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## MONTPELLIER VINE & WINE SCIENCES INTERNATIONAL SEMINAR



# SEMINAR PROCEEDINGS

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#### **INTRODUCTION**

In cooperation with scientists from USA (University of California Davis, Cornell University), South Africa (Stellenbosch University) and Chile (Universidad de Chile, INIA La Platina), the Key Initiative (KIM) **Montpellier Vine & Wine Sciences**, supported by the University of Montpellier, and its partners INRAE and Institut Agro Montpellier, organized a 3-days scientific seminar on the campus Institut Agro-INRAE of Montpellier in October 2022.

This event, which follows the remote seminar organized in June 2021, brought together involved world-class scientific speakers from local and international institutions with the aim of sharing knowledge to design cooperative research programs tackling some of the current challenges of the wine industry.

One hundred and twenty scientists and staffs from fourteen countries participated in conferences and workshops, organized around four key topics:

- Adaptation and mitigation of climate change issues;
- Reduction of chemical inputs;
- Building wine quality;
- Biodiversity, microbiomes and ecosystems.

Several initiatives were formalized to combine international expertise and construct international research partnerships.

## **CONFERENCES**

Climate	Change:	adaptation	and	mitigation
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#### A global overview on adaptive strategy to climate change of the wine industry

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The communication proposes a synthesis of the impacts of CC on wines and an exploration of the ways and conditions of their possible adaptation. It is based on the outcomes of the LACCAVE research program (2012-2021).

The increase in the average temperature, the changes in rainfall, the growing climate variability are radically changing the conditions of production:

- i) vine development stages are earlier, affecting berry ripening conditions and harvest dates;
- ii) water stress is more pronounced, especially in the Mediterranean area, affecting both the yields and the grape components;
- ii) the quality of wines is changing, with more alcohol, less acidity and new aromatic profiles;
- iv) the increasing climatic variability accentuates the effects of vintages;
- v) the risks linked to bio-aggressors or extreme events threaten the economic viability of wine growers;
- vi) the CC effects on ecosystems, landscapes or fire also affect the vineyards and their image;
- vii) New possibilities of planting vines in regions that have become warmer modify the competition between wine regions.

#### However, there are many solutions to adapt:

- i) new vine varieties (later, more resistant to drought, high temperatures and diseases...);
- ii) the evolution of agronomic practices (soil and canopy management, planting density, irrigation...);
- iii) the modification of oenological practices to correct the impacts of CC on wine quality;
- iv) the reorganization of plantations in space, within the same production area, or through the creation of new vineyards in regions with no wine-growing tradition;
- v) the development of insurance tools and strategies;
- vi) taking into account the expectations of consumers;
- vii) the revision of institutions, in particular the codes of practices.

#### CC therefore places producers in front of two contrasting paths:

- i) a conservative strategy would result in limited effects, given the acceleration of CC. The wines will be too variable, no longer having the qualities expected by the consumer and affecting the economic results of the producers...
- ii) Conversely, a highly innovative adaptation strategy would result in costly innovations, an artificialization of production systems that would risk strongly reducing the links to territorial resources and would not necessarily be accepted by the consumer.

Would there then be no future for wines? A third way is possible under a set of conditions:

- i) the most moderate global warming, close to the COP21 targets which would provide more options for adaptation;
- ii) a redefinition of the principles that underpin Geographical Indication, moving from a conservative definition to a procedural definition promoting a specific quality based on adaptive management of local resources;
- iii) The inclusion of mitigation actions in both the specification and voluntary actions;
- iv) The development of R&D projects between researchers and economic actors with "participatory approach";
- v) A new "engineering of terroirs", combining skills in diagnosis, spatial analysis, climate simulation and adaptive management of local projects.

#### Characterization of genomic diversity in Vitis

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More extensive exploitation of the existing cultivar diversity and the development of improved germplasm will be critical to achieving sustainable viticulture under climate change. Both precision breeding that minimizes genetic drag and genome editing require the identification of specific DNA sequences to target. Unfortunately, in Vitis the knowledge of candidate genes responsible for important traits, including responses to biotic and abiotic stresses, is still minimal. Using a combination of reference genome construction, genetic association analysis, and transcriptomics, we have been dissecting loci associated with genetic resistance to powdery mildew, Pierce's Disease, and other viticulture traits related to abiotic stress tolerance and berry quality. The entry point of these studies has been the development of phased chromosome-scale reference genomes for the accessions that carry the trait of interest. For example, the phased chromosome-scale assembly of the muscadine grape Trayshed was instrumental in identifying the four candidate alleles for the two powdery mildew resistance loci Run1.2 and Run2.2. To achieve the highest possible quality with the current sequencing technologies, we developed HaploSync, an open-source package that scaffolds, refines, and fully phases diploid and chromosome-anchored genomes. HaploSync allows to incorporate genetic map information into the scaffolding process. HaploSync is novel because it leverages the relationship between haplotypes to improve the contiguity and accuracy of phased homologous chromosomes. To date, as part of these projects we have generate over fifty diploid reference genomes, many of which are publicly available through our web portal – www.grapegenomics.com.

#### Genetic diversity of the local criolla germplasm and adaptation strategies to a changing environment in Chile

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Up to recent years, climate was supposed to be rather stable, even when geologically Earth climate oscillations is the norm. What is new for all is the dynamics of current warming, and the effect on the planet this could have. For the ones interested in fruit and wine production, the horizon is complex, but also an invitation to adapt and change to new genetics and management styles. In the case of Chile, during last decades a systematic reduction in the pluviometry has paralleled an increase in average temperatures. In this scenario, the development and evaluation of alternative genotypes are mandatory, for both wine and table grapes. At the same time, new cultural practices or revisiting others are under evaluation, such as the use of plastic covering, the optimization of watering, the expansion of the cultivated area to new regions, etc. In the case of wine, a group of interesting genotypes are criolla varieties, derived mainly from 'País' (syn. 'Listán Prieto') and 'Muscat of Alexandria', the two "founder varieties" brought by Spaniards to America. As criollas were raised in the Region (southern South America), they could be better adapted to local conditions compared to foreign materials. Up to now, we have found 32 new criolla genotypes, additional to the ones already described and not included in the VIVC catalogue. Interestingly, also over 20 European varieties have been identified in old vineyards, which are not listed in the Chilean official registers for wine varieties. In addition to the founder varieties, other possible parents are proposed based on SSR allelic matching, including 'Muscat à petits grains' and 'Mollar Cano', as well as other criolla varieties ('Canela', 'Italiona', etc.). Their prevalence is quite variable, some corresponding to a single vine, meanwhile others are repeatedly present in different valleys, indicating they were propagated on purpose. The discovery and documentation of these new genotypes is the first step of a long-term work that must be followed by evaluations of their enological characteristics, productivity, and management systems, in the search for a diversification of the local wine industry. An example of new alternatives is the possible use of some of these "patrimonial genotypes" as rootstocks, considering their adaptation to grow under desertic conditions and in the presence of high concentration of salts such as Boron.

## Temperature dynamics and impact on cold hardiness, deacclimation, and budbreak phenology

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Grapevine winter physiology is a dynamic and critical time during the lifecycle of grapes grown in all climates, but particularly cool climate regions. During early winter, grapevines use temperature cues to gain freeze resistance in preparation for deep cold in mid-winter. During mid-winter, temperatures may approach or exceed dormant bud cold hardiness, leading to damage and loss of harvest potential or entire vines. Intertwined with changes in cold hardiness are critical temperature sensing components associated with dormancy regulation, deacclimation sensitivity, and ultimately budbreak phenology. I will discuss our current work and results which center on determining cultivar specific responses during mid and late winter physiology and applying that knowledge to develop predictive models. Additionally, I will introduce some concepts around how climate change is shifting winter temperatures and influencing cold hardiness, dormancy, and budbreak phenology in the Northeastern US, and some attempts to model similar shifts in Europe.

#### **Breeding of drought tolerant cultivars**

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The change of the climate is now very well documented and its consequences on plants relatively clear. The predicted climate changes will concern two main components, temperature and precipitation: mean air temperature will increase but at a variable rate depending on the greenhouse gas emission scenarios, while evolution of rains is more difficult to predict. However, the south of France will certainly be more impacted by summer drought than the northern parts of France. These climate changes will have direct effect on phenology of the vine but also on grape and wine composition, yield and plants sustainability. The risk of late spring frosts may also be more important, especially in region of the south rarely concerned until now.

Among the various ways to mitigate these unwanted effects, adopting new innovative material as rootstocks and scions appears as relevant approach. The use of the new rootstocks will help in maintaining the current cultivars in agreement with consumers' expectations or specifications for protected designation of origin which are a master piece of the French appellation concept. The selection of clones more adapted to new conditions, i.e. with modified phenology and/or fruit composition, could offer the same benefits. However, more drastic evolution of the plant material will probably be needed to cope with climate change, which ultimately will require the creation of new scion cultivars.

The first step in the breeding process is the definition of the ideotypes to be targeted. Different declinations of the ideotype can be considered from the plant material by itself to the whole cultivation system. This step needs to be accomplished by a multidisciplinary scientific panel, and preferentially in a participatory way, involving professional partners from upstream to downstream of the production chain including socio-economists.

As only traits with natural variation can be improved by classical genetics, the knowledge of diversity for the different traits of interest, either existing or obtained through crosses, is also very important to take into account in the strategy of selection.

Considering the tolerance to drought, plants can adopt different behaviours: tolerance, escape or avoidance. Behind these different scenarios lie different traits and their interaction. Identifying the main traits involved in the plant response by (eco)physiological studies and analyse their genetic diversity is needed to understand their genetic architecture as well as to develop marker assisted or genomic selection. These tools will help the breeder in the selection of the best progenitors and in early screening of deriving breeding progenies. The presentation will consider these different points which will eventually lead to more drought tolerant varieties.

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## Integrated grape-to-wine studies to evaluate adaptation and resilience of grapevine scion / rootstocks to water limitation

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Climate change is significantly impacting prominent grape-growing regions around the world. In the South African grape growing areas the climate is predicted to become hotter and dryer, thus exposing grapevines to harsher conditions. There is a wealth of information on the water use efficiency of scion/rootstocks for current vineyard production systems and irrigation norms around the world, but in most of this research it was assumed that there will be enough water for the plants when needed. This is no longer true considering the "new reality" of extreme and frequent droughts. As a result, we must (re)assess grapevine responses under these conditions to comprehend the intrinsic and extrinsic factors that may influence adaptation and resilience.

Here we describe the development of a model vineyard in the Stellenbosch region of South Africa to describe and study the impacts of various levels of water limitation on different grapevine cultivars and rootstock combinations to investigate the potential adaptation and/or resilience of the vines to potential drought conditions. In our approach we consider scenarios modulated by different water regimes, multiple plant conditions (rootstock and scion's combinations), comprehensive and integrated phenotyping, epigenetics, and microbiomic analysis, complemented with innovative technologies of stress detection that can be implemented at the vine level.

The benefit of this approach is that the limiting factor (water) is implemented form the establishment phase of this vineyard and that a record of the responses of the vines over the life-time of the vineyard should provide information about the response and resilience of grapevines to limiting water conditions.

**Reduction of inputs** 

#### Microbiological approaches to reduce bentonite requirements

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#### **Objective**

Regardless of its origin, haziness is considered as a fault in wine. One source of haziness, especially in white and rosé wines, is that induced by grape pathogenesis-related (PR) proteins, including chitinases and thaumatin-like proteins. Upon sub-optimal ageing conditions, they may denature and aggregate, eventually forming a cloudiness visible with the naked eye. Bentonite has been used since the 1960's to remove PR proteins. Although very efficient, this treatment has several drawbacks including non-specificity, yield reduction, cost and sustainability. In this context, we endeavoured to explore the yeasts as a biological alternative to bentonite. The following aspects were investigated: secretion of acid protease, mannoproteins and chitins.

#### Methods

Wine yeast species were screened for their ability to secrete acid protease. The protease of the strongest producing yeast was identified at gene level, the enzyme activity and its impact on wine proteins characterised. In parallel, the mannoproteins of various yeast species were purified, characterised and their impact on wine haze investigated. Finally, yeast cell wall chitin levels were assessed and the potential of yeast chitin to reduce wine haze was investigated.

#### **Results**

Metschnikowia pulcherrima was identified as one of the only wine yeasts secreting a protease active at wine pH. The data showed that it is produced, secreted and active against PR-proteins. These results are promising, but the concentrations of protease remain too low to yield a significant reduction in wine haze. The cell wall mannoproteins of four different wine yeast species were extracted, purified, and characterised. Although some of them displayed interesting properties, their ability to reduce wine haze was found limited. Finally, the concentration of yeast cell wall chitin was found to be strain-dependent with some species such as Saccharomyces paradoxus consistently displaying higher concentrations. A correlation between yeast cell wall chitin levels and the level of chitinase in wine was established. The data suggest that grape chitinases bind to lateral chitin, thereby reducing wine haze potential.

#### Conclusion

Taken together, the data showed that yeasts could be used to prevent wine haze or at least reduce bentonite requirements. Further research is currently underway to enhance protease production in *M. pulcherrima* and chitin levels in the cell wall of commercial wine strains of *S. cerevisiae*.

#### How to cultivate grape without pesticide, the VITAE project

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Growing vine without chemical pesticides is a big challenge for this emblematic crop. Eliminating pesticides requires multiple management solutions – biological regulation, plant immunity stimulation, genetic resistance, for example – each of which yields only partial effects. The goal is to move to an agroecological approach based on prophylaxis, monitoring and better resilience of winegrowing systems. These control methods must be integrated into new protection strategies that maximize their combined effects while adapting them to local environmental factors, socio-economical contexts, and market issues.

VITAE is an interdisciplinary project that adopt a pesticide-free paradigm to address economic and technological conditions that will favor the transition in winegrowing systems. Founded by the National Research Agency (3 M€, 2021-2026), research fronts addressed in Vitae are related to four strategies: (1) mobilizing microbiota and diversifying biocontrol strategies, (2) broadening the scope of grape breeding towards durable resistance (3) redesigning cropping systems to enhance prophylaxis and biodiversity (4) elaborating the structural alternatives and economic/regulatory incentives that will support the transition. VITAE also carries out foresight studies with stakeholders to generate scenarios for pesticide-free vine growing at the regional level. These scenarios will help organizations and policy makers to implement pesticide-free strategies with appropriate incentive programs.

## Developing a satellite image-based sampling protocol for leaf blade nutrient monitoring in vineyards

Presented by Justine VANDEN HEUVEL, in collaboration with Manushi TRIVEDI, James MEYERS and Terry BATES

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In the United States, extension specialists recommend collecting a large, random sample of petioles or leaf blades at bloom and/or veraison for each vineyard block. However, many growers do not sample tissues for nutrient content due to the time and effort required to complete the task, or they sample in a biased manner (i.e., collect a non-random sample) resulting in inaccurate recommendations for fertilization. A large, random sample is usually recommended to account for unknown variability, but variability can be assessed using remotely-sensed images.

Spatially explicit sampling protocols were developed using readily available Sentinel-1 Synthetic Aperture Radar (SAR) images as a proxy of soil characteristics and Sentinel-2 Normalized Difference Vegetation Index (NDVI) as a proxy of canopy characteristics. This study tested sampling for leaf blade nutrient concentration using two spatial sampling protocols: grower path (GP) & NDVI+SAR3 against a control of a computer-generated random sample called Random20 (R20). Field-specific sampling protocol trials were conducted at bloom and veraison in vineyards located in New York and Washington State. Two of the eight bloom-sampled blocks were sampled intensively to understand the nutrient variability, while one of the three veraison-sampled blocks was sampled intensively (i.e., each pixel was sampled). R20 explained substantial variation with <1% error in bloom for all macro & micro-nutrients at bloom compared to intensive sampling, whereas GP explained 5.7% and 11.1% error for macro and micronutrients, respectively. GP worked well at bloom for some micro-nutrients like Cu (5.8%) & Zn (6.2%), but not for B or Fe. At veraison, GP and NDVI+SAR3 respectively explained variation with 11.4% and 6.18% error for macro and 19.4% and 17.5% for micronutrients, with sample collection taking only a fraction of the time of R20. This study proves the usefulness of multispectral & moderate spatial resolution images for improving the efficiency of vineyard nutrient sampling.

#### Tools to discover resistance phenotypes and haplotypes in diverse germplasm

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Over the past decade, the VitisGen grape breeding project has developed and provided centralized tools for powdery mildew (PM) resistance breeding. A VitisGen PM phenotyping center has provided laboratory-based, single isolate inoculation and standardized evaluation of mapping family leaves shipped from across the U.S. (and internationally). These protocols have been optimized and automated via the Blackbird microscopy robot, with computer vision analysis that exceeds the accuracy of manual microscopy while providing a 60-fold increase in sample throughput. The VitisGen genotyping center developed a multi-tiered haplotype marker system: core genome rhAmpSeq markers for QTL analysis, whole genome skim-seq of recombinants for fine mapping, and PacBio genome assembly for candidate gene identification. The rhAmpSeq platform has been used for over 50,000 samples by over 20 geneticists across 4 continents since VitisGen made it available in 2020. When a new trait is mapped with rhAmpSeq, we notify breeders and repositories of germplasm that has this trait in their program, providing a rapid opportunity for validation and utilization. Over the next few years, VitisGen will focus on functional validation of candidate resistance genes; vineyard computer vision for disease detection and quantification; low-cost DNA markers for selection; vineyard practices optimized for resistant grapevines; and marketing wines from disease resistant grapes. We welcome collaboration in these and related areas.

#### Reducing inputs, adapting to climate change and supporting the agroecological transition of grapevine systems through biodiversity management

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Reducing pesticide use is a major issue to enhance agriculture sustainability. The wine sector is one of the most intensive agricultural sectors in terms of pesticide use (Urruty et al., 2016). For instance, the average treatment frequency index (TFI) for French vineyards was 12.4 in 2019, whereas the average TFI for wheat (a major annual crop in France) was 4.2 in 2017. Systemic approaches at both farm and field levels are relevant to better understand and support agroecological transition in viticulture (Meynard, 2017). At farm level, the analysis of 244 cropping systems from DEPHY farm network spread across 12 winegrowing regions showed that pesticide use within the network decreased over the 10-year period of the study following three distinct trajectories (Fouillet et al., 2022). Interestingly, the use of biocontrol products, the reduction of sprayed doses of fungicides before flowering and more largely a progressive transition towards organic practices (Merot et al. 2020) were often associated with an important pesticide reduction even if it raised equipment, labor and learning issues (Merot et al., 2019). The analysis of yield evolution showed a significant mean reduction, although it was smaller than the pesticide use reduction (Fouillet et al., 2022). At field level, introducing biodiversity, and particularly service crops in vineyards is often promoted as a way to decrease herbicide use and increase soil related ecosystem services (Garcia et al., 2018). For instance, we found experimental evidence that service crops improved aggregate stability under Mediterranean conditions (Garcia et al., 2019). However, associating service crops with grapevines may also generate disservices and impair grape production due to competition for soil resources with the grapevine. Experimental studies in the Mediterranean region highlighted: i) the need to adapt the design of winegrowing systems associated with service crops in both time (Fernandez-Mena et al., 2020) and space (Delpuech et Metay, 2018) and ii) showed that a functional approach of service species may help to select species or communities service crops that could perform interesting trade-offs between multiple services due to a suited combination of related markers, and provide insights for plant selection in order to breed plant varieties and cultivars with the aim of providing agroecosystem services (Garcia et al., 2020). The prospect of developing multicriteria evaluation tools such as DEXI PM (Gary et al. 2020), integrating agroecological transition levers, is promising for assessing their sustainability and co-designing innovative winegrowing systems with winegrowers.

## **Building quality**

## Building authenticity in the wine sector: a narrative analysis of newcomers' stories

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**Objectives:** This research focuses on new winegrowers. In France, two-thirds of installations are done by newcomers in the wine sector. These new winegrowers must be identified as winegrowers and endorse the social norms of the sector. This research explores the stories they tell to become true winemakers, and namely how they learn to be part of the wine business network that will condition success and access to resources; how they narratively construct their brand biographies to position themselves favorably; and how they create an identity.

**Methods:** For that, we conducted a narrative analysis in two phases. First, we collected the websites of 14 new winegrowers in Saint Chinian, to analyze the way they present their story to the different stakeholders. Then, we conducted 13 interviews with new winegrowers in Val de Loire, to explore their identity, its construction, their storytelling, and the way it is integrated into their strategy. For these two phases, we developed a coding grid based on these themes. Our analysis led to understand how this storytelling was a way to build authenticity.

**Results:** The stories found on the websites are following the actantial model and often include ideas of destiny (love at first sight) and passion for the land and the winemaking. They also show winemakers as heroes of their own stories. Winemakers can highlight different types of authenticity in their stories (Thurnell-Read, 2019): procedural; material; geographical; temporal; oppositional; and biographic. The results underline a deep attachment to the vineyard, which, with the choice to use ancestral methods, is a way to show their identity but also a form of geographic and procedural authenticity. It is supplemented, for some, by a biographical authenticity created by the story of their conversion and their life plans told in the form of adventures.

**Conclusion:** The choice to become a winegrower is not insignificant and passion guides most of the stories collected. Part of this transition implies integrating a new culture and proving the stakeholders and admission of social norms. In that context, storytelling is used to build an identity, integrate the sector, and conquer consumers.

#### Varietal thiols: from vine to wine

Presented by Aurélie ROLAND<sup>1</sup> in collaboration with:

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**Objectives:** Varietal thiols such as 3-sulfanylhexan-1-ol (3SH) and its acetate (3SHA) are powerful aroma compounds in wines with notes of grapefruit and passionfruit. Arising from odorless grapederived precursors, the thiols are released during alcoholic fermentation (AF) by yeast enzymes. This keynote will review our latest research on the fate of thiol precursors from the vine to the wine, focusing on the following topics: water deficit and organic viticulture, copper stress during AF, and reactions between thiols and acetaldehyde.

**Methods:** A study of water deficit was conducted on fungus-resistant varieties at the INRAE experimental center of Pech-Rouge (France) and organic viticulture was studied with Colombard and Gros Manseng varieties in the Côtes de Gascogne (France).

**Results:** Red resistant varieties (3176, Artaban and G14) exhibited higher levels of precursors than white varieties (3159, Floréal and G5) with concentrations reaching 538 μg/kg for 3SH precursors in grapes of 3176. At the physiological ripe stage (phloem unloading), water deficit decreased thiol precursor levels in grapes only in red varieties where precursors were more abundant, meaning that biosynthesis was affected.(1) Organic viticulture modulated the concentration of thiol precursors only in Gros Manseng grapes, with a decrease of 30% at harvest in comparison to grapes from non-organic management. The levels of copper did not correlate with thiol precursor concentrations, suggesting a multi-factorial phenomenon.(2) During AF, thiol precursors undergo interconversion mechanisms leading to the release of 3SH/3SHA. Under copper stress conditions in a synthetic medium, a new intermediate precursor called 3-S-(N-acetyl-cysteinyl)-hexan-1-ol was identified. Although absent in grapes, this compound released 3SH during AF, thus providing new insight into the metabolism of precursors by yeast.(3) Notably, 3SH is a very reactive compound that can combine with acetaldehyde during AF to form the corresponding odoriferous cis-2-methyl-4-propyl-1,3-oxathiane (cis-2-MPO), which was identified in wine for the first time. cis-2-MPO was produced at the beginning of AF and then constantly decreased during the aging process, depending on storage conditions.(4)

**Conclusion:** Having an integrated vision of the production and reaction of varietal thiols, beginning with precursors in the vineyard and continuing through to thiols in the bottle, is necessary to manage the organoleptic profile of wines.

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#### Enzyme treatment enhances astringency through decreasing polysaccharide and increasing polyphenolic content in Cabernet Sauvignon wines

Presented by John P. MOORE<sup>1</sup>, in collaboration with Brock KUHLMAN<sup>1</sup>, Jeanett HANSEN<sup>2</sup>, Bodil JØRGENSEN<sup>2</sup> and Wessel DU TOIT<sup>1</sup> moorejp@sun.ac.za

**Objective:** Assess the impact of pectolytic enzyme treatment on bitterness and astringency perception in relation to the polysaccharide-polyphenol content of Cabernet Sauvignon wines produced from grapes of different degrees of ripeness

**Methods:** Glycan microarray technology with cell wall probes was used to evaluate polysaccharide changes and pholoroglucinolysis-spectrophotometry assessed polyphenolic changes linked to enzyme action. Sensory testing for bitterness and astringency descriptors in the enzyme and non-enzyme wines was performed

**Results:** Enzyme treatment enhanced astringency irrespective of ripeness level but did not affect bitterness perception. Enzyme crafted wines had higher levels of polyphenols which were more galloylated and polymerised. In contrast non-enzyme wines had higher levels of arabinogalactan proteins present. The enzyme produced wines were characterised as grippy, grainy, chalky, hard and dry. In contrast, the non-enzyme wines were described as soft, fine and velvety.

**Conclusion:** Enzyme treatment produced wines of lower polysaccharide and higher polyphenolic composition which were harder, more astringent compared to non-enzyme treatment suggesting a role for arabinogalactan proteins in modulating astringency in red wines.

#### **Reference:**

Brock Kuhlman, Jeanett Hansen, Bodil Jørgensen, Wessel J. du Toit, John P. Moore. 2022. The effect of enzyme treatment on polyphenol and cell wall polysaccharide extraction from the grape berry and subsequent sensory attributes in Cabernet Sauvignon wines. Food Chemistry 385, 132645

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#### Interplay of water deficit and grape berry chemical composition

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Water deficit leads to reductions in the capacity for growth and yield, but when imposed under safe thresholds, a trade off favouring grape berry composition has been observed. Such beneficial results derive from the interaction of synergistic signals, mainly the plant hormone ABA and sugar concentration at the onset of veraison. The phenylpropanoid pathway responds to early signals lasting along the ripening stage, upregulating gene expression and metabolites concentration. At the transcriptional level, ABA around veraison transiently fastens ripening, and induces an immediate increase in the grape transcript abundance of VvDFR, VvANS, VvUFGT and VvMybA1. A long-lasting effect, for nearly 40 days, is also observed in VvPAL, while late in ripening, VvLAR2 and VvMyb4A are increased. Metabolites associated to the activity of the enzymes coded by those genes such as anthocyanins and flavanols are also increased in concentration. Beyond some thresholds, the potential ABA and sugar synergistic modulation of the phenylpropanoid pathway upregulation, resulting from water deficit, could be compromised because of source carbon limitation, however. Such effect, even for a specific grape vine variety, is difficult to predict due to the high variability in the stomatal sensitivity to water deficit. Also, indirect effects of water deficit could affect the grape berry metabolism, for instance through a vigour mediated improvement on the microclimate of the fruiting zone. Higher cluster illumination levels result in lower pyrazine concentrations, higher condensed tannin concentration with higher levels of polymerization in grape berries with a higher skin to pulp ratio.

## Metabolic QTL analysis for characterisation of the genetic potential of grapevine flavour formation

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In grapevine (Vitis vinifera L.) volatile terpenoid aroma compounds (such as monoterpenes and sesquiterpenes) and phenylpropanoids (such as flavonoids and hydroxycinnamic acids), play a vital role in plant development and plant-environment interactions such as pollinator attraction and pathogen/herbivore defence. However, in the context of viticulture their significant contributions to both the flavour and aroma profiles of table grapes and wine are particularly significant. In this work, metabolic QTL analysis was performed in a grapevine cross population, using chemical profiling data over two seasons. Data was collected via GC-MS analysis, and QTLs were identified for individual as well as classes of compounds. Several significant QTLs were found for volatile terpenoids, and the genomic regions were then investigated after mapping of the QTLs to the grapevine physical map. A QTL region associated with total monoterpene accumulation was identified on linkage group 5, and contains the VvDXS gene, which is understood to regulate flux though the methylerythritol phosphate (MEP) pathway, leading to terpenoid precursor formation. While work here confirms the importance of a single SNP in VvDXS for monoterpene accumulation further QTL regions were identified that associated with the accumulation of specific terpenes. Genomic screening identified several putative terpene synthase genes (TPSs) in each region. These were then further investigated via a range of molecular techniques and in silico approaches, including gene expression analysis, gene sequencing, and molecular phylogenetic analysis. The results indicated a large amount of copy number variation for these genes in the population, which was further explored through the analysis of recently available diploid genome sequences of a number of grapevine cultivars. It was found that gene duplications for TPSs in Vitis have occurred recently, and in some cases in the individuals of cross population under investigation. Overall, the role extensive gene duplication events have played in the diversity of TPS gene function in grapevine is described, and the importance of cultivar specific genome sequencing highlighted.

#### Phenolic compounds, from grape to wine

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Wine color is due to the presence of phenolic compounds extracted from grape or formed from them during wine-making and ageing. Major grape phenolics are anthocyanins, the red grape pigments, localised in berry skins, and colorless compounds, namely hydroxycinnamic acids, present in skins and pulps, and flavanols, including monomers but also oligomers and polymers refered to as condensed tannins, which are very abundant in skins and seeds.

The wine-making process in addition to grape characteristics determines the proportions of anthocyanins, flavanols, and hydroxycinnamic acids in musts and wines. Thus, white musts obtained with no skin contact contain mostly hydroxycinnamic acids and their enzymatic oxidation products while red musts show large concentrations of anthocyanins and flavanols. Rosé musts are intermediate but show large composition differences, responsible for their large color variations.

Wine pigments include anthocyanin reaction products with hydroxycinnamic acids, flavanols, and yeast metabolites such as acetaldehyde and pyruvic acid but also brown pigments resulting from oxidation. Their proportions in wine are largely determined by those of their precursors; this will be illustrated with recent findings on the drivers of rosé wine color. In spite of the important progress achieved on identification of phenolic reaction products and their formation mechanisms over the last decades, known compounds acount only for the emerged part of the "wine polyphenol iceberg". The missing part results from random reaction cascades, involving grape phenolics and intermediate products formed from them, leading to high complexity, as demonstrated in simple model systems. Characterisation of such highly complex media is needed to establish links between grape characteristics, wine-making process and final wine quality and address current challenges in enology. This requires development of multiple analytical approaches providing complementary information.

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## Conserving biodiversity and ecosystem services in vineyards through agroecology

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New agricultural systems are needed to balance the dual sustainability goals of food production and biodiversity conservation. Scientists around the globe have generated growing evidence that agroecological management practices that enhance reliance on ecological processes to maintain or increase production while reducing or replacing external inputs are a promising route toward sustainable food production. Greater reliance on ecological processes in agroecosystems necessitates integrated frameworks and assessments of ecosystem services, the benefits provided to and by biodiversity in agroecosystems such as pest management, food production, and biodiversity conservation, as well as potential ecosystem disservices, the detrimental effects of biodiversity in agroecosystems.

Vineyards in Mediterranean climate biomes ideally pursue a dual goal of wine production and wildlife conservation. A series of studies evaluates how remnants of native vegetation within vineyards impact bird communities, species composition, and ecosystem services provision. We assessed the relevance of these remnants in Chilean vineyards along a gradient of increasing natural vegetation in the surrounding landscape (N=120). We found that overall total detections of birds and species richness were significantly greater in continuous mixed Mediterranean forest. 84.9% of these species were also detected in forest remnants within vineyards. Endemic birds, insectivores, granivores, and omnivores were all more abundant in vineyards with remnant native vegetation than in vineyards without remnant native vegetation. Our results show the value of maintaining and restoring natural vegetation remnants in vineyards as a tool for bird conservation and potential ecosystem service provision. An ongoing Before-After-Control-Impact experiment seeks to assess the impact of bird perches on bird communities in vineyards to quantify bird community changes and potential disservices (grape damage). Preliminary results will be presented.

We hope that results will provide scientific guidance on the impacts of these agroecological practices in vineyards landscapes and provide data to support stakeholders in meeting their goals of sustainable food production and biodiversity conservation. Findings will guide future research, agricultural management, and sustainability monitoring in vineyards and other perennial cropping systems in Mediterranean climates.

## Microbial interactions between grapevine pathogens and the leaf microbiome

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#### **Objective**

Grapevine leaves are colonized by a multitude of microorganisms that affect, positively or negatively, plant health and growth. Pathogen species, including *Erysiphe necator* (powdery mildew) and *Plasmopara viticola* (downy mildew), are among the key players. When these pathogens colonise the grapevine leaf, they encounter the resident microbiome, which can act as a barrier to their growth through direct ecological interactions, such as exploitative competition, interference competition (antibiosis), and hyperparasitism. A current challenge is to identify members of the resident microbiome that naturally regulate powdery and downy mildews, and identify the biodiversity management practices that favour these natural antagonists. Such results are urgently needed to move towards pesticide-free viticulture, as mildews are currently controlled by chemical pesticides whose use should be drastically reduced to preserve environmental and human health.

#### Methods

To decipher ecological interactions between leaf pathogens and the resident microbiome, we inferred microbial networks from metabarcoding datasets describing microbial communities associated with symptomatic and asymptomatic leaf samples. Two methods were used to generate hypotheses of ecological interactions: a joint species distribution model adapted to sequence counts modeling and a machine-learning method that simultaneously detects interactions and classifies them into ecological types. The underlying assumption of both methods is that since past or ongoing interactions between microbial taxa may impact their frequencies, variations in microbial community composition may provide information about the structure of the ecological interaction networks.

#### **Results**

Preliminary interaction networks were computed and revealed subnetworks of interest, centered on pathogen species (*E. necator* or *P. viticola*) and their direct antagonists or facilitators. Some of the antagonistic interactions were known, others are new and could be options to control disease development.

#### Conclusion

Inferring microbial networks from metabarcoding data is an option to guide future research in microbial biocontrol of grapevine diseases. It can be used to highlight small microbial consortia of interest, whose dynamics and interactions should then be studied experimentally.

#### Microbial flux from grape to wine

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The quality of wines results from the combination of the quality of the grapes and from its optimal transformation during fermentation and aging. The fermentation step is performed by a microbial community whose main actor is the yeast *Saccharomyces cerevisiae*. However, the origin of this yeast has been intensely debated along the last 50 years: vineyard or cellar? The initial concept of the correspondence between the yeast populations of these two compartments was contradicted by the first tracking experiments of Saccharomyces cerevisiae in the cellars (Rosini et al., 1982). Since, the development of more and more precise molecular tools for S. cerevisiae has progressively changed the vision that we have of S. cerevisiae ecology. This model species enables us to draw a new scheme in which vineyards and cellars are connected with bidirectional flux and in which insects also participate in the vection.

The actual trends of many winemakers towards minimal practices including as little additive as possible has provoked the abandon of selected yeast starters and a progressive switch-back to "spontaneous fermentations". Such practice leads to more complex microbial dynamics during alcoholic fermentation and more variable microbiota in vats. This raise at the same time the question of the effective contribution of the grape's microbiota to the fermentation, and how it impacts the metabolic and aromatic profiles of wine.

A better comprehension of the global microbial flux between the vineyards and cellars, focusing on the major players of the wine ecosystem, is thus a major priority for the wine industry.

## Effects of regenerative management on vineyard soil biodiversity and climate change mitigation

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**Objective:** Regenerative management takes advantage of the potential synergistic effects between well know soil conservation practices to increase soil organic matter and soil health. Increases in soil organic matter, could also lead to C sequestration and therefore climate change mitigation although changes in microbial activity could trigger increases in decomposition rates and the production of greenhouse gasses. As winegrape producers throughout the world are embracing the principles of regenerative farming, a more thorough investigation of this approach and the efficacy to build soil health and mitigate climate change is needed. Here we evaluated the effects of no-till and cover crop grazing by sheep on the structure and function of the soil microbiome as well as changes in soil C and greenhouse gas emissions in a regenerative vineyard.

**Methods:** We conducted a three-year field study in a Vitis vinifera L. cv. Syrah biodynamic vineyard to evaluate grazing and tillage in a full factorial design. Throughout the experiment we measured the fluxes of CO2 and N2O using static chambers and we determined changes in grape quality and yield. The third year, we collected soil samples to analyze changes in the diversity (16SrRNA and ITS) and function (potential enzyme activity) of the soil microbiome, as well as changes in soil C and N fractions at two different depths (0-15, 15-30 cm) and locations (tractor and vine row).

**Results:** Grazing and tillage had contrasting effects on the soil microbiome. While tillage had a stronger effect on microbial alpha and beta diversity, grazing seemed to stimulate microbial function. None of the treatments resulted in increases in total soil C. Sheep grazing resulted in sporadic and localized peaks in daily N2O, and CO2 emissions specially when combined with tillage. Nevertheless, emissions were not significantly larger than non-grazed soils when extrapolated to the cumulative emissions of the whole study. Tillage increased cumulative soil CO2 emissions during the wet seasons as compared to no-till soils. Sheep grazing and tillage did not have a significant effect on the yield and quality of the grapes during the two years of the study.

**Conclusion:** That the combination of conservation practices has short term beneficial effects for soil biodiversity and climate change mitigation by reducing vineyard greenhouse gas emissions. Nonetheless, more time may be needed for the practices to sequester C in the soil.

#### Inter-row cover crop applications and their influence in the soil microbiome

Presented by M (Evodia) SETATI, in collaboration with Ricardo V. SMART, Erna H. BLANCQUAERT and Florian F. BAUER

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Cover crops are increasingly integrated into vineyard floor management practices due to their efficiency in providing ecosystems services such as weed suppression, reducing soil erosion and enhancing soil water-holding capacity and microbial activity. However, the efficiency of cover crops may be influenced by edaphic factors as well as attributes associated with soil development. Consequently, one cover crop type many not yield similar results on different soil types. We investigated the impact of Vicia faba (faba bean) and x Triticosecale (triticale) compared to natural vegetation on the soil microbiome and physicochemical properties over two consecutive seasons of cover crop application (2018/2019 and 2019/2020) in three vineyards characterized by Duplex, Oakleaf, and Tukulu soil types. Automated Ribosomal Intergenic Spacer Analysis (ARISA) as well as rRNA gene amplicon sequencing were used to analyse microbial community dynamics and identify microbial taxa. Overall, cover crop performance varied between the two seasons depending on soil type, with 100% soil coverage in duplex in both seasons and ≈50% coverage in the second season Oakleaf and Tukulu for faba bean and Triticale. Canonical Correspondence Analysis revealed a clear separation between microbial communities of the three soil types mainly driven by soil EC, pH, Mg, and K. Each cover crop differentially influenced the abundance of a few minor taxa. The interaction between soil type and cover crop, explained 10% and 8% of the fungal and bacterial community variance, while soil type alone explained 32 and 23% of the variance. Overall, our data suggest that cover crops may introduce unique taxa depending on soil type without major perturbations on the core microbiome.

#### **Ecological patterns and molecular signatures in fermentation ecosystems**

Presented by Florian F BAUER<sup>1</sup>, in collaboration with Cleo CONACHER<sup>1</sup>, Mathabatha E. SETATI<sup>1</sup>, Natasha LUYT<sup>1</sup>, Eleonore POURCELOT<sup>2</sup>, Thibault NIDELET<sup>2</sup> and Virginie GALEOTE<sup>2</sup>

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Wine fermentation represents a global, anthropic, but evolutionarily relevant ecological niche for yeast and bacterial species. Compared to other evolutionarily relevant microbial ecosystems, wine fermentation is characterised by a core ecosystem - species found consistently, and actively participating, in fermentations globally – that is relatively low in species diversity, while the "sequential batch" character of the system allows relatively straightforward monitoring of species succession. The system therefore represents an excellent model to investigate the molecular mechanisms governing interactions between microbial species and the evolutionary importance of biotic selection pressure. Here we report on several data sets investigating interactions between two species or within larger consortia, as well as an evaluation of the relative importance of the taxonomic level of "species" vs "strain" in such interactions. The data suggest both species-specific and ecosystem-wide generic interactions. Transcriptomic data suggest that higher order interactions lead to different transcription profiles than binary interactions with the same species. These data provide fundamental insights into the molecular nature of yeast species interactions and of the evolutionary adaptation of species to the wine ecosystem. High-throughput monitoring of consortia dynamics will provide baseline data for predictive modelling of this system. Such models may support the application of natural fermentation-based wine making.

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## **POSTERS**

## New plant biopolymers for the colloidal stability of the coloring matter of red wines

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**Objective:** In a global perspective of limitation and reduction of additives in the field of oenology, one of the objectives of the wine industry is notably (i) to reduce the use of additives in wines, by their number and/or their quantity, and (ii) to favor the use of natural additives while preserving the organoleptic and sensory qualities of wines. The aim of this work is to identify some plant biopolymers, other than Acacia *senegal* gum, allowing the colloidal stability of the coloring matter of red wines, and satisfying the technical (solubility and non-clogging) and sensory requirements of wine making. The selected plant biopolymers should also significantly improve the coloring matter colloidal stability.

**Methods:** Nine natural different plant biopolymers were used in this study. Their biochemical composition (protein and carbohydrate contents, amino acids and sugar compositions) and structural properties (Molar mass, intrinsic viscosity, hydrodynamic and gyration radii) were characterized. The colloidal stability properties of all biopolymers were evaluated in comparison to Acacia senegal gum on three different matrices: a mineral-hydro-alcoholic solution corresponding to the test recommended by the oenological codex (COEI-1-GOMARA:2000), a hydro-alcoholic-grape marc solution and unstable red wines.

Evaluation of the colloidal stability of the coloring matter was conducted by turbidity measurements on the red wine after 48 h at +4  $^{\circ}$ C and by kinetic measurements of transmittance of the hydro-alcoholic-grape marc solution during 48 h at 10  $^{\circ}$ C using a Turbiscan tower.

**Results:** The use of nine natural different plant biopolymers allowed to identify their intrinsic biochemical and structural properties essential for the colloidal stability of the coloring mater. Among these nine plant biopolymers, a new promising one presents interesting colloidal stabilization properties towards the coloring matter. This plant biopolymer possesses superior colloidal stability properties than Acacia *senegal* gum and good clogging index (under 66 using a PALL membrane, pore size  $0.65 \mu m$ ). Its quantity in red wines can be reduced between 5 and 10 while maintaining the colloidal stability of the coloring matter and allowing the filtration of red wines.

**Conclusion:** The identification of a new promising natural exudate is therefore of interest for its use in oenology.

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## Selected Ion Flow Tube Mass Spectrometry: a promising technology for the high throughput phenotyping of grape berry volatilome

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#### **Objectives**

Wine grapes breeding has been concentrating a lot of efforts within the grape research community over the last decade. The quick phenotyping of genotype quality traits including aroma composition remains challenging. Selected Ion Flow Tube Mass Spectrometry (SIFT-MS), a technology first available in 2008, could be particularly valuable for this usage. The aims of this study were i) to use SIFT-MS, to analyze the whole volatilome from different grape varieties, ii) to assess the ability of this technology to discriminate varieties according to their grape aroma composition, and iii) to study the stability of SIFT-MS signal over maturation to define a sampling strategy.

#### Methods

23 grape varieties were sampled at 40 days after mid-veraison in a germplasm collection, and 50 g were subsampled and crushed to be directly analyzed in the SIFT-MS using  $H_30^+$ ,  $NO^+$  and  $O_2^+$ . Straightforward chemometrics techniques were used to analyze spectral data and for each homogenous group of cultivars made using hierarchical ascendant classification (HAC), one grape variety was chosen to perform analyzes by gas chromatography–mass spectrometry (GC-MS). At the same time on a neighboring vineyard, samples of Syrah were collected at 7 times between mid-veraison and 50 days after mid-veraison.

#### **Results and conclusion**

Based on their SIFT-MS volatilome scan, different grape variety groups could be identified.  $O_2^+$  reagent ions should be given preference due to their propensity to ionize the majority of compounds, effectively fragment them to produce ions with distinct m/z ratios, and improve the distinction of compounds with comparable masses. As demonstrated by headspace GC-MS investigations, the technique permitted the differentiation of high and low aroma compound producers. Analyzes performed on Syrah berries highlighted the stability of SIFT-MS volatilome from 28 days after mid-veraison.

SIFT-MS has proved to be an interesting instrument, enabling a rapid analysis of berry aroma composition in less than 2 minutes with  $O_2^+$ , with potential applications in many viticultural areas, including the phenotyping of grape varieties. For this use, berries shall be collected from 28 days after mid-veraison, a time point allowing a signal stability.

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## Impact of VvTPS24 genetics on farnesyl pyrophosphate bonding and production of $\alpha$ -guaiene, the rotundone direct precursor.

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#### **Objectives**

Rotundone is an interesting compound for the winemaking industry imparting distinctive peppery notes, detected in 2008 for the first time in Australian Shiraz wines. Rotundone biosynthesis elucidation has proven to be quite challenging.

Recently, a new terpene synthase (TPS) was discovered by sequencing the Pinot noir genome. This TPS, known as VvTPS24 was functionally characterized and was able to produce  $\alpha$ -guaiene, rotundone direct precursor. VvTPS24 is at the time, the only TPS known to transform farnesyl diphosphate (FPP) into  $\alpha$ -guaiene. It was also recently demonstrated that VvTPS24 was polymorphic and that variants produced more or less  $\alpha$ -guaiene depending on their sequence which implies that slight variations in sequence could lead to difference in  $\alpha$ -guaiene production and *in fine* to rotundone accumulation. This makes the TPS the perfect study subject to understand rotundone accumulation in grapevine.

In this research work, we investigated how *VvTPS24* sequence impacted FPP-enzyme docking i) via the sequencing of multiple *VvTPS24* alleles belonging to a large range of rotundone producing grapevine varieties, and ii) via the modelling of ligand-protein interactions.

#### Methods

PCR followed by cloning was used to amplify *VvTPS24* alleles in rotundone producers Syrah, Pinot Noir and Tardif. Sanger sequencing of these sequences were analyzed by SEAview (PRABI-Doua, Lyon, France) and Blasted (National Library of Medecine NCBI, Bethesda, Maryland, USA) to compute differences between sequences. Consensus alleles were then translated to the corresponding protein and protein tertiary structure was simulated using Phyre2 (Structural Bioinformatics Group, Imperial college, London, Great Britain). Then, these simulated proteins were used to mimic protein-ligand docking and to calculate docking properties.

#### Results and conclusions

Protein sequence directly influence FPP docking. Pinot noir was homozygous for *VvTPS24* while Syrah and Tardif were heterozygous and had an allele in common with Pinot noir. Syrah and Tardif exhibited both a specific *VvTPS24* variation.

Syrah and Tardif alleles were more prone to FPP docking, indicating that these alleles may have a greater affinity to FPP than the other allele corresponding to the non  $\alpha$ -guaiene producing allele.

Enzyme-ligand affinity might be one of the ways to explain  $\alpha$ -guaiene production and accumulation in the grape berries. More research notably expression experiments are needed to comprehend  $\alpha$ -guaiene accumulation and on how these new alleles might regulate  $\alpha$ -guaiene production.

### The impact of Saccharomyces yeasts on wine varietal aroma, wine aging and wine longevity

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#### **Objectives**

The objective of the present work is to evaluate the impact of different *Saccharomyces* strains on wine varietal aroma and on its evolution during wine aging.

#### Methods

Different types of musts, natural or semisynthetic supplemented with specific precursors of varietal aroma extracted from grapes, were fermented with different yeast strains. Wines were further submitted to a process of accelerated aging, heating the wine in complete anoxia at 50 or 75°C. The aroma profiles of the wines were analyzed during alcoholic fermentation, in the recently fermented wine and after accelerated aging. A wide range of volatiles was analyzed using several extraction methods including LLE, SPE, SBSE, derivatization, combined with different chromatographic techniques such as GC, GC-MS, GC-SCD, GC-GC-MS.

#### **Results and conclusion**

Overall, these experiments led to the identification of several pathways by which yeast can modulate wine varietal aroma, its evolution, and hence its longevity and sensory quality.

The first type of modulation is the direct action of yeast on the aroma precursors. Our results showed that yeasts could i) accelerate their hydrolysis, which advances the development of varietal aroma but does not change the final quantity formed ( $\beta$ -damascenone, geraniol), ii) metabolize some aroma precursors, so reducing the final quantity of aroma formed. This was particularly relevant in the case of aging off-odors (TDN, massoia lactone) or iii) metabolize grape components into aroma precursors, increasing the final aroma quantity (vanillin derivatives).

The second type of modulation proposed is indirect and consists in the biosynthesis by yeasts of some reactive species that can degrade varietal aroma. The best example is that of vinylphenols, which are reactive toward polyfunctional mercaptans.

The third type of modulation is via the formation of Strecker aldehydes, responsible for the oxidative degradation of wine aroma. These compounds were detected in recently fermented wines, supporting a fermentative and strain-related origin, and their levels increased during aging, despite the strict anoxia conditions, also in a strain-related way. Dicarbonyls left by fermentation may trigger the Strecker degradation of amino acids.

Finally, yeasts also deeply affected aging aroma by determining the levels formed during fermentation of the acid precursors of relevant esters (branched organic acids, leucidic and cinnamic acid).

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## Soil and climate zoning determining grapevine resource yield-gaps in Languedoc-Roussillon vineyards

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#### **Abstract**

Grapevine yield has been historically overlooked, assuming a strong trade-off between grape yield and wine quality. At present, menaced by climate change, many vineyards in Southern France are far from the quality label threshold, becoming grapevine yield-gaps a major subject of concern. Although yield-gaps are well studied in arable crops, we know very little about grapevine yield-gaps. In the present study, we analysed the environmental component of grapevine yield-gaps linked to climate and soil resources in the Languedoc Roussillon. We used SAFRAN data and IGP Pays d'Oc wine yields from 2010 to 2018. We selected climate and soil indicators proving to have a significant effect on average wine yield-gaps at the municipality scale. The most significant factors of grapevine yield were the Soil Available Water Capacity; followed by the Huglin Index and the Climatic Dryness Index. The Days of Frost; the Soil pH; and the Very Hot Days were also significant. Then, we clustered geographical zones presenting similar indicators, facilitating the identification of resources yield-gaps. We discussed the number of zones with the experts of IGP Pays d'Oc label, obtaining 7 zones with similar limitations for grapevine yield. Finally, we analysed the main resources causing yield-gaps and the grapevine varieties planted on each zone. Mapping grapevine resource yield-gaps are the first stage for understanding grapevine yield-gaps at the regional scale.

**Keywords:** grapevine yield-gaps, climate and soil indicators, vineyard regional mapping, yield declining.

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### Investigating the conceptualization and practices linked to peppery notes in Syrah red wines by French winemakers from different regions

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#### **Objectives**

The peppery attribute is often used to describe the aroma of Syrah wines. Rotundone was identified as the main aroma compound responsible for these notes. A significant percentage of anosmic respondents to this molecule was reported in previous studies. However, in most cases, these anosmic respondents, formally tested through three-alternative forced choice (3AFC), frequently declare being able to perceive peppery notes in wines. The main objective of this study was to investigate how anosmic French producers from two different regions conceptualize the peppery notes in Syrah red wines, and how they link it to production practices in comparison with non-anosmic producers. The first hypothesis was that anosmic and non-anosmic producers differently conceptualize the peppery perception and consequently, their practices are differently oriented to produce or avoid these notes. The second hypothesis was that this conceptualization may differ according to the studied region.

#### Methods

A total of 101 experts from two French wine regions involved in the winemaking of Syrah wines (n = 54 for the Northern Rhone valley (NRV), n = 48 for Languedoc-Roussillon (LR)) conducted two tasks. In the first task, experts were evaluated for their ability to detect rotundone using two consecutive 3AFC tests. Then, they participated in individual face-to-face semi-structured interviews consisting in three main parts. Firstly, they were asked to describe the last peppery wine they tasted, and they further had to provide technical information about it; secondly, they had to indicate the producing practices enhancing peppery notes in Syrah red; finally, they provided information about the positive or negative role played by this sensory on overall wine quality. Terms were lemmatized and grouped in categories and subcategories using a triangulation procedure, then the frequency of citation for each category was calculated.

#### **Results and conclusion**

The proportion of anosmic respondents who failed in both 3AFC tests were 45 % and 44 % for NRV and LR, respectively. Differences in the conceptualization of peppery notes were observed among anosmic and non-anosmic experts from NRV, but the region effect was greater. "Spicy," "freshness" and "length/aftertaste" were among the most frequently cited terms in the description task. The wine described by experts was in most cases made from Syrah grapes produced in specific regions (i.e., Rhone valley) or during particular seasons (i.e., 2021). In both regions, these notes were related to cool and wet conditions of climate. In most cases, experts from LR positively perceived peppery notes and associate them with high maturity grapes while for NRV, the perception was more mitigated as these notes were frequently related to a lack of maturity. The same study is being conducted in Australia and the results will be available soon.

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### Impact of different commercial Saccharomyces cerevisiae strains in Savatiano wines harvested at two ripening stages

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The ability of S. cerevisiae to synthesize or release volatile compounds varies significantly between yeast strains and strain specific differences in secondary metabolite composition in the fermented products have been observed. Although grape composition is also an important source of varietal characters and metabolites for yeast during fermentation. Savatianio variety is the second most planted variety in Greece and is considered to be one of the most ancient grape varieties in the world. The aim of this work was to study how grape maturity and yeast strain affect volatile profile and sensorial quality of Savatiano wines. Four commercial S.cerevisiae strains were used in order to compare and evaluate their impact on the chemical composition and sensorial characteristics of the wines produced from grapes harvested at two stages of grape ripening. The fermentation kinetics were monitored daily throughout the experiment. All the produced wines were analysed for the main oenological parameters (total acidity, volatile acidity, residual sugars, alcohol, malic acid, phenolic content), as well as for volatile compounds, such as higher alcohols, acetate esters, ethyl esters, volatile acids, and terpenes. In addition, sensory analyses were performed in all fermentation trials using a trained panel. Phenolic content and classic oenological characteristics showed no specific trend related to yeast strain but regarding these parameters, wines could be clearly separated according to the harvest date. Fermentation-derived compounds, contributed most to the differentiation of the samples with the wines from the second maturity stage characterized by higher volatile compounds concentrations. The aromatic profile of the produced wines was highly affected from the yeast strain. This study demonstrated the impact of the yeast strain and maturity level to wine volatile and sensory profile.

#### Volatile and phenolic composition of Agiorgitiko wines from fifteen different regions of PDO Nemea zone

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Agiorgitiko (Vitis Vinifera L. cv.) is the most cultivated red variety in Greece, mainly in Nemea region, the largest PDO zone of Greece. Although Agiorgitiko wines generates a great interest, in global market, there is a lack of information regarding the phenolic and aromatic profile of the variety. Fifteen vineyards, from different regions of the PDO Nemea zone, were selected in order to characterize the phenolic and aromatic composition of the variety and the heterogeneity among the different regions inside the PDO zone.

Vines from the fifteen vineyards were selected according to the common selection protocol and 60 kg of grapes were harvested from each one at the technological maturity. Microvinifications were conducted, in duplicate, following the same winemaking protocol. The produced wines were analyzed for their main oenological parameters and for their phenolic and volatile composition. Moreover, the wines were sensory evaluated by a trained panel.

Phenolic compounds and anthocyanidins content of wines ranged from medium to high levels in comparison to other red wine varieties. Also, the aroma compounds profile presented differences among the wines (p<0.05) from different regions. The sensory evaluation shows that Agiorgitiko wines were described mostly by red fruit attributes.

The present study provides a useful approach on the characterization of the phenolic and aromatic profile of Agiorgitiko wines, which is a great tool to improve the quality of the PDO Nemea wines and to gain knowledge on sub-zones in the general Agiorgitiko zone. Also, proved that the variability of Nemea's zone pedoclimatic conditions were affected on wine characteristics from different areas, which implies the need of further research on the impact of "terroir" in Agiorgitiko wines.

Keywords: red wine phenolics, aromatic content, Agiorgitiko, Nemea terroir

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#### Exploring the microbiota of resistant varieties in organic farming

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Grape varieties resistant to mildew and powdery mildew make it possible to reduce the use of phytosanitary products both in conventional and organic farming systems and as such can provide a solution to health and environmental concerns associated with fungicide use. The aim of the MicroVarioR project is to explore the microbiota of resistant grape varieties in comparison to conventional Vitis vinifera cultivars in various viticulture systems: conventional, organic and absence of treatment. Grapes from experimental parcels set up on two INRAE agricultural devices in Gruissan and Bordeaux, and on the Coupe Roses vineyard in Minervois, were harvested. This includes four resistant hybrid cultivars (Floréal, G5, Artaban, G14) and five traditional cultivars (Muscat, Grenache, Syrah, Carignan, Merlot). Microbiota on the surface of grape berries and at different time-points of spontaneous fermentations were analyzed with a metabarcoding approach. Relative abundance and diversity of yeast and fungal genera were compared and relative impact of different parameters were estimated. The strongest effect on microbiota diversity was linked to the geographical region, probably due to abiotic factors such as climate and soil. No statistical differences were observed between the microbiota of varieties cultivated in different viticulture systems. However, in each experimental device, cultivars were distinguishable, suggesting an influence of grape variety on the microbial community of grape berry surface. Ongoing research focuses on the effect of vintage on a broader set of resistant cultivars, and on berry composition as a determinant of microbiota variation.

Thanks are expressed to David Blon for providing grapes from the Coupe Roses vineyard. The MicroVarioR project is supported by the Métaprogramme METABIO of INRAE.

### Diffusion of phenolic compounds during a model maceration in winemaking: role of skins, flesh, and seeds.

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During red winemaking, the diffusion of phenolic compounds from the grape berry cells into the liquid phase occurs simultaneously with adsorptions of the same compounds onto the flesh cell walls. Previously, we quantified the proportions of polyphenols diffusing from the skins and assessed the amounts that can be fixed by the flesh [1, 2]. We showed that tannin adsorption, which can reach 50% of the initial amount in solution, depends on flesh cell wall composition. Higher molecular weight tannins and para-coumaroylated anthocyanins are selectively adsorbed [2].

In this study, we aimed at assessing (i) the impact of seeds, also present during winemaking, and (ii) the impacts of diffusion and adsorption when they occur concomitantly. This was done by carrying out macerations in a model medium with the following berry compartments: skins, seeds, skins + seeds, skins + seeds + pulp of two *Vitis Vinifera* grape varieties (Carignan, Car and Grenache, Gre), at two levels of maturity (D+ and D-). Microvinifications were performed in parallel with 900 g of grapes.

The seeds alone in solution released a rather high amount of tannins. However, as soon as they were in the presence of skin or flesh, and/or anthocyanins, the concentration of seed tannins in solution dropped abruptly, due to a combined effect of adsorption onto flesh or skin cell walls, and/or precipitation and/or chemical reactions. Some molecules disappeared preferentially: (i) a slight decrease in color was observed, attributable to a loss of anthocyanins and/or polymeric pigments; (ii) a decrease in the percentage of galloylation of the tannins that remain in solution (measured by HPLC after their depolymerization); (iii) a loss of higher molecular weight tannins (observed by Size Exclusion Chromatography).

When all the compartments of the berry were placed in solution together, the flesh adsorbed a significant amount of tannins as observed in previous studies [2], however, the amounts observed may suggest that the presence of flesh shifts the extraction equilibrium and that more polyphenols are extracted in the presence of flesh, even if they subsequently adsorb on it. Polyphenol amounts extracted in model systems with skins + seeds+ pulp were close to what was extracted in microvinification experiments. These model experiments reflect relatively well real winemaking and constitute a small volume tool for the characterization of new grape varieties.

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### Characterising innovations and sustainability in wine firms. An exploratory study of French wine industry.

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#### **Objective**

The objective of this study is to understand how the characteristics of the different types of firms, i.e. wine estates, wine cooperatives and wine merchants, and their forms of governance, can influence innovations and sustainability decisions in the wine industry.

This study has been run as part of the action plan of the Chaire Inq'Faaqt (Innovating in Agricultural and Agrifood chains, Quality and Territories), and more particularly the Axis 4 "firms and jobs".

#### Methods

Our methodology is qualitative and based on semi-structured interviews with the three main types of French wine firms: wine estates, wine cooperatives and wine merchants. A total of 16 businesses, located in Occitanie, Provence and Alsace, were interviewed. These interviews were quasi totally recorded and transcribed. These firms have very different production orientation, size and strategies.

In this poster, as a first exploratory step, we analyzed in deep one of each type of firms. This will allow us to perform a subsequent analysis of the full sample.

#### **Results**

We observed that process-related innovations were the most frequent, followed by organizational ones, related to governance. Marketing and logistic innovations were more frequent in the biggest and more decentralized firms (wine co-op and wine merchant). Also, sustainability-related innovations were very contrasted from a firm to another, the wine estate showing a more balanced sustainability strategy on the economic, environmental and social pillars.

Standards seem to be the most spread means to signal sustainability, especially for the wine co-op. The sustainability performance is not easy to measure because of its complexity. Firms preferred to implement more flexible voluntary commitment strategies (partnerships with stakeholders, returnable or recycling bottles systems, among others).

#### Conclusion

This exploratory study underlined the great diversity of the wine firms in terms of types of innovations and sustainability orientation. It is difficult to design a typology of wine firms, but it is now possible to have a more detailed idea about the main concerns, as water management, pesticides shortening or the relevance of organic wines.

#### LiDAR, a tool to inform sits-specific spraying: Application in a New York Concord grape production area

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In viticulture, linking a LiDAR, (Light Detection And Ranging) sensor with spraying devices, makes it possible to scan the vegetation while the tractor is passing through the rows and to adjust spray volumes to considerably improve the efficacy of the application of phytosanitary products. The objective of this work, carried out as continuation of the Efficient Vineyard project, led by Cornell University and supported by INRAE, was to test this in Concord vineyards and to propose a framework and workflow to analyse LiDAR sensor to evaluate, by simulation, the application of a variable spray dose according to the vegetative development of the vine. The plot to be treated is represented in a 3D image in the form of a point cloud representing the height, thickness and porosity of the foliage.

For this purpose, the research presented here was based on predictive modelling of the quantities and profiles of spray deposits using models developed within the UMR ITAP. These models take into account the architecture of the canopy, the characteristics of the sprayers and the spatial precision of the application when modelling deposition rates. To this end, four acquisitions with a vehicle-mounted LiDAR sensor were carried out in the 2022 production system at the Cornell Lake Erie Research and Extension Laboratory vineyard (~20 ha). This field data was used to evaluate, through simulations, the value of developing precision spraying techniques associated with data-intensive digital agriculture, particularly for the Concord production case in western New York State, through simulations.

This work resulted in the development of a coherent set of research tools that are eing incorporated nto the MyEV toolkit to generate maps of deposition patterns in Concord vineyards based on the spatial variability in the canopy. These will ultimately be used to inform spray management programs in these vineyards. The introduction of technologies for quantitative vegetation diagnosis (e.g. LiDAR), as well as an approach for calculating and managing an application rate (a functional dose relative to the leaf area), are essential to support site-specific management of agri-chemical application rates. It is now possible to use sprayers that implement variable rate technology to modulate the doses of plant protection products to be applied at fine spatial scales.

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#### New glutathionylated precursors of polyfunctional thiols in grapes: focus on Chardonnay and white interspecific cultivars grown in Belgium

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#### **OBJECTIVES**

16 Years ago, our laboratory evidenced a large number of pleasant polyfunctional thiols in Sauternes wines (1), and their degradation through aging (2). In the last decade, a big part of our research focused on the evidence of bounded forms of thiols in hop and in grapes (>20 references available on http://www.chair-de-clerck-2018.com), and the way to release this huge aromatic potential (3). So far, the number of S-conjugates identified in grapes remains limited to the cysteinylated and glutathionylated forms (+ dipeptide) of a few sulfanylalkyl alcohols and one sulfanylalkyl ketone.

Interspecific grape cultivars are highly represented in Belgian vineyards due to their resistance to fungal diseases and cold tolerance as compared to traditional cultivars. Recently, our group evidenced an exceptional polyfunctional thiol diversity in Belgian Solaris wines (4). Unfortunately, nothing is known concerning the availability of their precursors.

#### **METHODS**

In the present work, glutathionylated precursors were studied by HPLC-ESI(-)-MRM in Belgian interspecific white grapes (Johanniter, Solaris and Bronner; harvests 2000/very hot and 2001/very wet), in comparison to Belgian Chardonnay. A silver extraction method was used to analyze the free polyfunctional thiol content of wines, while the SAFE method enabled the isolation of the global aromatic fraction (GC-MS, GC-NPD and GC-O-AEDA).

#### **RESULTS**

G-3SHol was found at significant concentrations in Belgian interspecific grapes (5), with strong differences between both harvests (3 mg/kg in Johanniter 2020 versus 0.1-0.3 mg/kg in 2021). Unexpected high levels (4 mg/kg) were also found for Chardonnay issued from the same Belgian terroir, together with traces of the corresponding ester (G-3SHA) (also evidenced in one sample of Johanniter) (5).

For the first time, two additional sulfanylalkyl alcohol precursors (leading to the lemon-like 3-sulfanyl -pentanol and -heptanol) were identified in grapes (Solaris and Johanniter samples): G-3SPol (up to 7 mg/kg in Johanniter 2020 (5)), and G-3SHptol.

The SAFE extractions of the corresponding wines confirmed a very strong impact of the harvest year (generally lower aroma concentrations for 2021), with interesting molecules for some interspecific varieties (ethyl leucate, furaneol, ..).

#### **CONCLUSION**

High diversity and concentrations of pleasant thiols and their precursors can be brought by the interspecific grape varieties cultivated in Belgian vineyards. Yet additional results are needed to understand how climate and terroir will influence the potential of such wines.

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### Potential of N-CovSel for variable selection: a case study on time-series of multispectral images

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Multispectral image time-series have been promising for some years; yet, the substantial advance of the technology involved, with unprecedented combinations of spatial, temporal, and spectral capabilities for remote sensing applications, raises new challenges, in particular, the need for methodologies that can process the different dimensions of satellite information. Considering that the multi-collinearity problem is present in remote sensing time-series, regression models are widespread tools to model multi-way data. This paper presents the results of the analysis of a high order data of Sentinel-2-time series, conducted in the framework of extreme weather event. A new feature extraction method for multi-way data, N-CovSel was used to identify the most relevant features explaining the loss of yield in Mediterranean vineyards during the 2019 heatwave. Different regression models (uni-way and multiway) from features extracted from the N-CovSel algorithm were calibrated based on available heat wave impact data for 107 vineyard blocks in the Languedoc-Roussillon region and multispectral time-series predictor data for the period May to August. The performance of the models was evaluated by the r<sup>2</sup> and the root mean square of error (RMSE) as follows: for the temporal N-PLS model ( $r^2 = 0.62$  - RMSE = 11%), for the spatial N-PLS model ( $r^2 = 0.61 - RMSE = 12\%$ ) and the temporal-spectral PLS model ( $r^2 = 0.61 - RMSE = 12\%$ ) = 0.63 - RMSE = 11%). The results validated the effectiveness of the proposed N-CovSel algorithm in order to reduce the number of total variables and restricting it to the most significant ones. The N-CovSel algorithm seems to be a suitable choice to interpret complex multispectral imagery by temporally discriminating the most appropriate spectral information.

#### **Keywords:**

Feature extraction, Multi-way, Covariance selection, Remote sensing-, Times-series, Grapevine

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