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Varietal Thiols and Green Characters

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Compounds responsible for the green character

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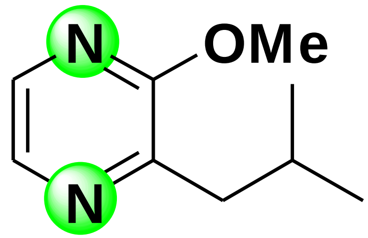
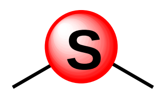
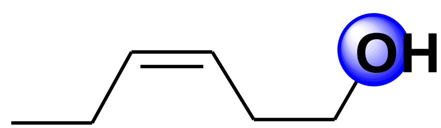
- Methoxypyrazines
  - **IBMP**, SBMP, IPMP
- Sulfur compounds
  - **DMS**, DES, DMDS
  - 2-Isobutylthiazole
- C6 compounds
  - ***(Z*)-3-Hexen-1-ol**
  - (*E*)-2-Hexenal
  - (*Z*)-3-Hexenal
  - Hexanal
  - 1-Hexanol
  - Hexyl esters



3-Isobutyl-2-methoxypyrazine (IBMP)



Dimethyl sulfide (DMS)



(*Z*)-3-Hexen-1-ol (*cis*-3-Hexen-1-ol)

Grape sources of C6 flavours in wine

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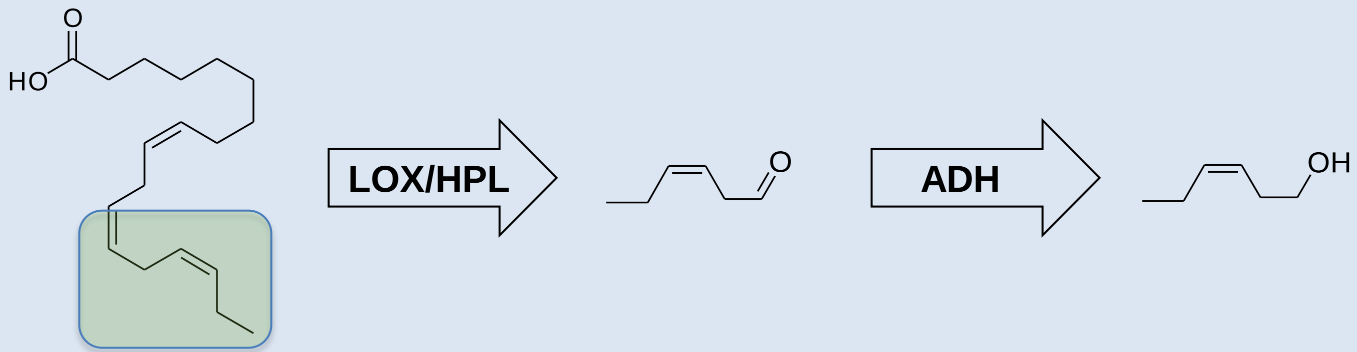
- *cis*-3-Hexen-1-ol precursors
  - Formed from unsaturated fatty acids after berry damage (usually upon crushing)
  - Derived from linolenic acid through enzyme cascade



Linolenic Acid (*Z*)-3-Hexenal (*Z*)-3-Hexen-1-ol

Lipoxygenase → hydroperoxide lyase → alcohol dehydrogenase

Modulating factors – *cis*-3-Hexen-1-ol

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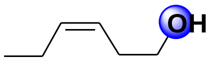
- Viticulture
  - Enzymatic formation via LOX pathway leads to C6 compounds
  - Differs between varieties and during berry development (e.g. Riesling vs Cabernet Sauvignon)
  - Highest at pre-veraison in line with unsaturated fatty acid levels – decline in linolenic acid with ripening
  - Higher in skin (from press cake) than must at all ripening stages



(*Z*)-3-Hexen-1-ol
(*cis*-3-Hexen-1-ol)

Modulating factors – *cis*-3-Hexen-1-ol

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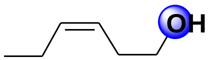

(*Z*)-3-Hexen-1-ol
(*cis*-3-Hexen-1-ol)

- Winemaking
  - Time and temperature of skin contact – similar extraction from 15-28 ^{o}C with max after 10-15 h, continual increase during contact time at 10 ^{o}C after 25 h
  - Relatively stable but SO_2 and enzymatic activity have effects – O_2 needed for formation
  - Esterification to the acetate – from green (alcohol) to green/floral/fruity (ester)
- Storage
  - Not affected by storage in presence of oxygen
  - Minimal change with storage on lees for up to seven months
  - Unaffected by short-term oxidative storage in presence of phenolics
  - Slow decline with storage for 210 days but no impact from different SO_2 levels

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Summary

- Green flavours in wine are caused by a number of different compound classes, with vastly different potencies
- Compound origins are in the grape, often in precursor form
- Viticultural practices and harvesting decisions can impact on green flavours
- Green flavours may be desirable, adding complexity or typicity to wine styles

Sensory impact of *cis*-3-Hexen-1-ol

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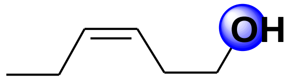

(*Z*)-3-Hexen-1-ol
(*cis*-3-Hexen-1-ol)

- *cis*-3-Hexen-1-ol – cut grass, herbaceous, leafy; 400 µg/L threshold
- Typically not found above threshold in most studies
- Found in wine up to
  - 650 µg/L in young red wines (highest in Tempranillo)
  - 800 µg/L in aged red wines
  - 75 µg/L in Gewurztraminer
  - 600 µg/L in some Italian and Spanish white wine varieties (Falanghina and Macabeo)

Varietal thiols – impact odorants

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- Polyfunctional thiols are especially potent and have some of the lowest aroma thresholds of any food odorant
- Varietal thiols are important impact odorants in some wines e.g. Sauvignon Blanc

Thiol	Perception threshold	Aroma	OAV
4-MMP	3 ng/L	blackcurrant box tree passionfruit	Up to 30
3-MH	60 ng/L	grapefruit passionfruit	Up to 210
3-MHA	4 ng/L	passion-fruit box tree sweaty	Up to 195

Darriet et al. Flavour Fragr. 1995, 10, 385-392
Tominaga et al. Vitis 1996, 35, 207-210
Tominaga et al. Flavour Fragr. 1998, 13, 159-162

Volatile thiol sensory descriptors

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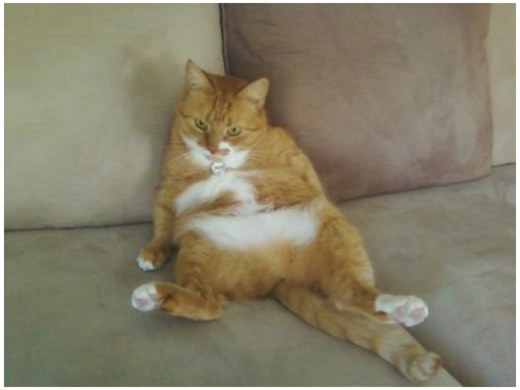
- Individual volatile thiols contribute *tropical* aromas to wine, 3MH also *citrus* aroma
- Volatile thiol combinations had aromas of *tropical* & ***cooked green vegetal*** at both levels, and at high levels also *cat urine/sweaty*
- 4MMP does not contribute any distinctive sensory properties at high levels
- At high concentrations 3MHA is responsible for *cat urine/sweaty* aromas



Consumer preference of volatile thiols

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- There was an optimal level of *cat urine/sweaty* attribute for one group of consumers identified

- The majority of consumers preferred the samples with ‘green’ attributes, with a minority strongly preferring the ‘fruit’ and ‘estery’ flavours

- Clear linking of volatile thiols in Sauvignon Blanc wines, their associated sensory attributes and effects on consumer preference

Grape varieties containing volatile thiol compounds

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White varieties

Sauvignon Blanc
Chardonnay
Chenin Blanc
Colombard
Gewürztraminer
Gros Manseng
Koshu
Maccabeo
Muscat
Muscadet
Petit Arvine
Petit Manseng
Pinot Blanc
Pinot Gris
Riesling
Scheurebe
Semillon
Sylvaner
Tokay

Red varieties

Cabernet Franc
Cabernet Sauvignon
Grenache
Merlot
Pinot Noir

Volatile thiol concentrations in wines from around the world

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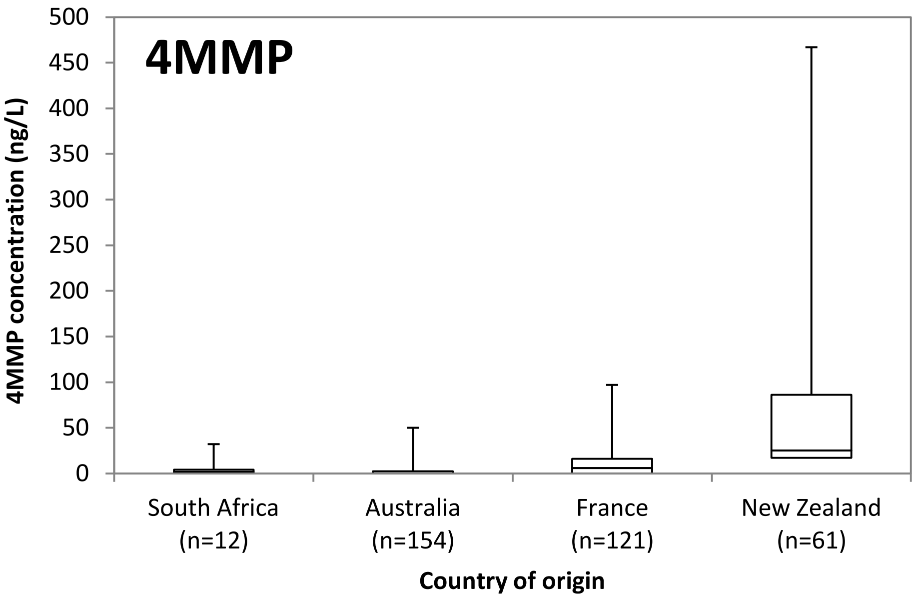

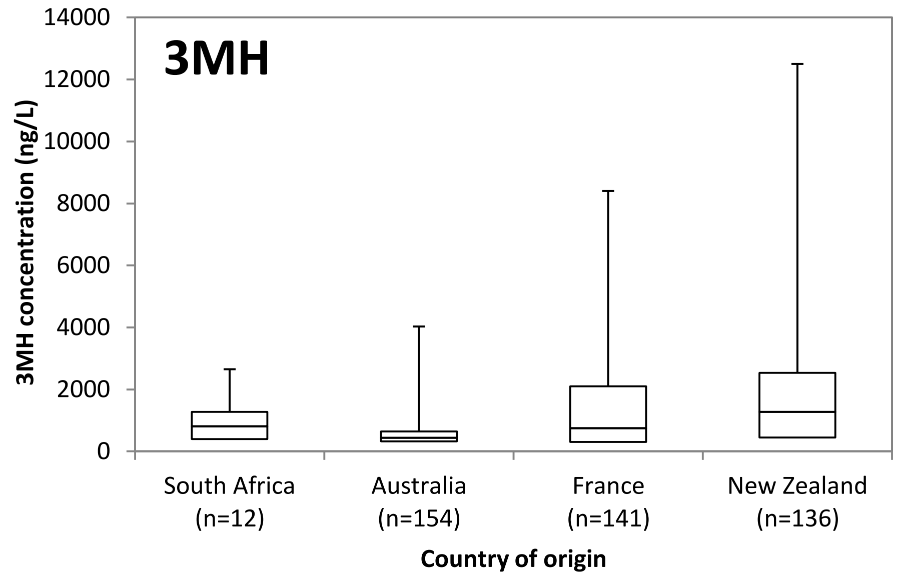

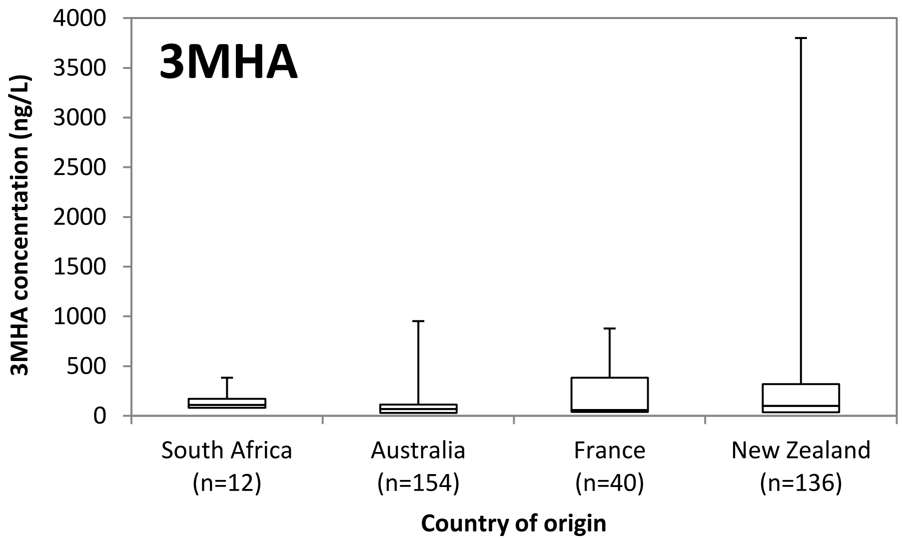

Varietal thiol formation

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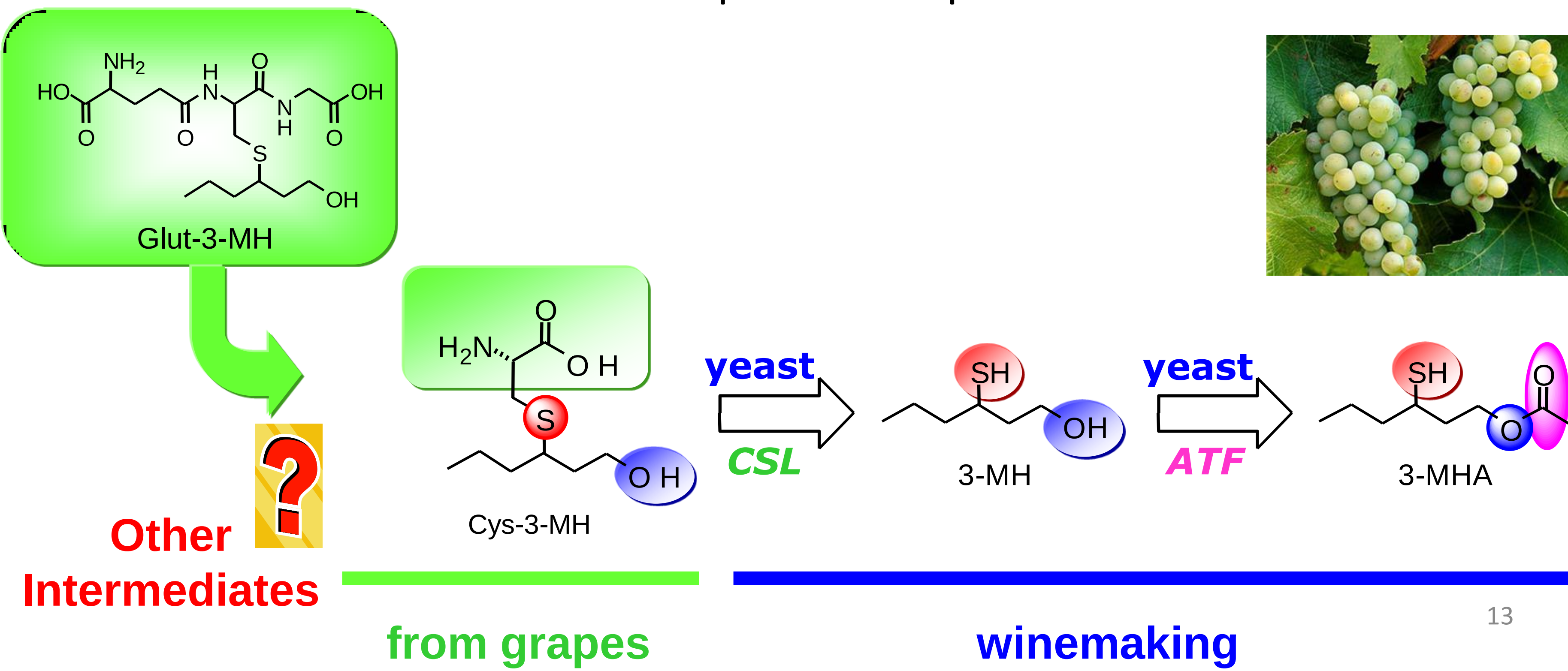
- Optimise formation and maximise stability of varietal thiols
- Need to further understand precursor formation (Stress response : Kobayashi et al)
- Yeast plays a key role in thiol release into wine
- Need to understand relationship between precursors and free thiols



Modulation of volatile thiol precursors

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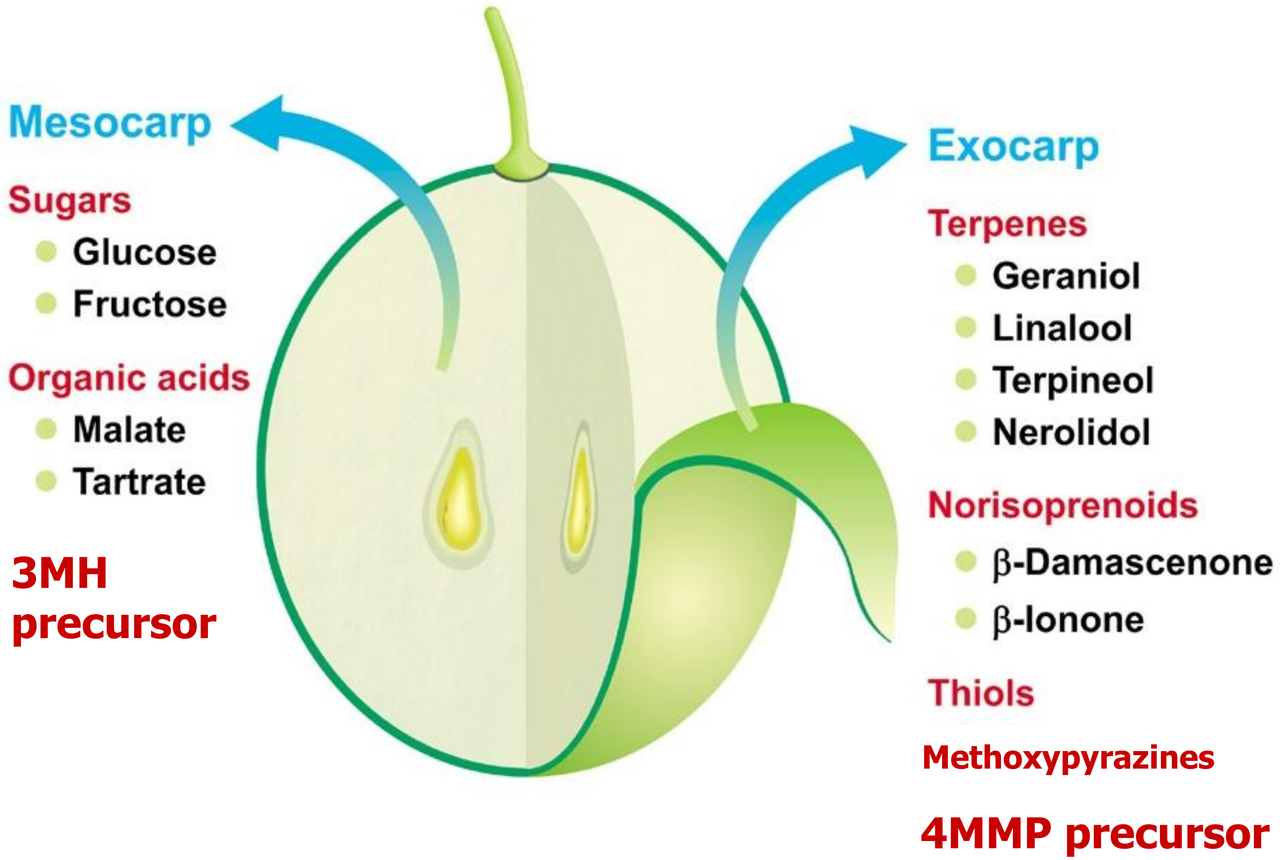


- 3MH precursors are mainly found in the skins of grape berries
- 4MMP precursors are mainly found in the flesh of grape berries

HPLC-MS/MS analysis of precursors

- Amount of precursors measured in SAB juice:

Cys-3-MH 21 – 55 µg/L

Glut-3-MH 245 – 696 µg/L

- Also found precursors in other varieties (in the juice) generally:

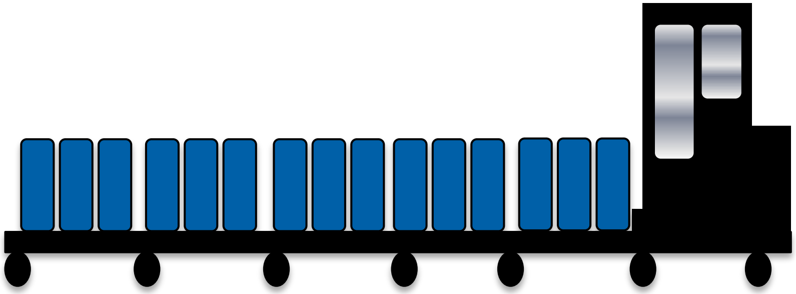
Sauvignon Blanc > Pinot Gris > Chardonnay > Riesling

Capone et al. JAFC 2010, 58, 1390-1395

Precursor Grape studies

- 5 different SAB clones in the same location in Adelaide Hills of South Australia
  - **Ripening**

  - **Transportation / Holding**



Concentration of 3MH during ripening: Clone effects

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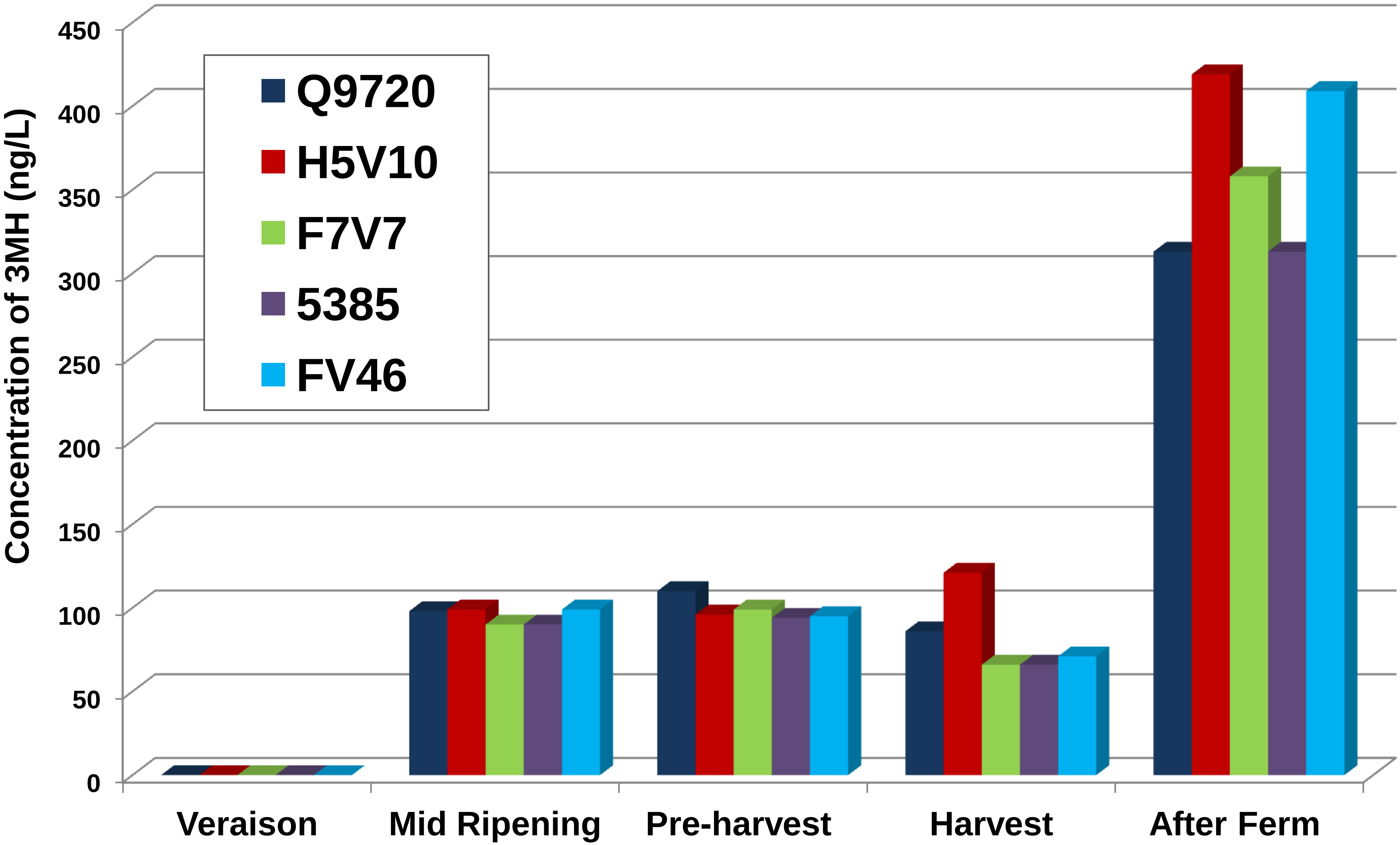

Capone et al. 2011, JAFC. 59: 4649-4658

Amount of 3-MH precursors during ripening

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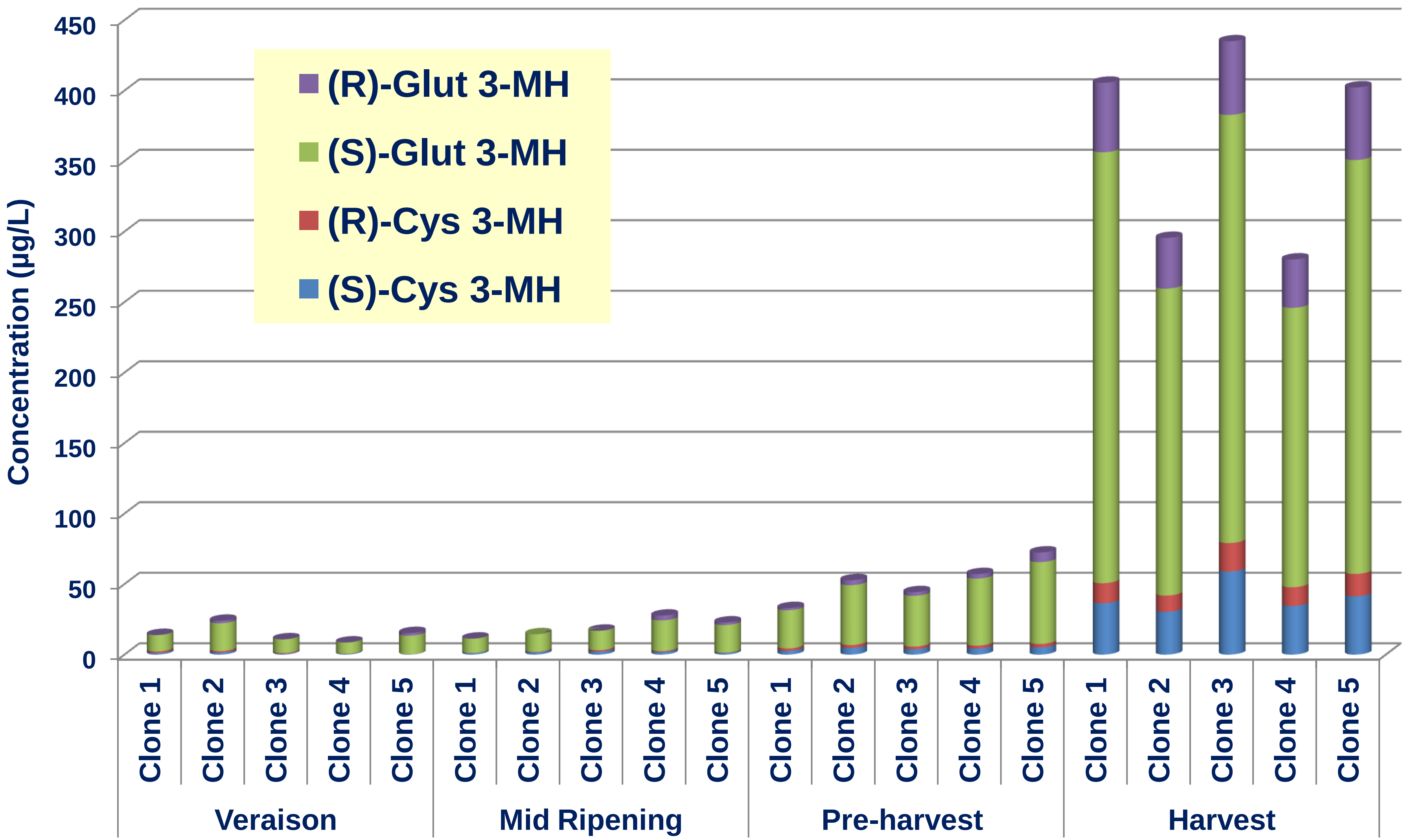

Capone et al. 2011, JAFC. 59: 4649-4658

Effect of transportation on precursor concentration

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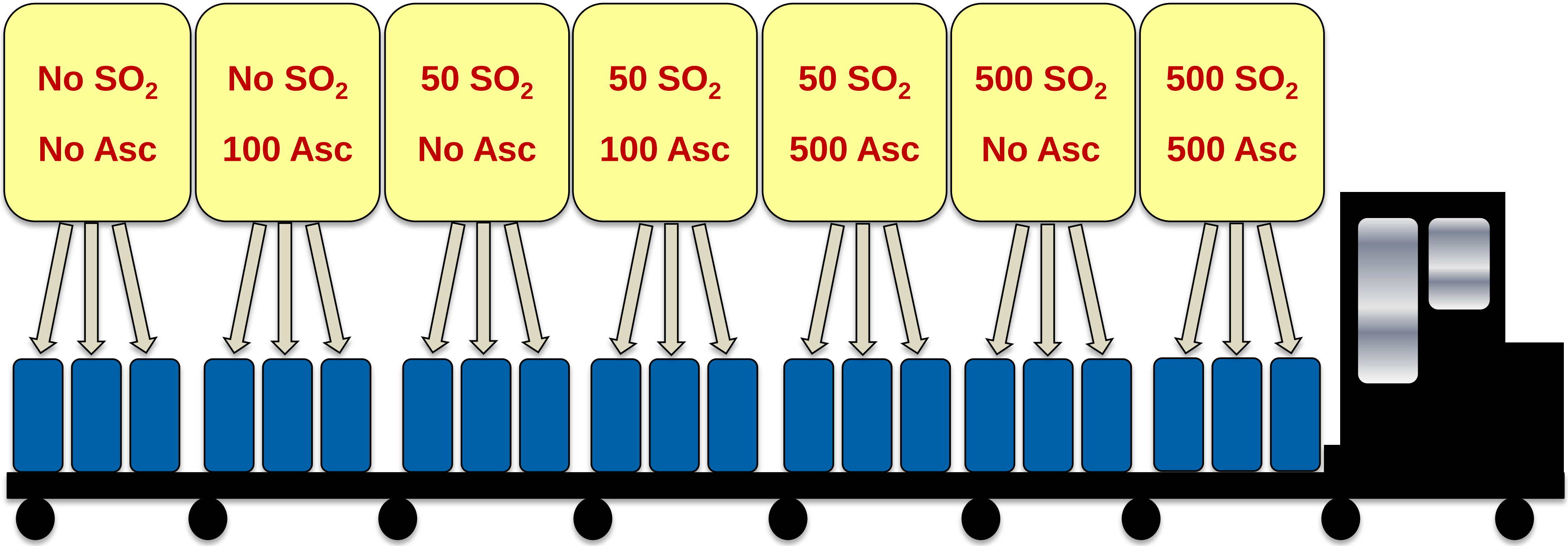

Analysed shortly after machine harvesting then ……..


Arafura Sea
Torres Straits
Timor Sea
Darwin
Gulf of Carpentaria
Coral Sea
Northern Territory
Cairns
Western Australia
South Australia
Indian Ocean
Perth
Great
794 km
New South Wales
Sydney
Canberra
Victoria
Melbourne
Adelaide
South Pacific Ocean
Hobart
Tasmania

Ti


Whyalla
B82
B80
B82
B85
B84
A1
B81
A20
Renmark
Mildura
79
75
24
End
Griffith
20
87
Start
B10
Murray Bridge
M1
Spencer Gulf
B12
Swan Hill
A79
B400
39
A8
Kingscote
Victor Harbor
Encounter Bay
B1
Bordertown
B200
B220
B260
Echuca
A39
58
Shepparton
A300
Coast
Indian Ocean
Perth
Great Australian Bight
794 km
New South Wales
Sydney
Canberra
Victoria
Melbourne
Adelaide
South Pacific Ocean
Southern Ocean
Hobart
Tasmania

Effect of transportation on precursors

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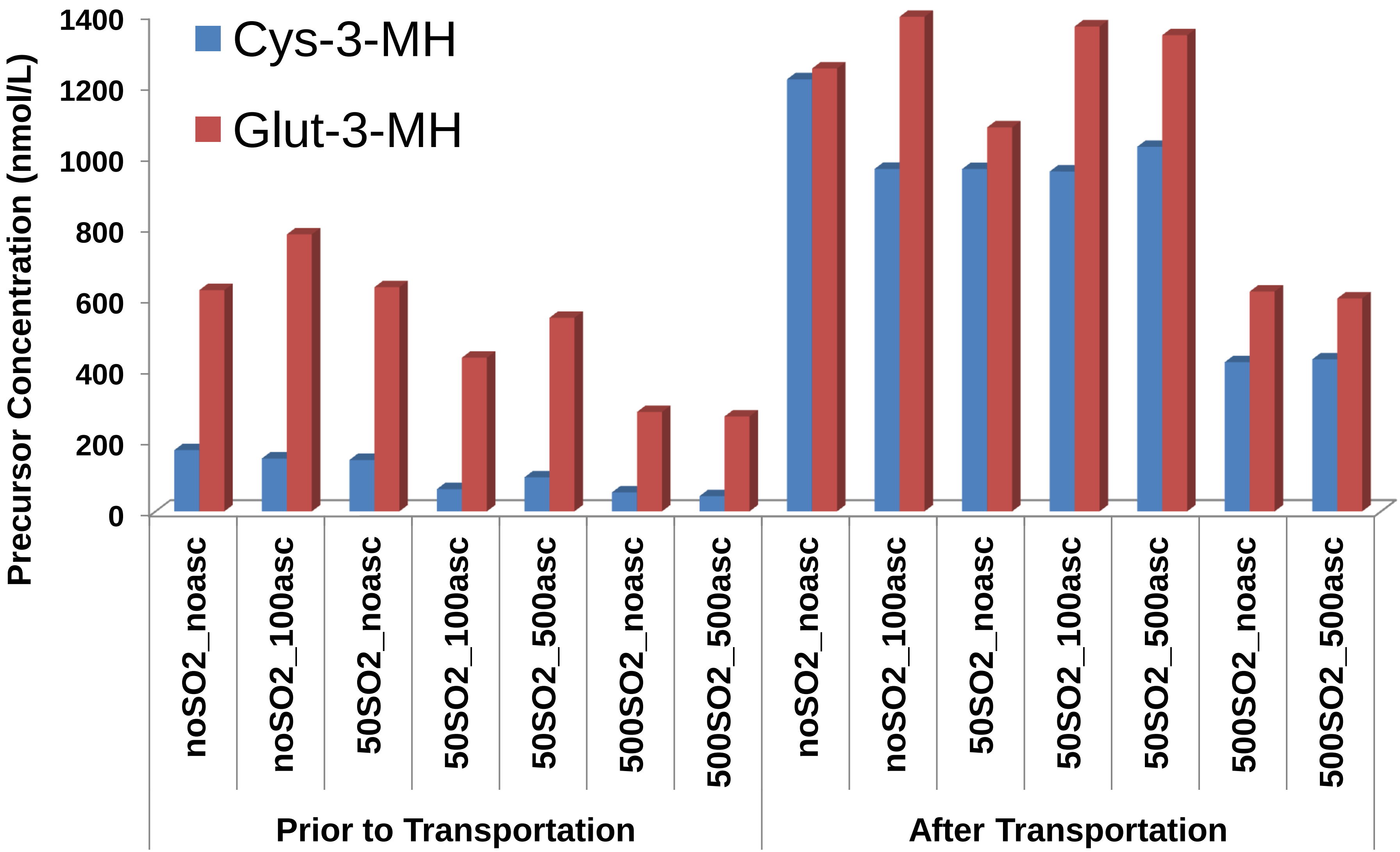

Capone et al. 2011, JAFC. 59: 4659-4667

Modulation of volatile thiol precursors

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- Glutathione 3MH precursor is more abundant than Cysteine 3MH precursor, regardless of grape variety
- 3MH precursors are affected by ripening.
- 4MMP precursor peaked early in ripening season, at approx. 10° Beame
- Mild water stress & moderate Nitrogen supply increased volatile thiols in wine
- Foliage Copper spray pre-veraison decreased volatile thiols in wine
- Foliar Nitrogen fertiliser with & without Sulfur increased volatile thiols in wine
- Botrytis infection affects the levels of volatile thiols in the wine
- 4MMP precursor found in free run juice & light pressings
- 3MH precursor extracted mainly during skin contact – particularly longer periods of maceration and higher temperatures (18-20° C)

Formation pathway to 3-MH

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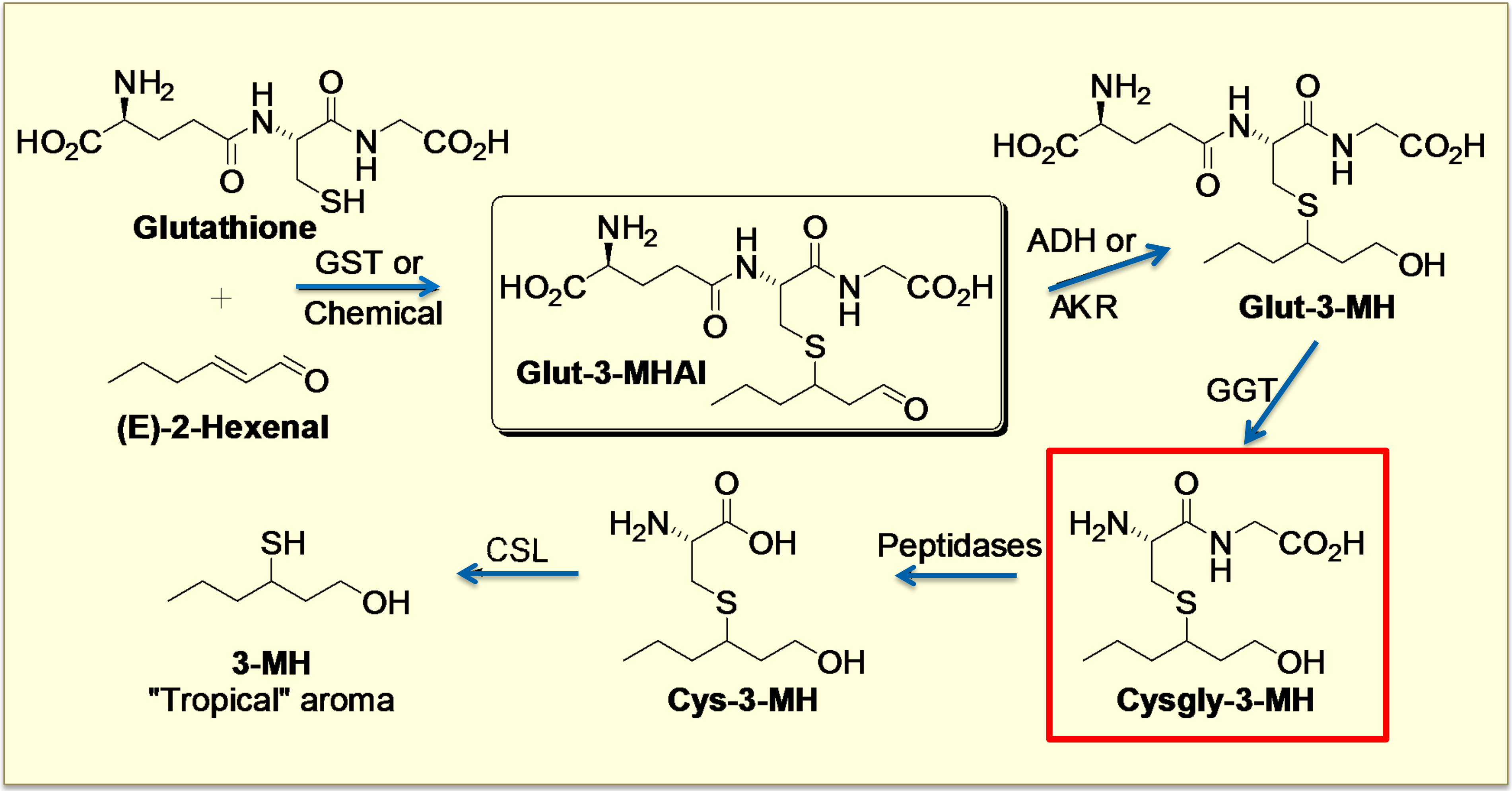

Conclusions – Factors affecting precursor concentration in fruit

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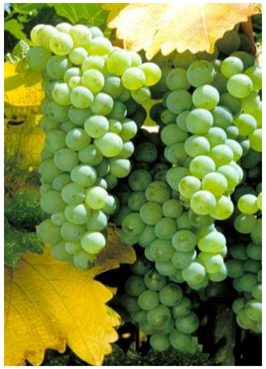
- **Ripening** - Low levels of precursors until commercial harvest

- **Transportation** – inc. precursor for Cys and Glut

  SO_2 and Ascorbic acid – a combination of both optimum – very high SO_2 suppresses conjugate formation

- **Glut-3-MHAl** – tentatively identified as intermediate between (hexenal + glutathione) and Glut-3-MH for the first time

- **Cysgly-3-MH** – Confirmed presence, is short lived



Modulation of volatile thiols

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- Yeast selection
- Higher fermentation temperatures increased volatile thiol levels (20° C compared to 13° C)
- 3MH decreased during malolactic fermentation and barrel ageing
- The addition of Sulfur dioxide stabilised 3MH and 4MMP levels in wine
- Cork closures decreased the levels of 3MH and 3MHA in wine
- 3MHA levels decreased dramatically within the first year of bottling
- Addition of Copper as a wine fining agent decreased volatile thiol levels
- In-mouth release of volatile thiol precursors by saliva bacteria

Yeast strains can release differing levels of volatile thiols

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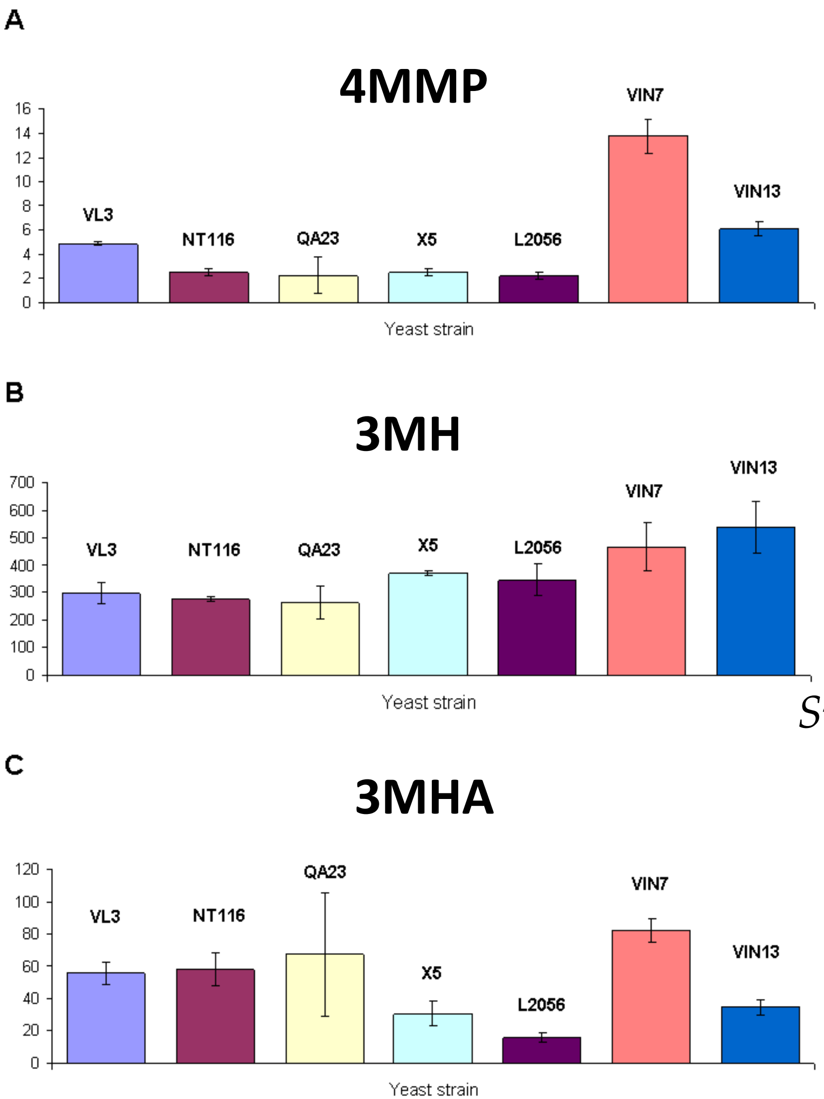

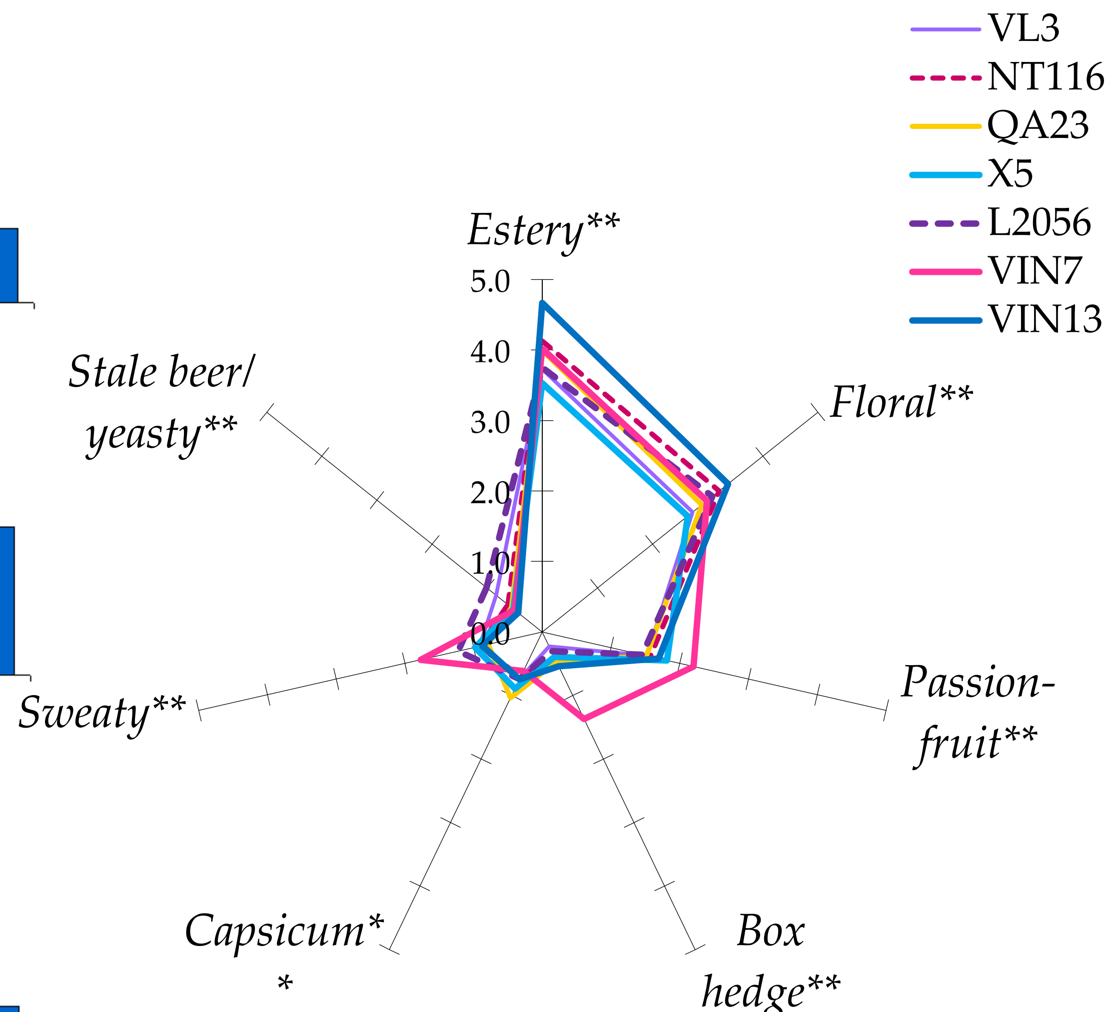

Modified from Swiegers et al. (2009)

Modulation of volatile thiols

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Flavour optimisation – the future

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- Be able to predict concentrations of volatiles from:

Acknowledgements

AWRI

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Australia's grapegrowers and winemakers through their investment body, the Grape and Wine Research Development Corporation, with matching funds from the Australian government

Sensory impact of 3-MHA

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- 3-mercaptohexyl acetate
  - *passionfruit, box tree, sweaty*

- 4 ng/L threshold
- Found in Aust. wine up to 3,000 ng/L
- Found in NZ wine up to 12,000 ng/L
- Final concentration in your glass - 740 ng/L

Sensory impact of 3-MH

The Australian Wine Research Institute

- 3-mercaptohexen-1-ol
  - *grapefruit, passionfruit, leafy*
- 60 ng/L threshold
- Found in wine up to 210 ng/L
- Final concentration in your glass - 7040 ng/L

Sensory impact of thiol mix (3-MHA + 4-MMP + 3-MH)

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- Individual aroma characteristics
  - *grapefruit, passionfruit, leafy, box tree, sweaty*

- Combined aroma characteristics
  - cooked green veg, tropical

Spiked levels in your wine:

- 3MHA - 740 ng/L

- 3MH – 7040 ng/L

- 4MMP – 40 ng/L