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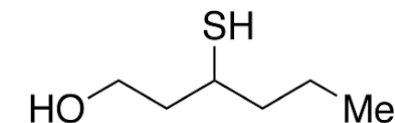
Rotundone in red wines

An overview of the impact of plant material,
environmental factors and viticultural techniques

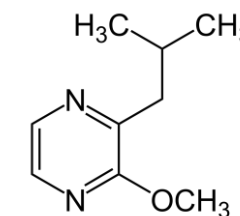


A few introductive words on wine aroma

- More than 800 volatile compounds
- About 10% of them have a sensory impact
- Odorants belong to several chemical families
- Concentration levels from ng/L to hundreds of mg/L
- If the varietal aroma of white wines is quite easy to apprehend (monoterpenols, varietal thiols...)
- Those of red wine remained more mysterious or limited to undesirable molecule such as IBMP....
- Until the discovery of rotundone in 2008 !



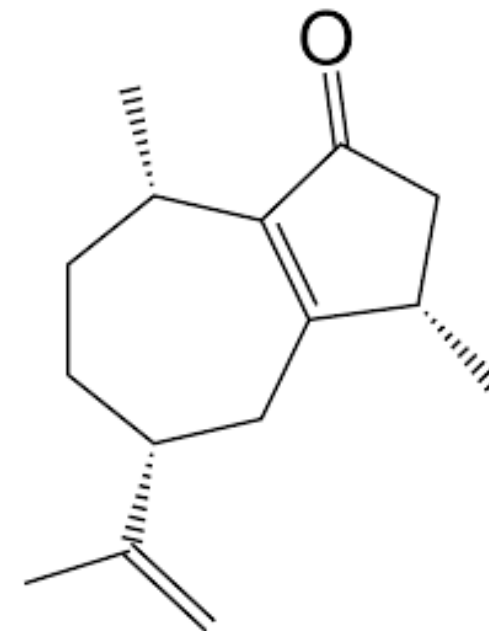
Varietal thiol
Ex. 3-sulfanylhexanol



Alkyl-méthoxypyrazines
Ex. IBMP

Interesting facts about rotundone (1/2)

- Sesquiterpènes ($C_{15}H_{22}O$) = active compounds of essential oils
- Found in *Cyperus rotundus*, pepper and aromatic plants
- Odour threshold of 8 ng/L (water) and 16 ng/L (wine)
- Heavy compound less volatile than monoterpenes
- Produced by enzymatic oxydation of α -guaiene
- 20-25% of specific anosmia reported
- Positively perceived except by young wine consumers
- Particularly appreciated by wine connoisseurs who usually spent more than 10 euros for a bottle wine



Wood et al. (2008), JAFRC

Interesting facts about rotundone (2/2)

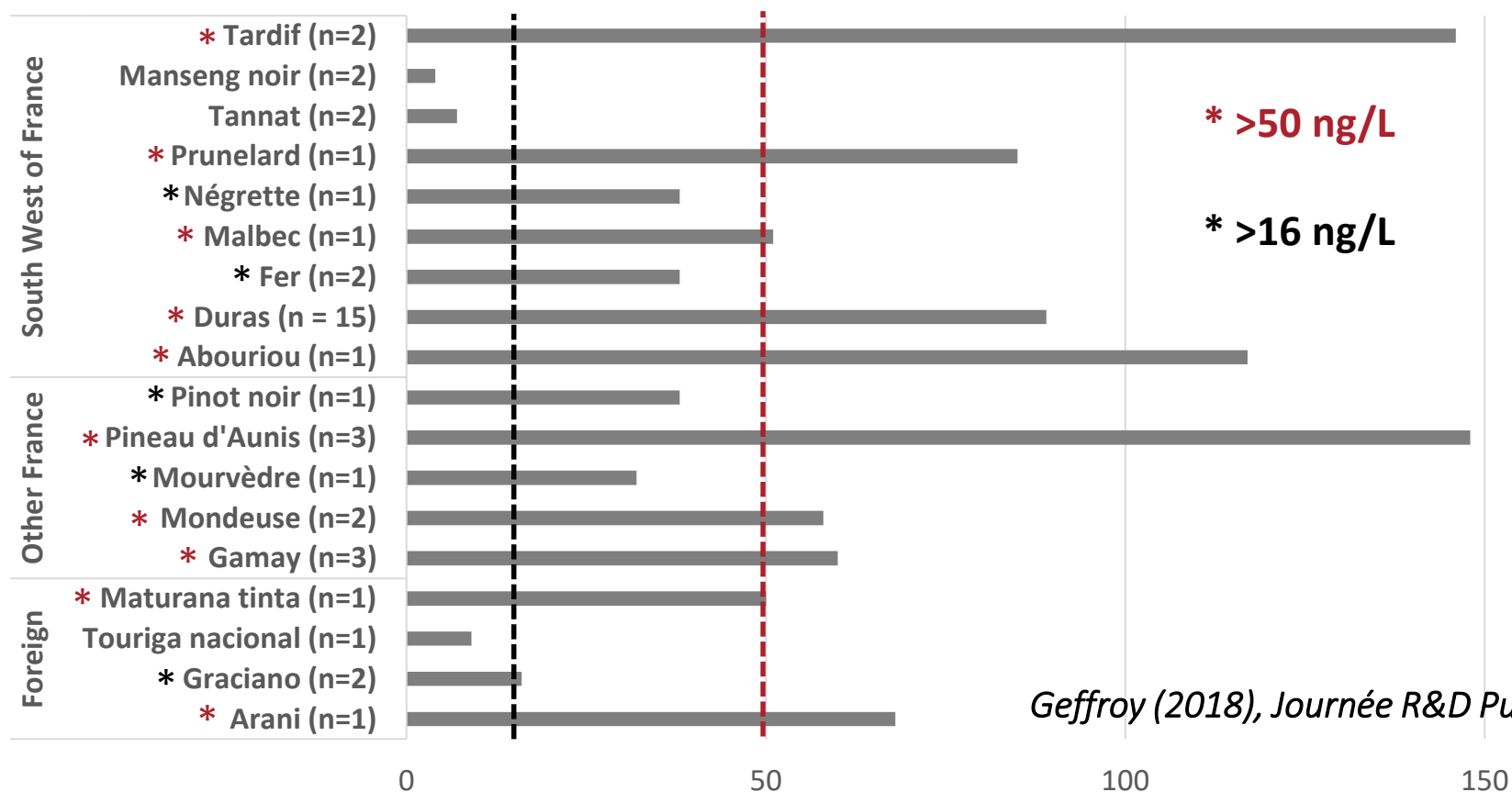
- 96-99% of rotundone is located in the skin
- Hydrophobic compound ($\approx 10\%$ extracted)
- Strong affinity with particles
- Winemaking practices have been identified to lower rotundone in wine (*Saccharomyces uvarum*, carbonic maceration)...
- But not to enhance rotundone, including the increase in maceration temperature and the use of pectolytic enzyme
- Effort to maximize rotundone in wine must be undertaken in vineyard !



Caputi et al. (2011), JAFC

Impact of genotype

Occurrence in wines frequently described as peppery (2008-2015)



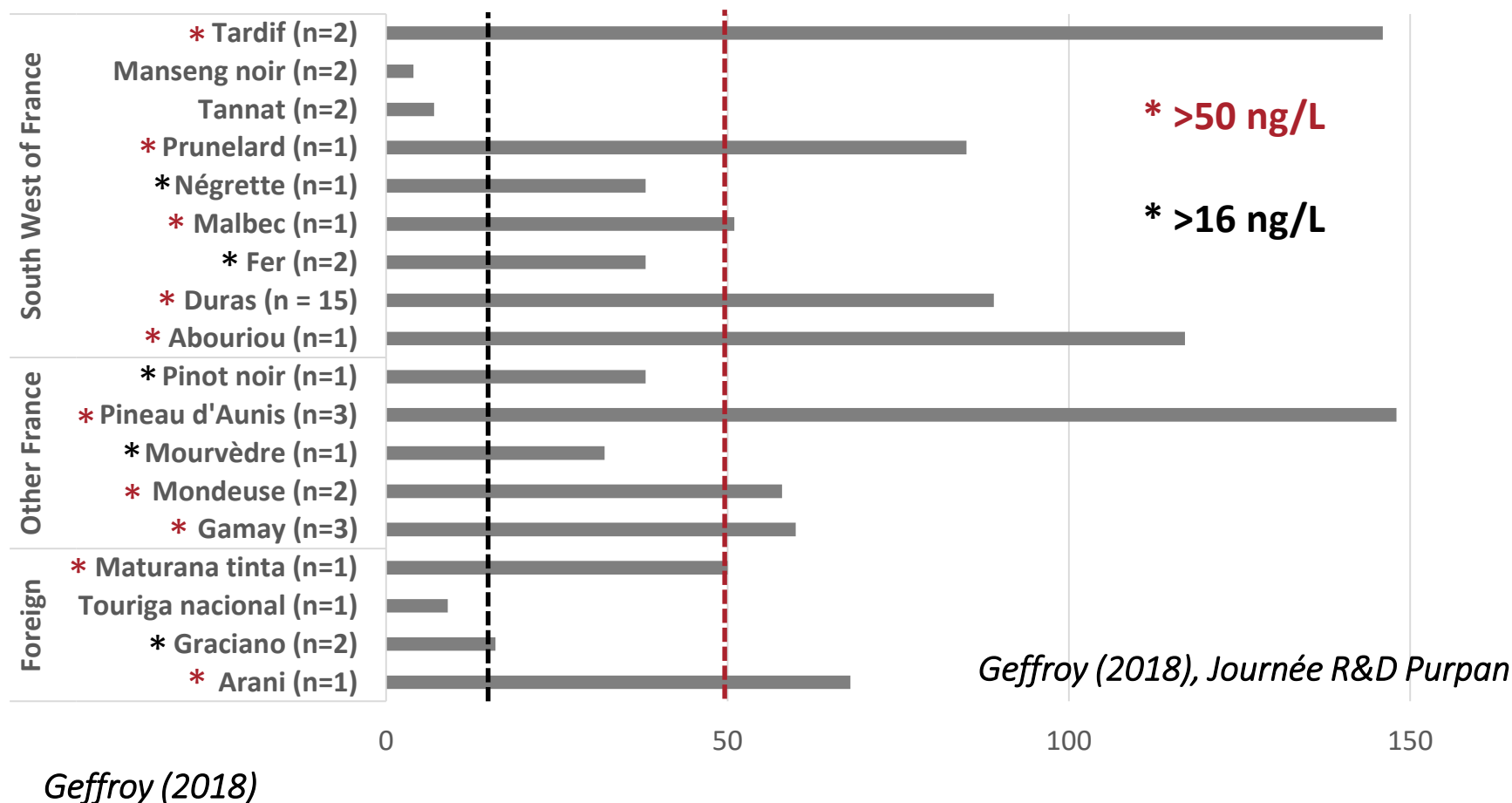
Geffroy (2018), Journée R&D Purpan



Geffroy (2018)

Impact of genotype

Occurrence in wines frequently described as peppery (2008-2015)



Merlot
19-62 ng/kg

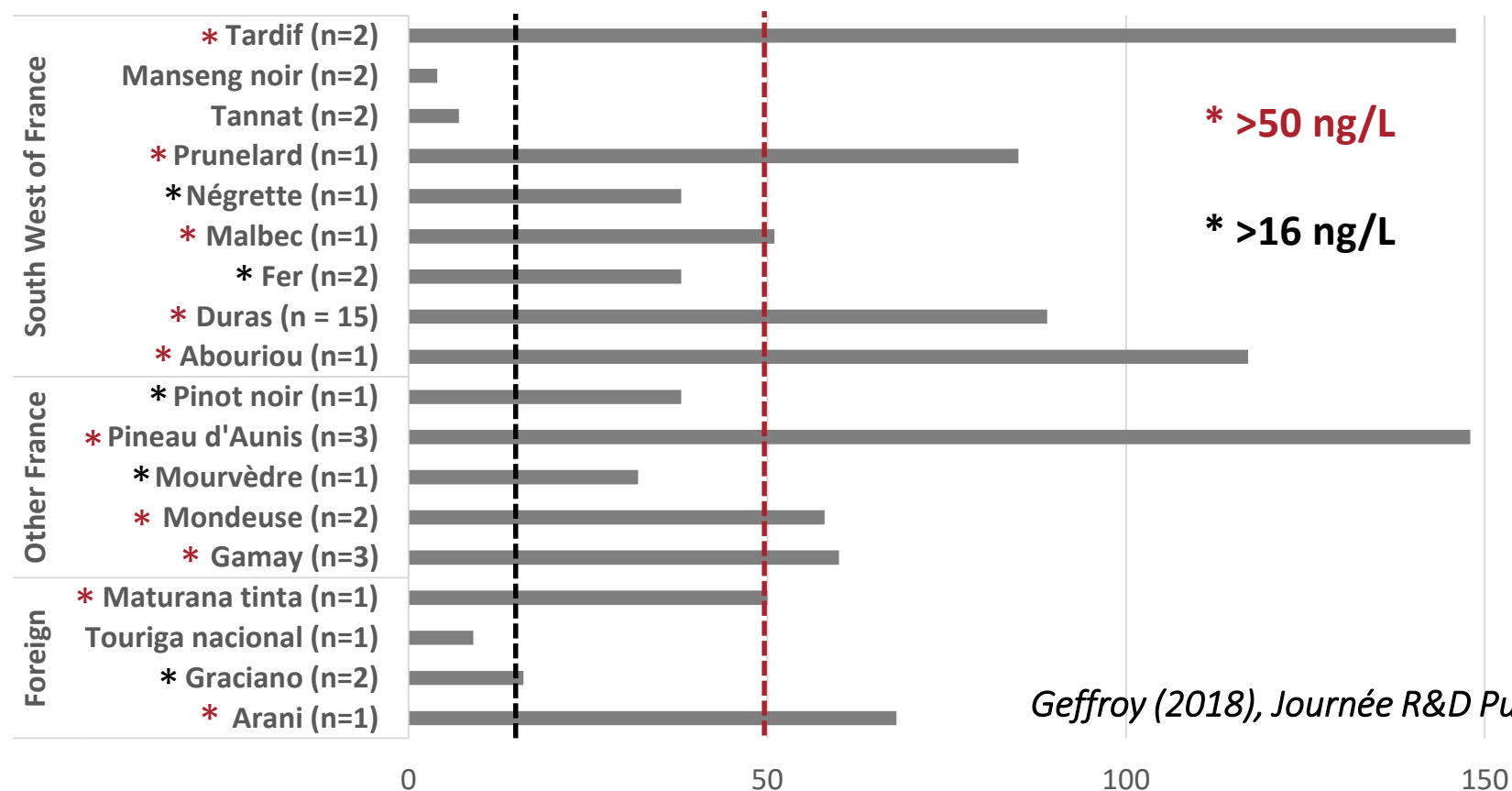
Cabernet Franc
243 ng/kg

Cabernet Sauvignon
21 ng/kg

Takase et al. (2015), AJEV
Logan (2015), Doctoral Thesis

Impact of genotype

Occurrence in wines frequently described as peppery (2008-2015)



Geffroy (2018)

Geffroy (2018), Journée R&D Purpan



*Up to 1345 ng/L
in spirits (whisky,
tequila, rum)*

*Concentrations
increase with barrel
aging time*

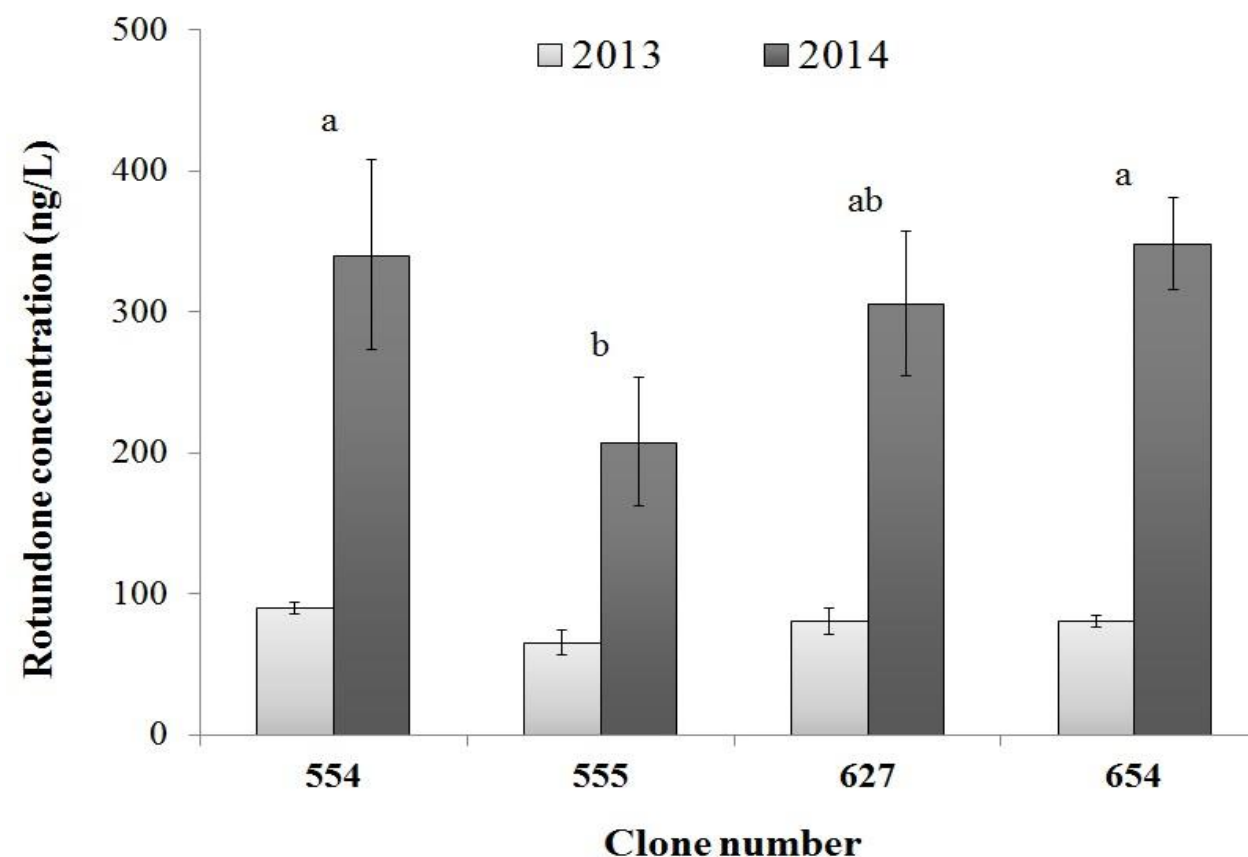
Genthner (2014), Master Thesis

Impact of genotype

Clonal differences observed for Duras

- Clones 554 and 654 showed the greatest concentrations...
- While clone 555 the lowest
- In accordance with other works conducted on Shiraz (Siebert and Solomon 2011) and Grüner Veltliner (Caputi et al. 2011)
- How to explain the extremely high levels for the 2014 vintage ?

Geffroy et al. (2015a), JISVV

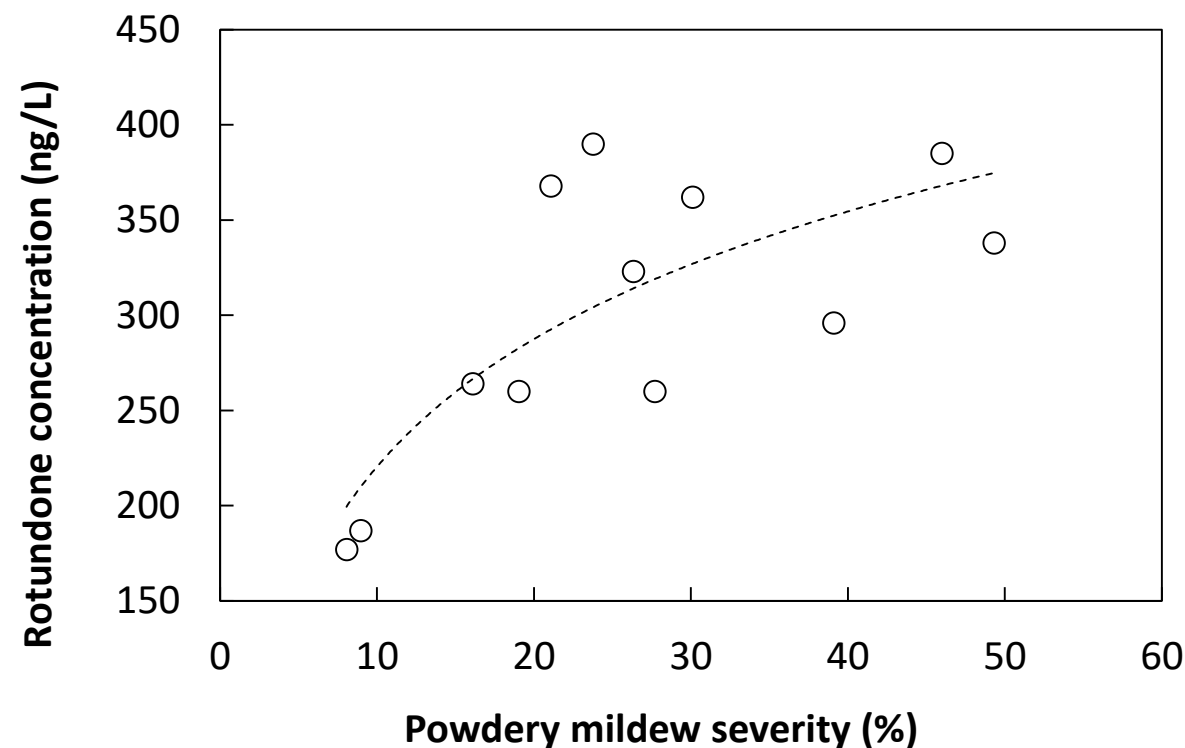


Impact of biotic factors

Powdery mildew (Erysiphe necator)

- Identification of a positive correlation between PM severity on bunches and rotundone concentration
- Rotundone could be produced in response to PM infection
- Systemic effect as wines were produced from healthy berries ?

Geffroy et al. (2015a), JISVV

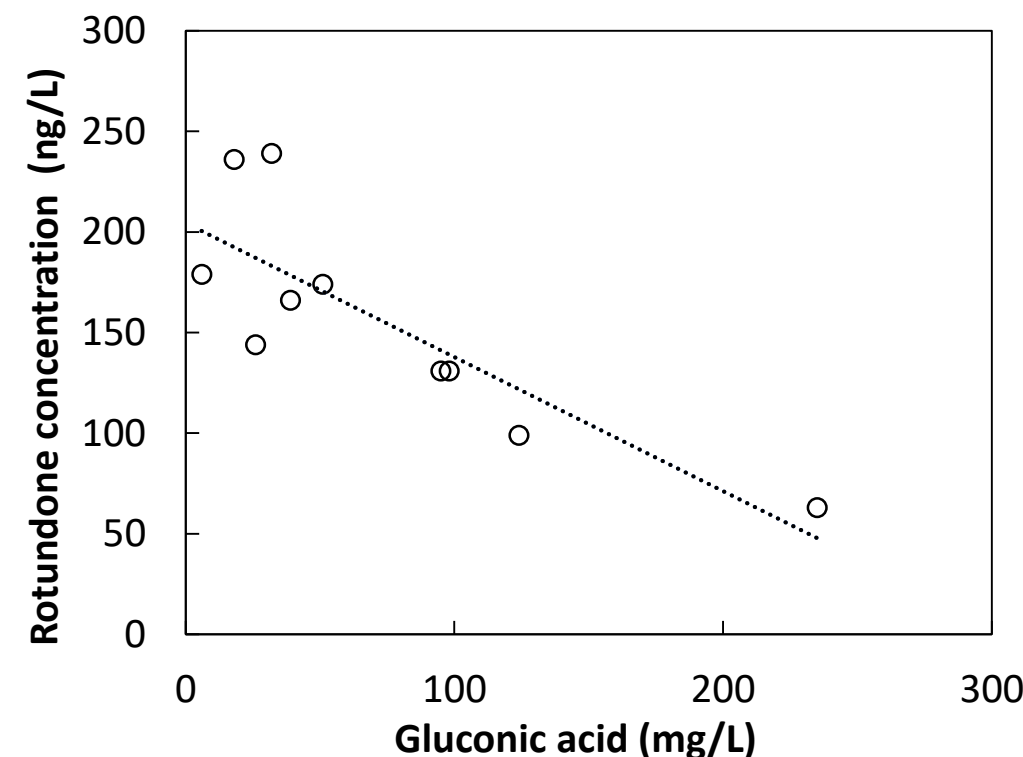


Impact of biotic factors

Bunch rot (Botrytis cinerea)

- Negative correlation between rotundone concentration and levels of gluconic acid
- Due to the fungus or to its laccase ?
- Rather unexpected as laccase has the ability to convert α -guaiene into rotundone (Schilling et al., 2013)
- However, laccase can neutralize the toxic effect of sesquiterpenes through oxydation (Mayer et Staples, 2002)

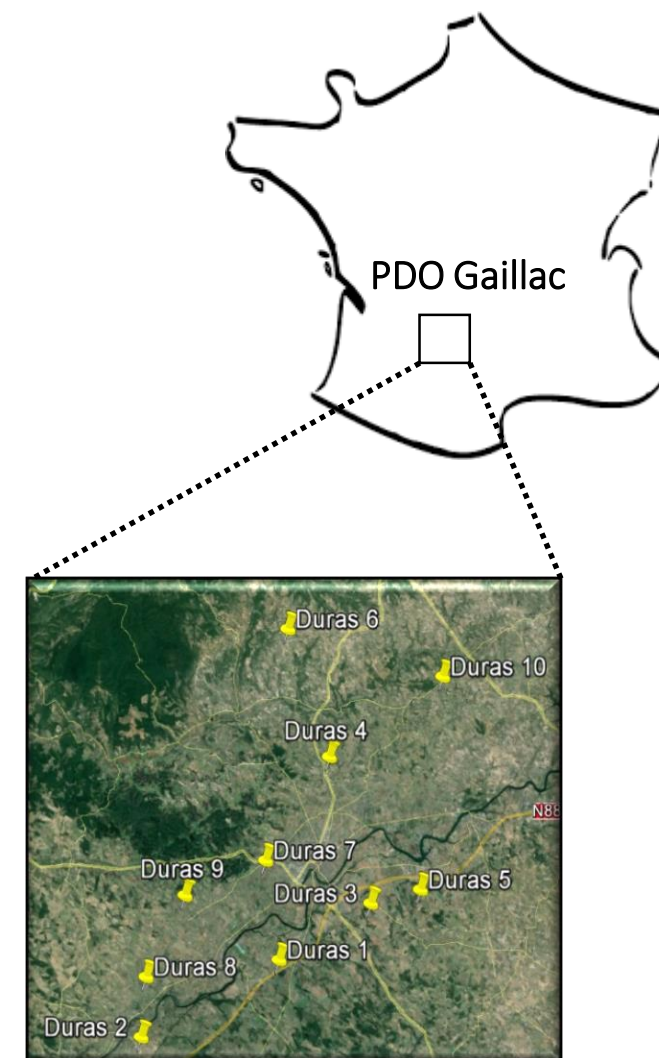
Geffroy et al. (2019a), OENO One



Impact of abiotic factors

Modeling rotundone in Duras wines

- 10 Duras blocks were monitored in 2013 and in 2014
- + 50 variables were collected (fruit, vine, climate) and Ψ_{stem} was modeled
- Rotundone was determined in wines made from microvinification techniques (1L Erlenmeyer flasks)
- PLSR models were built to model rotundone in 2013, 2014 and in both vintages

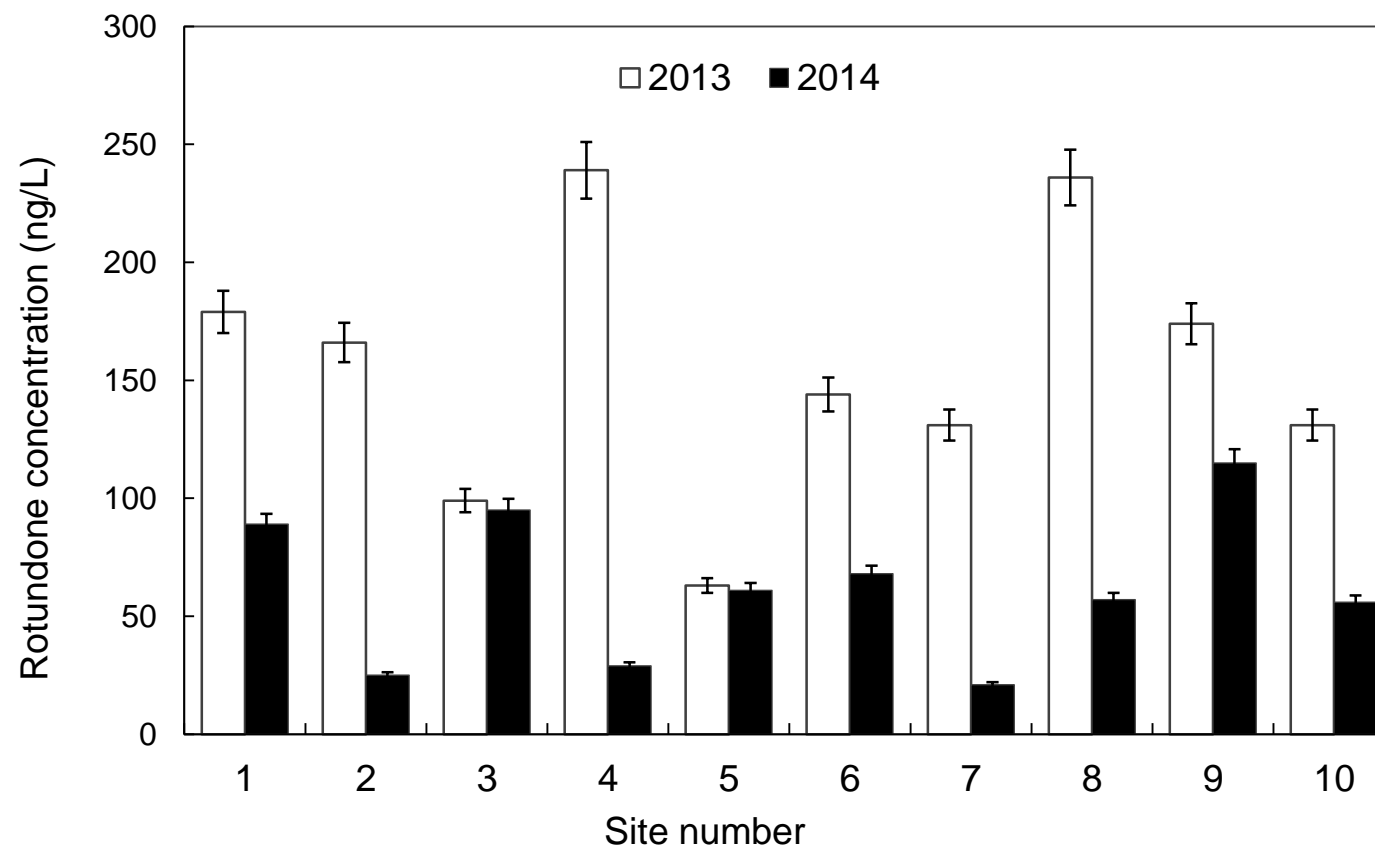


Impact of abiotic factors

Modeling rotundone in Duras wines

Geffroy et al. (2019a), OENO One

- Some blocks with high rotundone levels in 2013 (i.e. n°2 and 4) exhibited low levels in 2014 and conversely (i.e. n°3)
- Fixed variables (topography, plant material...) do not allow to explain the differences between blocks



Impact of abiotic factors

Modeling rotundone in Duras wines

Geffroy et al. (2019a), OENO One

- Positive contribution of cumulative rainfall, daily irradiation and hours of sunshine to the models
- Negative contribution of the Huglin index
- Water supply and light stimulate rotundone production while temperature has a depreciative effect ?

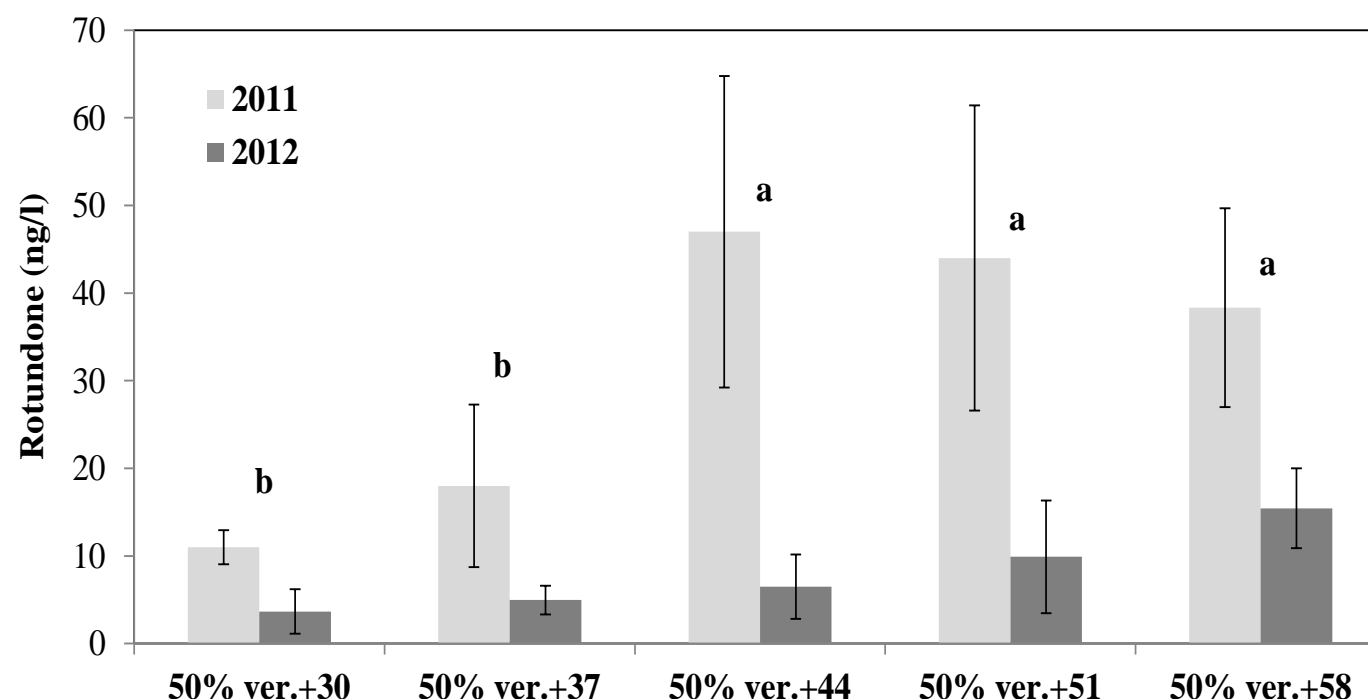
Variable	Model		
	2013	2014	2013–2014
Cumulative rainfall, veraison – harvest (mm)	−0.90 ^a	NI	NI
+ Hours of sunshine	0.72 ^a	NI	0.61 ^a
Gluconic acid concentration (mg/L)	−0.83 ^a	NI	−0.001 ^a
+ Cumulative rainfall, 1 April – 30 September (mm)	NI	0.85 ^a	0.77 ^a
− Huglin index	NI	−0.63 ^a	NI
+ Cumulative rainfall, 1 January – 31 December (mm)	NI	NI	0.66 ^a
+ Mean daily irradiation, veraison – harvest (W/m ²)	NI	NI	0.59 ^a
No. of latent variables	2	2	2
Root mean square error	11.4	14.1	28.5
R ² (calibration)	0.95	0.78	0.81
Root mean square error of cross-validation	14.9	20.8	41.3
R ² (validation)	0.92	0.53	0.59

How to manipulate rotundone concentration in wine ?

Throught harvesting date

- Kinetic of accumulation slightly differ between the two vintages
- In 2011, fast increase and then reach a plateau
- In 2012, steady increase
- To enhance rotundone, dont pick grapes too early !

Geffroy et al. (2014), AJGWR

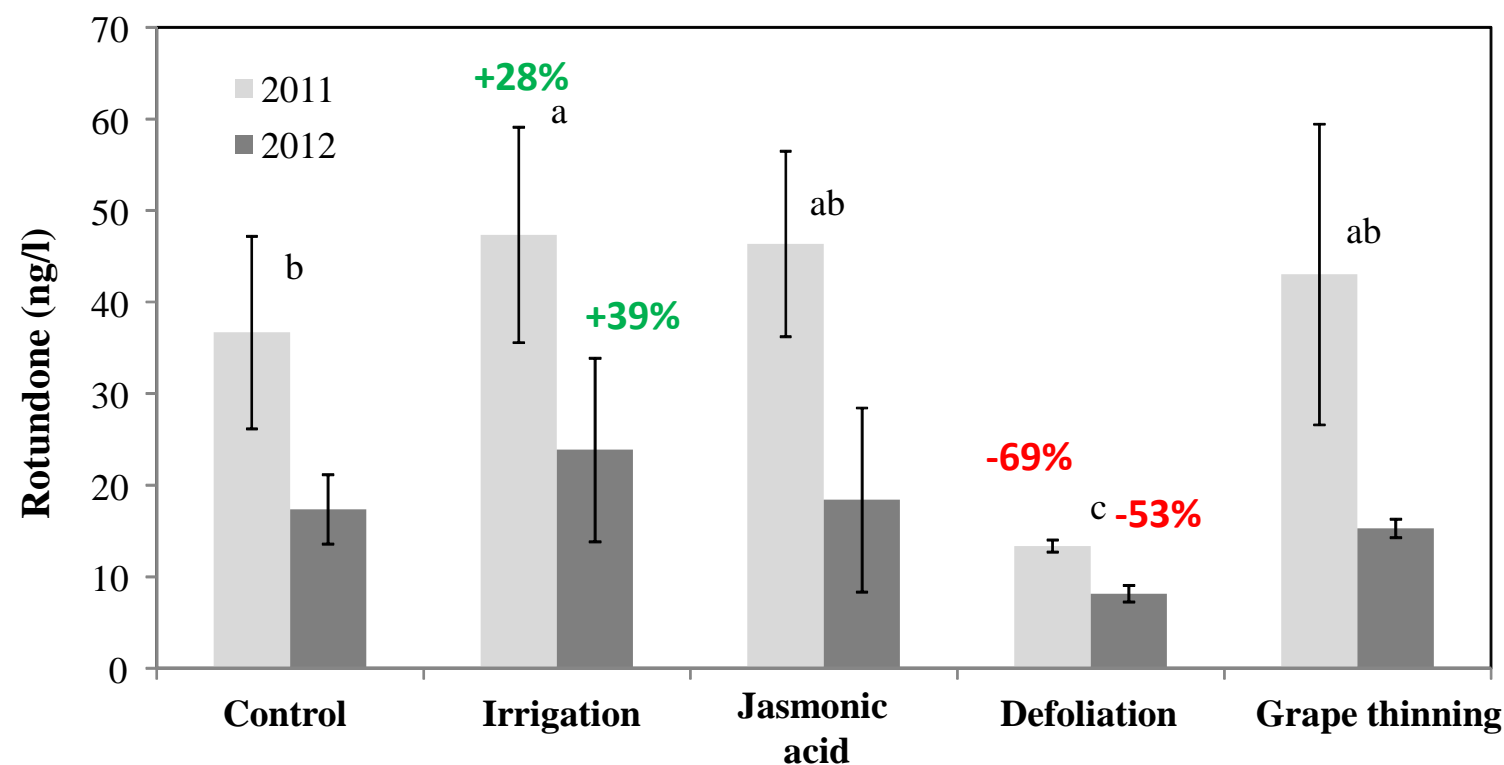


How to manipulate rotundone concentration in wine ?

Through irrigation and defoliation

Geffroy et al. (2014), AJGWR

- No impact of grape thinning (40%) and jasmonic acid spraying on rotundone
- Positive effect of irrigation (4x10mm before veraison)
- Depreciative effect of defoliation (2 faces at mid-veraison)

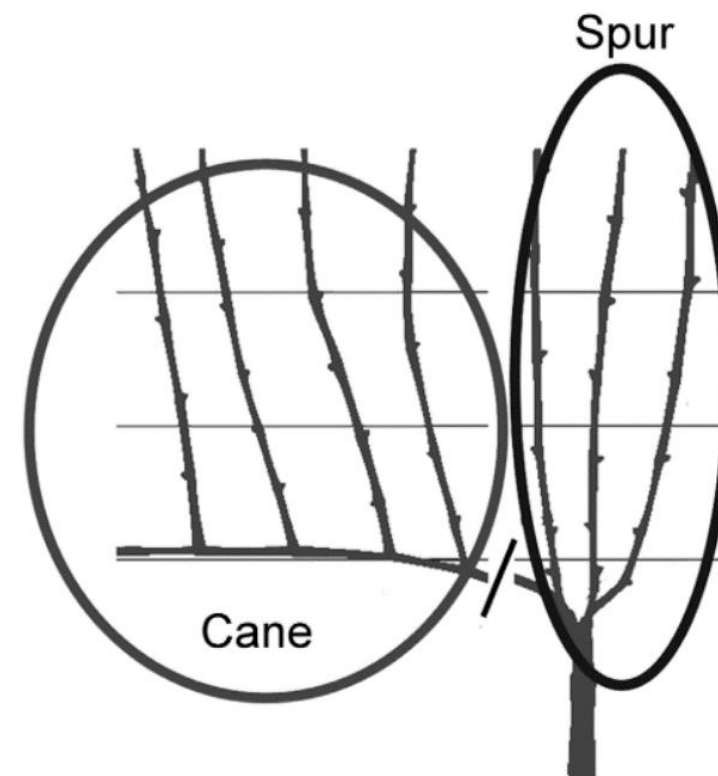


How to manipulate rotundone concentration in wine ?

Enhancing rotundone through irrigation while mitigating the depreciative impact on phenolics

Geffroy et al. (2016), Scientia Horticulturae

- While enhancing rotundone accumulation, irrigation is likely to have a depreciative impact on phenolic compounds (direct and indirect)
- Can a cutting of the fruit bearing cane (PES) 20 days before harvest compensate this loss ?
- We showed in 2013 and in 2014 that PES had a négligeable impact on rotundone accumulation
- In 2014, implementation of a viticultural system combining irrigation (5x14mm) + PES



How to manipulate rotundone concentration in wine ?

Enhancing rotundone through irrigation while mitigating the depreciative impact on phenolics

Parameter	Control	Irrigation + PES	Δ / control
Alcohol (% Vol.)	12.9 b	14.4 b	+1.5 % vol.
Total acidity (g/L tartaric acid)	5.59 b	6.51 a	+0.92 g/L
pH	3.60 a	3.61 a	=
Tartaric acid (g/L)	1.21 a	1.22 a	=
Volatile acidity (g/L acetic acid)	0.47 a	0.55 a	=
Anthocyanins (mg/L)	1061 b	1199 a	+138 mg/L
Total Phenol Index (TPI)	69.6 b	85.0 a	+15.4 points
Yield (kg/vine)	2.12 a	1.78 b	-16%
Rotundone (ng/L)	30 b	44 a	+47%

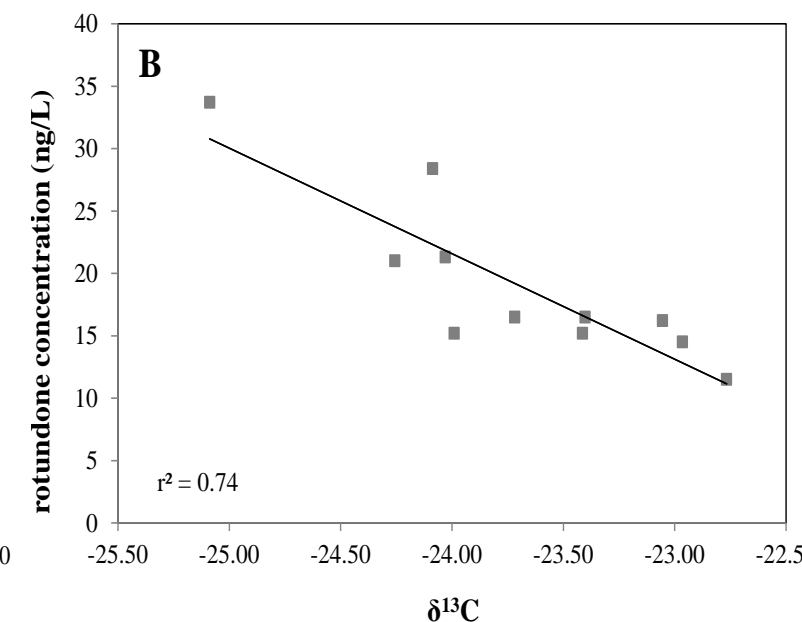
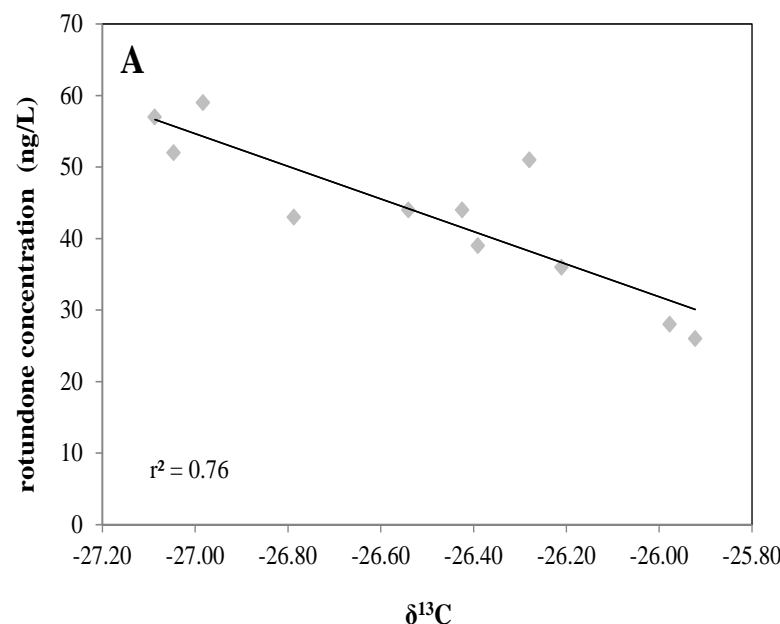
Geffroy et al. (2016), Scientia Horticulturae

How to manipulate rotundone concentration in wine ?

Throught differential harvest

Geffroy et al. (2014), AJGWR

- Correlation between rotundone and $\delta_{13}\text{C}$
- Rotundone is sensitive to weak variation of water constraint
- Possibility to organize differential harvest in order to produce wines with distinct levels of peppery intensity ?

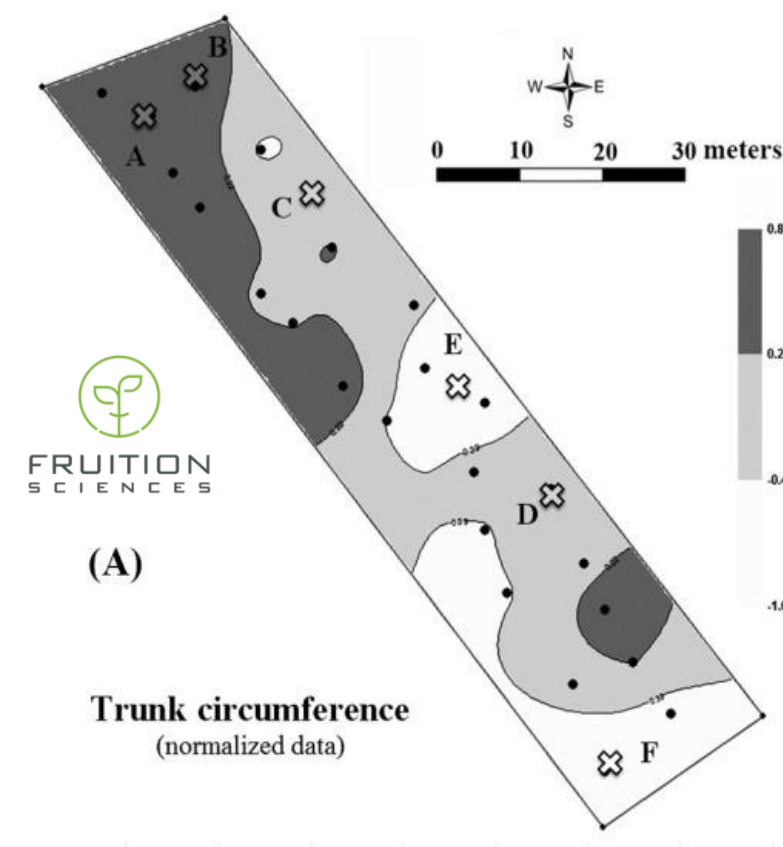


How to manipulate rotundone concentration in wine ?

Throught differential harvest

- We set up a methodology based on stratified sampling to get access to rotundone spatial distribution
- 25 measurements of Trunk Circumference (TC) on a 0.40-ha block
- Positionning of 6 smart points each one composed of 50 vine plants
- For each smart point, we monitored fruit and plant variables

Geffroy et al. (2015b), GiESCO meeting

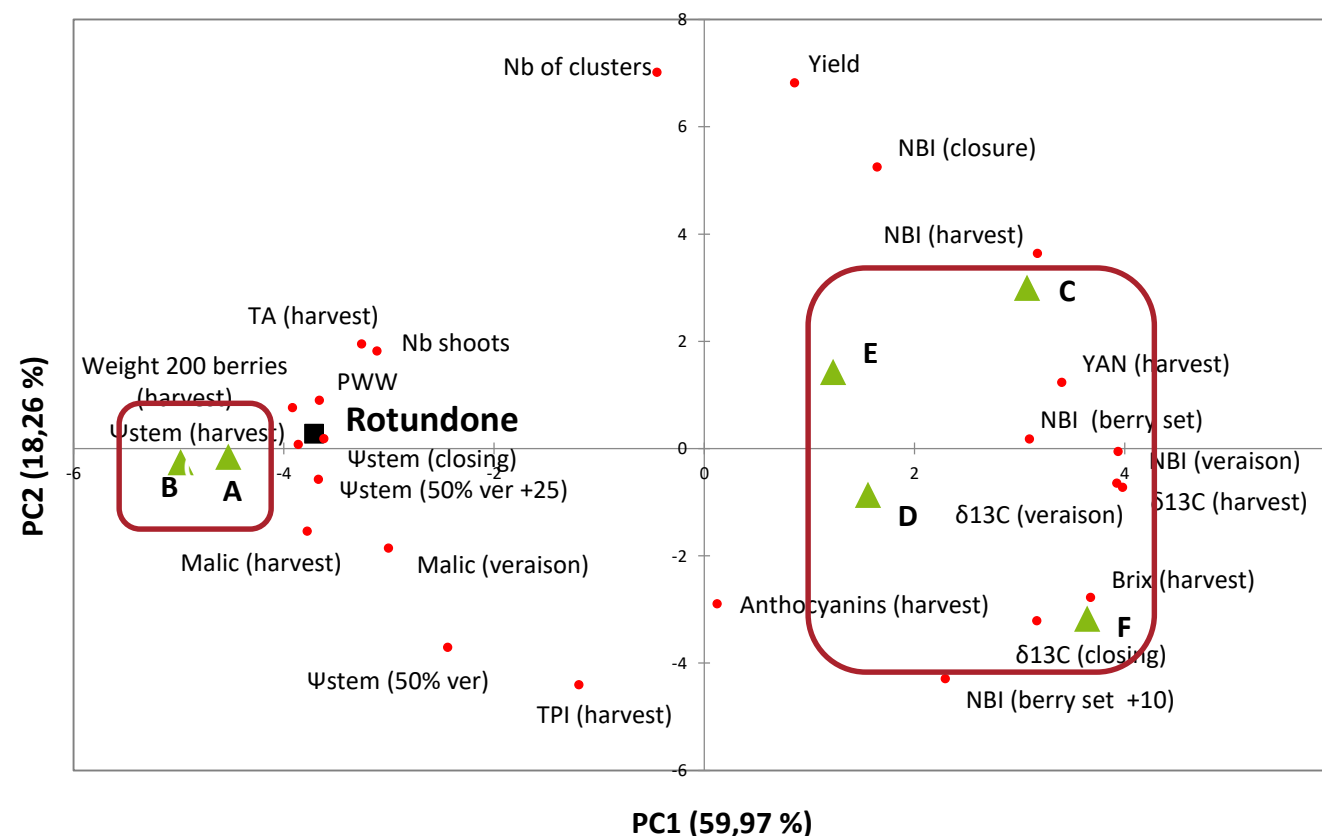


How to manipulate rotundone concentration in wine ?

Throught differential harvest

- Average TC class has more similarity with the weak TC class and these two classes should be merged
- Rotundone is well correlated with $\delta_{13}\text{C}$ at veraison ($r^2 = 0.93$) and harvest ($r^2 = 0.82$),
- 94.5 ng/L (large TC), 60.5 and 61 ng/L (average and weak TC)
- TC can be used to map rotundone

Geffroy et al. (2015b), GiESCO meeting

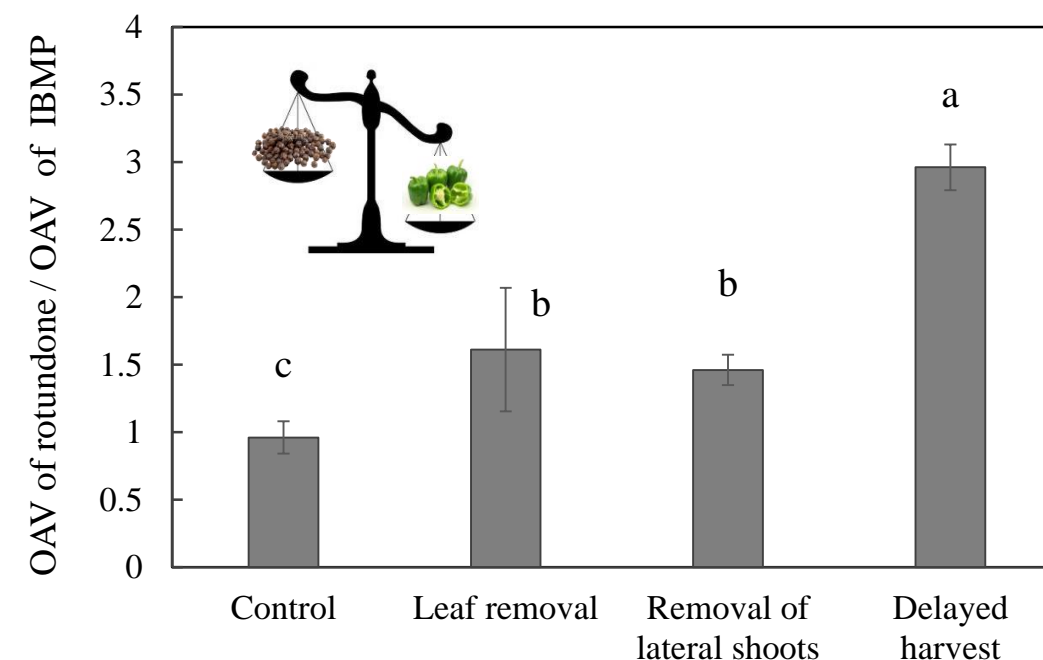


How to manipulate rotundone concentration in wine ?

Enhancing rotundone in Fer wines while decreasing IBMP

Geffroy et al. (2019b), OENO One

- Fer represents ≈ 1500 ha in the SW of France
- Within the PDO Marcillac, Fer wines exhibit intense green and peppery notes imputable to IBMP and rotundone
- Leaf removal, removal of lateral shoots and delayed harvest can be used to modulate the aroma of Fer wines towards an increase in $OAV_{\text{rotundone}} / OAV_{\text{IBMP}}$ ratio

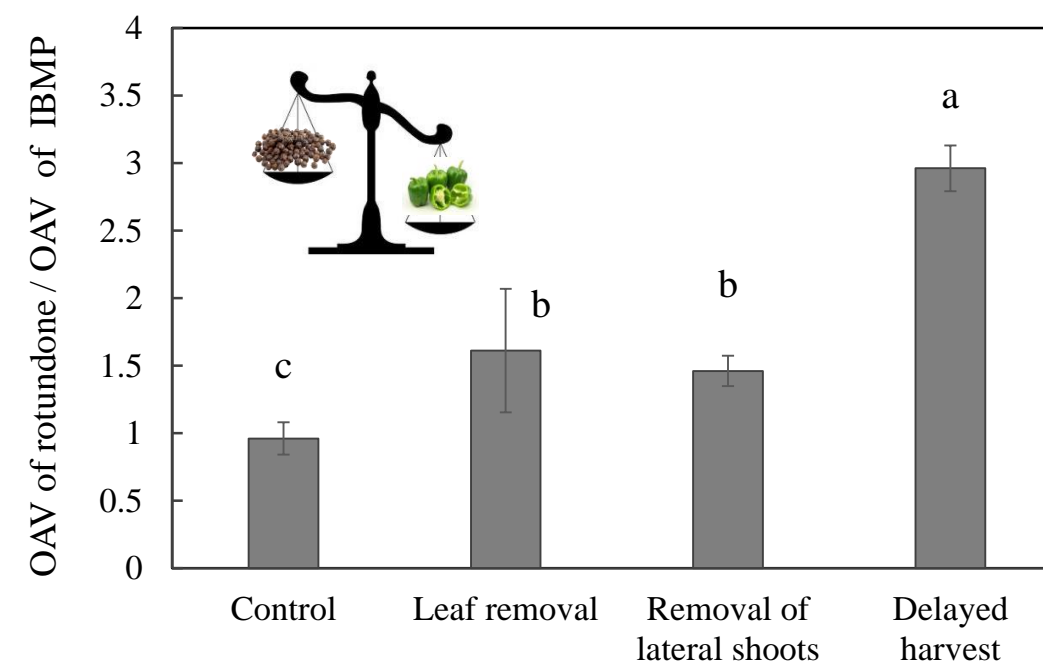


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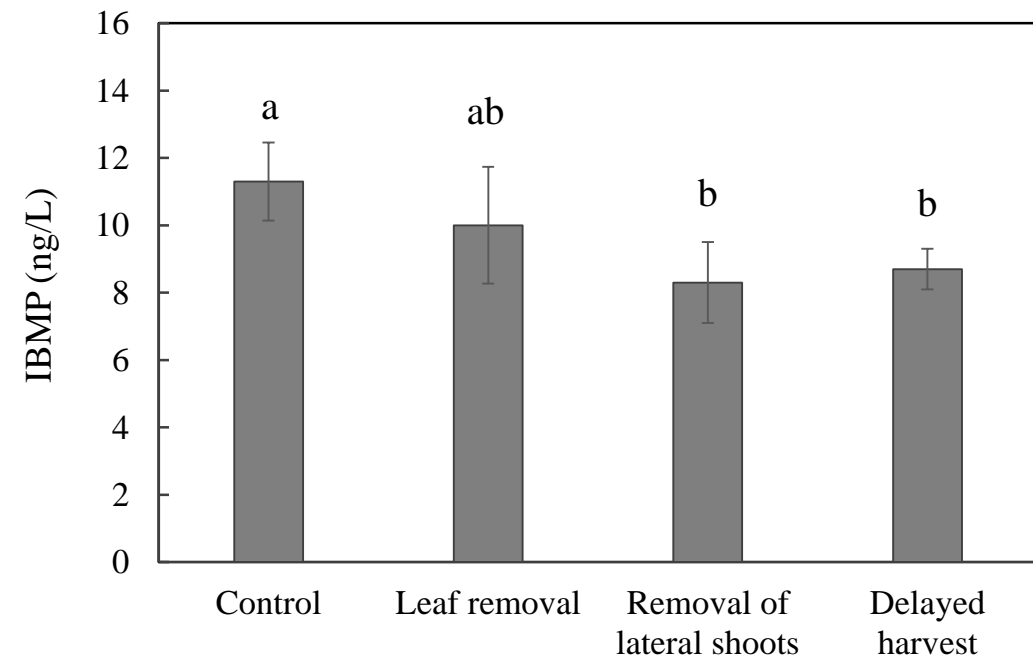
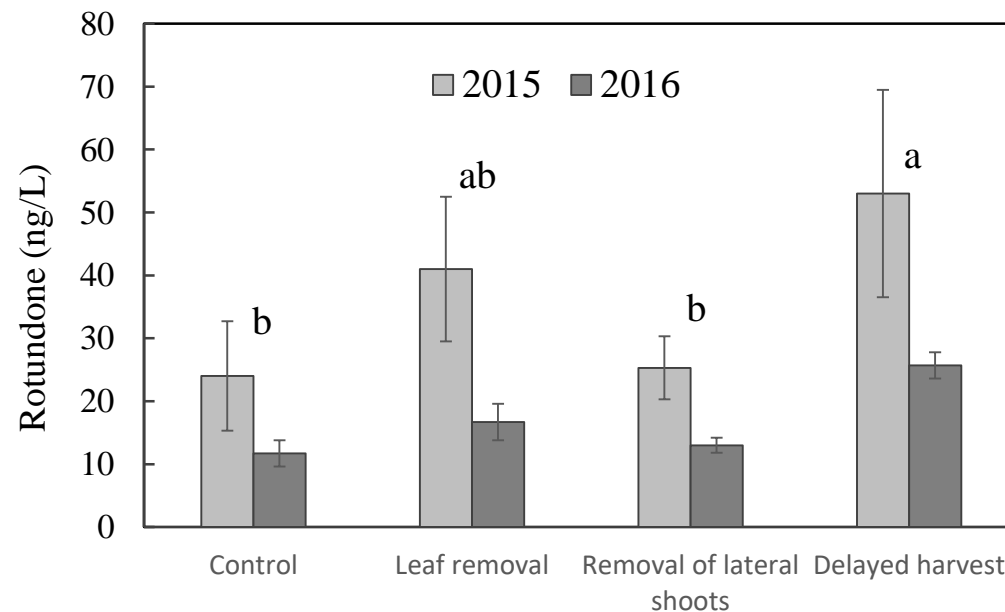


These results might be transposable to Cabernet Franc ?

How to manipulate rotundone concentration in wine ?

Enhancing rotundone in Fer wines while decreasing IBMP

Geffroy et al. (2019b), OENO One



- Significant decrease in IBMP observed for removal of lateral shoots and delayed harvest
- Highest rotundone levels found for delayed harvest while levels for leaf removal were intermediate

Take home message

- Rotundone is a positive aroma compound
- It is a rather ubiquitous molecule
- Certified clones have an impact on rotundone concentration in wine
- The production of rotundone by the plant could be a response to biotic stress (i.e. PM)
- *Botrytis cinerea* has a negative impact on rotundone in wine
- Rotundone accumulation is also affected by abiotic factors (↗ with water supply and light, ↘ with temperature)
- The compound is not impacted by grape thinning and crop load
- Harvesting date, irrigation (+PES), defoliation and selective harvest are leverages to manipulate rotundone levels in wines



Thanks to your attention and to the partners



Technical partners



The Australian Wine
Research Institute



Funding partners



What did I bring for tasting ?



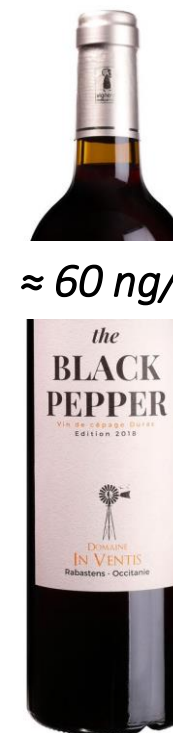
Sample n°1



200 ng/L

*Rotundone
solution*

Sample n°2



≈ 60 ng/L

*Gaillac PDO
100% Duras
No defoliation*

Sample n°3



≈ 20 ng/L

*Gaillac PDO
100% Duras
Intense defoliation
Aged in 2nd hand barrels*