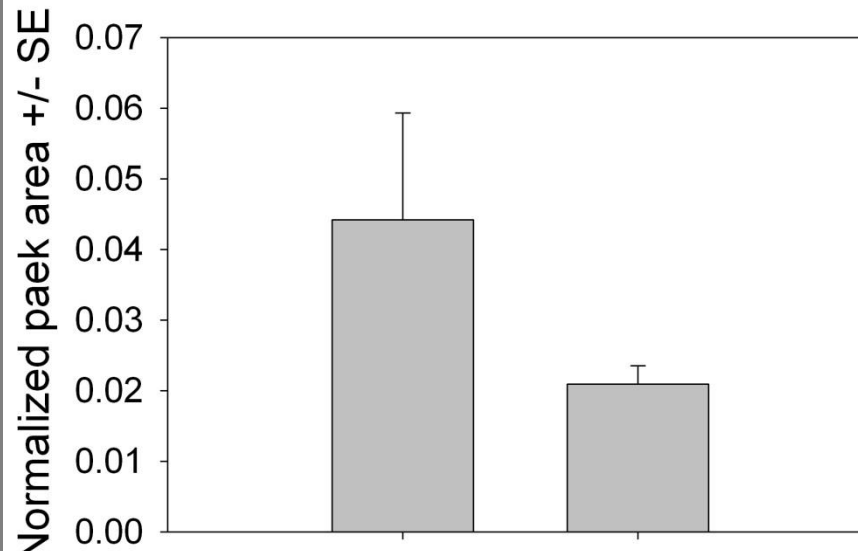


# $\beta$ -Damascenone from Napa in 2009



# Linalool [and Ionone] are increased in SAD fruit

Sonoma site



Napa site

