



USE OF ENZYMES IN WINEMAKING



**Enzymes form an essential
tool of any winemaking
process !**

3 main types of objectives
Process improvement
Wine quality enhancement
Problem solving

ongoing discussions

Wine enzyme Regulation

**OIV International Enological
Codex and Code**

EU regulation

Application of enological enzymes is defined by OIV International Enological codex and Code

Till 2013	Since 2013
Pectinases for clarification of grape juices	Pectinases, cellulases, glucanases, xylanases for several application : maceration, yield, clarification , ...
Beta-glucanases for filtration of wines	Beta-glucanases for filtration and for ageing on lees
	Glycosidases for hydrolysis of grape aroma precursors
Urease for prevention of ethyl carbamate in wines	Urease for prevention of ethyl carbamate
Lysozyme for prevention of microbial spoilage during fermentation	Lysozyme for prevention of microbial spoilage

Enological enzymes

Enzymes	Production micro-organisms	Production technique	Formulations
Polysaccharide-hydrolases : pectinases, cellulases, glucanases, xylanases ...	<i>A. niger</i> , <i>A. aculeatus</i> , <i>T. longibrachiatum</i> ...	Submerged or surface fermentation	Liquid glycerol/ KCl Avoid Sorbate No benzoate
Glycosidases	<i>A. niger</i>	Mainly Classical , use of self-cloned still very limited	Microgranulates maltrodextrin
Glucanases	<i>T. harzianum</i> , <i>T sp.</i>		
Urease	<i>L. fermentum</i>	?	?
Lysozyme	Egg white	Purified by Chromatography	Pure Microgranulate

Processing enzymes

Quality enhancers

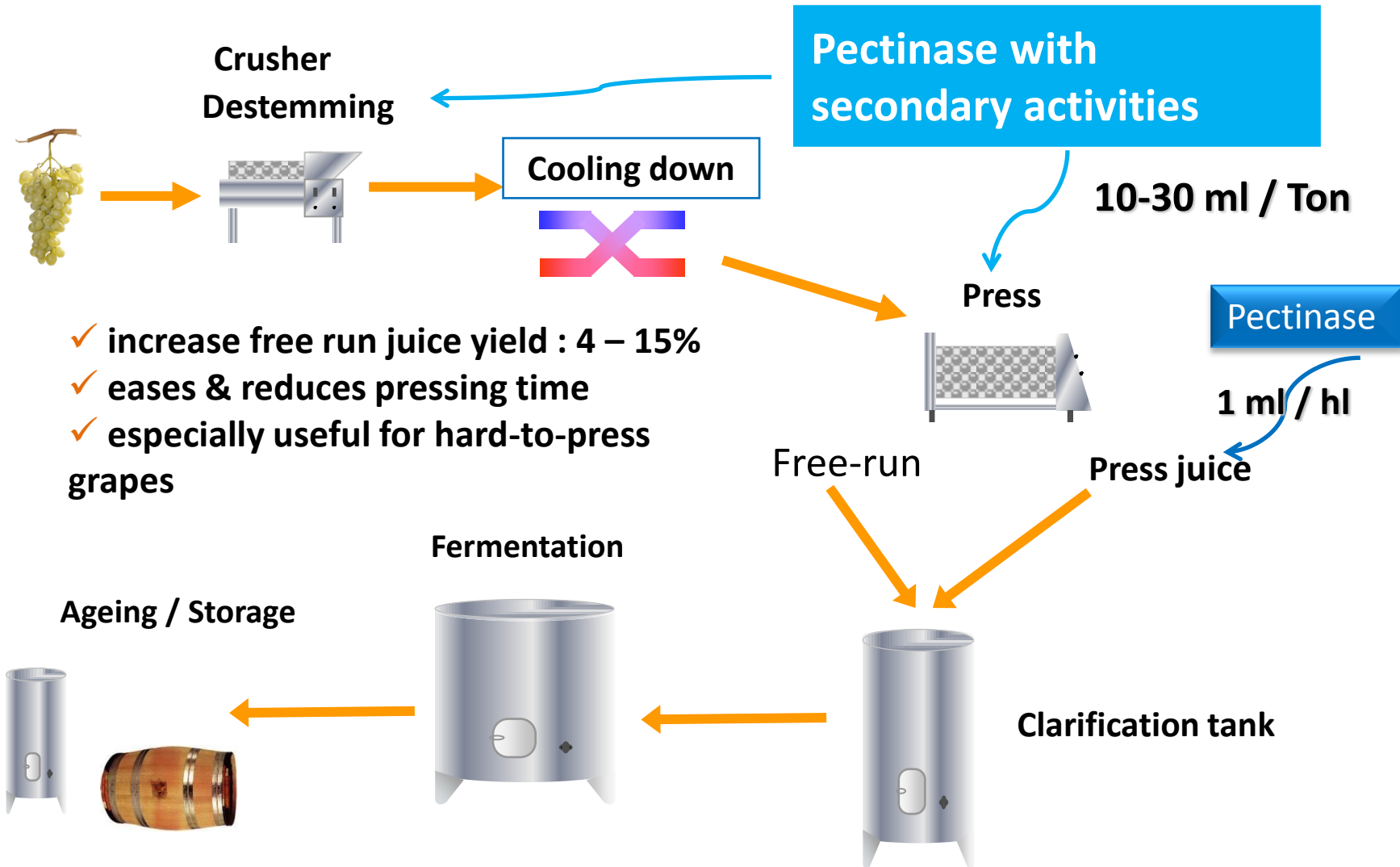
Problem solving tools

Some debates ongoing

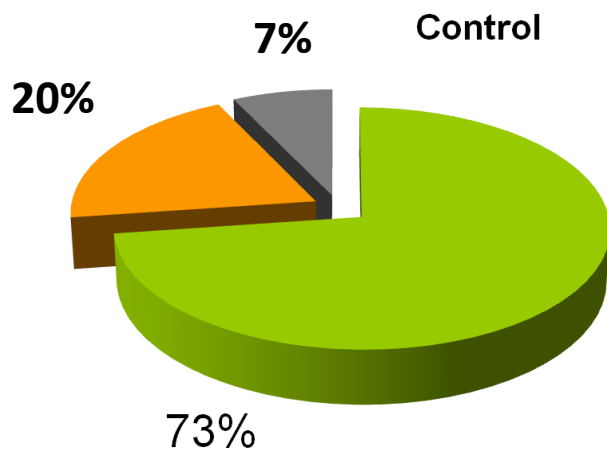
Processing enzymes

Process improved	Activity profile	Benefits for winemakers
Pressing & yield improvement	Pectinases active on homogalacturonans and arabinan side chains	Cost savings , more juice yield
Clarification of grape musts by Flotation	Pectinase active on homogalacturonans	Time and labour savings
Clarification of grape musts by static settling	Pectinase active on homogalacturonans	Energy & water savings
Clarification of grape musts and wines by filtration	Pectinase active on homogalacturonans Glucanases	Sustainability : less vine land used for same production volume
Thermovinification	Thermostable pectinases	More quality juice : drain juice
		Reduced oxidation due to shorter processing time

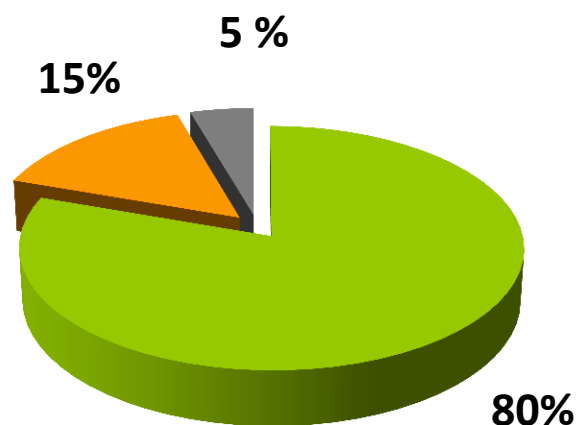
Application of pectinases for pressing & yield improvement



Pressing yields with pectinase



Pectinase with secondary activities



Total yield : 790 Liters/Ton

Total yield : 825 Liters/Ton

- Free run juice
- Low pressure
- High pressure

Chardonnay, Coop winery Languedoc France

Enzyme : 20 g/Ton

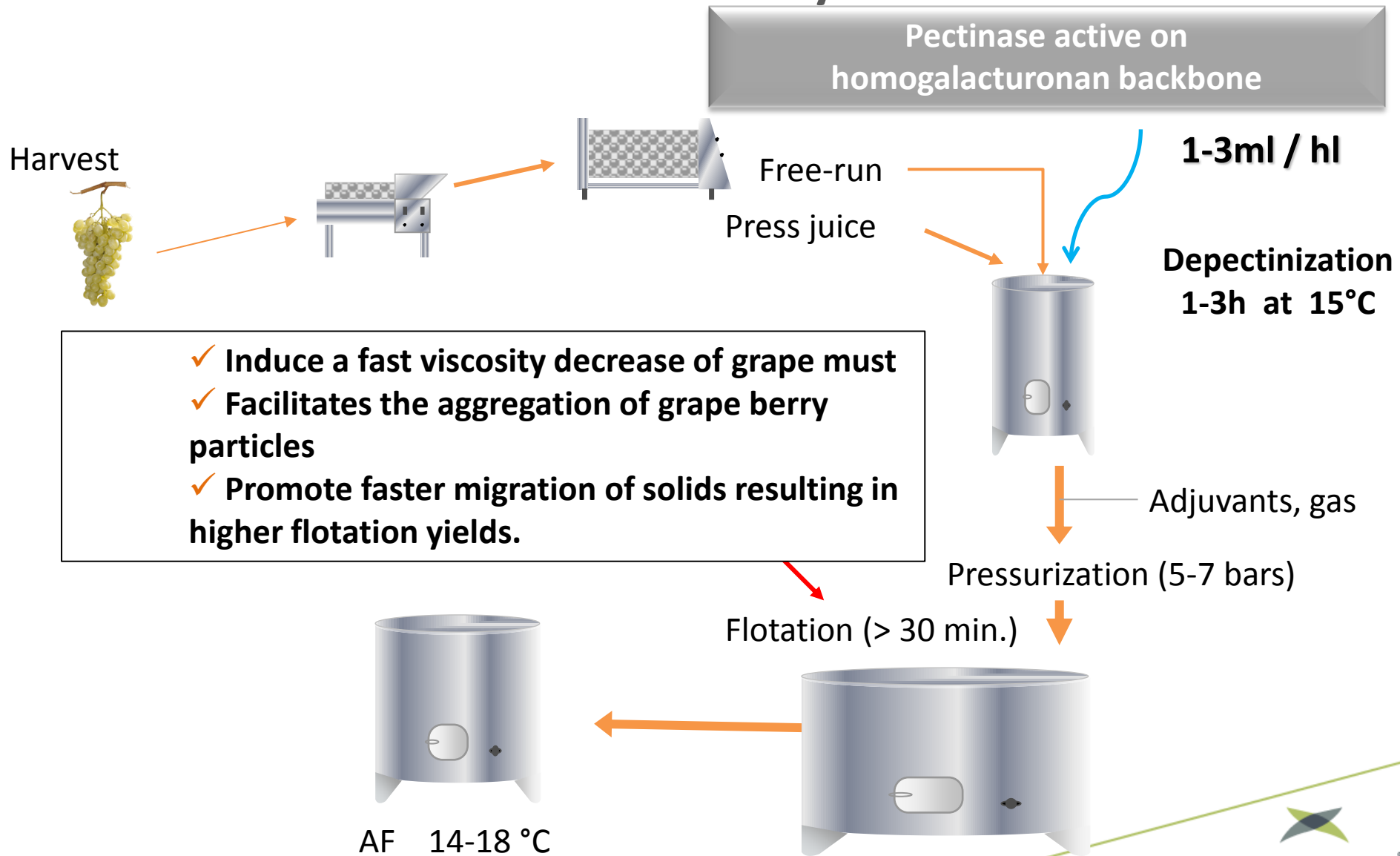
2 h at 20 ° C

Pneumatic press

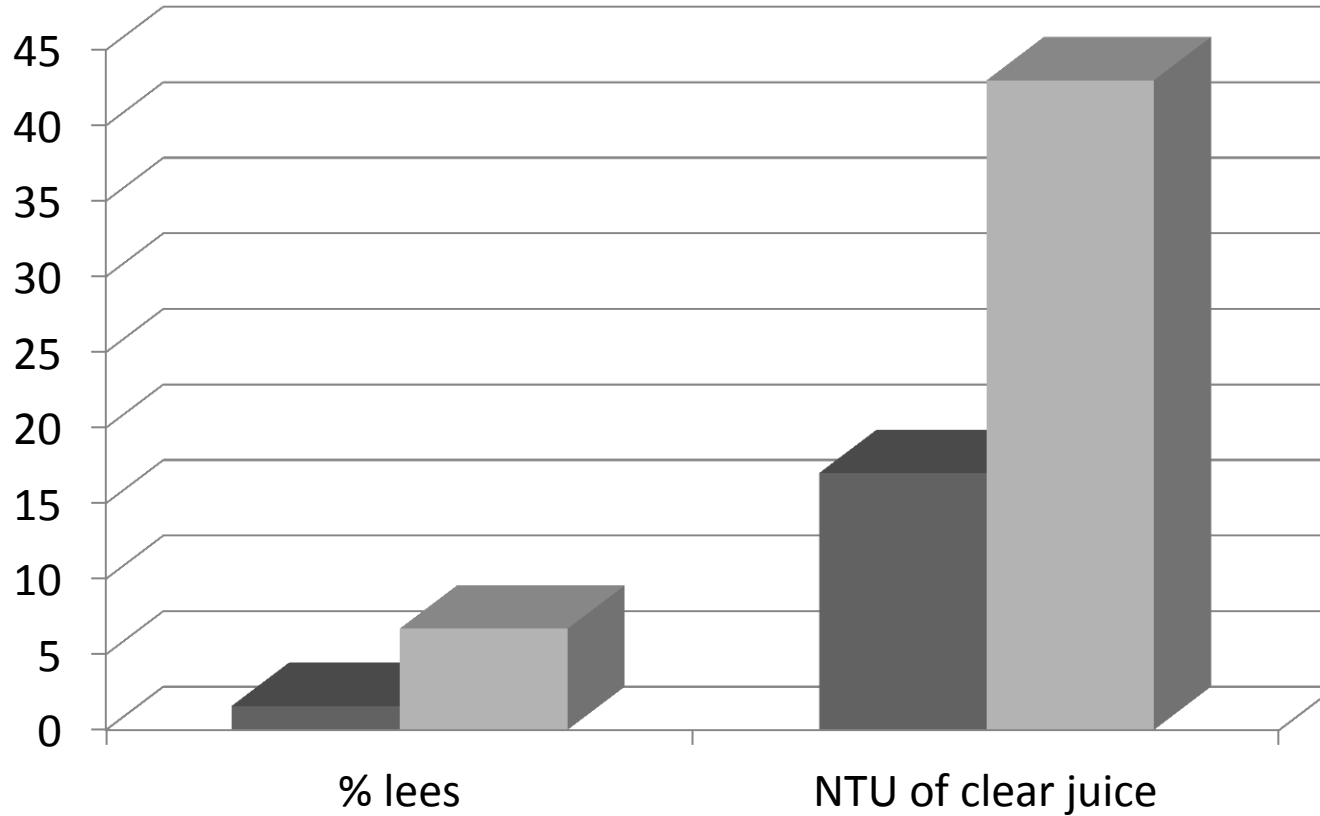
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Clarification of grape musts and wines by filtration	Pectinase active on homogalacturonans Glucanases	Sustainability : less energy used
Thermovinification	Thermostable pectinases	More quality juice : lower turbidity for better wine quality
		Reduced oxidation due to shorter pre-AF processing time

Application of pectinases for grape juice clarification by Flotation

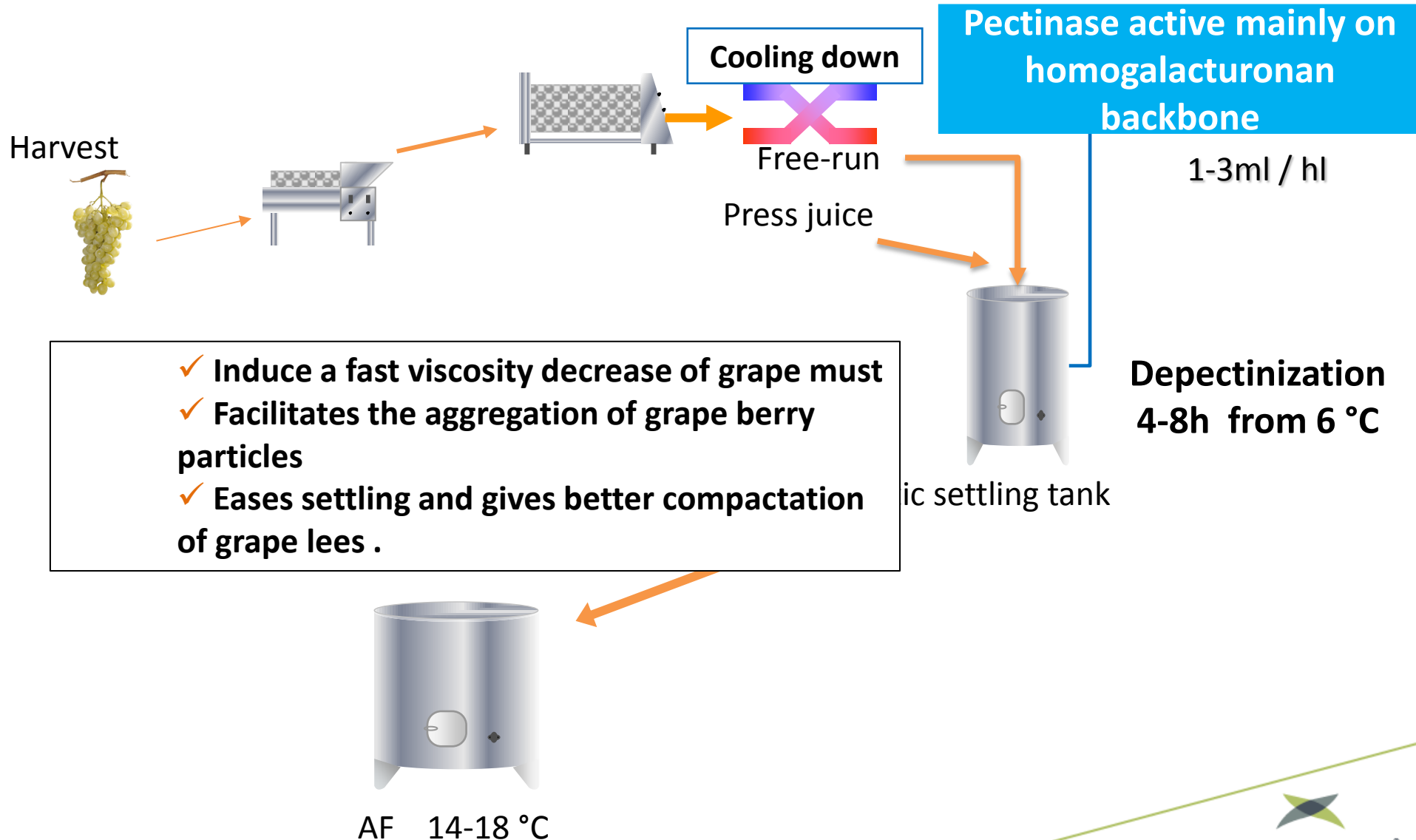


Flotation in Chile, Muscat (Pisco area)



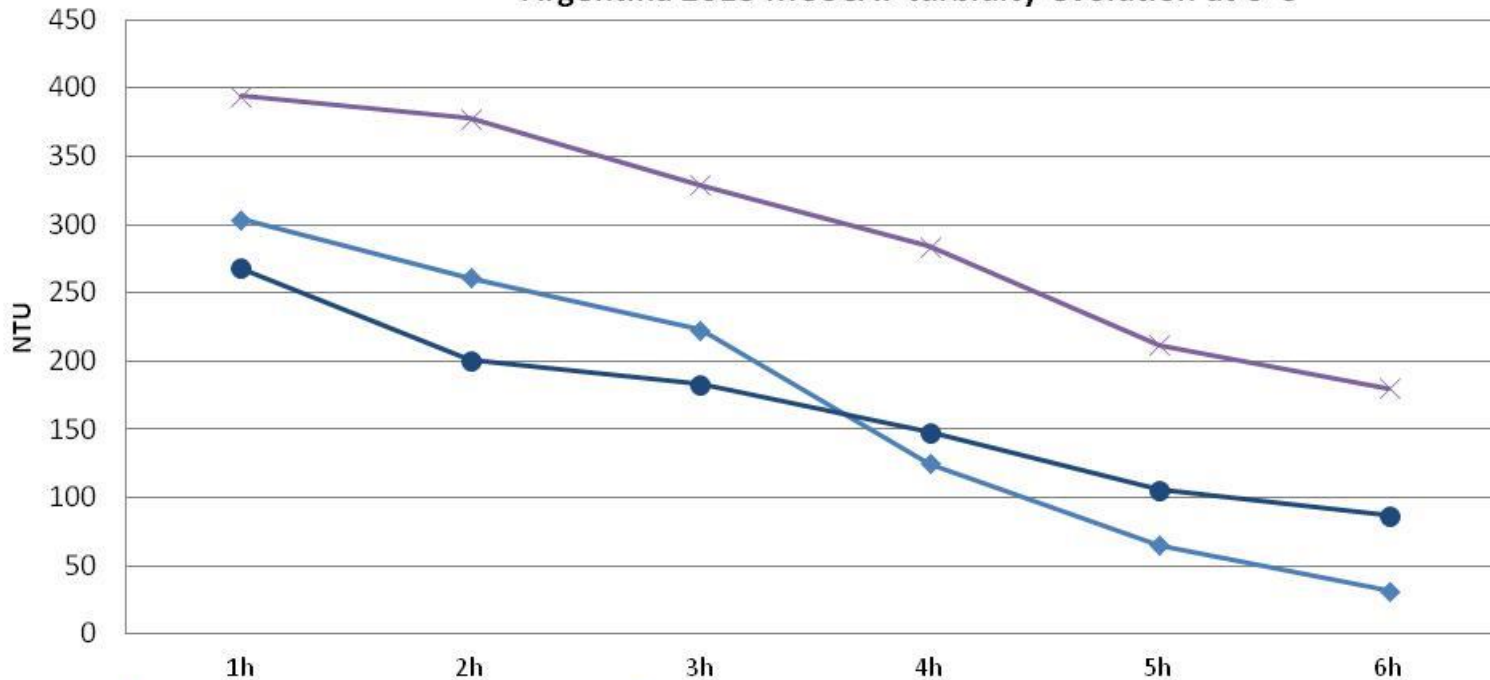
Comparison of 2 Flotation enzymes

Application of pectinases for grape juice clarification by static settling



Static settling in extreme conditions

Argentina 2013 MUSCAT turbidity evolution at 6°C



Different pectolytic enzymes compared

winemaker testimonial: *“The enzyme used resulted in faster, more efficient settling. Depectinisation was completed in 2 hours at 6 °C! In the case of Torrontes, (a variety known for its difficult clarification), we obtained both clearer musts and more compact lees*

Processing enzymes

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Application of beta-glucanases for wine clarification by filtration

Grapes affected by grey or noble rot (*B. cinerea*)



Press



Clarification tank

Fermentation

- ✓ Fast hydrolysis of fungal glucans
- ✓ Faster filtration rates
- ✓ Avoid filter plugging

2-3 ml / Hl

Beta-(1,3-1,6)-glucanases
active on Botrytis glucans

storage

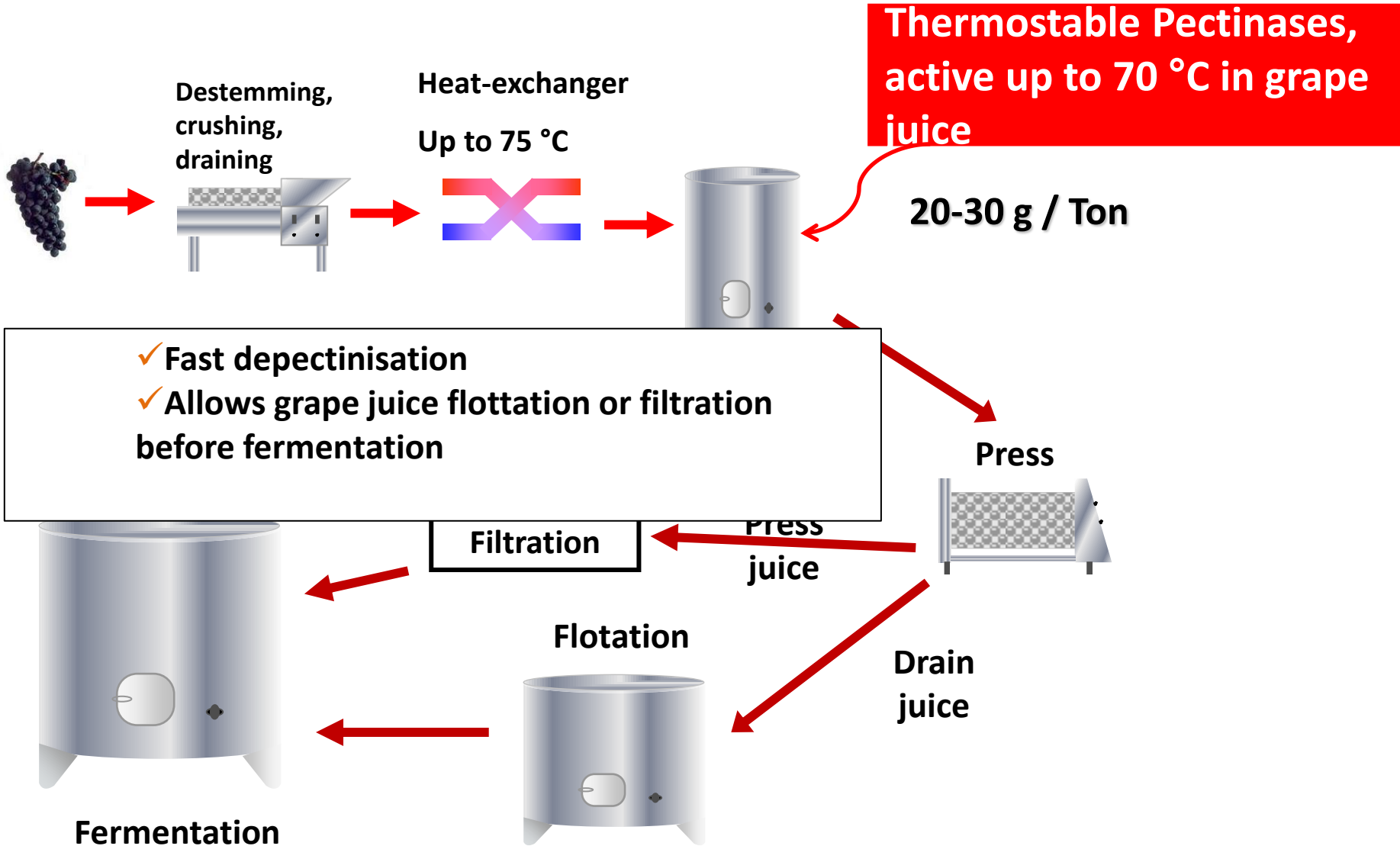
Filtration



Processing enzymes

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Thermovinification	Thermostable pectinases	Process not applicable without enzyme

Application of pectinases on thermovinification



Application of pectinases on thermovinification

Pectin Test	Enzyme maceration time			
	0 min	60 min	120 min	240 min
Thermostable pectinase	3	2/1	0	0
Standard pectinase	3	3	2	1

Carignan grapes
Languedoc 2012

Processing enzymes

Quality enhancers

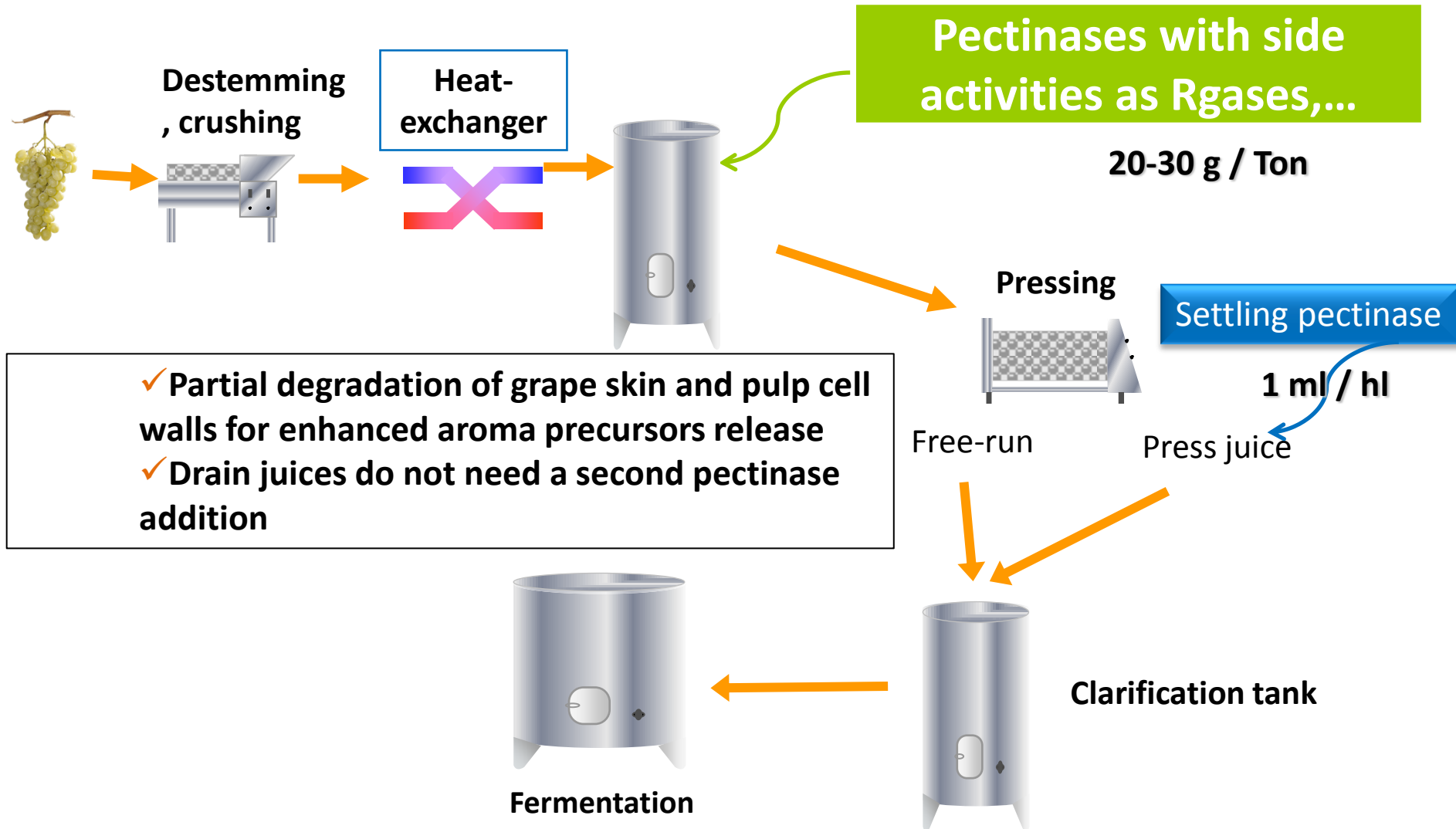
Problem solving tools

Some debates ongoing

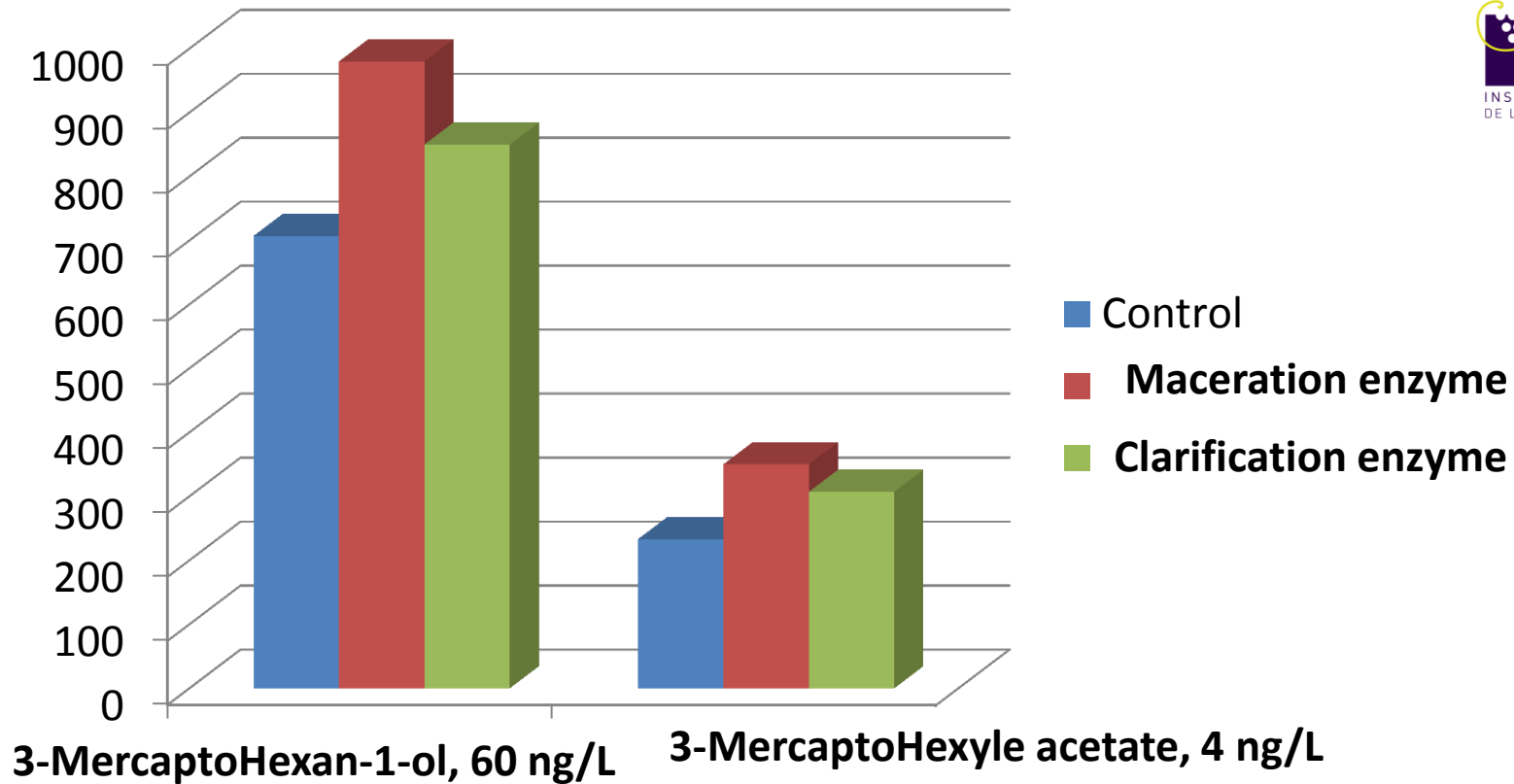
Quality enhancer enzymes

Quality enhancers	Activity profiles	Applied to processing step	Benefits for winemakers
Aroma precursors released from grape skins	Pectinases , Rgases,	Skin contact of white and red grapes	Increase in grape aromas as thiols, terpenes,
Tannins and other polyphenols concentration	Pectinases , Rgases, Xylanases, cellulases	Maceration of red grapes	Increase in color and polyphenols, essential for red wine quality
Non volatile glycosylated grape aroma turned into volatile flavors	Glucosidase, arabinosidase, rhamnosidase, apiosidase	Applied to wine	Increased in terpenes and norisoprenoids
Release of specific yeast compounds ()	Beta-(1,6)-glucanases	Ageing on lees, batonnage	mouthfeel, aroma, stability improvement

Application of complex pectinases for grape skin contact

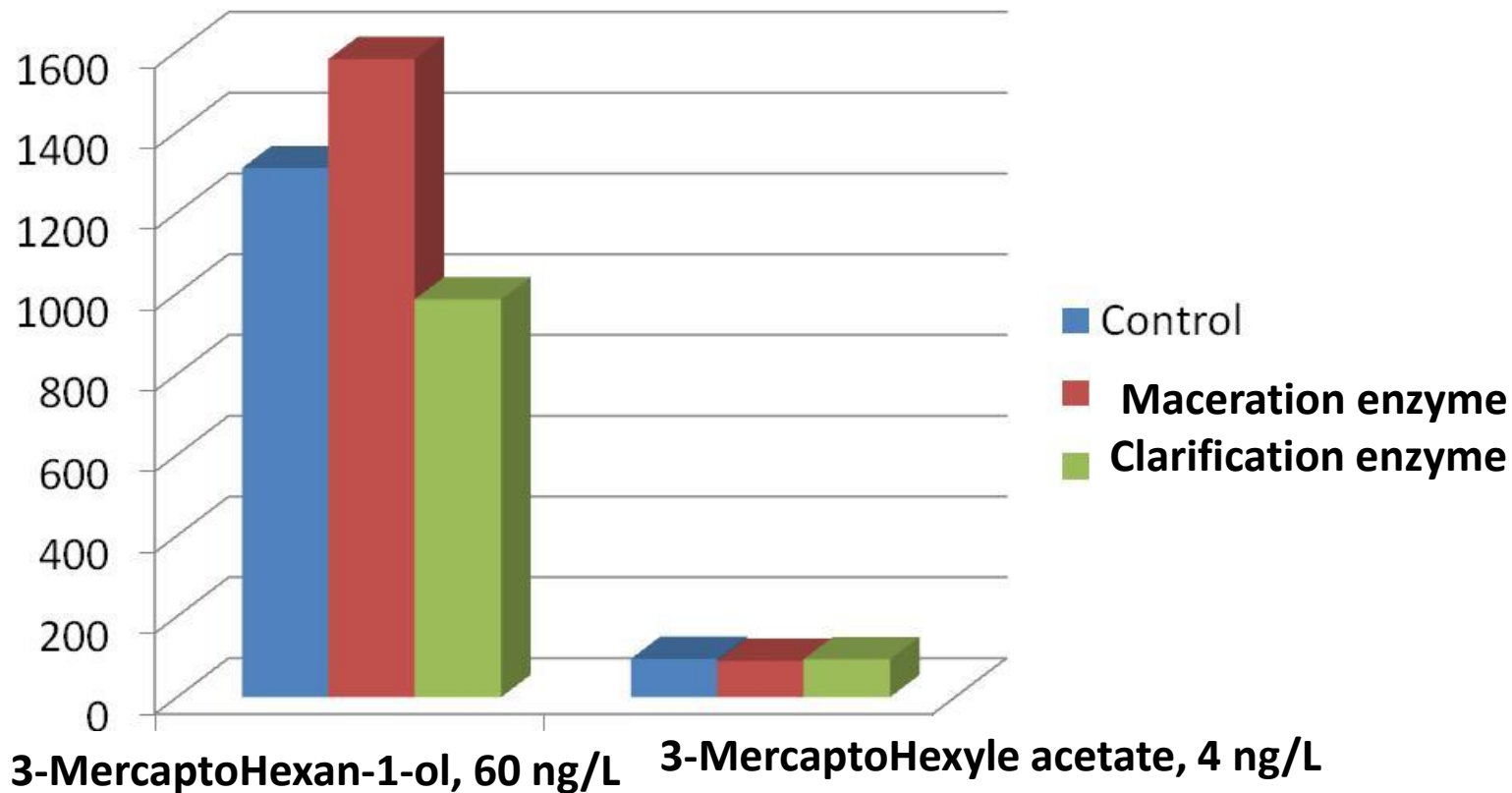


Maceration enzymes and their impact on white wine aroma : example Sauvignon blanc



With thiol converting yeast

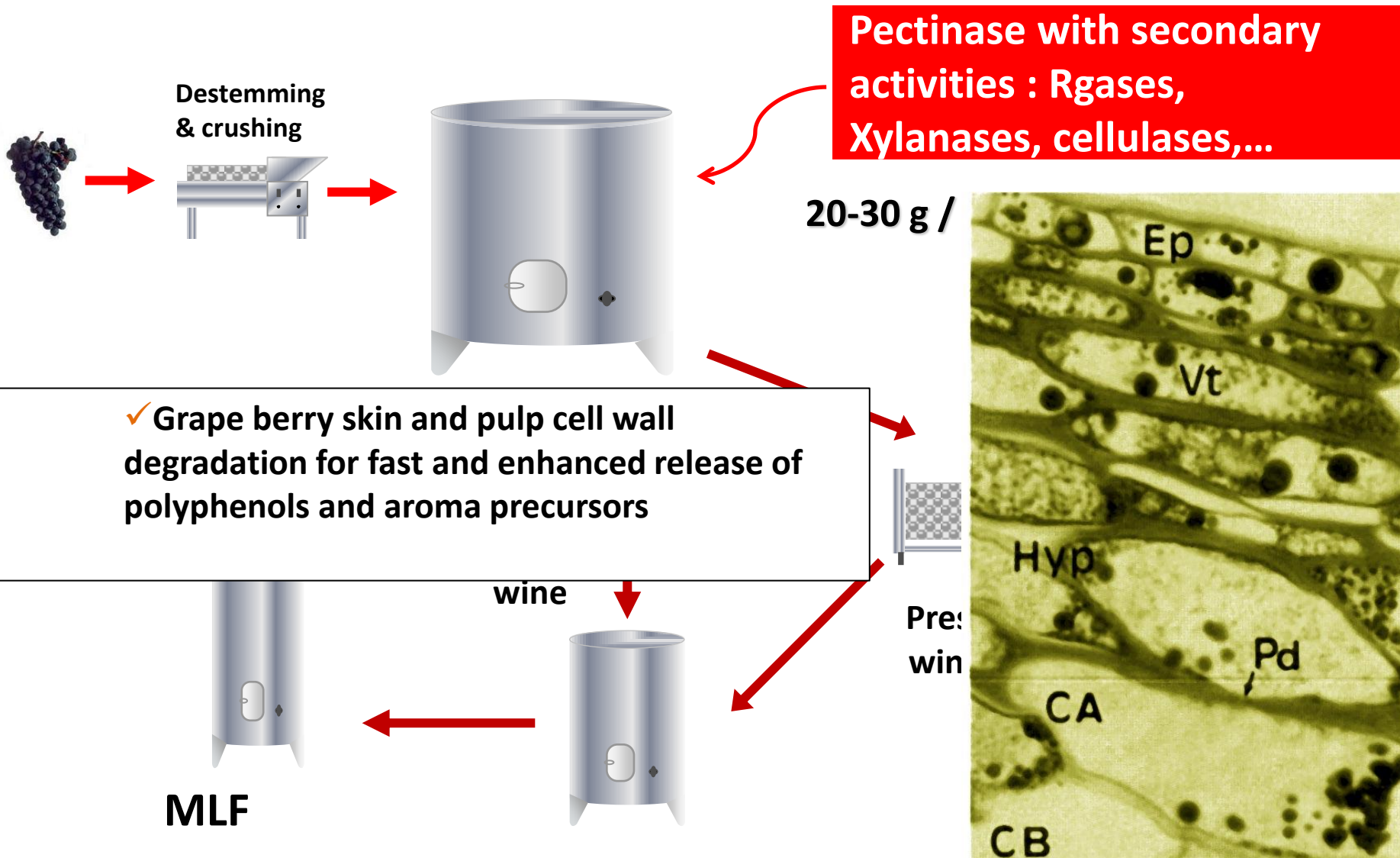
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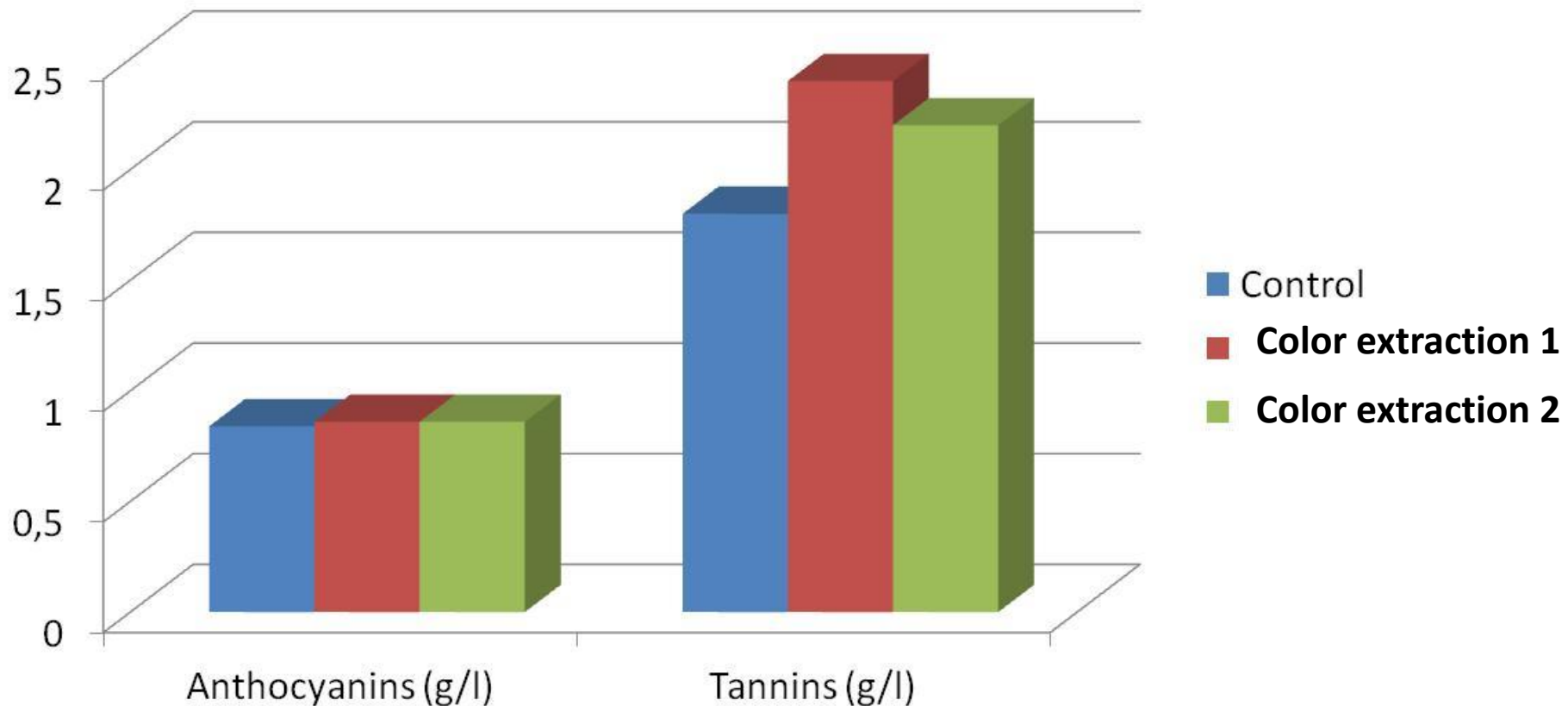
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Application of red wine maceration enzymes



Maceration enzymes and their impact on red wine polyphenols

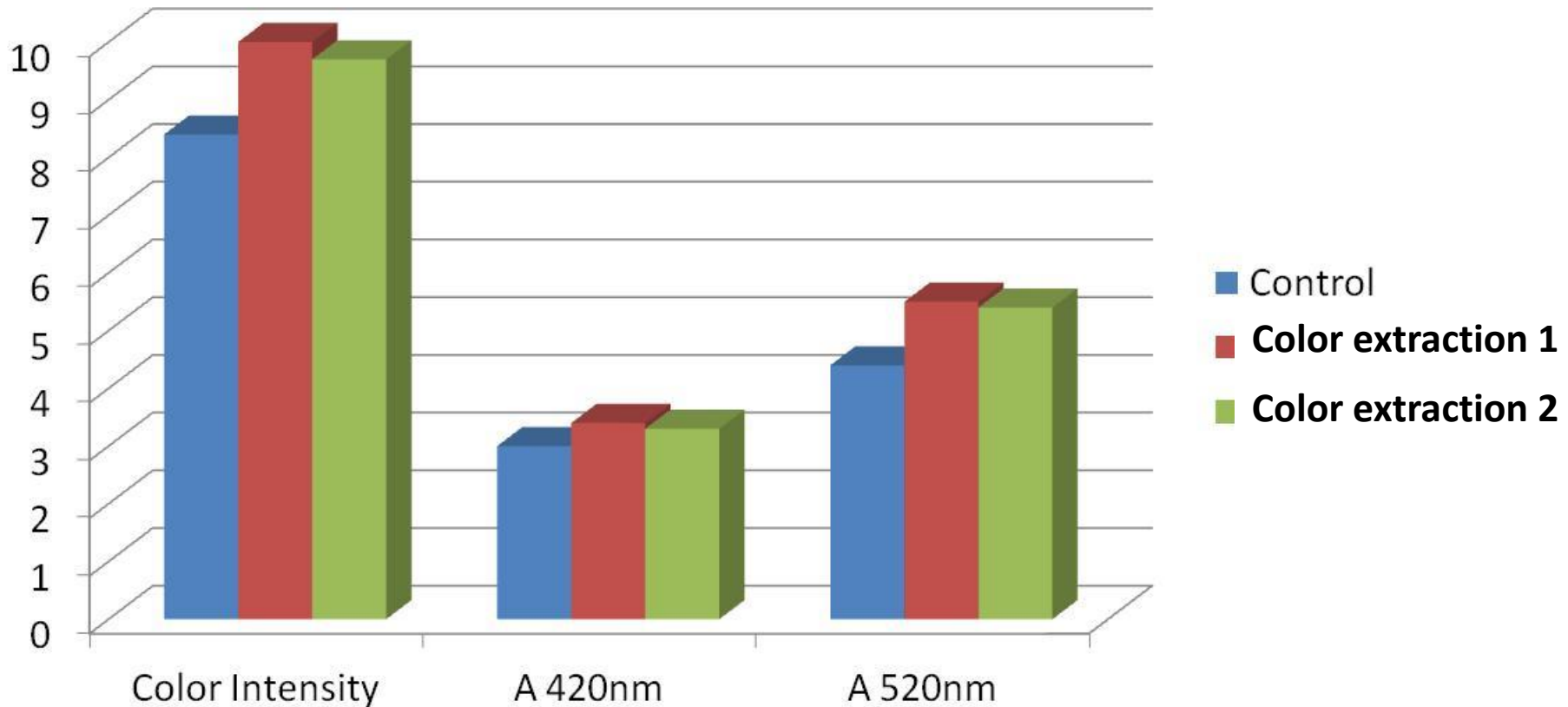


⇒ Tempranillo wines, 12 days maceration

⇒ AF complete

⇒ Before MLF

Maceration enzymes and their impact on red wine color

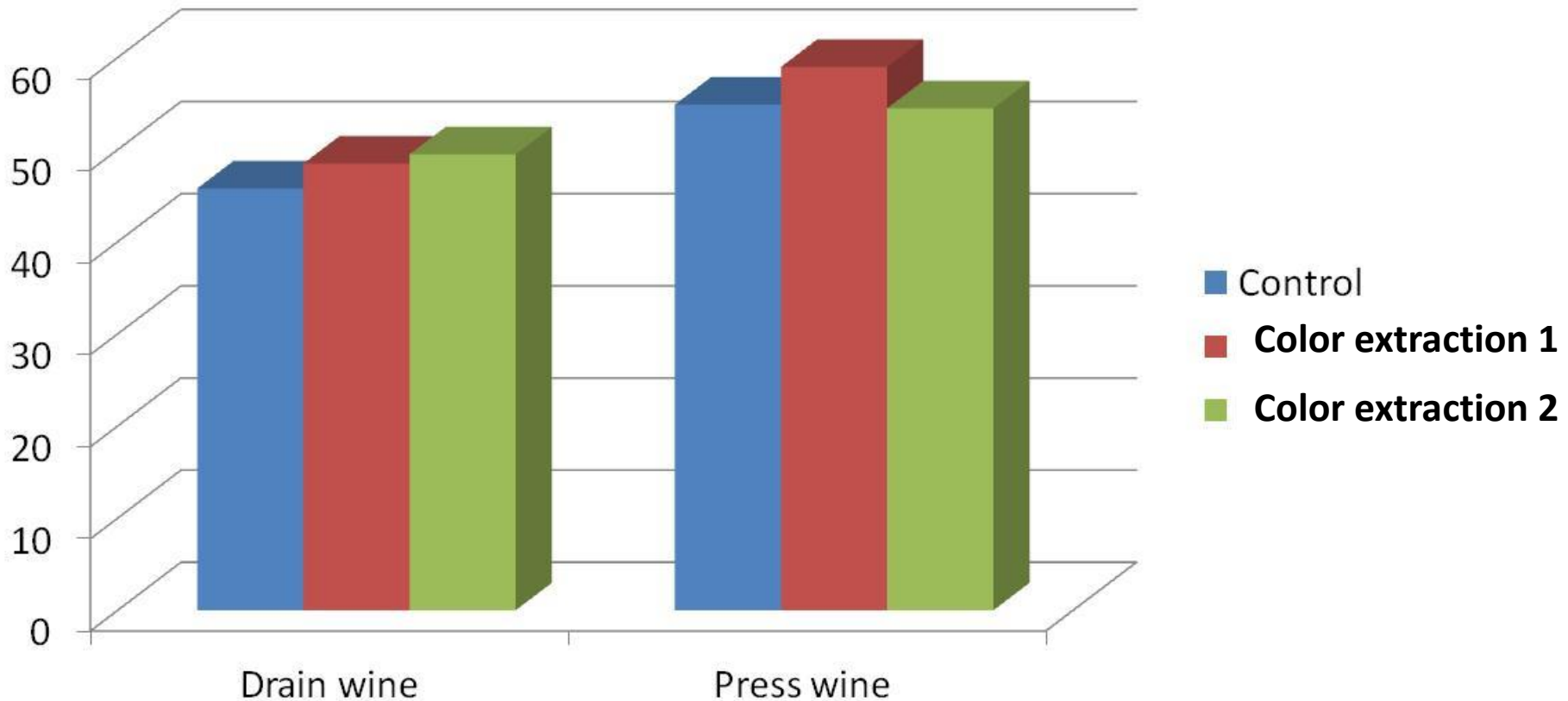


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⇒ Before MLF

Maceration enzymes and their impact on red wine polyphenols Index



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Glycosylated grape aroma precursors

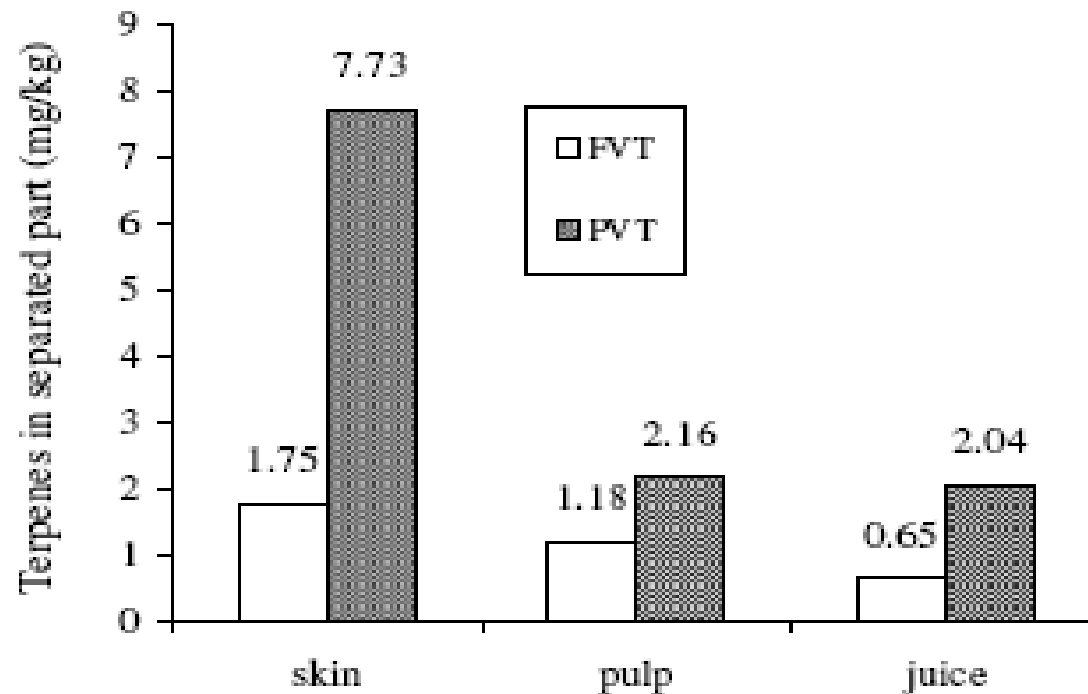
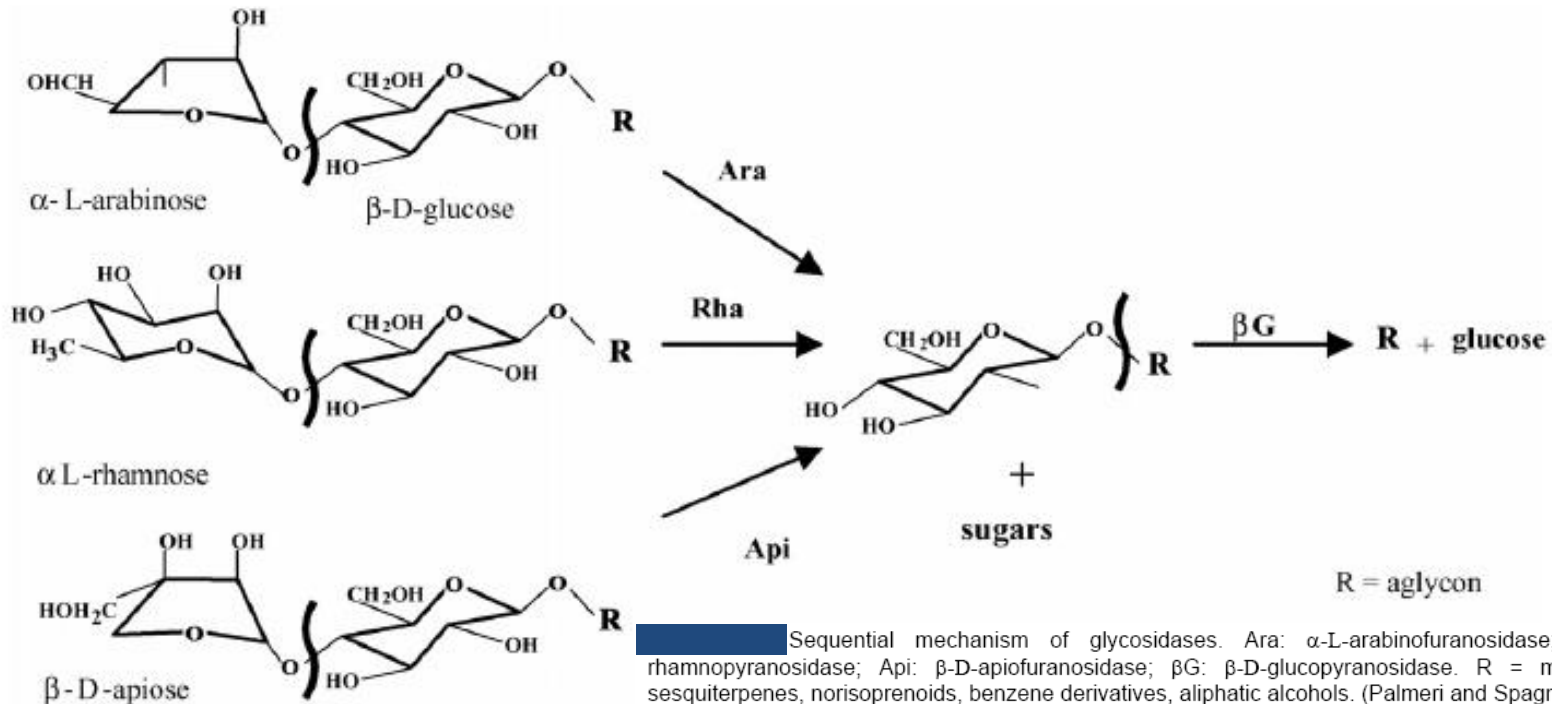
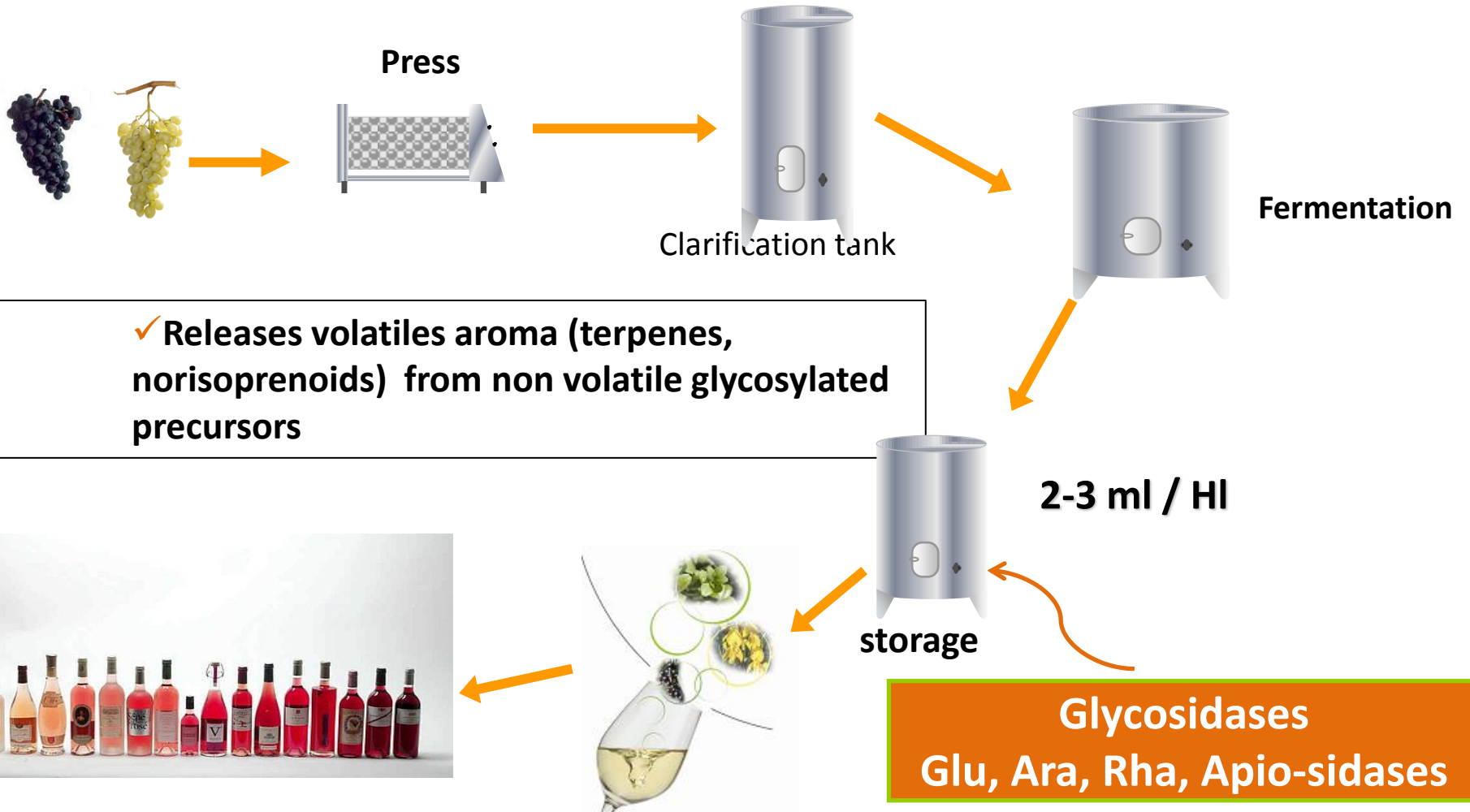


Fig. 1. Content of free and potentially volatile terpenes in grape variety Muscat Italia

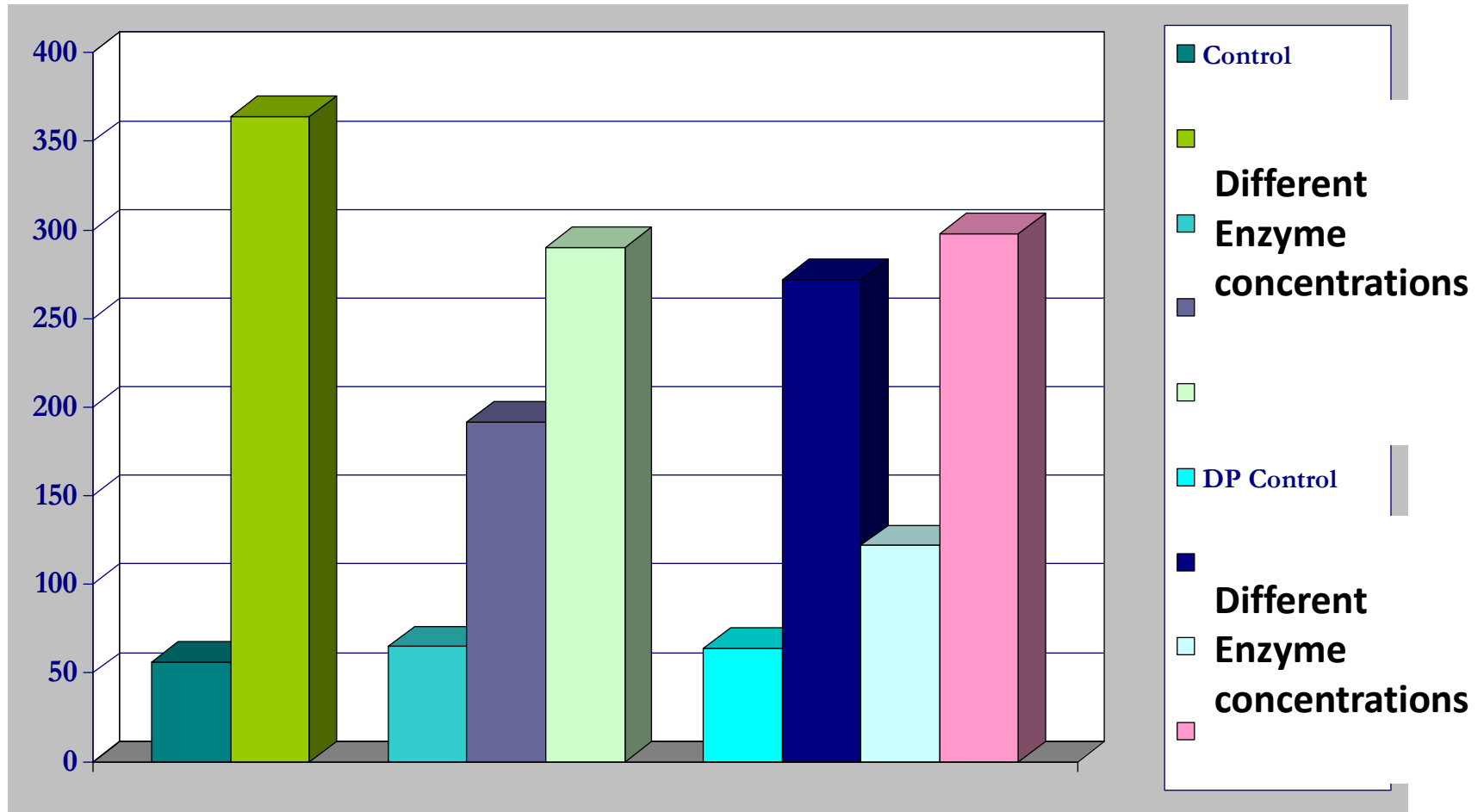
Glycosylated grape aroma precursors : sequential enzyme hydrolysis



Application of Glycosidases



Enzyme induced release of geraniol from its glycosylated precursors in a Muscat wine



Processing enzymes

Quality enhancers

Problem solving tools

Some debates ongoing

Problem solving enzymes

Problem solved	Enzyme	Applied to processing step	Benefits for winemakers
Spoilage by lactic bacteria	Lysozyme	Fermentation	Avoid spoilage due to heterofermentative lactic bacteria
Excess of ethyl carbamate	Urease	Wine	Analytical profile No real market

Processing enzymes

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Some debates ongoing

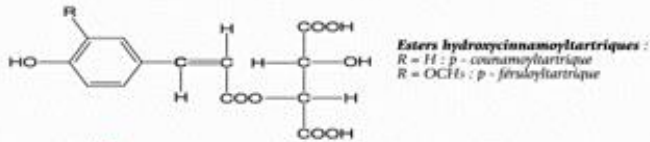
New regulation developments and ongoing Discussions

Proposed for aproval	Debate ongoing
Proteases and the prevention of protein haze in wines	Cinnamyl esterase and the prevention of spoilage by <i>Brettanomyces</i>

No consensus found

ongoing discussion : Cinamyl esterase and red winemaking

SYNERGIE LEVURE/ENZYME : stabilisation de la couleur



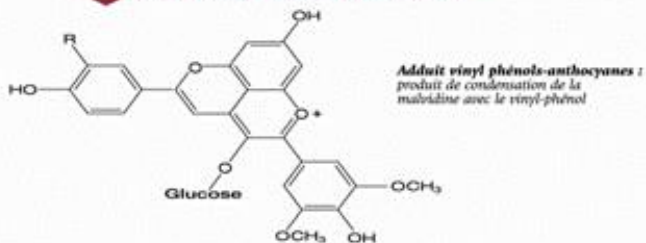
Cinnamyl esterase



HCDC+ yeast



Cyclo-addition, réaction spontanée des
vinyl phénols avec les anthocyanes



? Formation of stable pyranoanthocyanins

- cinnamyl esterase and HCDC activity of **POF(+)** yeast form vinyl phenols
- In red winemaking spontaneous cycloaddition with anthocyanins to form stable pigments
- partial prevention of ethyl phenol formation by the spoilage yeast *Brettanomyces bruxellensis*

Conclusion

enzymes have bright future in winemaking

better understood molecular mechanisms allow design of more “specific” products



- RAPIDASE® Expression Aroma ● RAPIDASE® Batonnage
- RAPIDASE® Revelation Aroma ● RAPIDASE® Extra Color
- RAPIDASE® Thermoflash ● RAPIDASE® Extra Fruit Rouge
- RAPIDASE® Rosé ● RAPIDASE® Extra Press
- RAPIDASE® Flotation ● RAPIDASE® Clear Extreme
- RAPIDASE® Clear



Les enzymes DSM vous apportent la tranquillité d'esprit
 Les enzymes Rapidase sont produites par DSM, le leader mondial sur le marché des enzymes perfectionnées alimentaires. Parmi les entreprises DSM, les plus grandes industries de production d'enzymes, nous disposons de l'équipe la plus expérimentée dédiée à DSM. Ce engagement nous assure que nos produits de DSM peuvent répondre aux besoins de vos clients, de la récolte à la production finale de vin. Les enzymes de DSM sont disponibles en formats adaptés à vos besoins. Les enzymes de DSM sont disponibles en formats adaptés à vos besoins. Les enzymes de DSM sont disponibles en formats adaptés à vos besoins.

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