Role of glutathione in winemaking

Legalisation of pure glutathione usage in winemaking

Carien Coetzee

Engela Kritzinger

Wessel du Toit

Industry WORKSHOP

6 May 2015

GSH in general

"Master antioxidant" – protection against oxidative stress

- Detoxification
- Defence molecule

Glutathione (Y-L-glutamyl-L-cysteinylglycine) : tripeptide of:

1. L-glutamate
2. L-cysteine
3. glycine

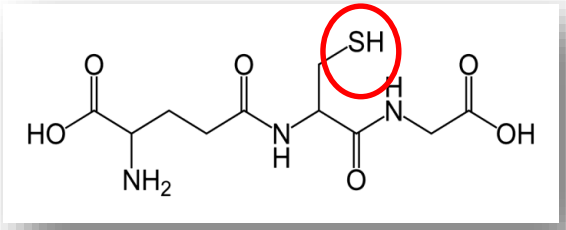
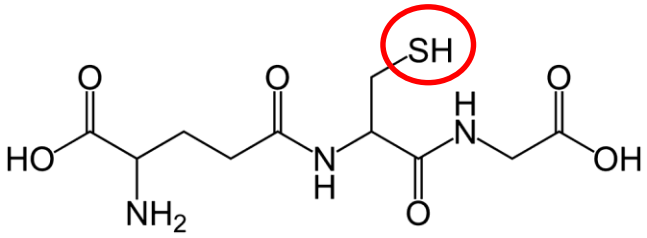


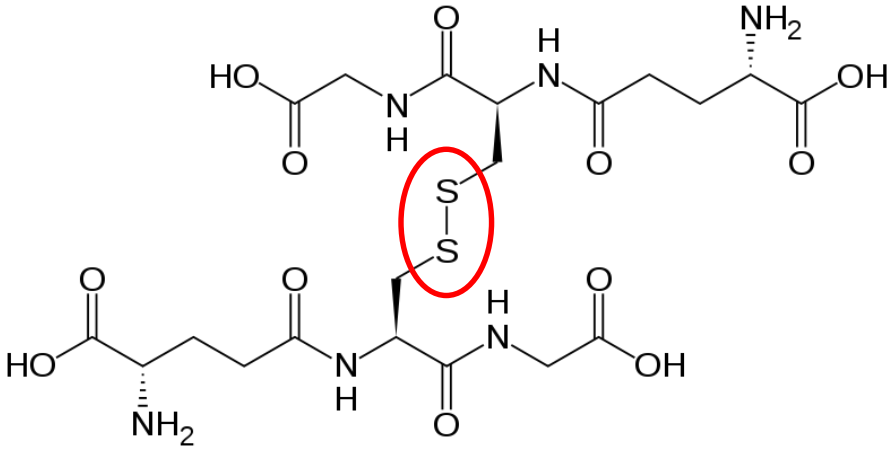
Figure 1: GSH

GSH in general

Three forms of GSH in the cell:

- Reduced form (GSH) (90%)
- Oxidized form (GSSG)
- Mixed disulfides





Role of GSH

Strategies to **improve** and **preserve wine quality** would confer a competitive advantage to the wine producer.

↑ **glutathione (GSH)** levels in wine possible strategy

Protective effects of GSH in must and wine:

1. **limits oxidative colouration** in grape juice and wine
2. **protective effect** on:
   - volatile thiols
   - esters and terpenes
3. **impedes** development of **atypical ageing flavours**
   - sotolon (curry)
   - 2-aminoacetophenone (wool, fusel alcohol, naphtalene)

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GSH traps *o*-quinones formed during phenolic oxidation

↓

Limits further oxidation reactions

Antioxidant activity of glutathione in must and wine

Hydroxycinnamic acid : GSH

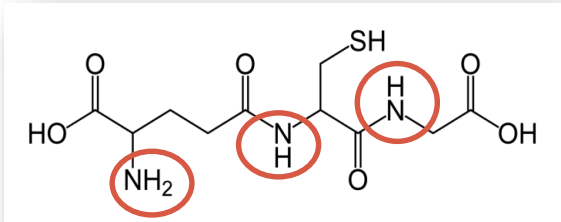
- ratio – indication of oxidation susceptibility
  - Higher ratio = darker must

0.9-2.2	1.1-3.6	3.8-5.9
light	medium	dark

GSH in grapes and wine

- Levels in South African juice ranged from 1-70 mg/L
- Average of 28 young Sauvignon blanc wines 13 mg/L

- Content in grapes – closely related to vine nitrogen status (YAN)
- Nitrogen fertilized vines had higher content of GSH



Factors influencing GSH levels in juice and wine

Grape ripeness level	Increase	Suklje et al., 2013
Oxidative state of the juice	Decrease	Coetzee et al., 2013
Pressing of skins	Decrease	Maggu et al., 2007
Anti-oxidant additions such as SO_2 to juice	Stabilise levels	Coetzee et al., 2013
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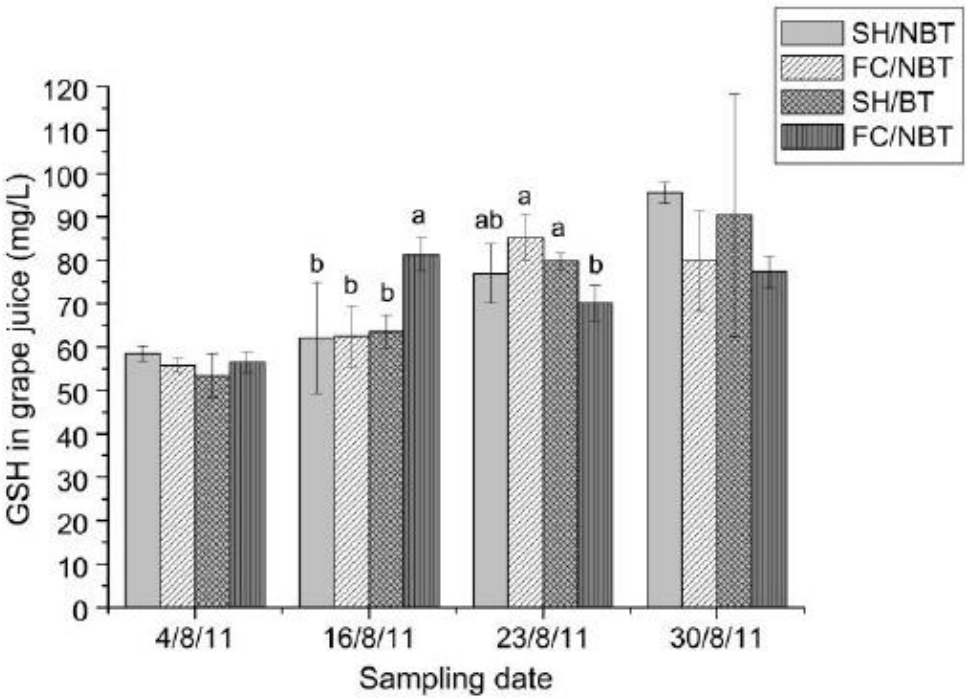
- Increase during ripening up to about 16˚B – stable thereafter
- > 90% in the reduced form



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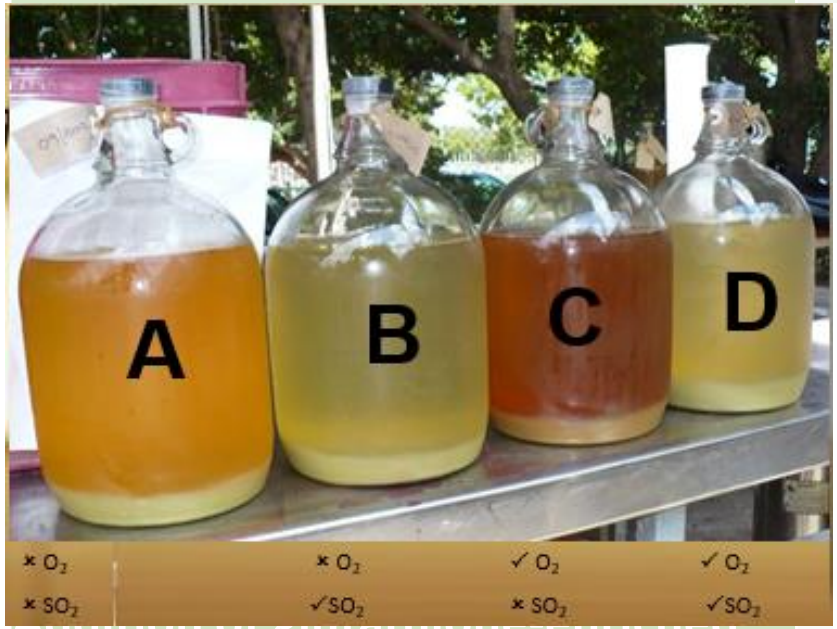

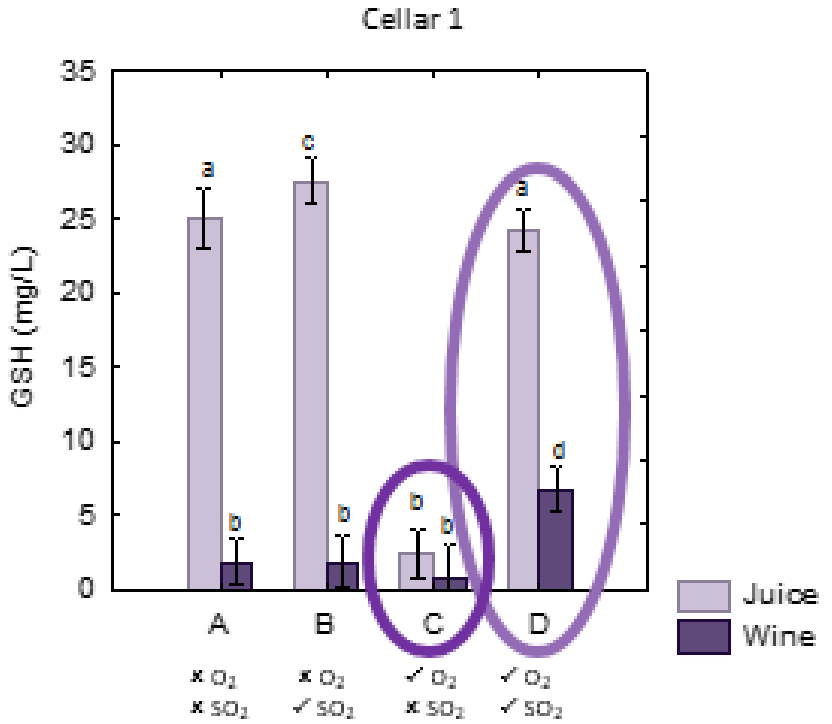

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Oxidative state of the juice	De
Pressing of skins	**De**
Anti-oxidant additions such as SO_2 to juice	Sta
Oxidation of wine	De
Bottle ageing	De
Yeast strain	Inc dec
Addition of glutathione-enriched inactive dry yeast preparation	Inc
YAN levels	Inc decrease

	glutathione (mg/L)
retention time (min)	5.4
λ_{max}	
free run	39.6 (±0.8) a
1 h of skin contact	23.3 (±0.6) b
0.4 atm	ND[b]
1.3 atm	ND
2.0 atm	ND
free run	36.5 (±1.3) a
1 h of skin contact	23.1 (±0.4) b
0.4 atm	8.9 (±0.3) c
1.2 atm	ND
2.0 atm	ND
free run	37.5 (±0.7) a
1 h of skin contact	16.7 (±0.2) b
0.4 atm	ND
1.2 atm	ND

Kritzinger et al., 2013

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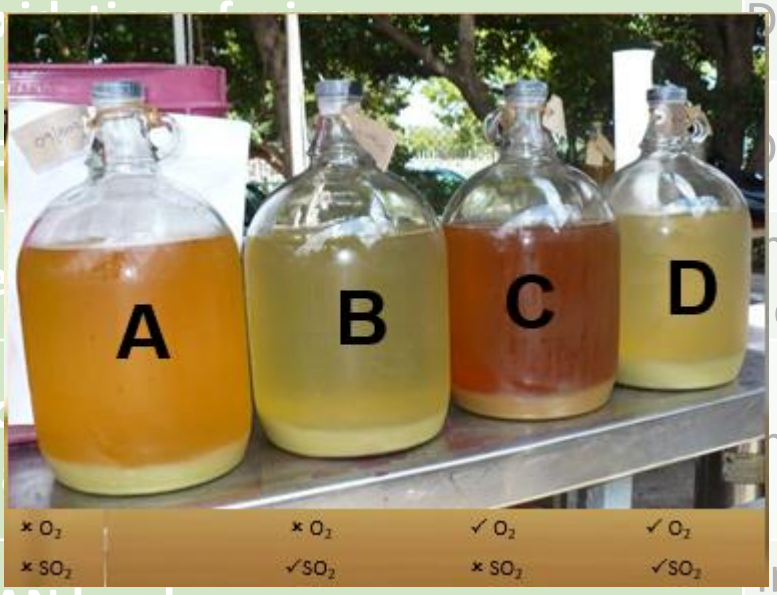

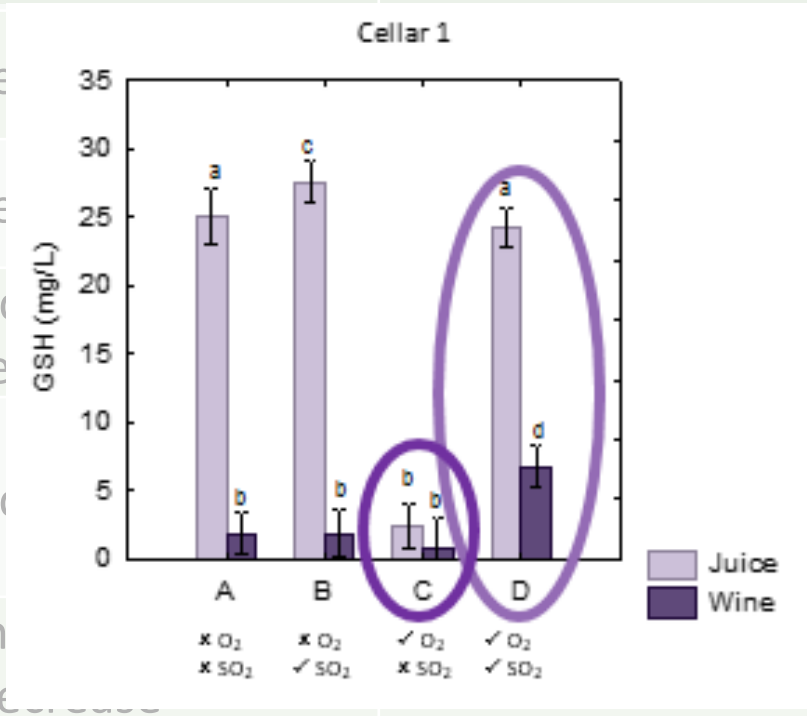

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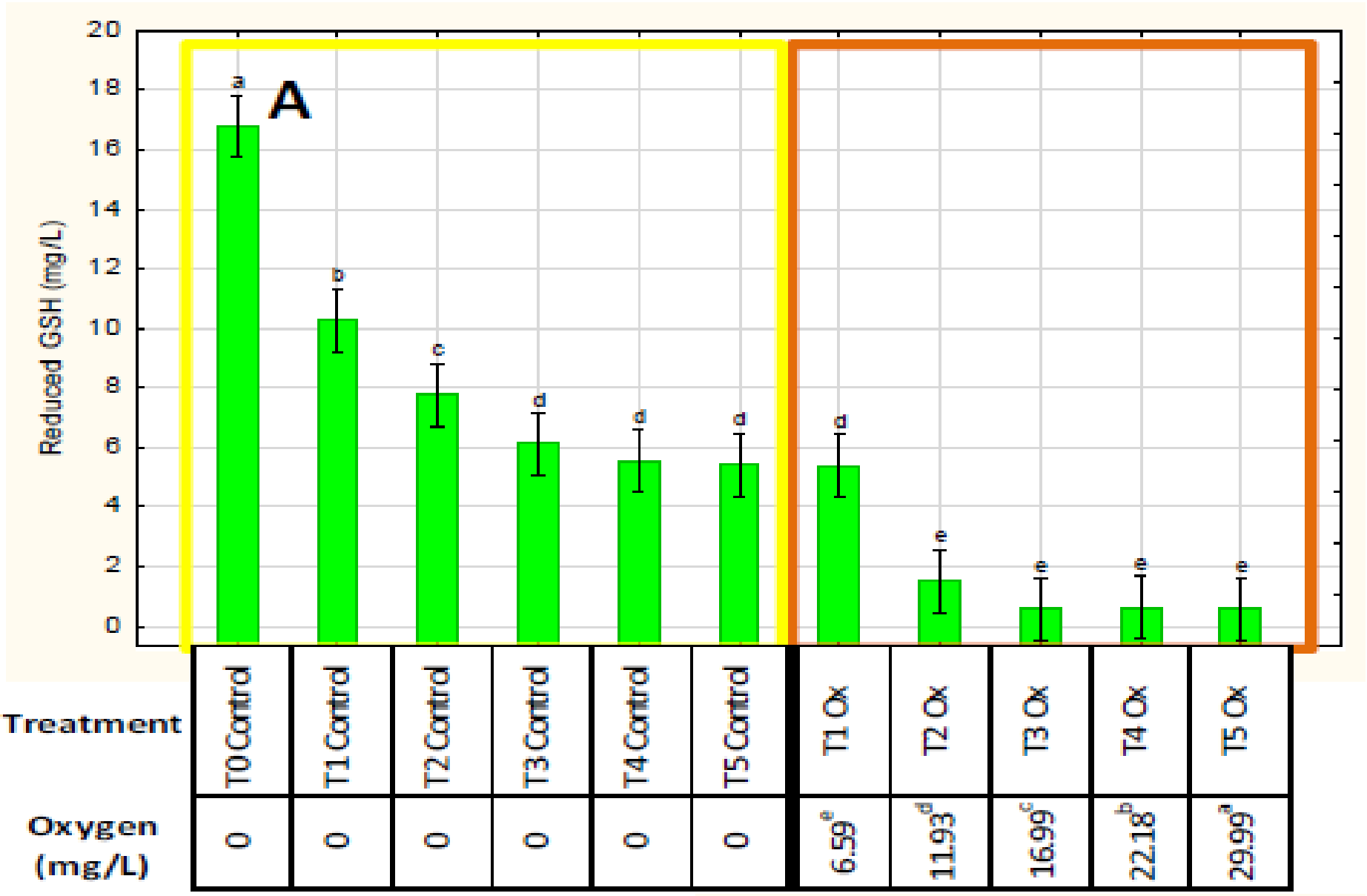

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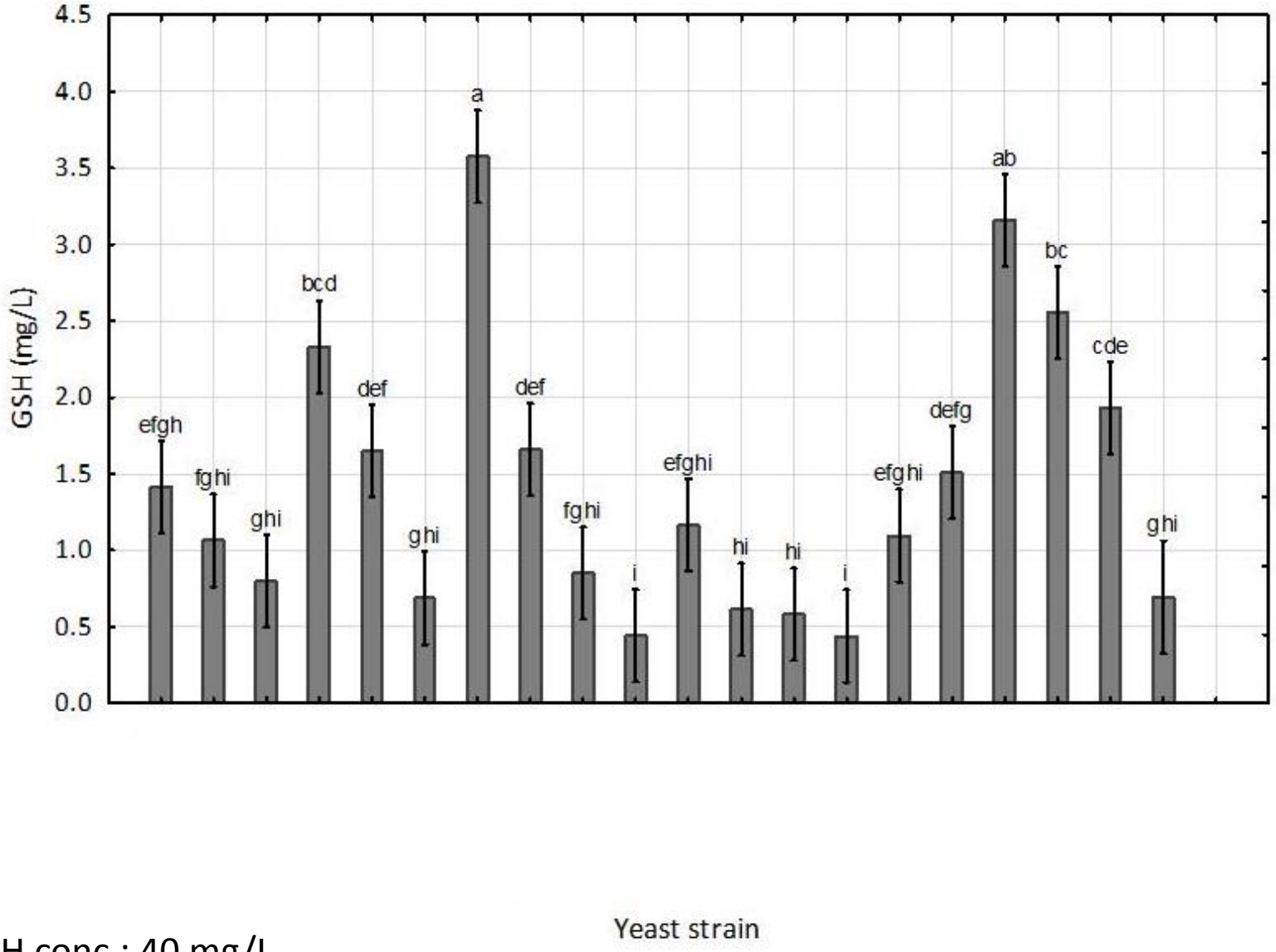
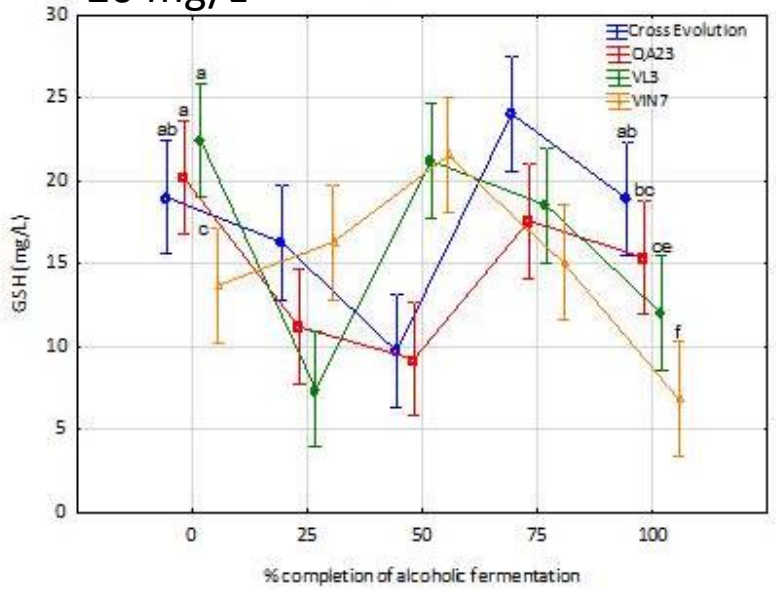


Initial GSH conc.: 40 mg/L

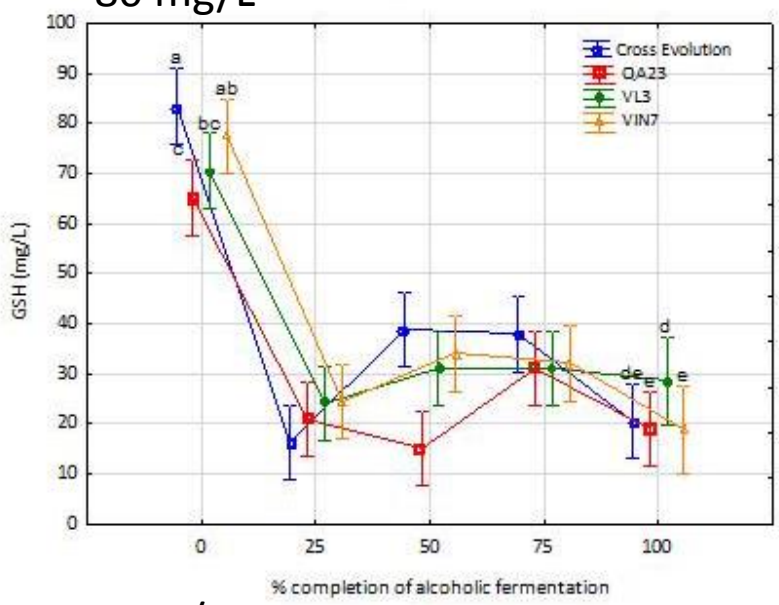
Adapted from Kritzinger et al (2013). *Australian Journal of Grape and Wine Research* 19, 161–170.

Factors influencing GSH levels in juice and wine

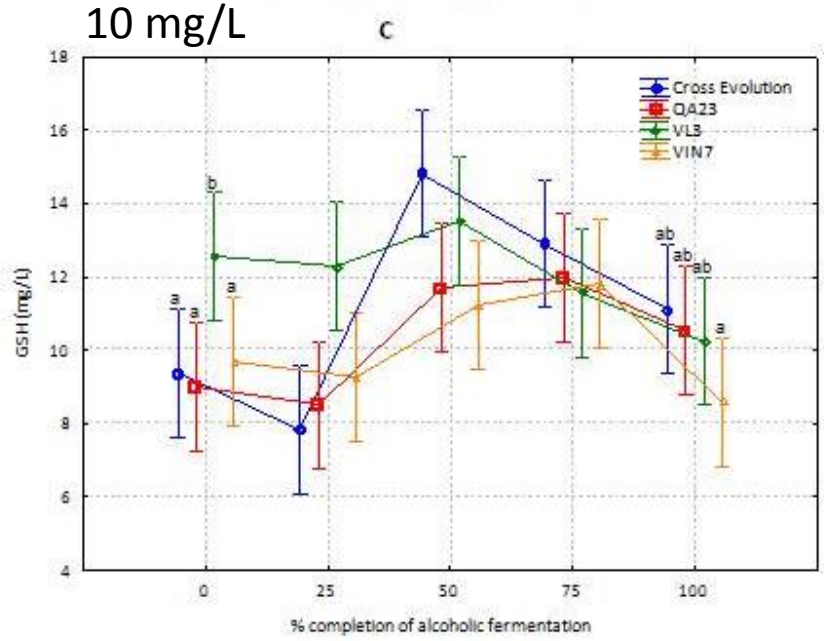
20 mg/L



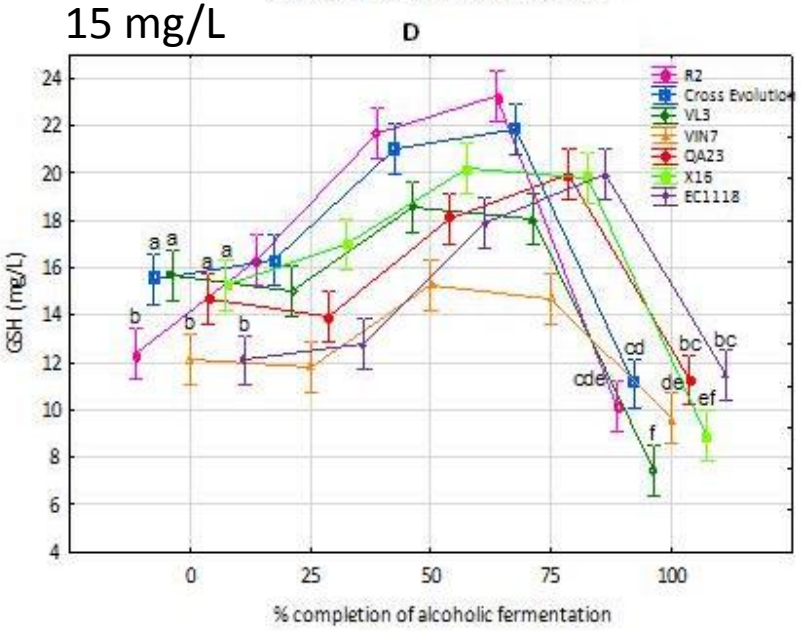
80 mg/L



10 mg/L



15 mg/L



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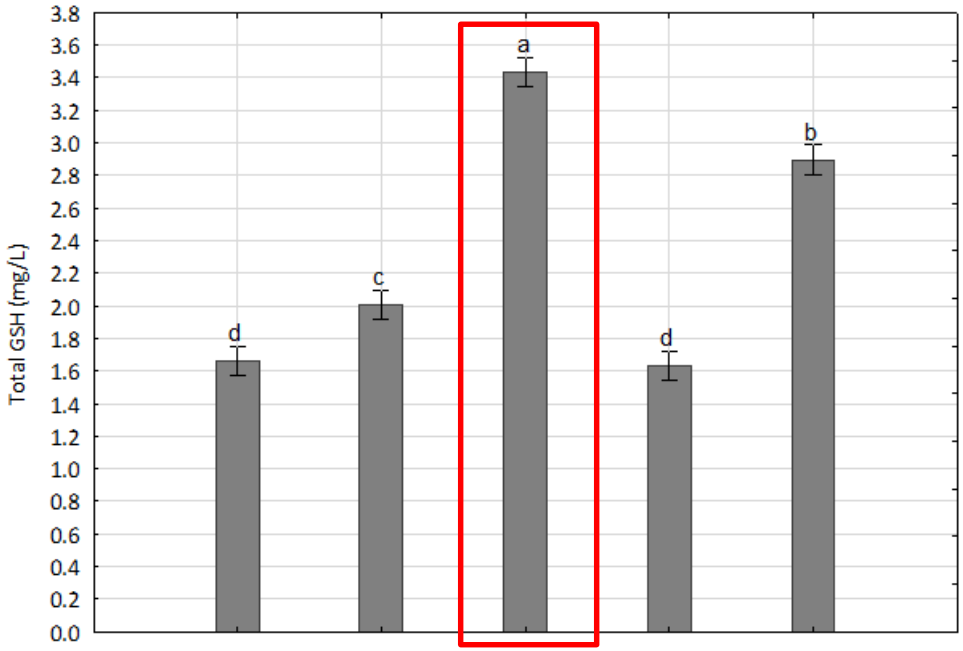
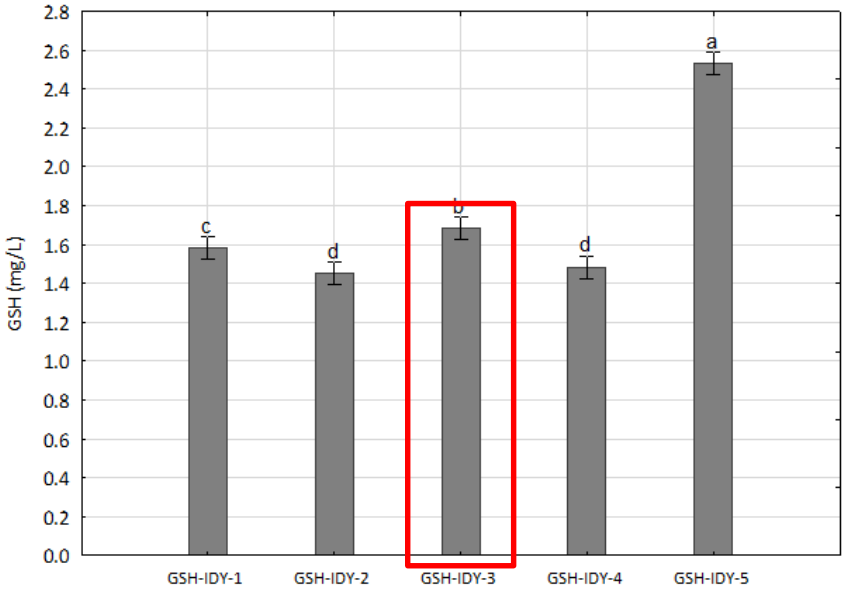
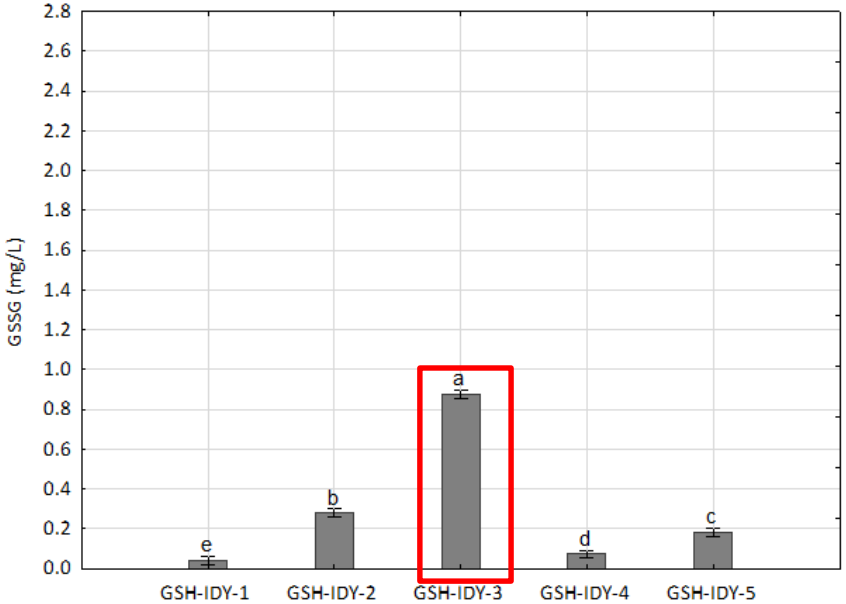
Total GSH



Reduced GSH



GSSG (oxidized)



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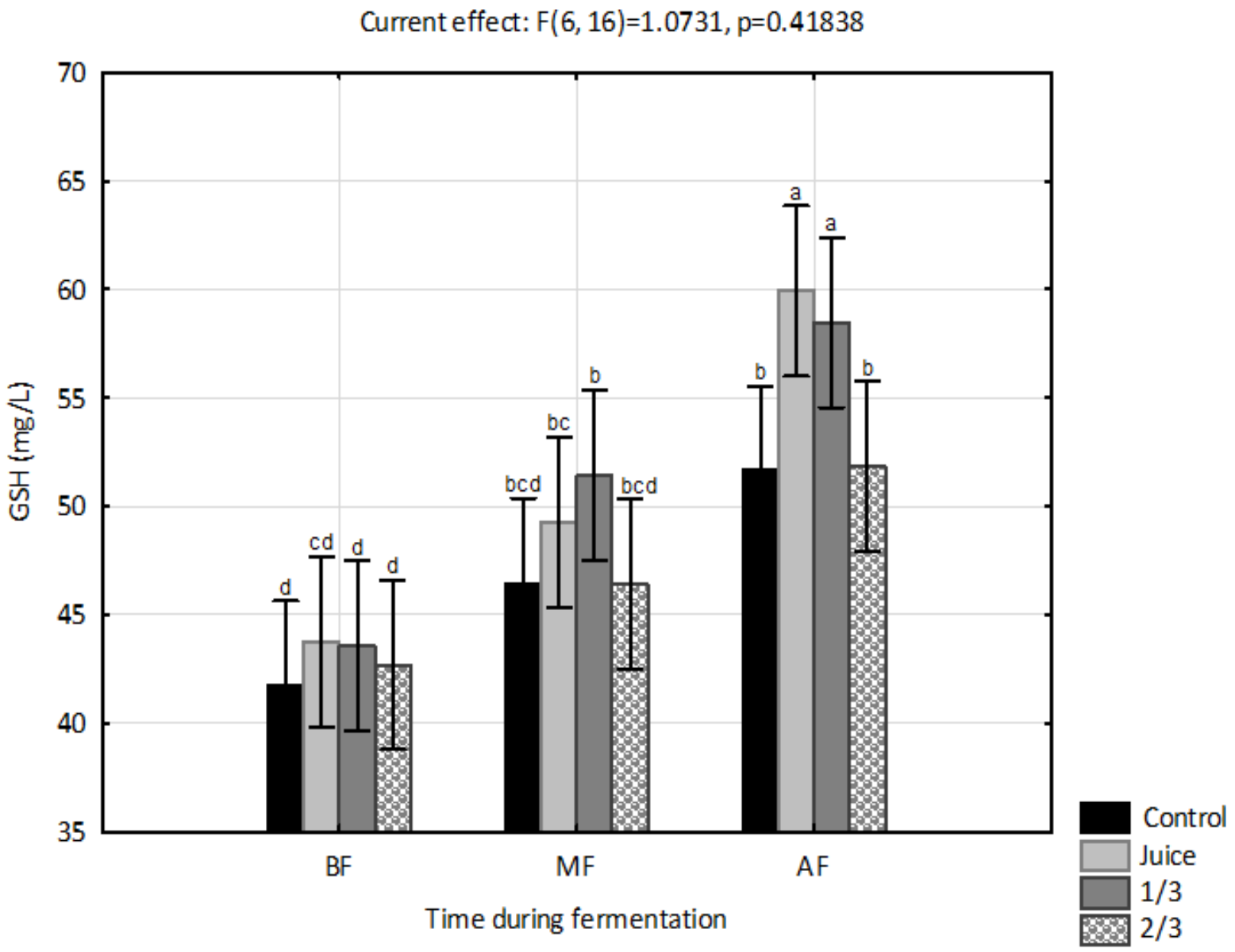

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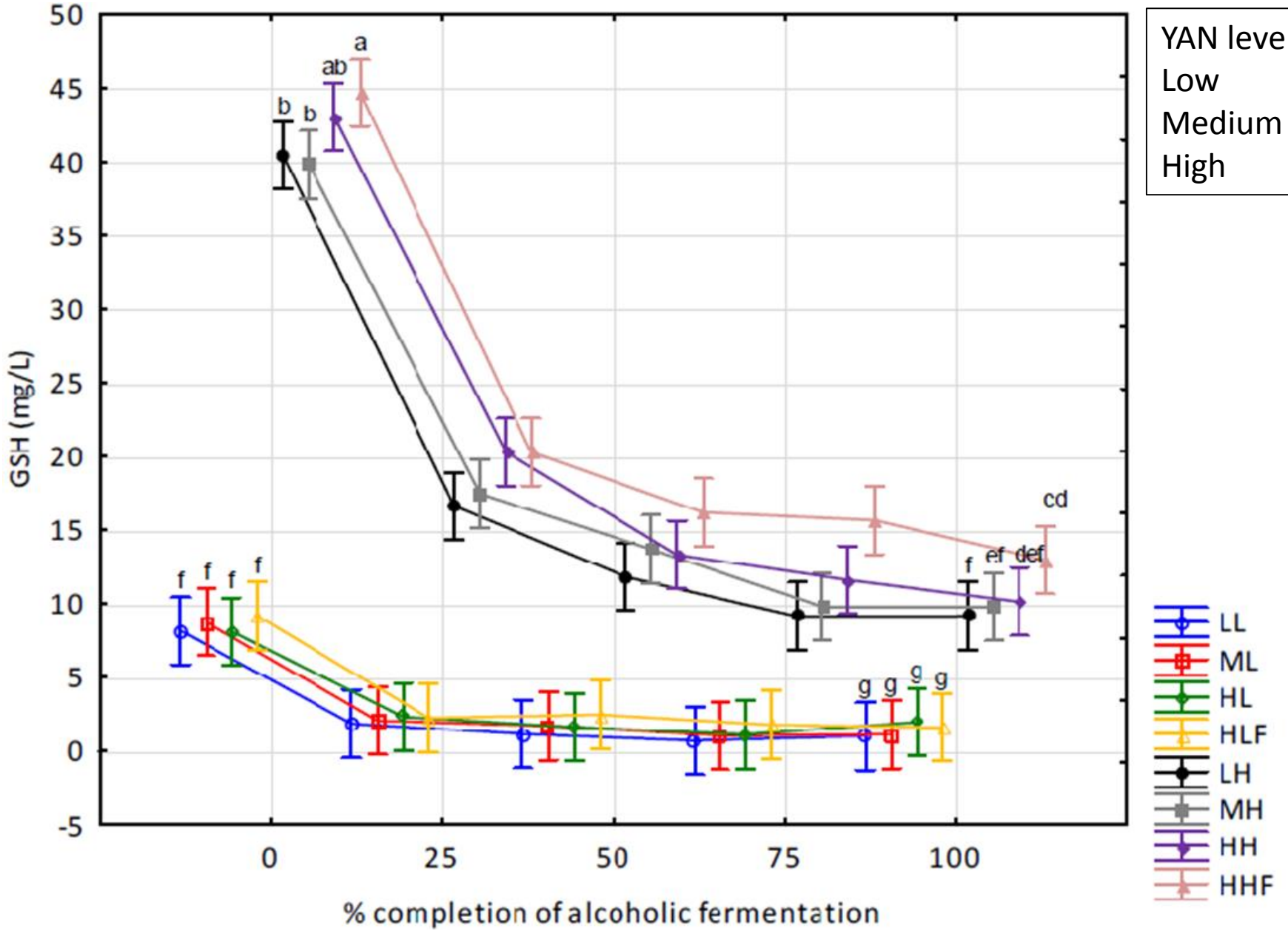

Table 2. Concentration[a] of Sulfur-Containing Volatile Compounds in Sauvignon blanc Wines after 6 Months of Bottle Storage

treatment	3-MH (ng/L)	H_2S (μg/L)	MeSH (μg/L)
high GSH, high Cu A/A	507 ± 1	1.1 ± 0.2	0.5 ± 0.1
high GSH, high Cu A/N	556 ± 4	3.2 ± 0.4	0.6 ± 0.2
high GSH, high Cu N/N	676 ± 2	4.5 ± 0.4	0.7 ± 0.2
high GSH, low Cu A/A	602 ± 8	1.3 ± 0.4	0.7 ± 0.0
high GSH, low Cu A/N	663 ± 5	1.5 ± 0.1	0.6 ± 0.1
high GSH, low Cu N/N	721 ± 2	1.5 ± 0.4	0.6 ± 0.1
low GSH, high Cu A/A	241 ± 3	tr	0.3 ± 0.1
low GSH, high Cu A/N	260 ± 6	0.3 ± 0.1	0.3 ± 0.0
low GSH, high Cu N/N	341 ± 6	2.5 ± 0.0	0.4 ± 0.0
low GSH, low Cu A/A	511 ± 7	0.2 ± 0.2	0.5 ± 0.1
low GSH, low Cu A/N	568 ± 5	0.6 ± 0.2	0.6 ± 0.1
low GSH, low Cu N/N	665 ± 5	1.2 ± 0.0	0.5 ± 0.1

[a]Values are the average of three wines analyzed in duplicate. "tr" denotes value below the limit of quantification of 0.2 μg/L.

High GSH = 20 mg/L

Low GSH = 0.1 mg/L

High Cu = 0.3 mg/L

Low Cu = 0.1 mg/L

Begin 3MH = 1000 ng/L

Ugliano et al., 2011. Journal of Agricultural and Food Chemistry. 59, 2564-2572.

Legalisation of pure glutathione usage in winemaking

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Current status

- Approval of GSH addition to must AND wine in stage 7 of OIV approval process (April 2015, OIV meeting, Paris)

- **Addition of GSH to must and wine:**
  - Prior to fermentation or at bottling
  - Max dosage: 20 mg/L
  - GSH

- **Addition of GSH-IDY to must and wine:**
  - Addition prior to fermentation or during fermentation
  - Addition prior or during storage
  - Product must contain minimum concentration of 8 mg/g

± R 5000 per kg
To increase GSH to 20 mg/L
= 10 c/L

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reduced l-glutathione food grade(gsh)

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