



BOOK OF ABSTRACTS

Eno Macrowine 2023 Bordeaux KEDGE Businnes School 680 cours de la Libération 33405 Talence

ENO MACROWINE 2023







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Sous le patronage de **l'Organisation Internationale de la Vigne et du Vin**

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COMMUNICATIONS LINKED TO R&D PROJECTS SUPPORTED BY BIOLAFFORT® IN 2022 **34** SCIENTIFIC PUBLICATIONS SINCE 2018







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Welcome to the participants of the joint congress ŒnoMacrowine 2023 12th International Symposium of Oenology of Bordeaux and 9th edition of the Macrowine symposium

On behalf of the Oenology Research Unit at ISVV, we are delighted to welcome you in Bordeaux for the 60th anniversary of *Œno* symposium.

You are more than 300 delegates, mainly scientists, from 24 countries.

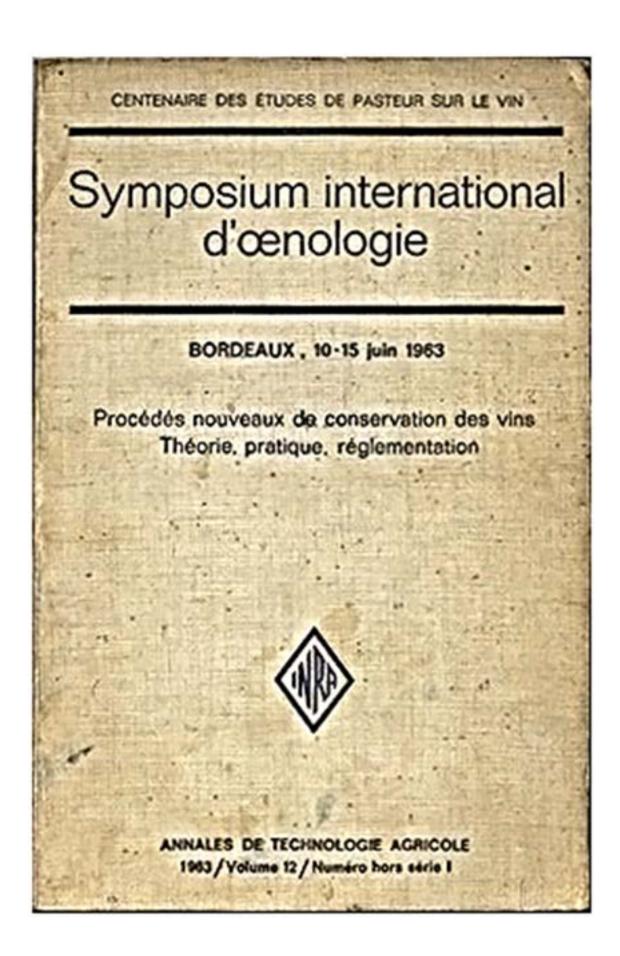
During these 4 days of scientific presentations at Kedge Business School building, we wish you pleasant and studious time, as well as fruitful exchanges and discussions. We also hope that you enjoy the welcome cocktail at the Palais de Rohan (Bordeaux City Hall) and the Gala evening at Château Luchey–Halde in Mérignac city as well as the tour of your choice. Do not hesitate to have a tour in our beautiful Bordeaux city, whose docks have been listed as a World Heritage Site.

This congress would not have been possible without the support of the Conseil regional de la Nouvelle Aquitaine (New Aquitaine council), Institute of wine and vine science, Bordeaux University, Bordeaux INP, Department of Environmental Sciences from Bordeaux University and Bordeaux Sciences Agro as companies sponsoring us : Biolaffort, DIAM, Cork Supply, Seguin-Moreau, BioSystems, Lallemand, Fermentis, Bellot, Hennessy.

We would also to extend our special thanks to Alexandre de Navailles, Managing Director of Kedge Business School, for kindly making his school's premises available to us. Eventually we want to thank sincerely the wine and spirits providers for all the wine provided during the congress

We will do our best to make sure that you will enjoy these days and wish you a very pleasant congress

The ŒnoMacrowine 2023 Organizing Committee



International Symposium of Enology of Bordeaux (Eno)

In 1963, with the objective to celebrate the 100th anniversary of Louis Pasteur's research on wine, the 1st International Symposium of Œnology of Bordeaux was created by Pr. Jean Ribéreau-Gayon and Dr. Emile Peynaud. After the first event, the symposium was again held in 1967, 1977, 1989 and then in 1995, 1999, 2003, 2007, 2011, 2015 and 2019. *Œno* symposia act as forums of exchange within the multidis-ciplinary scientific community dedicated to vine and wine sciences. *Œno* symposia are founded on traditional themes: plant environment, grape and wine quality; grape and wine microorganisms; enological practices and Processes; wine chemistry, wine components with physiological effects; sensory perception (psychophysics, cognitive psychology, experimental economy, connections with neurosciences). The research questions have evolved: research initially focused on wine aspect and stability problems. More recent questions focus on fermentation control, evaluation of grape qualitative potential, evolution during vinification and aging, including the parameters of sensory perception. The June 2019 edition was a joint collaboration with *In Vino Analytica Scientia* symposium under the denomination *ŒnoIVAS* 2019, a congress with a specific focus on grape, wine, wine-spirit analysis.

Macrowine congress

The Macrowine congress, International Conference of Macromolecules and Metabolites, was initiated in 2006 at the University of Reims-Champagne-Ardenne by Pr. Philippe Jeandet, Pr. Christophe Clément and Pr. Alexandra Conreux. Macrowine is a prestigious international conference initially focusing on wine macromolecules. "The core topic has also been the understanding of the structure, evolution, role and physico-chemical interactions of grape, vine and wine macromolecules, expanding to grape vine and wine secondary metabolites. The general purpose of the conference is to bring scientists closer and build international networks in order to help the wine industry to become more sustainable and competitive, able to produce better, richer, more appealing, safer, healthier and more stable and diverse products in a context strongly restricted by climate change, and the conference stimulates the presentation of results from international collaborations and networks. The congress, held every 2 years, constitutes a platform for exchange and presentation of the latest advances concerning macromolecules and compounds of secondary metabolism. "

Macrowine congress, as itinerant congress, was first held in Reims, France, (2006) then in Montpellier, France, (2008), Turino, Italy, (2010), Bordeaux, France, (2012), Stellenbosch, South Africa, (2014), Changins, Switzerland, (2016), Zaragoza, Spain, (2018) and Verona, Italy, (2021) before the Bordeaux Research unit Enology obtained the approval to organize the 9th edition in 2023 in Bordeaux.

Local Organizing Committee

Emmanuel Baugier, Marion Breniaux, Margaux Camelevre, Kleopatra Chira, Céline Cholet, Olivier Claisse. François Clavero, Marie Courregelongue, Philippe Darriet, Manon Ferreira. Céline Franc. Marine Gammacurta, Anne-Laure Gancel. Alicia Iouin. Michael Jourdes, Soizic Lacampagne, coordinator Marie Le Scanff. Axel Marchal. Stéphanie Marchand, Cécile Miot-Sertier, Julie Miranda, Lebna Mizani, Pierre Moulis. Claudia Nioi. Benjamin Poulain, Pascaline Redon, Laurent Riquier, Emilie Suhas. Pierre-Louis Teissedre Cécile Thibon. Sandra Vanbrabant, Sara Windholtz, Delphine Winstel,

ISVV, University of Bordeaux

UMR INRAE 1366 OENOLOGIE, ISVV, University of Bordeaux UMR INRAE 1366 OENOLOGIE, ISVV, University of Bordeaux

Dear colleagues,

It is a great pleasure for us to welcome you in Bordeaux for this joint scientific meeting, **ŒnoMacrowine 2023**, organized under the patronage of OIV. Oenology in its scientific dimension makes it possible to bring together a large community of researchers from various fields of expertise to share their knowledge and experience around wine and spirits.

Thus, as part of the organization of the 12th International Oenology Symposium, the Œnology Research Unit, Institute of Vine and Wine Sciences Bordeaux-Aquitaine, is hosting this year the 9th edition of the Macrowine symposium.

As a result, during 4 days, from July 10th to July 13nd, 2 international congresses are exceptionally held in the context of a joint organization with the following themes : Plant environment, Grape and Wine quality ; Grape and wine microorganisms ; Œnological Practices and Processes ; Wine chemistry, wine components with physiological effects ; Sensory properties : psychophysics, cognitive psychology, experimental economy, connexions with neurosciences. A particular emphasis has been given to communications regarding Macromolecules and secondary metabolites of grapes and wines.

Also, the wine sector is facing a number of challenges in the context of the agroecological transition and societal demands. **EnoMacrowine 2023** will explore aspects related to these challenges as the evaluation of ripening characteristics due to climate change, or the effect of viticultural and oenological practices on wine composition and quality, including adaptations, as well as opportunities given by oenological practices and processes or study of some chemical and sensory aspects.

7 speakers have been invited to introduce the different thematic fields of the congress and the evaluation of the papers by the scientific committees led to a scientific program of 63 oral communications, including 29 short communications. Then, 181 poster communications have been selected that is in total more than 250 communications.

We believe that the diversity of these communications will create a favorable context for scientific exchanges, through enabling our international community to contribute to the knowledge on wine as well as to support the wine sector in the face of its challenges.

We wish you an excellent symposium **ŒnoMacrowine 2023** rich in discoveries and fruitful exchanges!

On the behalf of ŒnoMacrowine scientific committee

Scientific committee

Philippe DARRIET, Stéphanie MARCHAND-MARION, Pierre-Louis TEISSEDRE, Conference Chair

ALBERTIN Warren - UMR INRAE 1366 OENOLOGIE, ISVV, University of Bordeaux, France BARBE Jean-Christophe - UMR INRAE 1366 OENOLOGIE, ISVV, University of Bordeaux, France BASTIAN Sue - School of Agriculture, Food and Wine, University Adelaide, Australia CHEYNIER Véronique - UMR INRAE 1083, Sciences pour l'œnologie, Montpellier, France CLUZET Stéphanie - UMR INRAE 1366 OENOLOGIE, ISVV, University of Bordeaux, France DARRIET Philippe - UMR INRAE 1366 OENOLOGIE, ISVV, University of Bordeaux, France De FREITAS Victor - FACULTY OF SCIENCES, University of Porto, Porto, Portugal De REVEL Gilles - UMR INRAE 1366 OENOLOGIE, ISVV, University of Bordeaux, France DELMOTTE François - UMR INRAE 1065 Santé Agro-écologie du vignoble, ISVV, Villenave d'Ornon, France Da SILVA FERREIRA Antonio Cesar - Portuguese Catholic University, Porto, Portugal FERREIRA Vicente - LAAE, Faculty of Sciences-University of Zaragoza, 50009 Zaragoza, Spain FISHER Ulrich - Institute for Viticulture and Oenology, DLR RHEINPFALZ, Neustadt-an-der-weinstraße, Germany GARDE Teresa – Institute of Wine Vine Sciences, CSIC – University of La Rioja, Logroño, Spain GHIDOSSI Rémy – UMR INRAE 1366 OENOLOGIE, ISVV, University of Bordeaux, France GIRAUD-H. Eric - Bordeaux School of Economics, ISVV, University of Bordeaux, France GOUGEON Régis - UMR 102, PAM Food and Wine Sciences Technology, University of Burgundy, Dijon, France HERDERICH Markus - The Australian Wine Research Institute, Adelaide, Australia HEYMANN Hildegarde - University of Missouri, Columbia, United States of America JOURDES Michael - UMR INRAE 1366 OENOLOGIE, ISVV, University of Bordeaux, France KILMARTIN Paul - School of Chemical Sciences, University of Auckland, Auckland, New Zealand KOTSERIDIS Yorgos - Dept Food Science and Nutrition, Agricultural University of Athens, Athens, Greece LE HENAFF-LE MARREC Claire - UMR INRAE 1366 OENOLOGIE, ISVV, University of Bordeaux, France LUCAS Patrick - UMR INRAE 1366 OENOLOGIE, ISVV, University of Bordeaux, France MARCHAL Axel - UMR INRAE 1366 OENOLOGIE, ISVV, University of Bordeaux, France MARCHAND Stéphanie - UMR INRAE 1366 OENOLOGIE, ISVV, University of Bordeaux, France MARULLO Philippe - UMR INRAE 1366 OENOLOGIE, ISVV, University of Bordeaux, France MASNEUF-POMAREDE Isabelle - UMR INRAE 1366 OENOLOGIE, ISVV, University of Bordeaux, France MAURY Chantal - GRAPPE research unit USC INRAE, Ecole Supérieure d'Agriculture d'Angers, France MOURET Jean Roch - UMR INRAE 1083, Sciences pour l'œnologie, Montpellier, France NIOI Claudia - UMR INRAE 1366 OENOLOGIE, ISVV, University of Bordeaux, France OLLAT Nathalie -UMR INRAE 1287 Ecophysiology and Functionnal Genomics of the Vine ISVV, University of Bordeaux, France RABOT Amélie - UMR INRAE 1366 OENOLOGIE, ISVV, University of Bordeaux, France RAUHUT Doris - Dept of Microbiology and Biochemistry, Geisenheim University, Geisenheim, Germany REGUANT Cristina - Dept of Biochemistry and Biotechnology, University Rovira I Virgili, Tarragona, Spain RICARDO da SILVA Jorge - School of Agriculture - University of Lisbon, Lisbon, Portugal RICHARD Tristan - UMR INRAE 1366 OENOLOGIE, ISVV, University of Bordeaux, France ROLLE Lucas - Department of Agricultural, Forestry and Food Sciences, University of Torino, Torino, Italy TAILLANDIER Patricia - UMR CNRS 5503, Chemical Engineering Laboratory, University of Toulouse Paul Sabatier, INP Toulouse, France TEISSEDRE Pierre-Louis - UMR INRAE 1366 OENOLOGIE, ISVV, University of Bordeaux, France TEMPERE Sophie - UMR INRAE 1366 OENOLOGIE, ISVV, University of Bordeaux, France UGLIANO Maurizio - Department of Biotechnology, University of Verona, Verona, Italy VAN LEUWEN Kees - UMR INRAE 1287 Ecophysiology and Functionnal Genomics of the Vine ISVV, University of Bordeaux, France VARELA Cristian - The Australian Wine Research Institute, Adelaide, Australia VERNHET Aude - UMR INRAE 1083, Sciences pour l'œnologie, Montpellier SupAgro Montpellier, France WAFFO-TEGUO Pierre - UMR INRAE 1366 OENOLOGIE, ISVV, University of Bordeaux, France WEIDMANN Stephanie - UMR 102 PAM, Food and Wine Sciences Technology, University of Burgundy, Dijon, France

ZAMORA Fernando - Dept of Biochemistry and Biotechnology, University Rovira I Virgili, Tarragona, Spain

ŒNO Macrowine 2023 PROGRAMME

Monday July 10th DAY 1

8:00 REGISTRATION

9:00 OPENING SESSION

10:00 KEY NOTE - Yvette Wohlfahrt Hochschule Geisenheim University, Department of General and Organic Viticulture, Von-Lade-Str. 1, 65366 Geisenheim, Germany

10:45 COFFEE BREAK + POSTERS

11:30 TOPIC I : Plant and Environment - Grape and wine quality - SESSION 1

- 11:30 I.O.4 Aaron Fait Rootstock mediated responses of grapevine (Vitis vinifera L.) meta bolism and physiology to combined water deficit and salinity stress in Syrah grafts -Albert Katz Department of Dryland Biotechnologies, French Associates Institute for Agriculture and Biotechnology of Drylands, Jacob Blaustein Institutes for Desert Research, Ben-Gurion University of the Negev, Israel
- 11:50 **I.O.3 Liliana Martinez** Overhead spray water treatment as a mitigation strategy for reducing vine stress and preserving grape quality during heatwaves National Univer sity of Cuyo, Argentina
- 12:10 **I.SC.4- Soeren Otto** Emergence of inorganic phosphonate residues in grapevine plant parts, berries and wines from sources other than foliar spraying – Geisenheim University, Germany

I.SC.7- Gabriele Valentini – An Automatic Canopy Cooling System To Cope With The Thermal-Radiative Stresses In The Pignoletto White Grape – University of Bologna, Department of Agricultural and Food Sciences (DISTAL), Viale Giuseppe Fanin 46, 40127 Bologna, Italy

I.SC.1 - Kenneth Olejar - Anti-transpirant modulation of grape ripening: Effects on Merlot vine development and Rosé wine phenolic and aromatic profiles. Appalachian State University, United States of America

12:30 LUNCH TIME + POSTER SESSION

14:30 TOPIC I : Plant and Environment - Grape and wine quality - SESSION 2

- 14:30 I.O.5 Laura Mezei Sip and Save the Planet: A Sensory and Consumer Exploration of Australian Wines made from potentially Drought-Tolerant White Wine Grapes -The University of Adelaide, Australia
- 14:50 **I.O.1- Marc Plantevin** Grouping *Vitis vinifera* grapevine varieties based on their aromatic composition Institut des Sciences de la Vigne et du Vin, France
- 15:10 **I.SC.8 Emilie Bruez** Effects Of Different Pruning Types On Charente Ugni Blanc Grape And Wine Quality - UMR 1366 OENOLOGIE, , Institut des Sciences de la Vigne et du Vin, France

I.SC.3 – Erika Jez – Do microplastics in vineyard soil affect the bioavailability of vine nutrition? – University of Nova Gorica, Slovenia

I.SC.9 - Céline Cholet - Impact Of Harvest Date On The Fine Molecular Composition Of Must And Bordeaux Red Wine (Var. Merlot, Cabernet Sauvignon). Focus On Acidity And Sensory Impact After Five Years Of Aging. - UMR 1366 OENOLOGIE, , Institut des Sciences de la Vigne et du Vin, France

15:30 COFFEE BREAK + POSTERS

16:30 TOPIC I : Plant and Environment - Grape and wine quality SESSION 3

- 16:30 **I.O.6 Vincent Carré** Probing grapevine-*Botrytis cinerea* interaction through mass spectrometry imaging LCP-A2MC, Université de Lorraine, France
- 16:50 I.SC.2 Liang Jiang Discrimination of Botrytis cinerea infected grapes using un targeted metabolomic analysis with direct electrospray ionisation mass spectrometry – Charles Sturt University, Australia
- 16:55 **I.O.2 Peter Schumacher** Influence of Agrophotovoltaic on Vine and Must in a Cool Climate – ZHAW Zurich University of Applied Sciences, Switzerland
- 17:15 **I.SC.6 Dimitrios** Evangelos Miliordos Influence of Chitosan, Abscisic Acid and Benzothiadiazole treatments on Savvatiano (Vitis vinifera L.) wines volatile composition profile - Agricultural University Athens, Department of Food Science and Human Nutrition, Laboratory of Oenology and Alcoholic Beverage Drinks, Greece
- 17:20 **I.SC.5 Ágnes Dienes-Nagy** Impact of must nitrogen deficiency on white wine composition depending on grape variety Agroscope, Switzerland

19:30 WELCOME AT "CITY HALL"

Tuesday July 11th DAY 2

9:00 KEY NOTE - Florian Bauer

South African Grape and Wine Research Institute, Department of Viticulture and Oenology, Stellenbosch University, Stellenbosch, South Africa

9:45 TOPIC II : Grape and wine microorganisms : diversity and adaptation - SESSION 1

- 9:45 **II.O.6 Mitja Martelanc** Yeast-produced volatiles in grape based system model acting as antifungal bioagents against phytopathogen *Botrytis cinerea* - University of Nova Gorica, Wine Research Centre, Slovenia
- 10:05 **II.O.3 Adrien Destanque** Fungal diversity and dynamics in Champagne vineyards: from vine to wine - Univ Brest, INRAE, Laboratoire Universitaire de Biodiversité et Ecologie Microbienne, France
- 10:25 II.O.2 Camille Eicher Changes in metabolic fluxes under low pH growth conditions: Can the slowdown of citrate consumption improve *Oenococcus oeni* acid-tolerance? UMR A. 02.102 Procédés Alimentaires et Microbiologiques, L'institut Agro Dijon, Université de Bourgogne Franche-Comté, Dijon, France

10:45 COFFEE BREAK + POSTERS

11:30 TOPIC II : Grape and wine microorganisms : diversity and adaptation - SESSION 2

- 11:30 **II.O.4 Jules Harrouard** Large-scale phenotypic screening of the spoilage yeast *Brettanomyces bruxellensis*: untangling patterns of adaptation and selection, and conse quences for innovative wine treatments – UMR 1366 OENOLOGIE, , Institut des Sciences de la Vigne et du Vin, France
- 11:50 I**I.O.5 Fanny Bordet** Metabolic interactions of *Saccharomyces cerevisiae* cocul tures: a way to extend the aroma diversity of Chardonnay wine - Univ. Bourgogne Franche-Comté, Institut Agro Dijon, PAM UMR A 02.102, France
- 12:10 **II.O.1 Charlotte Vion** Acidic and demalic *Saccharomyces cerevisiae* strains for managing problems of acidity during the alcoholic fermentation - Biolaffort, UMR 1366 OENOLOGIE, , Institut des Sciences de la Vigne et du Vin, France

12:30 LUNCH TIME + POSTER SESSION

14:00 KEY NOTE - Maria Tiziana Lisanti

Università degli Studi di Napoli Federico II, Italy

14:45 TOPIC IV : Œnological Practices and Processes - SESSION 1

 14:45 IV.SC.4 - Sara Windholtz - Bioprotection by adding non-Saccharomyces yeasts : advanced research on this promising alternative to SO₂ - UMR 1366 OENOLOGIE, , Institut des Sciences de la Vigne et du Vin, France
 IV.SC.1 - Edouard Pelonnier-Magimel - Bordeaux red wines without added sulfites specificities: compositional and sensory approaches towards highlighting and explaining their specific fruitiness and coolness - UMR 1366 OENOLOGIE, Institut des Sciences de la Vigne et du Vin, France
 IV.SC.7 - Fernando Zamora - Inoculation of the selected Metschnikowia pulcherrima MP1 as a bioprotective alternative to sulfites to prevent browning of white grapemust

MP1 as a bioprotective alternative to sulfites to prevent browning of white grapemust Departament de Bioquímica i Biotecnologia, Facultat d'Enologia de Tarragona, Universi tat Rovira i Virgili, Spain

- 15:05 **IV.O.6 Milena Lambri** Monitor some key parameters through the implementation of continuous control systems of the must-wine during maceration-fermentation in red winemaking to manage operations in "automation" Department for Sustainable Food Process DiSTAS, Università Cattolica del Sacro Cuore, Italy
- 15:25 **IV.O.3 Xavier Hastoy** Eugenol as quality marker of wines and spirits from hybrid vines: impact of different winemaking and distillation processes UMR 1366 OENOLOGIE, Institut des Sciences de la Vigne et du Vin, France
- 15:45 IV.SC.5 Liming Zeng Development of distillation sensors for spirit beverages pro duction monitoring based on impedance spectroscopy measurement and partial least squares regression (PLS-R) - Changins Viticulture and Enology College, University of Applied Sciences and Arts of Western Switzerland (HES-SO), Nyon, Switzerland IV.SC.6 - Almudena Marrufo-Curtido - Impact of the wines' quality on the wine distil lates' organoleptic profile - Departamento de Investigación y Desarrollo de BODEGAS FUNDADOR, Spain

15:55 COFFEE BREAK + POSTERS

- 16:30 TOPIC IV : Œnological Practices and Processes SESSION 2
 - 16:30 IV.0.1 Gauthier Lagarde A new strategy and methodology for the characterization of polyphenols in fining precipitate UMR 1366 OENOLOGIE, Institut des Sciences de la Vigne et du Vin, France
 IV.SC.10 Michaël Nigen New plant biopolymers for the colloidal stability of the colo ring matter of red wines UMR IATE, France
 - 16:55 **IV.O.7 Marie Le Scanff** Effect of whole bunch vinification on the abundance of a sweetening compound UMR 1366 OENOLOGIE, Institut des Sciences de la Vigne et du Vin, France
 - 17:15 **IV.O.2 Elisabetta Pittari** Effect of different temperature and water-loss dehydration conditions on the pattern of free and glycosylated volatile metabolites of Italian red grapes University of Naples Federico II, Italy
 - 17:35 **IV.O.4 Saul Assunção Bicca** -Exploring the influence of *s. cerevisiae* mannoproteins on wine astringency and the impact of their polysaccharide structure Institut Agro Montpellier, France
 - 17:55 **IV.SC.2 Bahareh Sarmadi** Analyzing the Role of Elemental Sulfur in Grape Juice on the Development of Polyfunctional Mercaptans in Sauvignon Blanc Wines - University of Auckland, New Zealand

IV.SC.3 - Florian Bahut - Antioxidant capacity of Inactivated non-*Saccharomyces* yeasts - Lallemand SAS, France

- 18:05 **IV.O.5 Patricia Taillandier** Production of a functional beverage from winemaking by-products: a new way of valorisation LGC/Université de Toulouse, France
- 18:25 IV.SC.8 François Clavero Intense pulsed light for vineyard wastewater: a promising new process of degradation for pesticides UMR 1366 OENOLOGIE, Institut des Sciences de la Vigne et du Vin, France
 IV.SC.12 Benjamin Poulain Sub-critical water: an original process to extract antioxidants compounds of wine lees UMR 1366 OENOLOGIE, Institut des Sciences de la Vigne et du Vin, France
 IV.SC.11 Silvia Rocha Pairing wine and stopper: an old issue with new achievements

Department of Chemistry & LAQV-REQUIMTE, University of Aveiro, Portugal

18:40 END OF THE SESSION

Wednesday July 12th DAY 3

8:30 KEY NOTE - Markus Herderich

The Australian Wine Research Institute - Urrbrae (Adelaide), Australia

9:15 TOPIC III : Wine chemistry, wine components with physiological effects - SESSION 1

- 9:15 **III.O.7 Alexandre Pouget** Revealing the origin of Bordeaux wines with raw 1D-chromatograms -Département des neurosciences fondamentales. Université de Genève, Switzerland
- 9:35 III.O.3 Giovanni Luzzini Aging patterns of varietal volatile profiles of white wines: a case study on 18 Italian varietal white wines -University of Verona, Department of Biotechnology, Italy
- 9:55 **III.O.10 Charlotte Maxe** White wines oxidative stability: a 2-vintage study of Char donnay champagne base wines aged on lees in barrels – Institut Universitaire de la Vigne et du Vin, UMR PAM Université de Bourgogne/Institut Agro Dijon, Jules Guyot, France
- 10:15 **III.SC.2 Alberto De Iseppi** Characterization and identification of yeast bioactive peptides released during fermentation and autolysis in model wine Department of Agronomy, Food, Natural Resources, Animals and Environment (DAFNAE), University of Padova, Italy

III.SC.5 -Aurélie Roland -New insights into the fate of markers involved in fresh mushroom off-flavours during alcoholic fermentation - SPO, Université de Montpellier, INRAE, Institut Agro, Montpellier, France

III.SC.7 - Daniela Fracassetti - Oenological tannins for preventing the light-struck taste in white and rosé wines - Department of Food, Environmental and Nutritional Sciences (DeFENS), Università degli Studi di Milano, Italy

III.SC.9 - Svetlana Cvetkova - Photochemical degradation of tryptophan in model wine: impact of heavy metals and oxygen on 2-aminoacetophenone formation - Wein campus Neustadt/DLR Rheinpfalz, Institute for Viticulture and Enology, Germany

10:45 COFFEE BREAK + POSTERS

11:15 TOPIC III : Wine chemistry, wine components with physiological effects - SESSION 2

11:15 **III.O.1 - Tracey Siebert** - 'Tropical' polyfunctional thiols and their role in Australian red wines - The Australian Wine Research Institute, Australia

- 11:35 **III.O.5 Emilio de Longhi** Contribution of volatile thiols to the aroma of Riesling wines from three regions in Germany and France (Rheingau, Mosel, and Alsace) Université de Bordeaux, Bordeaux INP, INRAE, OENO, UMR 1366, ISVV, France
- 11:55 III.SC.8 -Emilie Suhas Optimizing the identification of new thiols at trace level in aged red wines using new oak wood functionalisation strategy
 Université de Bordeaux, Bordeaux INP, INRAE, OENO, UMR 1366, ISVV, France

 III.SC.6 Cole Cerrato Novel benzenethiols with phenols cause ashy, smoke flavor perception in red wines MHCS, Epernay, France
 III.SC.4 Ruth Hornedo-Ortega -Neuroprotective and anti-inflammatory properties of hydroxytyrosol: a promising bioactive component of wine Departamento de
 Nutrición, Bromatología, Toxicología y Medicina Legal, Facultad de Farmacia, Universidad de Sevilla, Spain

12:15 LUNCH TIME + POSTER SESSION

13:30 KEY NOTE - Victor Freitas

LAQV-REQUIMTE - University of Porto, Faculty of Science, Portugal

14:15 TOPIC III : Wine chemistry, wine components with physiological effects - SESSION 3

- 14:15 **III.O.2 Vicente Ferreira** About the role played by the different polyphenols on oxygen consumption and on the accumulation of acetaldehyde and Strecker Aldehydes during wine oxidation – University of Zaragoza, Spain
- 14:35 III.SC.1 Isara Vongluanngam Changes in Cu fractions and riboflavin in white wines during short-term light exposure: Impacts of oxygen and bottle colour, School of Agri cultural, Environmental and Veterinary Sciences, Charles Sturt University, Australia III.SC.10 - Luís Filipe-Ribeiro - Pinking phenomena on white wines: Relation between pinking susceptibility index (PSI) and wine anthocyanins content - CQ-VR-Chemistry Research Centre-Vila Real, Food and Wine Chemistry Laboratory, University of Trásos-Montes, Portugal
- 14:45 **III.O.4 Tom Estier** Assessing the role of 27 known bitter compounds in commercial white wines combining LC-MS quantification and sensory analysis –UMR 1366 OENOLOGIE, Institut des Sciences de la Vigne et du Vin, France
- 15:05 **III.O.9 Jean–Claude BOULET** The role of cell wall polysaccharides in the extraction of anthocyanins and tannins: results, perspectives of a more positive contribution Univ. Montpellier, SPO, INRAE, Institut Agro Montpellier Supagro, France
- 15:25 **III.O.8 Ulrich Fischer** Sensory profiles and European Consumer Preference related to Aroma and Phenolic Composition of Wines made from Fungus Resistant Grape Va rieties (PIWI) DLR Rheinpfalz, Institute for Viticutlure and oenology, Germany
- 15:45 III.SC.3 Marie Courregelongue Impact of climatic conditions on the seasoning quality of oak wood for oenological use (Quercus petraea) - UMR 1366 OENOLOGIE, Institut des Sciences de la Vigne et du Vin, France

15:50 END OF THE SESSION

Thursday July 13th DAY 4

9:00 KEY NOTE - María-Pilar Sáenz-Navajas

Instituto de Ciencias de la Vid y del Vino (ICVV) (UR-CSIC-GR), Carretera de Burgos Km. 6, Finca La Grajera, 26007 Logroño, La Rioja, Spain

9:45 TOPIC V : Sensory properties : psychophysics-cognitive psychology, experimental economy, connexions with neurosciences – SESSION 1

- 9:45 **V.O.2 Susan Bastian** Sensory Properties Important to Australian Fine Wine Consu mer Segment Perception of Chardonnay Wine Complexity and Preference. School of Agriculture, Food & Wine, Waite Research Institute, The University of Adelaide, Australia
- 10:05 **V.O.4 Léa Lecomte** Wine consumer trade-off between organoleptic characteristics and sustainable claims. An experiment on red wines from Bordeaux Region Univ. Bordeaux, CNRS, BSE, UMR 6060, INRAE, Bordeaux Sciences Agro, France
- 10:25 **V.O.3 Inès Elali** Wine as an emotional and aesthetic object: impact of expertise UMR 1366 OENOLOGIE, Institut des Sciences de la Vigne et du Vin, France

10:45 COFFEE BREAK + POSTERS

11:15 TOPIC V : Sensory properties : psychophysics-cognitive psychology, experimental economy, connexions with neurosciences - SESSION 2

- 11:15 **V.O. 5 Margaux Cameleyre** Which impact for proanthocyanidic tannins on red wine fruity aroma? sensory and physicochemical approaches UMR 1366 OENOLOGIE, Institut des Sciences de la Vigne et du Vin, France
- 11:35 **V.O.1 Lisa Käppler** Sensory improvement of dealcoholised wines DLR Rheinpfalz, Neustadt/Weinstraße, Germany
- 11:55 **V.SC.1 Kleopatra Chira** Sensory characterization of Cognac eaux-de-vie aged in barrels representing different toasting process Univ. Bordeaux, Bordeaux INP, INRAE, OENO, UMR 1366, ISVV, France
- 12:00 **V.SC.2 Florian Lecasse** Wine swirling: A first step towards the unlocking of the wine's taster gesture Groupe de Spectrométrie Moléculaire et Atmosphérique (GSMA), UMR CNRS 7331, UFR Sciences Exactes et Naturelles, France

12:05 CLOSURE KEY NOTE - Graham HARDING Oxford college (UK)

12:50 CLOSURE OF THE EVENT + NEXT MACROWINE PRESENTATION

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I-SC 02. DISCRIMINATION OF *BOTRYTIS CINEREA* INFECTED GRAPES USING UNTARGETED ME-TABOLOMIC ANALYSIS WITH DIRECT ELECTROSPRAY IONISATION MASS SPECTROMETRY Liang Jiang, Morphy C. Dumlao, William A. Donald, Christopher C. Steel, Leigh M. Schmidtke

I-SC 03. DO MICROPLASTICS IN VINEYARD SOIL AFFECT THE BIOAVAILABILITY OF VINE NU-TRITION?

Erika Jez, Elisa Pellegrini, Maria De Nobili, Marco Contin

I-SC 04. EMERGENCE OF INORGANIC PHOSPHONATE RESIDUES IN GRAPEVINE PLANT PARTS, BERRIES AND WINES FROM SOURCES OTHER THAN FOLIAR SPRAYING Sören Otto, Randolf Kauer, Yvette Wohlfahrt, Beate Berkelmann-Löhnertz, Bianca May, Ralf Schweiggert

I-SC 05. IMPACT OF MUST NITROGEN DEFICIENCY ON WHITE WINE COMPOSITION DEPENDING ON GRAPE VARIETY Thibaut, Verdenal, Jean-Laurent Spring, Marie Blackford, Fabrice Lorenzini

I-SC 06. INFLUENCE OF CHITOSAN, ABSCISIC ACID AND BENZOTHIADIAZOLE TREATMENTS ON SAVVATIANO (*VITIS VINIFERA* L.) WINES VOLATILE COMPOSITION PROFILE. Miliordos Dimitrios Evangelos, Elli Gouliti, Kontoudakis Nikolaos, Kotseridis Yorgos

I-SC 07. AN AUTOMATIC CANOPY COOLING SYSTEM TO COPE WITH THE THERMAL-RADIATIVE STRESSES IN THE PIGNOLETTO WHITE GRAPE

Gabriele Valentini, Ginaluca Allegro, Chiara Pastore, Riccardo Mazzoleni, Massimo Noferini, Ilaria Filippetti

I-SC 08. EFFECTS OF DIFFERENT PRUNING TYPES ON CHARENTE UGNI BLANC GRAPE AND WINE QUALITY

Emilie Bruez, Céline Cholet, Patrice Coll, Mathilde Boisseau, Xavier Poitou, Pascaline Redon , Laurent Riquier, Ghislaine Hilbert-Masson, Sandra Vanbrabant, Soizic Lacampagne, Laurence Geny-Denis

I-SC 09. IMPACT OF HARVEST DATE ON THE FINE MOLECULAR COMPOSITION OF MUST AND BORDEAUX RED WINE (VAR. MERLOT, CABERNET SAUVIGNON). FOCUS ON ACIDITY AND SENSORY IMPACT AFTER FIVE YEARS OF AGING

Zoi Avramidou, Margaux Cameleyre, Alexandre Pons, Axel Marchal, Michael Jourdes, Taku Saito, Soizic Lacampagne, Pascaline Redon, Laurence Geny, Pierre-Louis Teissedre, Philippe Darriet, Céline Cholet

Topic III

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III-SC 03. IMPACT OF CLIMATIC CONDITIONS ON THE SEASONING QUALITY OF OAK WOOD FOR OENOLOGICAL USE (*QUERCUS PETRAEA*) Marie Courregelongue, Andrei Prida, Alexandre Pons

III-SC 04. NEUROPROTECTIVE AND ANTI-INFLAMMATORY PROPERTIES OF HYDROXYTYRO-SOL: A PROMISING BIOACTIVE COMPONENT OF WINE Hornedo-Ortega, Ruth, Gallardo-Fernández Marta, Cerezo Ana Belén, Troncoso Ana, Garcia-Parrilla Carmen

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IV-SC 07. INOCULATION OF THE SELECTED METSCHNIKOWIA PULCHERRIMA MP1 AS A BIO-PROTECTIVE ALTERNATIVE TO SULFITES TO PREVENT BROWNING OF WHITE GRAPE MUST Marco Bustamante, Pol Giménez, Arnau Just-Borras, Ignasi Solé-Clua, Jordi Gombau, José M. Heras, Nathalie Sieczkowski, Mariona Gil, Joan Miquel Canals, Fernando Zamora

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Maria Antonieta Anaya-Castro, Thierry Doco, Pascale Williams, Céline Charbonnel, Virginie Moine, Arnaud Massot, Philippe Louazil, Isabelle Jaouen, Christian Sanchez and Michaël Nigen

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Benjamin Poulain, Delphine Winstel, Axel Marchal, Virginie Moine And Claudia Nioi

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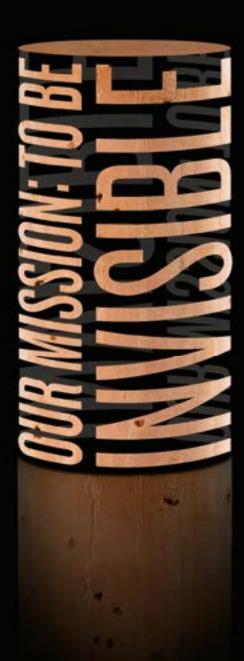
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Lectures

A SYNTHESIS APPROACH ON THE IMPACT OF ELEVATED CO₂ ON BERRY PHYSIO-LOGY AND YIELD OF VITIS VINIFERA

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Keywords: climate change, carbon dioxide (CO₂), grapevine physiology, berry development

Besides the increase in global mean temperature the second main challenge of a changing climate is the increase in atmospheric carbon dioxide (CO_2) in relation to physiology and yield performance of grape-vines. The benefits of increasing CO_2 levels under greenhouse environment or open field studies have been well investigated for various annual crops. Research under free carbon dioxide enrichment on field-grown perennial plants such as grapevines is limited to a few studies. Further, chamber and greenhouse experiments have been conducted mostly on potted vines under eCO_2 conditions.

In this synthesis approach, the output of different CO₂ enrichment experiments such as greenhouse and growth chamber trials will be compared to open top chamber (OTC) and Free Air Carbon dioxide Enrichment (FACE) studies. Furthermore, the regional climate in which single field studies have been conducted plays a major role in terms of up and down regulation of CO₂ induced processes, whereas in open or closed chamber systems a stable but artificial microclimate exists within the chamber.

Due to higher photosynthesis rates under eCO_2 mature field grown vines showed higher transport capacity and larger sinks for additional carbohydrates produced under eCO_2 , thus grapevines increased in vegetative and reproductive growth. During fruit ripening single berry weight, bunch architecture and bunch compactness altered similarly for vines under eCO_2 within the field and to a lower extent when it comes to short-term chamber and greenhouse trials. Regarding crop yield, no or little differences occurred for all varieties for the first year of investigation. Usually, higher yield emerged under eCO_2 in the following season as explained by the grapevine's reproductive cycle. Analyses of berries and must resulted mostly in alterations of malic and tartaric acid concentrations under eCO_2 and was close linked to berry size. Sugar accumulation in berries depended on climatic factors and differed if vines were grown under warm or cool climate conditions in combination with CO_2 enrichment. Elevated CO_2 was also described to modify some berry colour parameters like anthocyanins, but in the end both syntheses were induced – stimulation and inhabitation of anthocyanin accumulation.

Overall, eCO_2 resulted in a change of vegetative, generative and qualitative parameters of grapevines compared to an atmospheric CO_2 concentration without affecting wine quality in general. Nevertheless, as carbon dioxide is one of many influencing climate factors on fruit and berry development it needs to be discussed within the context of future wine quality.

MICROBIAL ECOSYSTEMS IN WINERIES – MOLECULAR INTERACTIONS BETWEEN SPECIES AND MODELLING OF POPULATION DYNAMICS

Florian F. BAUER

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Microbial ecosystems are primary drivers of viticultural, oenological and other cellar-related processes such as wastewater treatment. Metagenomic datasets have broadly mapped the vast microbial species diversity of many of the relevant ecological niches within the broader wine environment, from vineyard soils to plants and grapes to fermentation. The data highlight that species identities and diversity significantly impact agronomic performance of vineyards as well as wine quality, but the complexity of these systems and of microbial growth dynamics has defeated attempts to offer actionable tools to guide or predict specific outcomes of ecosystem-based interventions. The application of such tools in future will depend on our understanding of the physiological and molecular drivers that govern microbial ecosystems. Here we describe several integrated approaches to characterize the molecular interactions between species within the fermentation and the waste-water ecosystem and to model the development of these ecosystems. Binary (two species) and consortia-based approaches indicate ecosystem-specific developmental patterns in these systems. On a molecular level, data strongly support that cell-wall related properties of yeast species impact the development of fermentation ecosystems during wine making and highlight the importance of physical contacts between species in these ecological processes. To model the wine yeast fermentation ecosystem, high-throughput flow cytometry-based approaches were developed, and specific models based on a machine-learning approach were developed. In winery wastewater, laboratory-based evolution of two species exposed to biotic selection pressure in a synthetic environment, Saccharomyces cerevisiae and the microalga Chlorella sorokiniana, identified two specific genes involved in carbon and nitrogen catabolite repression that facilitate mutualistic behaviors between yeast and microalgae when inactive. Taken together the data suggest novel strategies for microbial ecosystem-based decision making in wine making and improved integration of natural microbial biodiversity in the process.

SEARCHING FOR THE SWEET SPOT: A FOCUS ON WINE DEALCOHOLIZATION

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It is well known that the vinification of grapes at full maturation can produce rich, full-bodied wines, with intense and complex flavour profiles. However, the juice obtained from such grapes may have very high sugar concentration, resulting in wines with an excessive concentration of ethanol. In addition, the decoupling between technological maturity and phenolic/aromatic one due to global warming, exacerbates this problem in some wine-growing regions. In parallel with the increase of the mean alcohol content of wines on the market, also the demand for reduced alcohol beverages has increased in recent years, mainly as a result of health and social concerns about the risks related to the consumption of alcohol. Moreover, an excessive ethanol content may result in wines with an unbalanced flavour. For this reason, wine dealcoholization is currently one of the most important issues for the wine industry and wine research.

Several dealcoholization techniques, mainly based on vacuum distillation and membrane separation techniques, are available to reduce wine alcohol content at different levels. However, the main concern about wine dealcoholization, most of all when it is applied as a corrective oenological practice, is the possible loss of sensory active compounds during the process. Considerable research has therefore been undertaken over the past ~15 years to understand the impact of wine dealcoholization on wine quality. This lecture will provide an overview on wine dealcoholization, with particular emphasis on its effects on wine chemical composition and sensory characteristics

METABOLOMICS FOR GRAPE AND WINE RESEARCH: EXPLORING THE CONTRIBUTIONS OF AMINO ACIDS TO WINE FLAVOUR

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A critical aspect of wine quality is the overall expression of wine flavour, which is formed by the interplay of volatile aroma compounds, their precursors, and taste and matrix components.

Grapes directly contribute to wine only a small number of potent aroma compounds, and the unique sensory attributes and perceived quality of a wine result from combining 100s of metabolites of grapes, yeast and bacteria, and oak wood.

So far only a relatively small proportion of flavour-active secondary metabolites in grapes has been extensively studied, including organic acids, polyphenols and some non-volatile aroma precursors such as glycoconjugates or glutathione.

In this presentation a summary will be presented about current technologies for metabolite analysis and key aspects for developing successful metabolomics applications in grape and wine research. Topics covered include methods for MS and NMR-based metabolomics and the application of metabolomics approaches to characterise the contributions of amino acids to wine flavour.

MOLECULAR APPROACHES FOR UNDERSTANDING AND MODULATING WINE TASTE

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Wine consumers generally demand wines having a perception of softer tannins and less ripe, having a heaviness and richness on palate (full-body wine) with a limpid and stable color. However, polyphenol (tannins)-rich wines have been also correlated with unpleasant taste properties such as astringency and bitterness when perceived at high intensities. Modulating these unpleasant properties could be important for consumer's approval of wines.

Indeed, polyphenols are usually associated with flavor, and particularly with astringency, due to their ability to complex with salivary proteins [1]. Saliva is rich in different SP families described to be involved in astringency, namely basic PRPs, glycosylated PRPs, acidic PRPs, statherin/P-B peptide and cystatins. However, due to saliva being a complex fluid, its protein profile may quantitatively and qualitatively vary under different conditions. Currently, astringency is recognized as a trigeminal sensation although the molecular pathway responsible for its onset is yet to be fully established. Moreover, it is unknown if the many different astringency mouthfeel sub-qualities such as velvet, puckering, harsh, among others, are perceived by different mechanisms. Besides the structural factors and medium conditions, there are some endogenous factors that affect astringency perception such as the physiological response, circadian rhythms, salivary flow rate and time of exposure. Indeed, astringency is perceived as a diffuse stimulus and dynamic process in the oral cavity that requires time to be elicited. It is known that astringency increases upon successive exposures to tannins [2,3].

Wine industry has some strategies to balance astringency and bitterness such as the use of some fining agents and also some winemaking practices (e.g. oak aging, batonnage and microoxygenations) leading to the loss of phenolics and also promoting the chemical change of some of them. While removing phenolic compounds is necessary to fulfill some organoleptic requirements of a beverage, the process must be controlled to avoid some collateral effects such as the loss of flavor [4]. Polysaccharides have been an emerging natural and sustainable option to be used on the modulation of taste properties. In fact, polysaccharides can influence salivary

protein-tannin interactions and they could be used to modulate astringency and bitterness.

^{1.} Soares, S., et al., Scientific Reports, 2020, 10, 12638.

^{2.} Lesschaeve, I. and Noble, C. A., Am. J. Clin. Nutr, 2005, 81, 330S-5S.

^{3.} Brandão, E.; Soares, S.; Mateus, N.; de Freitas, V., J. Agri. Food Chem. 2014, 62, 9562–9568.

^{4.} Francisco, T., et al., Food Res. Int., 2021, 143, 110261

BEYOND LIKING SCORES: THE IMPORTANCE OF THE DRINKING EXPERIENCE TO UNDERSTAND OUR CONSUMERS

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The presentation will approach the understanding of wine consumers 'perception based on the experiential model suggested by Warell (2008). In this framework, wine consumption gives rise to a variety of experiences related to the perception, understanding, and judgment of the product. These multidimensional facets of the drinking experience can be explored by measuring affective, cognitive, and sensory responses of consumers, which are shown to be stable regardless of the social context. The weight of each of these three dimensions varies according to consumers 'expertise and involvement in wine, which highlights the importance of cognitive drivers on the overall wine drinking experience. The understanding of differences in the weight of the affective, cognitive and sensory cues will be illustrated by the notion of "natural wine", where the sensory and cognitive cues show an interesting conflict.

Warell, A. (2008). Multi-modal visual experience of brand-specific automobile design. The TQM Journal, 20, 356–371.

MANAGING CHANGES IN TASTE: LESSONS FROM CHAMPAGNE IN BRITAIN 1800-1914

Dr Graham HARDING

Oxford college (UK)

This paper focuses on how taste in wine (and other foods) changes and the implications of this process for producers and merchants.

It draws primarily on the changing taste of and taste for champagne in Britain in the 19th century. Between 1850 and 1880 champagne went from a dosage level of around 20% (20 grams sugar / litre) to 0%. Champagne became the 'dinner wine of the elite – drunk with roast meat and savoury dishes. Contemporaries accepted that while most people could distinguish the taste of good champagne from that of bad, very few could distinguish very good from good. The shift to dryer champagne was driven less by an appreciation of a changing wine than by social pressure; by the wish of the elite users to differentiate themselves from newly-wealthy middle-class users.

There is plenty of evidence to support the view that the majority of people do not have the tasting skills or sensory sophistication to identify small changes in the taste of a product from one occasion to the next. What they are good at is acquiring the taste for a given substance if and when it becomes necessary.

Using primarily 19th and early 20th century cases my paper will look at how changes in taste are created and how they can be sparked by broader 'taste regimes' developing in other areas – such as the 19th century panic over product adulteration. Once sparked how are they fuelled and accelerated by social and media pressure. I will then focus on how producers might and should respond to such changes. When and how fast should they make changes to their style of wine? What are the branding implications of such changes? Changes to product formulation can happen fast; changes to brands generally cannot. Is there a broader positioning opportunity to gain competitive share? If so, the opportunity be maximised by migrating to a new brand? Or is the challenge best met by (temporary) changes to label copy?

Such challenges faced a number of 19th century producers and their successes and failures provide illuminating insights for modern producers and distributors.

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I.O.1

GROUPING VITIS VINIFERA GRAPEVINE VARIETIES BASED ON THEIR AROMATIC COMPOSITION

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Keywords: Aromatic Composition, Aroma Compounds, Climate Change, GC-MS

Context and purpose of the study

Climate change is likely to impact wine typicity across the globe, raising concerns in wine regions historically renowned for the quality of their terroir¹. Amongst several changes in viticultural practices, replacing some of the planting material (i.e. clones, rootstocks and cultivars) is thought to be one of the most promising potential levers to be used for adapting to climate change. But the change of cultivars also involves the issue of protecting the region's wine typicity. In Bordeaux (France), extensive research has been conducted on identifying meridional varieties that could be good candidates to help guard against the effects of climate change² while less research has been done concerning their impacts on Bordeaux wine typicity. Thus, the present study aims to characterize the aromatic composition of a large pool of *Vitis vinifera* cultivars through the analyses of some impacting aromatic compounds. Then, aromatic composition of traditional-Bordeaux varieties and non-Bordeaux varieties are compared.

Materials and Methods

A 2-hectares plot of 84 cultivars was planted in 2013, in the Médoc wine region (France) within the vineyards of a wine estate. Amongst this very large collection of cultivars, a pool of 25 red varieties was isolated, including traditional Bordeaux varieties and potential candidates for introduction in the Bordeaux varietal mix. Each of those varieties has been separately vinified since 2018 in 2hL stainless steel tanks, close to commercial wine production conditions. 46 major aroma compounds were then quantified in each variety for each vintage (from three to five vintages per cultivar) by gas chromatography and mass spectrometry (GC-MS). Statistical analyses, including hierarchical clustering analysis (HCA) and principal component analysis (PCA) was then performed on this unique dataset for aroma profile characterization and to discriminate and isolate varieties according to their aromatic composition.

Results

As expected, analyses resulted in a strong varietal characterization of the different wines with a significant vintage effect on some of the aroma compounds. Of the 46 aroma compounds analyzed, a select few appear to explain a large part of the Bordeaux wines aromatic composition. Clustering of cultivars was possible, and Bordeaux cultivars group well together into a unique cluster. Interestingly, a few non-traditional Bordeaux cultivars were close to some of the classical Bordeaux varieties in both the HCA and PCA analyses. These results enhanced the idea that some non-native cultivars could be introduced in the Bordeaux cultivar mix while maintaining some of the wine typicity. This methodology could help other established wine regions to identify varieties that could be potential candidates for adaptation to climate change.

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 Destrac-Irvine, A.; Van Leeuwen, K. VitAdapt, an Experimental Program to Study the Behavior of a Wide Range of Grape Varieties of Vitis Vinifera in a Context of Climate Change in the Bordeaux Vineyards, 2018. https://hal.archives-ouvertes.fr/ hal-03179912 (accessed 2023-02-13).

I.O.2 INFLUENCE OF AGROPHOTOVOLTAIC ON VINE AND MUST IN A COOL CLIMATE

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Keywords: Agrophotovoltaic, Plasmospara viticola, berry composition, water stress

The current energy crisis means that interest in agrophotovoltaics has increased significantly. The reason behind this is that the system aims to combine agricultural production with energy production. During the three-year period from 2020 to 2022, the effects of photovoltaic panels on the vine, the yield and the quality of the must were studied in Walenstadt in northern Switzerland, an area with a cool, humid climate. 65 Pinot noir vines were planted in the 160m² study area. Because of the large edge effects, only 3 repetitions with 4 vines each could be created. A significantly lower leaf infestation by Plasmopara viticola was observed among the panels in each of the three years. However, in 2021 a significantly lower grape infestation was observed with an extremely high infestation pressure. In contrast, significantly higher powdery mildew leaf infestations were observed in 2020 and 2022, with infestations well below the economic threshold. During the three-year study period, the sugar content among the panels was significantly lower at around 0.9 to 1.8 Brix, and the total acidity was significantly higher between 1 and 1.3 g/l. These results indicate an approximate 1-week delay in ripening. Growth was optically stronger underneath the panels throughout the dry years 2020 and 2022, but only in the latter was it possible to measure a significantly higher pruning weight. The significantly lower value of the relative carbon isotope composition (Δ 13C) in must sugars under the panels indicates greater water stress compared to the control, which can be attributed to reduced transpiration under the panels.

Conclusion: In the three years observed no significant negative impact of the photovoltaic panels could be determined, either on the yield or on the components of the must. The delay in ripening is usually a positive aspect, as is the reduced pressure from downy mildew. However, there is still a need for optimisation in construction so that unhindered mechanical cultivation is possible.

I.O.3

OVERHEAD SPRAY WATER TREATMENT AS A MITIGATION STRATEGY FOR REDUCING VINE STRESS AND PRESERVING GRAPE QUALITY DURING HEATWAVES

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Keywords: red-blended-wine , molecular marker , Aroma compound , Sensorial attribute

Changes in climate have been influencing the quality of wine grapes worldwide. The impact of extreme climate events over short periods is increasingly recognized as a serious risk to grape quality and yield quantity. In this study the mitigation effects of a pulsed water spray on vine canopy during heatwave events has been evaluated for maintaining vine condition during the growing season and grape quality. Vines of three varieties (Malbec, Bonarda, and Syrah) under drip irrigation in the UNCuyo experimental vineyard were treated with an overhead pulsed water spray. The spray was applied to the top of the vine canopy for 15 minutes per hour during 12 daylight hours over the course of heat waves occurring between veraison and harvest. Heatwaves were defined as days with a minimum temperature of no less than 21 °C and a maximum temperature of no less than 34 °C. Two heat waves were identified over the course of the growing season. Temperature was measured at the canopy level (CT) while a weather station provided multiple climate parameters of the vineyard (VT). Samples were collected at weekly intervals from veraison to harvest. During 5 sample dates Leaf and Stem Water Potential (LWP, SWP), Stomatal Conductance (SC), Leaf Temperature (LT), Berry Temperature (BT), Chlorophyll Content (CC), Fluorescence (FV/FM), and Performance Index (PI) were collected at several intervals during the day to evaluate physiological responses. Berries were collected at each sample date as well as at harvest. Berry weights, soluble solids content, and pH were measured. At harvest, anthocyanin profile, kg/plant, number of bunches and their average weight were also evaluated. LWP, SWP, FV/FM, PI, SC, CC, Kg/plant, and BW, were significantly higher while LT, BT, and CT were lower in treated vines as compared to the control during the second heatwave, which was longer and more intense than the first one. One week after the more severe heatwave, LWP, SWP and SC were still significantly different between treatment and control, displaying reduced physiological stress in the treated vines. No differences were identified in the sum of total anthocyanins. However, some individual anthocyanins were higher in treated vines. These results suggest that vines with the overhead water treatment during heat waves had reduced physiological stress and increased yield. As a consequence, this practice could be used as a mitigating tool to reduce the impact of heat waves.

ROOTSTOCK MEDIATED RESPONSES OF GRAPEVINE (*VITIS VINIFERA L.*) ME-TABOLISM AND PHYSIOLOGY TO COMBINED WATER DEFICIT AND SALINITY STRESS IN SYRAH GRAFTS

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I.O.4

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Keywords: combined stress, grafts, physiology, metabolite

Water deficit and salinity are increasingly affecting the viticulture and wine industry. These two stresses are intimately related; understanding the physiological and metabolic responses of grapevines to water deficit, salinity and combined stress is critical for developing strategies to mitigate the negative impacts of these stresses on wine grape production. These strategies can include selecting more tolerant grapevine cultivars and graft combinations, improving irrigation management, and using soil amendments to reduce the effects of salinity. For this purpose, understanding the response of grapevine metabolism to altered water balance and salinity is of pivotal importance. Hence, we used cv. Syrah grafted on rootstocks 1103 Paulsen and SO4, under a set of combinations of salinity (0.5 and 2.5 dS m-1) and differential irrigation levels (66%, 100% and 133% of the local recommended irrigation amount) in an experimental vineyard located on Sede Boger, Israel at 30051'22.37" N and 34046'52.98" E with an elevation of 480 m.a.s.l. SO4 grafts generally produced a higher yield than 1103Paulsen grafts, while accumulating more Cl⁻ ions in wine and leaves. These results may suggest different salt exclusion potentials. Spectrophotometric readings showed that high salinity with deficit irrigation increased tannins and reduced carotenoid content in the berries. In addition, a lower fluorescence and photosystem efficiency under stress were recorded in 1103 Paulsen vines. GC-MS-based profiling of central metabolism showed the accumulation of major sugars and amino acids. For example, under salinity stress, proline and alanine relative content increased while lysine, valine, and leucine content decreased irrespectively of the rootstock. Grafts of 1103 Paulsen showed greater accumulation of N-compounds being pyroglutamate, leucine, valine, ethanolamine, sugars including xylose and trehalose, and few other metabolites (cinnamate, lactate, and galactarate) when compared to SO4 grafts. Altogether, our results show multi-level differences in Syrah metabolism and physiology due to the rootstock mediation of salinity and water deficit combined stress.

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I.O.5

SIP AND SAVE THE PLANET: A SENSORY AND CONSUMER EXPLORATION OF AUSTRALIAN WINES MADE FROM POTENTIALLY DROUGHT-TOLERANT WHITE WINE GRAPES

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Keywords: Rate-All-That-Apply, drought tolerant, sensory profiles, consumer acceptance

In order to attenuate the effects of climate change on the ability to cultivate quality wine grape vines in Australia, it is essential to adapt to the projected less favourable Australian climate scenarios. One response may be to convert a portion of the current grapevine plantings to those varieties that demand less water and can tolerate increased heat. This investigation aimed to (i) generate sensory profiles and (ii) obtain knowledge about Australian wine consumers' preferences and opinions of Australian wines made from potentially drought tolerant, white wine grape varieties not traditionally cultivated in Australia. A Rate-All-That-Apply (RATA) sensory panel (n = 49) generated sensory profiles of 44 commercial white wines made from 7 different white grape varieties (Arinto, Fiano, Garganega, Greco, Verdejo, Verdelho and Vermentino), plus two benchmark examples each of an Australian Riesling, Pinot Gris and Chardonnay wine. All wines underwent basic chemical analyses and a subset of 10 wines was subjected to preference trials with Australian white wine consumers (n = 102) who consumed white wine at least monthly. Consumers liked 9 of the 10 wines, scoring them greater than 5 on a 9-point hedonic scale. Verdejo and Garganega, the second and third most liked wines, could potentially be offered as an alternative to Pinot Gris, due to their similar sensory attributes. Arinto and Greco could become alternative wines to Chardonnay; and Fiano to Riesling. These findings provide more accurate information about the potential performance of these new wines in the Australian market and suggest wine producers could provide alternative wine styles which meet the taste specifications of this competitive market, yet promote a more sustainable grape and wine industry as it faces the impact of climate change. The adoption of drought tolerant varieties will potentially reduce the amount of water needed for irrigation, increase yield and income. Moreover, sustainable Australian wine products may introduce a point of difference in the current global market.

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I.O.6

PROBING GRAPEVINE-*BOTRYTIS CINEREA* INTERACTION THROUGH MASS SPECTROMETRY IMAGING

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Keywords: Mass Spectrometry, Imaging Metabolomics, Plant–Pathogen Interaction, Stilbene phytoalexins

Plants in their natural environment are in continuous interaction with large numbers of potentially pathogenic and beneficial microorganisms. Depending on the microbe, plants have evolved a variety of resistance mechanisms that can be constitutively expressed or induced. Phytoalexins, which are biocidal compounds of low to medium molecular weight synthesized by and accumulated in plants as a response to stress, take part in this intricate defense system.^{1,2}

One of the limitations of our knowledge of phytoalexins is the difficulty of analyzing their spatial responsiveness occurring during plant- pathogen interactions under natural conditions.

Mass spectrometry imaging is an innovative analytical technique for the spatial mapping of molecules within a sample. It has the ability to provide valuable insights into the molecular processes underlying the interaction between a plant and a pathogen. Despite its advantages, to date, the studies of the application of MALDI-MSI to plants is not extensive, even more the ones investigating grapevine compounds.³⁻⁵

In a first part, the presentation will focus on the principle and characteristic data provided by this methodology involving a localized ion source and a high-resolution mass spectrometer. The following part will be dedicated to our work on the application of this methodology to investigate phytoalexin accumulation and distribution in leaf tissues of *Vitis vinifera* cv Chardonnay, infected with *Botrytis cinerea*, the causal agent for gray mold. We specifically investigate the accumulation and spatial distribution of resveratrol and its derivative oligomers through a time course infection using matrix-assisted laser desorption ionization – mass spectrometry imaging (MALDI-MSI). Our results show that production of stilbene phytoalexins is rapidely activated by the presence of the pathogen, with a high molecular diversity as evidenced by the detection of various oligomeric forms. Moreover, the specific behavior of these compounds towards the propagation of the infection by the pathogen is clearly highlighted at the micron scale.

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Grape and wine microorganisms : diversity and adaptation

II.0.1

ACIDIC AND DEMALIC *SACCHAROMYCES CEREVISIAE* STRAINS FOR MANAGING PROBLEMS OF ACIDITY DURING THE ALCOHOLIC FERMENTATION

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Keywords: Acidity perception, Malic acid, Wine yeast

In a recent study several genes controlling the acidification properties of the wine yeast *Saccharomyces cerevisiae* have been identified by a QTL approach [1]. Many of these genes showed allelic variations that affect the metabolism of malic acid and the pH homeostasis during the alcoholic fermentation. Such alleles have been used for driving genetic selection of new *S. cerevisiae* starters that may conversely acidify or deacidify the wine by producing or consuming large amount of malic acid [2]. This particular feature drastically modulates the final pH of wine with difference of 0.5 units between the two groups. Such extreme strains called ACIDIC or DEMALIC have been compared in several grape juices in order to evaluate their technological and sensorial impact on the resulting wines. Beside routine phenotypic characterization (fermentation kinetics and basic enological analyses), targeted NMR metabolomic as well as LC-MS non targeted metabolomics were used for characterizing such group of strains. These ACIDIC or DEMALIC strains definitively challenge a wide range of industrial starters and provide new tools for managing the rising problem of acidity in the context of global warming change.

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II.O.2

CHANGES IN METABOLIC FLUXES UNDER LOW PH GROWTH CONDITIONS: CAN THE SLOWDOWN OF CITRATE CONSUMPTION IMPROVE *OENOCOCCUS OENI* ACID-TOLERANCE?

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Keywords: Oenococcus oeni, Citrate, Metabolic fluxes, Acid-tolerance

Oenococcus oeni is the main Lactic Acid Bacteria responsible for malolactic fermentation, converting malic acid into lactic acid and carbon dioxide in wines. Following the alcoholic fermentation, this second fermentation ensures a deacidification and remains essential for the release of aromatic notes and the improvement of microbial stability in many wines. Nevertheless, wine is a harsh environment for microbial growth, especially because of its low pH (between 2.9 and 3.6 depending on the type of wine) and nutrient deficiency. In order to maintain homeostasis and ensure viability, *O. oeni* possesses different cellular mechanisms including organic acid metabolisms which represent also the major pathway to synthetize energy in wine. Indeed, uptake and consumption of malate and citrate by this bacteria enables to activate a proton motive force (PMF) hence maintaining an intracellular pH by proton consumption¹².

Citrate is found in wine at small concentrations (0.13 to 0.90 g/L). It can be metabolized by *O. oeni* into acetate, pyruvate and then aromatic compounds such as diacetyl, acetoin and 2,3-butanediol. The ability of citrate metabolism to activate the PMF could play a central role in the acid-tolerance of this bacterium. Nevertheless, a previous study has described an inhibition of *O. oeni* growth at low pH in presence of high amounts of citrate3. This toxic effect could come from the synthesis of one of the citrate metabolites as acetate.

In order to understand how citrate metabolism can be linked to acid tolerance of this bacterium, consumption of citrate was investigated in a great diversity of *O. oeni* strains. In addition, malate and sugar consumptions were also followed, as they can be impacted by citrate metabolism. These experiments enabled to draw metabolic fluxes in *O. oeni* according to the pH of the medium. In most cases, malate is first metabolized, then citrate and sugars, sequentially, proving that the priority is given to organic acid consumption at the expense of sugars in this bacterium. However, this experiment revealed different citrate consumption profiles which may be correlated to a greater or lesser acid tolerance according to the strain. Furthermore, a genomic comparison demonstrated the presence of mutations in the citrate operon of acid-tolerant industrial strains. Hence, acid tolerance could be linked to a change in the rate of citrate consumption in *O. oeni*.

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II.O.3 FUNGAL DIVERSITY AND DYNAMICS IN CHAMPAGNE VINEYARDS: FROM VINE TO WINE

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Keywords: Mycobiota, Diversity, Metagenetics, Co-occurrence networks

Champagne is a well-known wine region in Northern France with distinct terroirs and three main grape varieties. As for any vineyard, wine quality is highly linked to the microbiological characteristics of the raw materials. However, Champagne grape microbiota, especially its fungal component, has yet to be fully characterized. Our study focused on describing this mycobiota, from vine to small scale model wine, for the two main Champagne grape varieties, Pinot Noir and Meunier, using complementary cultural and omics approaches.

Changes in microbial diversity and dynamics, especially mycobiota colonizing grapes, was evaluated at 5 berry ripening and vinification stages in 31 vineyards for two harvests. Grapes were collected at fruit set, veraison and harvest and micro-pressing (40 kg) and micro-vinifications (5L) of grape musts from each vineyard were performed.

For both harvests, fungal counts increased during berry ripening (although lower counts were observed in 2022 versus 2021), remained relatively high in musts before yeasts solely dominated in laboratory wines. Clear shifts in mycobiota diversity were observed from vine to laboratory wine for both years. Berries were dominated by yeasts (including filamentous *Aureobasidium* spp.) regardless of ripening stage with an increase in mold diversity during ripening. *Cladosporium* (7 species identified) were predominant in unmature berries before other molds colonized grapes, especially *Penicillium* (9 species identified) and *Botrytis cinerea*, from veraison onwards. Metagenetic analyses (equivalent to 450 samples in 2021) were well correlated with culture-dependent data. This approach confirmed the predominance of yeasts (*Aureobasidium* and *Vishniacozyma*) at both fruit set and veraison, in addition to *Cladosporium*, although higher fungal diversity and variability between musts was observed.

This unprecedent and thorough description of mycobiota from unmature berries to microvinified wines will clearly contribute to a better understanding of the fungal determinants of specific traits linked to wine quality or defects. Microbial co-occurrence networks and global analyses with the generated climatic data and vineyard practices is currently being explored to understand species interactions and identify factors shaping mycobiota composition.

II.O.4

LARGE-SCALE PHENOTYPIC SCREENING OF THE SPOILAGE YEAST *BRET-TANOMYCES BRUXELLENSIS*: UNTANGLING PATTERNS OF ADAPTATION AND SELECTION, AND CONSEQUENCES FOR INNOVATIVE WINE TREATMENTS

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Keywords: comparative phenotyping, local adaptation, UVC, Pulsed light

Brettanomyces bruxellensis is considered as the main spoilage yeast in oenology. Its presence in red wine leads to off-flavour due to the production of volatile phenols such as 4-vinylphenol, 4-vinylguaiacol, 4-ethylphenol and 4-ethylguaiacol, whose aromatic notes are unpleasant (e.g. animal, leather, horse or pharmaceutical). Beside wine, B. bruxellensis is commonly isolated from beer, kombucha and bioethanol production, where its role can be described as negative or positive. Recent genomic studies unveiled the existence of various populations. These genetic groups differ from each other by their ploidy level (diploid or triploid), their hybridization status (auto or- allo-triploid) and their ecological fermentation niches (wine, beer, tequila/bioethanol, etc.). While the genomic landscape of B. bruxellensis is nowadays clearer, its phenotypic diversity is still insufficiently assessed in the light of its genetic diversity. In this work, on one hand, we designed an experiment where 151 B. bruxellensis strains representative of the genetic diversity of the species were phenotypically characterized in five natural beverages (grape must, wine, wort, beer, kombucha wort). Various phenotypic traits were monitored: parameters of growth and fermentation ability, metabolites of technological interest... Signatures of local adaptation were investigated and showed that at least one allotriploid population of B. bruxellensis is specifically adapted to wine environment. Moreover, such large screening allowed the identification of ancestral traits like maltose and maltotriose consumption or nitrate metabolization that were randomly lost in specific populations, an evolutionary phenomenon called relaxed selection. On a second hand, two innovative control methods, continuous UV-C light and pulsed light, were tested on a large collection of *B. bruxellensis* (>100 strains) and other wine yeast species (14 species). These two stabilization treatments were deemed as particularly efficient on wine yeast spoilers (B. bruxellensis including) using i - a drop-platted system to screen various strains and conditions, and ii- lab-made reactors to stabilize several litters of red wines. Altogether, our results contribute to a deeper understanding of the wine spoiler B. bruxellensis both at the fundamental and applied levels.

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II.O.5

METABOLIC INTERACTIONS OF *SACCHAROMYCES CEREVISIAE* COCULTURES: A WAY TO EXTEND THE AROMA DIVERSITY OF CHARDONNAY WINE

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Keywords: fermentation, interactions, Saccharomyces cervevisiae, metabolomic

Yeast co-inoculations in winemaking have been investigated in various applications, but most often in the context of modulating the aromatic profiles of wines. Our study aimed to characterize S. cerevisiae interactions and their impact on wine by taking an integrative approach. Three cocultures and corresponding pure cultures of S. cerevisiae were characterized according to their fermentative capacities, the chemical composition and aromatic profile of the associated Chardonnay wines. The various strains studied within the cocultures showed different behaviors regarding their development. More than half of the 67 volatile compounds quantified were modulated by interactions, including 18 relevant wine aroma compounds. The main families affected were higher alcohols and their associated esters, vinyl phenols, and fatty acids. Coculture makes it possible to obtain new aromatic expressions that do not exist in the original pure cultures attributed to yeast interactions. The sensory profile of the wines related to the cocultures differed from the wines associated with the pure cultures. However, they also differed from the blends (50/50 v/v) of post AF wines from pure cultures. Based on the exometabolome, this was confirmed. The cocultures were revealed as not being simple additions of two wines represented by blend, thereby indicating complex interactions. High resolution mass spectrometry allowed to highlight thousands of cocultures biomarkers. Most of these biomarkers belonged to metabolic pathways involved in nitrogen metabolism. The latter is therefore a marker of changes associated with interactions between two strains of S. cerevisiae. Despite of preserved fermentative properties, the described interactions induced a modification of the chemical composition and sensory profile of the wines from the cocultures. A comprehensive approach by combining different techniques is essential to understand yeast interactions and describe the consequences on wine.

II.O.6 YEAST-PRODUCED VOLATILES IN GRAPE BASED SYSTEM MODEL ACTING AS ANTIFUNGAL BIOAGENTS AGAINST PHYTOPATHOGEN *BOTRYTIS CINEREA*

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Keywords: volatile organic compounds, HS-SPME-GC-MS, biocontrol, Botrytis cinerea

Botrytis cinerea Pers., the causal agent of grey mould disease, is responsible for substantial economic losses, as it causes reduction of grape and wine quality and quantity. Exploitation of antagonistic yeasts is a promising strategy for controlling grey mould incidence and limiting the usage of synthetic fungicides. In our previous studies, 119 different indigenous yeasts were screened for putative multidimensional modes of action against filamentous fungus *B. cinerea* [1]. The most promissing biocontrol yeast was *Pichia guilliermondii* ZIM624, which exhibited several anatagonistic traits (production of cell wall degrading enzymes, chitinase and β -1,3-glucanase; demonstration of in vitro inhibitory effect on *B. cinerea* mycelia radial growth; production of antifungal volatiles, assimilation of a broad diversity of carbon sources, contributing to its competitivnes in inhabiting grapes in nature). In addition, P. guilliermondii ZIM 624 possessed interesting enological traits, did not produce off-flavor related H₂S and appeared as β -lyase and β -glucosidase producer.

Accordingly, the aim of this researsch was to study the antifungal mechanisms by assessing the volatiles produced by P. guilliermondii ZIM624. Namely, a study was conducted to identify volatile organic compounds (4 higher alcohols, 6 volatile phenols, 23 esters and 27 terpenes) produced by antagonistic Pichia guilliermondii strain ZIM624 and to determine the efficacy of the chosen volatiles of P. guilliermondii in suppression of *B. cinerea* growth and control of *Botrytis* fruit rot of grape berries. Thereby, a comprehensive assessment of produced volatiles in the process of wine production was achieved using two validated analytical methods (one for terpenes and one for the rest of mentioned volatiles) comprised of automated headspace (HS) solid-phase microextraction (SPME) and gas chromatography coupled with mass spectrometric detection (GC-MS). Both methods were developed based on already published method for determionation of volatiles in wine samples [2]. Among identified volatiles, 13 yeast-produced volatiles were selected and their antifungal activity was tested against B. cinerea in the fumigation bioassay. Terpenes citronellol, geraniol, nerol, α -terpineol and linalool were the most effective against *B. cinerea* mycelium growth with the EC_{50} beetwen 6,6 to 32,8 μ L/L. 4-Vinyl phenol and isoamyl acetate also effectively inhibited mycelial growth of *B. cinerea*, EC_{50} being 48,6 and 63,3 μ L/L, respectively, followed by eucalyptol (EC₅₀ 201,6 μ L/L) and ethyl butyrate (EC₅₀ 238,4 μ L/L). 4-Vinyl guaiacol did not show any inhibitory effect, while the remaining tested compounds showed inhibition against B. cinerea growth, however we were not able to determine EC_{50} with the selected concentration ranges. Additionaly, exposure of B. cinerea-infected grape berries to the volatiles from P. guilliermondii cultures also lowered the number of infected grape berries, when applied to in vivo assay.

Herein presented novel research approach strongly suggests that yeast produced volatiles such as terpenes, volatile phenols and esters are one of the possible mechanisms for controlling *Botrytis* rot of fruit and promising biofumigants.

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Wine chemistry, wine components with physiological effects

III.O.1

'TROPICAL' POLYFUNCTIONAL THIOLS AND THEIR ROLE IN AUSTRALIAN RED WINES

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Keywords: red wine, 'tropical' aroma, thiols, sensory

Following anecdotal evidence of unwanted 'tropical' character in red wines resulting from vineyard interventions and a subsequent yeast trial observing higher 'red fruit' character correlated with higher thiol concentrations, the role of polyfunctional thiols in commercial Australian red wines was investigated.

First, trials into the known tropical thiol modulation technique of foliar applications of sulfur and urea were conducted in parallel on Chardonnay and Shiraz.¹ The Chardonnay wines showed expected results with elevated concentrations of 3-sulfanylhexanol (3-SH) and 3-sulfanylhexyl acetate (3-SHA), whereas the Shiraz wines lacked 3-SHA. Furthermore, the Shiraz wines were described as 'drain' (known as 'reductive' aroma character) during sensory evaluation although they did not contain thiols traditionally associated with 'reductive' thiols (H₂S, methanethiol etc.).

Secondly, a survey of over 100 commercial Australian red wines across 10 different varieties supported the outcome of the foliar application trial.² While all red wines contained 3–SH above the sensory detection threshold (60 ng/L, aqueous ethanol), no wines were observed to contain detectable concentrations of 3–SHA. As such, the acetylation of 3–SH to 3–SHA in red wine fermentations appears to be extremely limited.

Lastly, 3-SH and/or 3-SHA were spiked into four different varieties of red wine to understand the impact on sensory attributes.² Traditionally lighter varieties (Pinot Noir and Grenache) had increased 'red fruit' and 'lolly' ratings at low concentrations of 3-SH and 3-SHA but changed to 'tropical' at higher concentrations. For Cabernet Sauvignon, 3-SH and 3-SHA additions resulted in increases to 'blackcurrant' and 'tropical' attributes, whereas Shiraz additions of 3-SH resulted in 'sweaty' and 'tropical' descriptors.

The 'tropical' thiol, 3–SH, was ubiquitous in Australian red wines although the acetylation to 3–SHA was not commonly observed. The impact of these thiols in red wines differed by variety, and their presence was increased by vineyard foliar application treatments. As such, vineyard management practises might hold the key to avoiding undesirable expressions of 'tropical' characters in red wine.

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III.0.2 ABOUT THE ROLE PLAYED BY THE DIFFERENT POLYPHENOLS ON OXYGEN CONSUMPTION AND ON THE ACCUMULATION OF ACETALDEHYDE AND STRECKER ALDEHYDES DURING WINE OXIDATION

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Keywords: oxidation, acetaldehyde, Strecker aldehydes, polyphenols

In a previous work¹, it was suggested that the different contents in delphinidin and catechin of the grapes were determinant on the O₂ consumption and Strecker aldehyde (SAs) accumulation rates. Higher delphinidin seemed to be related to a faster O₂ consumption and a smaller SAs accumulation rate, and the opposite was observed regarding catechin.

In the present paper, these observations were fully corroborated by adding synthetic delphinidin to a wine model containing polyphenolic fractions (PFs) extracted from garnacha and synthetic catechin to a wine model containing PF extracted from tempranillo: The delphinin-containing garnacha model consumed O_2 significantly faster and accumulated significantly smaller amounts of SAs than the original garnacha model, and the catechin-containing tempranillo model, consumed O_2 significantly slower and accumulated significantly model.

The work was further expanded by studying the effects of 8 individual polyphenols (delphinidin, malvidin, caffeic and coumaric acids, catechin, epigallocatechin, quercetin and myricetin) in normalized wine models subjected to a forced oxidation procedure on the O₂ consumption rates (OCRs) and acetaldehyde and SAs accumulation rates.

Most surprisingly, all polyphenols but anthocyanins, initially slowed down OCRs observed in the plain wine model, which contained cation metals, hydrogen sulfide, cysteine and glutathione in reduced forms. Leaving aside anthocyanins, only B-ring tri-hydroxylated polyphenols were able to consume all O₂ supplied. Moreover, the polyphenol determined also the fraction of ethanol oxidized to acetaldehyde. Catechin is the strongest inducer of ethanol oxidation, while delphinidin and epigallocatechin were the weakest.

Regarding SAs, the surprising finding is that, considering formation per O₂ consumed, these are most efficiently formed in the basic wine model without polyphenols. Efficiency comes to a minimum with delphinidin, followed by coumaric acid and malvidin.

These set of results introduces a completely new perspective to wine oxidation kinetics and to the accumulation of aldehydes.

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III.O.3

AGING PATTERNS OF VARIETAL VOLATILE PROFILES OF WHITE WINES: A CASE STUDY ON 18 ITALIAN VARIETAL WHITE WINES

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Keywords: Volatile compounds, white wine, Aging pattern, Varietal typicality

During wine aging many compositional changes take place. In particular, aroma undergoes dramatic modifications through a wide range of reactions that to date are only partly understood. Italy owns one of the largest ampelographic heritages worldwide, with over three-hundred different varieties. Among these, many white grapes are employed for the production of dry still white wines. Some of these wines are consumed young while others are more prone to aging. For many of these wines, the aging patterns related to volatile composition are still unknown.

An extensive survey was conducted on 18 monovarietal Italian still white wines with the aim of elucidating the behaviours of different volatile compounds during aging. In particular, a range of volatile compounds including terpenes, norisoprenoids, benzenoids and volatile sulfur compounds was investigated. A total of 108 different samples were analysed, including Nosiola, Vermentino, Müller-Thurgau, Greco di Tufo, Garganega, Lugana, Erbaluce di Caluso, Pinot Grigio, Cortese, Arneis, Albana, Pallagrello, Falanghina, Fiano, Ribolla Gialla, Vernaccia, Gewürztraminer, Verdicchio. All wines were adjusted to 30 mg/L of free SO₂ and submitted to an accelerated aging protocol involving storage for 30 days 10 °C, 40 °C and 60°C in oxygen-free environment. Volatile compounds were analysed by means a combination of analytical methods based on SPME-GC-MS.

During aging some common trends were observed, among which a decrease in linear terpenes and an increase in bicyclic terpenes, non-megastigmane norisoprenoids and volatile sulfur compounds. From a quantitative point of view, the extent of these transformations varied significantly according to wine type. Besides Gewürztraminer, which was generally rich in terpenes, other wine types such as Vermentino Verdicchio and Lugana showed peculiar terpene patterns, for example accumulation of above-threshold levels of the bicyclic terpene 1,4-cineole. Regarding non-megastigmane norisoprenoids, Falanghina and Vermentino were found to accumulate high levels of TDN and vitispirane, which was not observed in other wines. Greco accumulated during aging the highest amount of DMS, showing an average content above the odor threshold. Müller-Thurgau, Nosiola and Vermentino also showed concentrations of DMS above the odor threshold after aging. In addition, the latter varieties also showed high accumulation of methanethiol.

As most of these patterns were not seen in young wines, this work highlights the important contribution of aging to the expression of aroma characters that are specific to the identity of individual varieties or wine types.

III.0.4 ASSESSING THE ROLE OF 27 KNOWN BITTER COMPOUNDS IN COMMERCIAL WHITE WINES COMBINING LC-MS QUANTIFICATION AND SENSORY ANALYSIS

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Keywords: LC-MS quantification, sensory analysis, bitterness, wine

The balance between the different flavours of a wine largely determines its perception and appreciation by the consumers. In white wines, sweetness and sourness are usually the two poles balancing the taste properties. The bitter flavour, on the other hand, is frequently associated with a loss of equilibrium and all white wines (dry and sweet, young and aged) are affected.

Several bitter compounds are already well-described in wines. Some are linked to microorganisms as acrolein (Bauer et *al.*, 2010) or oak wood, for example lyoniresinol (Cretin et *al.*, 2015), while others come directly from grapes: mostly phenolic (Hufnagel and Hofmann, 2008) and nitrogen compounds (Rou-dot-Algaron, 1996). Furthermore, the enhancing role played by ethanol has also been well established (Cretin et *al.*, 2018). The present study aims to determine the influence of twenty-seven known bitter compounds on the taste of various commercial white wines.

Thirty wines have been selected and submitted to sensory analysis by a trained panel. The various intensities of sourness, sweetness and bitterness have been determined for each wine. Jointly, five quantification methods have been developed and validated using liquid chromatography coupled with high resolution mass spectrometry (UHPLC-Exactive, Orbitrap analyzer) in order to determine the amount of the selected bitter compounds.

Potential correlations between the described tastes of the wines and concentrations of bitter molecules have been assessed. For the most relevant compounds, detection thresholds have been updated using the same trained panel, enabling a better understanding of the impact of various compounds.

This study enlightens the role of already known bitter compounds in bitter wine. It is also leading the way to further research as some wine's taste remain unexplained by the selected compounds, thus confirming the potential presence of still unknown bitter compounds.

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III.O.5

CONTRIBUTION OF VOLATILE THIOLS TO THE AROMA OF RIESLING WINES FROM THREE REGIONS IN GERMANY AND FRANCE (RHEINGAU, MOSEL, AND ALSACE)

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Keywords: Riesling wine aroma, Volatile thiols, Identification, Quantitation

Riesling wines are appreciated for their diverse aromas, ranging from the fruity fresh characters in young vintages to the fragrant empyreumatic notes developed with aging. Wine tasters often refer to Riesling wines as prime examples showcasing terroir, with their typical aroma profiles reflecting the geographical provenance of the wine. However, the molecular basis of the distinctive aromas of these varietal wines from major Riesling producing regions in Europe have not been fully elucidated. In this study, new lights were shed on the chemical characterization and the sensory contribution of volatile thiols to Riesling wines from Rheingau, Mosel, and Alsace. First, Riesling wines (n = 46) from the three regions were collected and assessed for their aroma typicality by an expert panel. Based on sensory assessment, selected wines were examined for their global aroma profile by sensory guided odorant screening techniques (preparative high pressure liquid chromatography; gas chromatography-mass spectrometry/olfactometry (GC-MS/O); sensory evaluation), and several odorous zones (OZs) of interest resembling the original olfactory notes (citrus, tropical fruits etc.) of the initial wines were noted. The aroma descriptors, linear retention index, and mass spectra of the suspected chromatography peaks and their accompanying OZs of interest revealed the presence and importance of volatile thiols in Riesling wines analysed. Hence, selective silver ion solid phase extraction and multidimensional GC-MS/O were applied for further characterization of targeted thiol-relevant OZs, allowing tentative identification of unknown thiols, with one new mercapto monoterpenoid confirmed by orthogonal approach. Following the sensory guided qualitative screening efforts, a new and highly sensitive quantitation method based on chemical derivatization and liquid chromatography quadrupole Orbitrap high-resolution MS was developed for the analysis of a substantial number of known and newly identified volatile thiols in the wine set. Quantitative results confirmed the relevance of 13 odorous thiols in Riesling, with several of them presented at concentrations well over their perception thresholds, as 3-sulfanylhexanol for instance. Thus, the combination of the chemical analysis of thiols and the sensory evaluation made it possible to draw up regional profiles according to the origin of the wines.

III.O.6 EFFECTIVENESS OF APPLIED MATERIALS IN REDUCING THE ABSORPTION OF SMOKE MARKER COMPOUNDS IN A SIMULATED WILDFIRE SCENARIO

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Keywords: Smoke taint, Barrier spray, Volatile phenols, Glycosylation

Smoke taint (ST) is a grape-wine off-flavour that may occur when grapes absorb volatile phenols (VPs) originating from wildfire smoke (1). ST is associated with the negative sensory attributes such as smoky and ashy notes. VPs are glycosylated in the plant and thus present in both free and bound forms (2; 3). Wildfire smoke has resulted in a decline in grape and wine quality and financial losses which has become a prominent issue for the global wine industry. This fact has highlighted the need to develop mitigation strategies to manage the impact of smoke exposure on grapes and the resulting wines (4). Currently there are no recommended mitigation action for grape smoke exposure.

The objective of this experiment was to compare the relative effectiveness of applying different potential protective barrier sprays to grapes in a simulated wildfire scenario. Twelve different material combinations were applied close to harvest as potential barrier sprays on three bunches each using four Cabernet Sauvignon vines. The vines were exposed to smoke for two hours three days after the barrier application. Control samples prior to smoke exposure was taken and smoked control (no barrier applied) grapes were sampled at different time points after exposure. For the smoke application a rectangular tent structure was built. The smoke was generated by a wood pellet smoker. Air samples were taken, and atmospheric parameters were monitored during the experiment. Grapes were analysed for free and acid-labile forms of VPs using a GC-MS and for glycosylated forms using UHPLC-qTOF MS.

Results show the presence of smoke and smoke derived compounds, however there was a strong stratification in the distribution of VPs within the structure. The heterogeneous distribution of smoke is reflected in the different concentrations of uptake of VPs in the grapes. Results from non-treated grapes indicate that glycosylation of the free volatile phenols takes place within hours, with significant increases in almost all glycosylated compounds. However, variation in smoke exposure will affect the glycosylation kinetics of VPs. The study indicated that some sprays exhibited some efficacy in reducing VPs absorption under these conditions. However, other treatments seemed to exacerbate the adsorption of VPs in grapes. In a next step, these barrier sprays will be studied further under field conditions. Acknowledgements: This work has been funded by the USDA-ARS.

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III.O.7

REVEALING THE ORIGIN OF BORDEAUX WINES WITH RAW 1D-CHROMATO-GRAMS

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Keywords: Machine learning, Wine composition, Sensorial classification, Terroir

Understanding the composition of wine and how it is influenced by climate or wine-making practices is a challenging issue. Two approaches are typically used to explore this issue. The first approach uses chemical fingerprints, which require advanced tools such as high-resolution mass spectrometry and multidimensional chromatography. The second approach is the targeted method, which relies on the widely available 1-D GC/MS, but involves integrating the areas under a few peaks which ends up using only a small fraction of the chromatogram.

Here, we employ state-of-the-art machine learning methods to optimize the analysis of 1-D GC/MS chromatograms. Specifically, we aim to determine whether these chromatograms contain valuable information beyond the manually extracted peaks typically utilized in the targeted approach.

To explore those questions, we analyzed 4 different types of 1–D raw chromatograms (3 SIM and 1 fullscan) of 80 wines (12 vintages from 7 estates of the Bordeaux area. We first applied nonlinear dimensionality reduction techniques (T-SNE and UMAP) to the chromatograms to obtain 2D maps. In the resulting maps, wines of the same estates across multiple vintages tended to form clear clusters, whose spatial distribution reflected the geography of the Bordeaux wine region. This indicated that, for this particular set of wine, the raw chromatograms are highly informative about terroir and wine identity.

Next, we applied cross-validated classifiers to the raw chromatograms and found that we could recover perfectly well estates identity independent of vintage. By contrast, performance on vintage classifica-tion was much lower with a maximum performance of 50% correct.

Crucially, we found that the entire chromatogram is informative with respect to both of these variables. Thus, the extraction of specific peaks of the chromatogram to quantify the concentration of 32 known chemical compounds--discarding the rest of the chromatograms--led to worse classification performance, suggesting that estate identity is distributed over a large chemical spectrum, including many molecules that have yet to be identified.

In addition, the GC raw data can be used to predict the ratings of a professional wine critic (Robert Parker) above chance, thus suggesting that GC might also contain information about the organoleptic properties of wine.

Overall, this study demonstrates the strong potential of raw chromatogram analysis for wine characterization and identification.

SENSORY PROFILES AND EUROPEAN CONSUMER PREFERENCE RELATED TO AROMA AND PHENOLIC COMPOSITION OF WINES MADE FROM FUNGUS RESISTANT GRAPE VARIETIES (PIWI)

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III.0.8

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Keywords: fungus resistant grape varieties, consumer preference, aroma compounds, polyphenols

Planting grape varieties with several resistance loci towards powdery and downy mildew reduces the use of fungicides significantly. These fungus resistant or PIWI varieties (acronym of German Pilzwiderstandsfähig) contribute significantly to the 50% pesticide reduction goal, set by the European Green Deal for 2030. However, wine growers hesitate to plant PIWIs as they lack experience in vinification and are uncertain, how consumer accept and buy wines from these yet mostly unknown varieties.

Grapes from four white and three red PIWI varieties were vinified in three vintages to obtain four different white and red wine styles, respectively plus one rosé. PIWI varieties as well as four reference *Vitis vinifera* varieties were planted in the same site. For each vintage, more than 70 winemakers compared the different PIWI styles with the *Vitis vinifera* reference wines. 70% of the comparisons yielded no significant differences, in 20% PIWI wines were superior and in 10% the reference wines. Regressing intensity ratings obtained by descriptive analysis with hedonic ratings from German, French, Italian, Danish and Dutch consumers, we could deduce drivers of liking in respect to the different cultural back ground. All consumers disliked sour, astringent and green expressions in wines while fruit and colour remained low. Floral and yellow fruits were preferred by French and German consumers, Danish liked thiol-derived flavours.

To unravel the molecular base aroma compounds were analysed non-targeted as well as targeted for monoterpenes, C_{13} -norisoprenoids and polyfunctional thiols applying SIDA-GC-MS or LC-MS. Analysis of phenolic compounds was done by indirect methods such as Folin-C or Harbertson-Adams-Assay as well as targeted analysis by LC-MS or LC-DAD. Comparing Muscaris (PIWI) versus Muskateller Muscaris wines were richer in cis-rose oxide, while linalool and α -terpineol were higher in Muskateller wines. So far, no specific off-flavour could be detected in wines from PIWI varieties as it was the case for old hybrid varieties. Although fungus resistance may be related to grape skin polyphenols, in general neither white nor red PIWI wines revealed higher polyphenol concentrations, except for Muscaris and Cabernet Cortis. A fact which needs to be addressed in winemaking.

In conclusion, applying targeted winemaking allows to produce PIWI wines, which meets the expectations of European consumers in sensory terms, but also in respect to improved sustainability.

III.O.9

THE ROLE OF CELL WALL POLYSACCHARIDES IN THE EXTRACTION OF ANTHOCYANINS AND TANNINS: RESULTS, PERSPECTIVES OF A MORE POSITIVE CONTRIBUTION

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Keywords: Yeast, New Zealand Pinot noir, Polysaccharides, Chemical profile

The composition of grape berry cell walls was studied on two grape varieties, two years and two maturation levels at the same time as the extraction of anthocyanins and tannins. The chemical composition of skins, seeds, and pulps, focused on polyphenols and polysaccharides, was compared to the chemical composition in polyphenols after extraction from the skins in model solutions or after wine making of the berries. Polyphenols were mainly characterized by UPLC-MS and HPLC-SEC. Polysaccharides were characterized by analysis of the neutral sugar compositions, and also by the CoMPP (comprehensive micropolymer profiling) analysis, a new method which targets the functional groups of cell wall polysaccharides.

The extractions rates showed huge differences between the non acylated and the para-coumaroylated anthocyanins. The former were much easier to extract than the latter. Particularily in model solutions, the extraction of p-coumaroylated anthocyanins was almost negligible. The extraction rate of tannins was between those of the two anthocyanin families. Moreover, in wines as in model solutions, the final concentrations in tannins, non acylated and p-coumaroylated anthocyanins showed correlations that did not exist in the berry compositions, suggesting a similar mechanism of extraction associating those three families of polyphenols. According to the CoMPPs, these mechanisms would mainly rely on poly-saccharidic families, namely hemicelluloses, homogalacturonans, rhamnogalacturonans, and extensins.

The major role of the cell wall polysaccharides in the extraction of tannins and anthocyanins was confirmed. CoMPPs revealed a much more complex mechanism than expected, e.g. homogalacturonans in skins and pulps associated to an increase and a decrease of the polyphenols extractibilities, respectively. Moreover, our study changed the standpoint on cell wall polysaccharides. Up to now, they were considered as detrimental since they bind polyphenols, and were thus expected to increase losses. But they also release soluble polysaccharides (PRAGs) which contribute positively to the colloidal stability of wines.

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III.O.10

WHITE WINES OXIDATIVE STABILITY: A 2-VINTAGE STUDY OF CHARDONNAY CHAMPAGNE BASE WINES AGED ON LEES IN BARRELS

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Keywords: Oxidative stability, Chardonnay, Phenolic compounds, Antioxidant metabolome

Ultra-premium champagne wines are characterized by a long stay on laths. The goal of the winemaker is to use all possible oenological techniques to keep the aromatic freshness of the future products. To that purpose, some champagne base wines can be aged on lees in oak barrels. However, if it is now acknowledged that such ageing practices contribute to the oxidative stability of dry white wines, no study has been done on Chardonnay champagne base wines designed for a long ageing on laths [1]. The antioxidant capacity of Chardonnay champagne base wines was measured by DPPH assay during barrel ageing for two successive vintages, 2020 and 2021. Regardless of the vintage, ageing in new oak barrels significantly improves the Chardonnay champagne base wines oxidative stability. Oak wood ellagitanins followed a linear extraction profile during barrel ageing on champagne base wines similar to that already reported for dry Chardonnay wines [2]. Moreover, Chardonnay champagne base wines aged in new barrels preserved at the end of ageing and important number of S-N containing compounds, which in addition to the known ellagitanins revealed wines better antioxidant stability [3]. A metabolomic approach based on an untargeted UHPLC-Q-ToF-MS/MS analysis allowed a clear discrimination of champagne base wines according to the ageing period on lees in new oak barrels undependably to the vintage. This result is very valuable for the future perspectives while it indicates that champagne base wines chemical composition is dominated essentially from the barrel ageing in new oak barrels than the vintage.

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Enological Practices and Processes

IV.0.1

A NEW STRATEGY AND METHODOLOGY FOR THE CHARACTERIZATION OF POLYPHENOLS IN FINING PRECIPITATE

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Keywords: Fining, Precipitate, Condensed tannins, Anthocyanin

Polyphenols are secondary metabolite widely distributed in plant kingdom such as in fruits, in grapes and in wine. During the winemaking process, polyphenols are extract from the skin and seed of the berries. Fining is an important winemaking step just before bottling which has an impact on wine stabilization and clarification. Most the time, fining agent are animal or vegetal protein while some of them can be synthetic polymer like PVPP or natural origin like bentonite. The aim of this study was better understand colloidals phenomenons involved in fining process and determine how polyphenols content and composition are impact by finning.

Different type and composition of fining agent were used during the investigation on each wine. Some of them were based on animal proteins, vegetal proteins, PVPP or bentonite alone while some mix with PVPP and vegetal proteins, PVPP and animal protein, and a mix with PVPP, vegetal proteins and bentonite were also used. On the wine before and after fining monomeric and total anthocyanins, monomeric, dimeric and total tannins, mDP (mean degree of polymerization), flavonol and phenolic acids content were measured. However in order to determine more precisely the content and composition of polyphe-nol lost during fining, a new strategy have been develop based on the re-solubilisation of the fining precipitate using some organic solvent.

For these experiments, this new method highlight some drastic differences between fining agents. Indeed, depending of the fining agent the amount and the composition of the polyphenols present in the fining precipitate change. For example, some fining agent don't remove anthocyanins while some other precipitate mainly the p-coumarolylated anthocyanins. Moreover, important differences are also observed for condensed tannins according to the nature of the fining agent. Indeed, fining agent without PVPP were able to precipitate monomeric or dimeric condensed tannins. Moreover, some fining agent are more selective of oligomeric tannins while some has tendency to precipitate tannins with higher mDP.

This new methodology allow a more precise and clear identification of the polyphenol precipitated by fining agent and will help to better understand impact of the fining the organoleptic properties of the wine. Similarly, a better characterization of the fining precipitate will also help better understanding the colloidal structure of the wine.

IV.O.2

EFFECT OF DIFFERENT TEMPERATURE AND WATER-LOSS DEHYDRATION CONDITIONS ON THE PATTERN OF FREE AND GLYCOSYLATED VOLATILE METABOLITES OF ITALIAN RED GRAPES

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Keywords: grapes dehydration, secondary metabolites, aromas, SPE/GC-MS

Post-harvest grape berries dehydration/withering are worldwide applied to produce high-quality sweet and dry wines (e.i., *Vin Santo, Tokaji, Amarone della Valpolicella*). Temperature and water loss impact grape metabolism [1] and are key variables in modulating the production of grape compounds of oenological interest, such as Volatile Organic Compounds (VOCs), secondary metabolites responsible for the aroma of the final wine.

The aim of this research was to assess the impact of post-harvest dehydration on free and glycosylated VOCs of two Italian red wine grapes, namely Nebbiolo and Aleatico, dehydrated in tunnel under controlled condition (varied temperature and weight-loss, at constant humidity and air flow). From these grapes *Sforzato di Valtellina Passito DOCG* and *Elba Aleatico Passito DOCG*, respectively.

The experimental plan followed a "Temperature (10°C, 15°C, 20°C, 25°C) x Weight loss (0%, 10%, 20%, 30%)" factorial design. Skin and juice free and glycosylated VOCs of grape berries were separately analysed by Solid Phase Extraction/Gas Chromatography–Mass Spectrometry (SPE/GC-MS) [2].

Results showed that skin and juice samples are well discriminated in both varieties, with skins exhibiting a greater aromatic richness, especially in terms of bound VOCs. In Nebbiolo grapes, weight loss showed a greater influence than temperature on free volatiles. This trend was not observed on free VOCs of Aleatico grapes, that were treated with more stressful dehydration conditions of temperature (15°C, 25°C) and weight loss (20%, 30%) compared to Nebbiolo grapes (10°C, 20°C; 10%, 20%).

Temperature seems to play an important role on bound VOCs of both grapes, albeit in a different form. In Nebbiolo grapes, low temperatures (10°C) showed positive correlations with the accumulation of aroma glycosidic precursors. In the case of Aleatico, which is a semi-aromatic variety, dehydration temperatures, appear to modulate terpenes pattern regardless of weight loss. Specifically, samples dehydrated at 15°C correlated with beta-linalool, epoxylinalool, cis- and trans-linalool oxide, and geranic acid, while 25°C ones with cis- and trans-geraniol, cis- and trans-citral, α-terpineol, and citronellol.

These results are of interest for optimizing the grape dehydration process not only in an optic of management of product characteristics and varietal oenology, but also in a prospective of management of energy resources needed under controlled dehydration conditions.

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IV.O.3

EUGENOL AS QUALITY MARKER OF WINES AND SPIRITS FROM HYBRID VINES: IMPACT OF DIFFERENT WINEMAKING AND DISTILLATION PROCESSES

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Keywords: micro-vinifications, micro-distillations, spirits quality, phenylpropanoids

Eugenol, widely spread in various plants notably cloves, basil and bay, was identified too in wines from hybrid grapes without contact with oak wood. This aromatic molecule presents a strong spicy note of clove and also antifongic properties. Eugenol was described as an endogenous compound of Baco blanc, from the grapes to the spirits of Armagnac area. Moreover, this compound is a chemical marker of Baco blanc products quality.

Influences of harvest time and different winemaking processes (settling, use of enzymatic preparations, lees content and stock time before distillation) on Baco blanc wine eugenol contents were explored using a two-levels full factorial Design of Experiments (DoEs). Each modality was twice-distilled at the same ABV (Alcohol By Volume) to produce white spirits. Quantified by HS-SPME-GC/MS with a proven methodology, eugenol is particularly concentrated in wines (median content 43 µg/L) and white spirits (median content 124 µg/L) made with Baco blanc cultivar. Eugenol content in wines and spirits were mainly determined (66%) by harvest time: the earlier the harvest date, the higher eugenol concentration in wines (variation from 28 to 126 µg/L) and spirits (variation from 57 to 317 µg/L). This observation was confirmed by measuring the concentration of eugenol in Baco blanc berries during their development. Our results also highlight the fact that the use of β -glucosidase enzymatic preparation after alcoholic fermentation (21% of determination) permits to enhance eugenol levels. At the evidence a bound fraction of eugenol in Baco blanc wines exists. The perspective of identification and evaluation of the eugenol precursor have been operated to improve quality of Baco blanc products. First approaches trends to identify a majority precursor (80% of the bound eugenol). Moreover the use of a full factorial DoE highlighted the key steps of winemaking process that most influence the concentrations of the different forms of eugenol. Sensory analyses carried out with a panel of professionals trends to show that eugenol is involved in perceptual interactions at the heart of Baco blanc spirits quality. Finally, the different forms of eugenol are not only a subject of study for Armagnac spirits, but of wider interest in the composition of hybrid and resistant vines and in the characterisation of the organoleptic quality of wine spirits.

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EXPLORING THE INFLUENCE OF *S. CEREVISIAE* MANNOPROTEINS ON WINE ASTRINGENCY AND THE IMPACT OF THEIR POLYSACCHARIDE STRUCTURE

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IV.0.4

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Keywords: Mannoproteins, Physico-chemical Interactions, Astringency, Condensed Tannins

Mannoproteins (MPs) are proteoglycans from the outmost layer of yeast cell walls released into wine during alcoholic fermentation and ageing on lees processes. The use of commercial preparations of mannoproteins as additives to improve wine stability with regards to the crystallization of tartaric salts and to prevent protein haze in the case of white and rosé wines is authorized by the OIV.

Regarding red wines and polyphenols, mannoproteins are described as able to improve their colloidal stability and modulate the astringent effect of condensed tannins. The latter interact with salivary proteins forming insoluble aggregates that cause a loss of lubrication in the mouth and promote a drying and puckering sensation. However, neither the interaction mechanisms involved in mannoproteins capacity to impact astringency nor the structure-function relationships related to this property are fully understood.

The aim of this study was to evaluate the impact of high molecular weight mannoproteins on tannin-protein interactions. To this end, experiments were performed in a model wine using tannins purified from a red Syrah wine and BSA. Tannin-BSA aggregation kinetics were followed for 1 hour through Dynamic Light Scattering measurements in the absence and presence of mannoproteins. To progress in the identification of structure-function relationships and on the part played by the polysaccharide part, mannoproteins fractions from four yeast strains were extracted and purified. Yeast Strains were selected according to their origin and specific mannoprotein polysaccharide structure: a commercial enological strain (MP-LMD47), the wild-type BY4742 strain (MP-WT), and two of its mutants Δ Mnn4 (MP-Mnn4, no mannosyl-phosphorylation) and Δ Mnn2 (MP-Mnn2, linear N-glycosylation backbone). A thorough characterization of mannoprotein fractions confirmed the structural differences between mannoproteins from each yeast strain.

MPs were capable of delaying tannin-BSA aggregation kinetics by preventing the formation of micron-sized particles within the hour of measurement but did not avoid the long-term precipitation of tannin-BSA aggregates. Experiments indicated that mannoproteins interfere with tannin-BSA enlarged aggregation through the formation of a ternary MP-Tannin-BSA system. To be able to prevent tannin-BSA particle growth, the density/compactness of the polysaccharide moiety of MPs was a key factor.

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PRODUCTION OF A FUNCTIONAL BEVERAGE FROM WINEMAKING BY-PRO-DUCTS: A NEW WAY OF VALORISATION

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IV.0.5

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Keywords: kombucha, grape pomace, anti-inflammatory, antidiabetic

In the challenge of transforming waste into useful products that can be re-used in a circular economy perspective, winery by-products can be considered as a source of potentially bioactive molecules such as polyphenols. The wine industry generates each year 20 million tons of by-products. Kombucha fermentation is an ancestral process which allow to increase the biological properties of tea by the action of a microbial consortium formed by yeasts and bacteria called SCOBY. It belongs to the field of healthy food for which the interest of consumers is growing. The objective of this work was to propose a new functional beverage made from winemaking by-products fermented by a Kombucha SCOBY.

In a preliminary step, 2 types of by products were evaluated for Kombucha fermentation, wine lees and grape pomaces in order to assess their ability to ferment. Then the work was focused on grape pomaces originated from red winemaking. Several parameters were varied during the fermentation process: temperature, pomace concentration, sugar concentration, temperature and duration. The fermentation kinetics and final composition of grape Kombucha were monitored. Several biological activities were assessed in vitro at the beginning and at the end of fermentation: antioxydant, antidiabetic and anti-in-flammatory. Depending on their physico-chemical and biological characteristics, some of the pomace Kombucha beverages were submitted to a sensory evaluation.

For all fermentation conditions the biological activities were increased, at least by a factor 2, at the end of fermentation compared to the non-fermented grape pomaces infusions. However according to their concentrations in sugar and total acidity, the grape pomace Kombuchas were not equally appreciated by the panellists. A majority of them preferred the Kombucha flavoured with natural fruity aroma.

This work confirmed the feasibility of making a grape pomace Kombucha beverage. Even if the kombucha fermentation improved the biological activities of this new beverage, results showed that there is no significant impact of the tested processing parameters on the biological activities in vitro. This new functional beverage consists into a new way of winemaking by-products valorisation. As for future perspectives, the organoleptic aspect must be improved.

IV.0.6

MONITOR SOME KEY PARAMETERS THROUGH THE IMPLEMENTATION OF CONTINUOUS CONTROL SYSTEMS OF THE MUST-WINE DURING MACERA-TION-FERMENTATION IN RED WINEMAKING TO MANAGE OPERATIONS IN "AU-TOMATION"

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Keywords: Automation 4.0, Digital winemaking, Automation, Red wines

This study is aimed to develop a complete tool for the winemaker with, complete and targeted "winemaking recipes" that can be adapted to criteria set by the winemaker, such as: grape variety, grape health status, degree of ripening, desired wine, redox status throughout the alcoholic fermentation.

To get such aim, specific sets of experiments using red grape juices from different varieties (Nebbiolo, Barbera, Pinot noir, etc.) collected at different technological and phenolic maturity points, will be held with "automatized 4.0 tanks" equipped with sensors for measuring: redox potential, dissolved oxygen, relative density, temperature, and color in order to collect a sufficient amount of data preparatory to the creation of operating models in the most widely winemaking situations in which the automatized 4.0 tanks "will be able to independently respond" with the right corrective actions (opening/closing aeration valve, execution/block pumping overs , etc.) if the key parameters exceed the limits of the recommended ranges set in the selected recipe.

To monitor every experimental winemaking, chemical and chemical-physical analyses according to OIV methods¹ and Glories' indexes², such as: density, sugars, total acidity, pH, yeast available nitrogen, acetic acid, ethyl alcohol, color intensity, anthocyanins, tannins, anthocyanin co-pigmentation indices, condensed tannins, astringent tannins, tannins combined with polysaccharides, will be daily provided.

Then, external monitoring of redox potential, T, dissolved O_2 , and relative density will be done in parallel to check the accuracy of the sensors.

Some claims from this research have been already included into Italian patent "PROCEDURE AND AP-PARATUS FOR THE VINIFICATION OF A GRAPE JUICE" no. 102022000023430 filed on 14 November 2022 in the name of GIMAR S.R.L. (Omnia Technologies Group, Della Toffola).

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IV.0.7

EFFECT OF WHOLE BUNCH VINIFICATION ON THE ABUNDANCE OF A SWEETENING COMPOUND

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Keywords: Stem, Whole bunch vinification, Sweetness, Astilbin

In classic red wine-making process, grapes are usually destemmed between harvest and the filling of the vat. However, some winemakers choose to let all or a part of the stems in contact with the juice during vatting, this is called whole bunch vinification. For instance, this practice is traditionally used in some French wine regions, notably in Burgundy, Beaujolais and the Rhone Valley. The choice to keep this part of the grape is likely to affect the sensory properties of wine, as its gustatory perception^{1,2}. Previous studies have focused on the chemical composition of stems and have shown that astilbin, a sweet molecule, is a one of the major phenolic compounds^{3,4}. The aim of this study is to investigate the effect of whole bunch vinification on the concentration of astilbin in wine.

Several experiments have been carried out in various French wine regions, in Burgundy, Beaujolais and Bordeaux over three vintages allowing to compare different grape varieties namely Pinot Noir, Gamay and Merlot. The addition of stems is carried out while filling the vats, according to different proportions ranging from 15 to 50 % of the total volume of introduced grape. For each experiment, the modality with addition of stems is compared with a vat of destemmed grapes coming from the same plot. Samples were taken throughout the wine-making process to be analyzed by liquid chromatography coupled with high resolution mass spectrometry (UHPLC-Exactive, Orbitrap analyzer).

The comparison of astilbin content in the two modalities showed that the addition of stems during vatting significantly increased astilbin concentration. Furthermore, this increase varies according to the grape variety. Indeed, the ratio between the astilbin concentration of wines from the two modalities is higher in Merlot than in Pinot Noir and Gamay. The localization of astilbin in the different components of Merlot and Pinot Noir bunch was also investigated in order to better understand this difference.

This study provides new insights on the contribution of stems to the concentration of a sweet compound, astilbin. Besides, these results bring new tools to better understand the practice of whole bunch vinification from a chemical perspective.

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Sensory properties : psychophysics-cognitive psychology, experimental economy, connexions with neurosciences

V.O.1 SENSORY IMPROVEMENT OF DEALCOHOLISED WINES

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Keywords: dealcoholised wines, sensory properties, winemaking, product development

Interest and willing-ness to buy alcohol-free wines by customers is increasing for several years [1]. Due to the rising relevance of dealcoholised wines it is the objective of this study to contribute to a better understanding of the flavor variation among dealcoholised wines and to explore enological measures, how to improve final quality.

First a range of commercial, alcoholfree white wines were analysed by the holistic sensory method projective mapping, including a question for hedonic acceptance. Based on the combination of a non-target-HS-SPME-GC/MS analysis with sensory analysis we obtained a clustering of the wines into three groups. They were characterised by varying degrees of positive, neutral and negative olfactory notes as well as the hedonic preference of the tasters.

For a targeted sensory improvement, a dealcoholised Riesling wine was processed using various oenological methods. Sweetening with grape juice obtained from a muscat variety enhanced fruity and floral aroma, which was backed by increasing concentrations of linalool, exceeding the sensory threshold. The addition of wooden chips imitated the character of a wine aged in barrels and stimulated higher preference ratings. A further improvement was tested by initiating a malolactic fermentation in the dealcoholised wines as well as a secondary alcoholic fermentation from 0 to 0.5 % vol. alc.. As dealcoholised wines lacks generally in body, mouthfeel and freshness due to the loss of ethanol, we explored the ability of yeast mannoproteins, carbonisation and prolonged yeast contact to improve these deficits. These variants were subjected to a descriptive analysis by a trained panel. It turned out that the tasters preferred wines with fruity and floral aroma, as well as a sweet, full-bodied taste. The use of grape juice as a sweetener, in combination with the ß-glycosidase activity to further release bound aroma compounds, as well as use of oak chips were the most successful treatments.

Modification of aroma compounds were investigated using a target HS-SPME-GC/MS-method for major wine aroma compounds. Fruity aromas were mainly linked to high concentrations of esters such as ethylbutanoate and ethylhexanoate and the floral notes with linalool and 2-phenylethanol. Especially addition of a grape juice instead of sucrose as well as use of ß-glycosidases yielded superior concentrations and sensory perception.

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V.O.2 SENSORY PROPERTIES IMPORTANT TO AUSTRALIAN FINE WINE CONSUMER SEGMENT PERCEPTION OF CHARDONNAY WINE COMPLEXITY AND PREFERENCE

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Keywords: wine expert, psychographic, Rate-All-That-Apply, consumer segmentation

Wine complexity is considered a multidimensional yet equivocal sensory percept. This project uncovered sensory attributes Australian Chardonnay wine consumers associate with Chardonnay wine complexity and correlations between expert and consumer perceived wine complexity and preference. A wine consumer test examined 6 Australian Chardonnay wines of three complexity levels designated low (LC1&2), medium (MC1&2), and high (HC1&2) by an expert panel (n = 8) using a benchtop sensory task. Consumers (n = 81) rated their perceived liking using a 9-point hedonic scale; wine complexity with a 5-point scale anchored "low", "low-medium", "medium", "medium-high", and "high" and lastly, profiled the wines using Rate-All-That-Apply (RATA). Psychographic segmentation with the Fine Wine Instrument (FWI) generated three segments; Wine Enthusiasts (WE n=29), Aspirants (ASP n=40) and No- Frills (NF n=12). Overall consumers liked all wines, but LC2 and MC2 were less liked and regarded as significantly lower in complexity which might be explained by these wines presenting less attributes overall with only citrus and green/grassy/leafy aromas and flavours plus higher acidity and astringency. In contrast, the HC1 and HC2 wines were more liked and regarded as more complex, showing grape-derived attributes of stone fruit flavours and winemaking-derived and developed characters including nutty, honey, vanilla, toffee, butterscotch and caramel, higher viscosity and body. Strong correlations between WE, ASP and expert complexity ratings and WE liking and WE complexity ratings were observed. However, correlations between the liking and complexity ratings of wines by NF and ASP were not found. ASP significantly preferred and rated MC1 more complex with cheesy, yeasty, nutty, bread, woody and toasty attributes and persistence of aftertaste; WE liked HC2; whilst NF in addition to HC1 also liked LC2 possibly due to it having more citrus properties and less woody notes. Consumers perceived complexity agreed with the widely accepted notion that a complex wine is considered to have balanced, multilayered flavours plus gustatory and mouthfeel attributes. Consumers perceived wines showing multilayered characters as more complex and liked those more compared to wines dominated by oak or fruit characters. The reported sensory attributes contributing to perceived complexity and consumer preference of Australian Chardonnay, could assist wine makers to produce wine styles consumers like.

V.O.3 WINE AS AN EMOTIONAL AND AESTHETIC OBJECT: IMPACT OF EXPERTISE

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Keywords: emotions, expertise, aesthetic, sensory analysis

Wine tasting has been shown to provide emotions to tasters (Coste et al. 2018). How will expertise impact this emotional response? Burnham and Skilleås (2012) reported that the cultural, experiential, and aesthetic competencies characterize an expert in wine compared to a novice. Although there is no consensual definition of an aesthetic experience, Burnham and Skilleås (2012) reported that aesthetic appreciation is "disinterested, normative for others and communicable" in comparison to sensory pleasure. In another sector where the emotional impact takes an important place, the artistic sector, Leder et al. (2014) demonstrated that expertise exerts an influence on cognitive and emotional processing, which results in attenuated emotional reactions. Paasschen et al. (2015) reported that the cognitive aspects of artistic evaluation, relating to the aesthetic aspects would strongly depend on expertise, but that the affective components would on the contrary be less affected by the expertise, and consistent between all the observers. These results are consistent with the Kantian notion that an aesthetic position is emotionally distanced. The purpose of this study is to determine the impact of expertise on emotions and aesthetic judgment in wine tasting.

To answer this issue, 20 oenologists, 20 musicians and 20 novices had tasted 6 red wines. Two kinds of emotional responses were measured and previously validated in a wine tasting context. The first one is a conscious and subjective response, the cognitive component of emotion (also called feelings). This response was measured using self-declarative questionnaires. The second kind of emotional response is an unconscious and objective response. The unconscious part of emotions was evaluated with measurement of the response of the autonomic nervous system (heart rate and electrodermal activity). The emotional responses were compared to the aesthetic judgments of wines evaluated with self-declarative questionnaires.

The aim was to evaluate whether there is a contradiction between the supposed distanced declarative response of the expert and the unconscious physiological emotional response. Moreover, the results obtained with musicians allowed to determine whether the impact of the expertise is specific or transposable to another aesthetic field.

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V.O.4 WINE CONSUMER TRADE-OFF BETWEEN ORGANOLEPTIC CHARACTERISTICS AND SUSTAINABLE CLAIMS. AN EXPERIMENT ON RED WINES FROM BORDEAUX REGION

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Keywords: Experimental Economics, Consumer preferences, Willingness to pay, Sustainability

In economics, the perception of wine quality is not limited to sensorial characteristics: an indication of the region of production significantly affects the perception of quality and consumers' WTP ([1]; [2]). However, [3] or more recently [4] show that even if a wine has an organic label, the taste of wine remains the predominant criterion in consumer preferences. The contribution of our experiment is to evaluate the impact of responsible attributes (organic label, Non Added Sulfites, HVE certification) on the appreciation of several red wines on the market. More than 280 consumers participated to the present study and they perform 25 tastings divided into 5 different sessions. 20 different red wines from Bordeaux Area are tasted.

We used sensory analysis and experimental economics techniques in order to evaluate the liking and willingness to pay of consumers, according to previous works describing this technique for wine market [5].

The main interest of this paper remains the evaluation of wine by sensorial analysis and experimental auctions. In a context of increasing responsible initiatives in the wine sector, the design and results of this study bring some insights on the important issue of consumer preferences for red wines and their trade-offs between the different wine attributes. Results show that consumer expectations for corporate responsibility labeling are growing. However, the share of consumers who actually value it is small. In addition, the sensory quality of the wine remains a primary expectation.

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V.O.5 WHICH IMPACT FOR PROANTHOCYANIDIC TANNINS ON RED WINE FRUITY AROMA? SENSORY AND PHYSICOCHEMICAL APPROACHES

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Keywords: Wine, Non-volatiles, Interactions

Previous research on the fruity character of red wines highlighted the role of esters. Literature provides evidence that, besides these esters, other compounds that are not necessarily volatiles may have an important impact on the overall aroma of wine, contributing to a modulation of its global aromatic expression. The goal of this work was to assess the olfactory consequences of a mixture between esters and proanthocyanidic tannins, through sensory and physico-chemical approaches.

Sensory analysis of numerous aromatic reconstitutions, including triangular tests, detection thresholds, and sensory profiles, were conducted in order to evaluate the sensory impact of tannins on red wine esters perception. Then, the impact of these non-volatile molecules on esters volatility, and thus taster stimulation, was evaluated thanks to the determination of partition coefficients.

Our results showed that the presence of tannins in the matrix significantly attenuated perception of fruity notes. In a consistant way, physico-chemical analysis demonstrated also that the presence of proanthocyanidic tannins in dilute alcohol solution resulted in a decrease in ester partition coefficients and thus in a decrease in ester contents in the headspace. This fact highlighted pre-sensory changes. Finally, a new sensory tool was developed, consisting in an ISO glass containing two identical compartments separated by a vertical glass wall, providing a way to compare perceived odours according to whether or not the components of the odour mixtures were actually mixed in solution. This new tool was used to demonstrate the impact of the physical mixture of proanthocyanidic tannins and esters in order to demonstrate the exclusive involvement of pre-sensory interactions.

These results confirmed the sensory impact of some non-volatile compounds on odor perception. Finally, esters partition coefficient evaluation revealed a decrease of the volatility of esters when tannins were present in the matrix, thus corroborating sensory evaluation results. Proanthocyanidic tannins decrease esters volatility when they are added in the matrix, thus reducing orthonasal taster stimulation and consequently reducing red wine fruity notes perception. Such a study should be extended to anthocyanins and other oenological tannins and, including their concentration ranges, to assess the impact of the phenolic matrix on red wines aroma perception.



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Plant and Environment, Grape quality

ANTI-TRANSPIRANT MODULATION OF GRAPE RIPENING: EFFECTS ON MERLOT VINE DEVELOPMENT AND ROSÉ WINE PHENOLIC AND AROMATIC PROFILES.

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Keywords: dissociatedripenin, glow-alcohol wine, wine sensory, wine aroma

Climate changes are impacting viticultural regions throughout the world with temperature increases being most prevalent.¹ These changes will not only impact the regions capable of growing grapes, but also the grapes that can be grown.² As temperatures rise the growing degree days increase and with it the sugar accumulation within the berries and subsequent alcohol levels in wine. Consequently, viticultural practices need to be examined to decrease the levels of sugars. Anti-transpirants have been used to some degree of success, however their benefits may be linked to the varietal and style of wine produced.³ With this in mind we undertook a study of anti-transpirant application to merlot grapes to determine its effectiveness for reducing alcohol in Rosé wines.

The trial was performed in a commercial vineyard in the Hawke's Bay region of New Zealand. The vines were two cane pruned and the vineyard was managed under conventional practices. The trial was setup as a randomized block design with five vines per block. Anti-transpirant was applied using a backpack sprayer to upper portion of the canopy to the point of run off at véraison. The berries were then harvested by hand at 18 °Brix and wine making using a standardized wine making protocol at the research winery.

The harvest dates were delayed between the treated and untreated vines. The treated wines were found to have a higher pH, lower titratable acidity, and increased total phenolics. The aroma compound analysis resulted in several significant differences that were noted in the sensory evaluation. In both vintages the control wines were found to be influenced by green, vegetal, and earthy notes while the treated wines were found to be influenced by fruit aromas. These sensory attributes were confirmed by examining the aromatic compounds by PCA. This resulted in the controls being influenced by methoxypyrazines and alcohols and a few esters, compared to treated wines which were influenced by esters and terpenoids.

In conclusion, we were able to show that the application of anti-transpirant was able to dissociate the ripening process of Merlot grapes. Its application decreased sugar production but allowed for aromatic compound production. This demonstrates the potential effectiveness for anti-transpirants to control sugar in grape production to mitigate increased temperatures. These results indicate that further research is necessary to optimize the application timing of the anti-transpirant.

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DISCRIMINATION OF *BOTRYTIS CINEREA* INFECTED GRAPES USING UNTARGE-TED METABOLOMIC ANALYSIS WITH DIRECT ELECTROSPRAY IONISATION MASS SPECTROMETRY

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Keywords: Rapid analysis, metabolomics work flow, high resolution mass spectrometry, fruit quality

Infection of grapes (*Vitis vinifera*) by *Botrytis cinerea* (grey mould) is a frequent occurrence in vineyards and during prolonged wet and humid conditions can lead to significant detrimental impact on yield and overall quality. Growth of *B. cinerea* causes oxidisation of phenolic compounds resulting in a loss of colour and formation of a suite of off-flavours and odours in wine made from excessively infected fruit. Apart from wine grapes, developing post-harvest *B. cinerea* infection in high-value horticultural products during storage, shipment and marketing may cause significant loss in fresh fruits, vegetables and other crops. A rapid and sensitive assessment method to detect, screen and quantify fungal infection would greatly assist viticultural growers and winemakers in determining fruit quality.

In this study metabolites were extracted from homogenate samples using acetonitrile with the data set comprising 140 healthy and infected grapes representing different vintages, cultivars, regions and maturity stages. Sample extracts were randomly analysed by direct injection into a LTQ ion mass spectrometer, operating in negative mode, including regular quality assurance samples with data collected from 50–2000 m/z for 1 minute. Molecular feature abundances were summed between 0.1–0.4 minutes and minmax normalised prior to PCA for quality control. Samples were randomly assigned to a calibration and independent test data set, with feature reduction, a two-class model PLS-DA, cross validation and permutation testing performed with the calibration data set. Prediction of sample class in the independent test samples demonstrated an overall predictive error of less than 5%. Feature importance was assessed using a combined VIP and selectivity ratio plot which demonstrated a high level of correlation with standard volcano plots. Annotation of important molecular features was undertaken using a high resolution Orbitrap MS detector, and LCqTOF of selected samples from healthy and infected extracts.

DO MICROPLASTICS IN VINEYARD SOIL AFFECT THE BIOAVAILABILITY OF VINE NUTRITION?

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Keywords: Soil, microplastics, makronutrients, micronutrients, availability

Microplastics can alter physicochemical and biogeochemical processes in the soil, but whether these changes have further effects on soil fertility, and if so, whether these effects vary depending on the type of soil in the vineyard and the type of plastic used in the vineyard. Knowing what types of plastics are currently used in vineyards in Slovenian viticultural regions as strings to tie vines to the stake, the aim of our study was to assess the effects of microplastic particles from polypropylene (PP) and polyvinyl chloride (PVC) on the availability of macro (potassium (K), Potassium (K), calcium (Ca), magnesium (Mg) and phosphate (P)) and micronutrients (iron (Fe), copper (Cu), manganese (Mn) and zinc (Zn)) in two vineyard soils contrasting in pH and mineralogy. For this purpose, a short-term soil incubation experiment (120 days) was carried out in which the soil samples were enriched with micro-PP and micro-PVC particles. After the incubation period, macro- and micronutrient availability were measured. The results show that micro-PP particles have a stronger influence on the availability of macronutrients in the soil. Phosphate availability decreased by up to 30%, potassium availability by 20% and magnesium by 10%. However, the macronutrient most affected was nitrate, as the availability of this element decreased by more than 90% with the presence of micro-PVC particles in the soil. These results were observed in both soil types (calcareous and acidic soils). On the other hand, the presence of micro-PP particles in the soil had a greater effect on the availability of micronutrients, but not to the same extent as micro-PVC - the availability of iron was reduced by 10% and that of Cu by 10%. If we assume PVC and PP contamination of vineyard soils, we can assume that nutrients should be supplied by spraying vines with soluble fretilizers in higher concentrations, as this is the most common method to specifically minimise micronutrient deficiencies in vineyards. On the other hand, Cu, which is present in fungicidal sprays and may be present in toxic concentrations in vineyard soils, was the least affected micronutrient and could also be active in the presence of microplastic particles. Although research on the effects of microplastics on nutrient cycling in soils is still in its infancy, microplastics directly affect some soil properties that may also have indirect effects on soil nutrient cycling, e.g. cycling of C, N, P and other elements.

EMERGENCE OF INORGANIC PHOSPHONATE RESIDUES IN GRAPEVINE PLANT PARTS, BERRIES AND WINES FROM SOURCES OTHER THAN FOLIAR SPRAYING

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Keywords: phosphonic acid, contaminants, IC-ICP-MS, organic viticulture

Inorganic phosphonates are known to effectively support the control of grapevine downy mildew in viticulture. Their application helps the plant to induce an earlier and more effective pathogen defense. However, inorganic phosphonates have been banned in organic viticulture due to their classification as plant protection products since October 2013. Despite the ban, phosphonate has been recently detected in organic wines. Winemakers often assured that they had not applied the fungicide, however, without providing solid proof. This development has fueled the need to better understand potential phosphonate sources and, in particular, phosphonate uptake and distribution in vines. For this purpose, we set up an isolated test field with container vines, allowing to investigate different routes of uptake and the subsequent mobility of phosphonate over two consecutive years after defined applications. Samples of leaves, stems and berries were analysed by IC-ICP-MS, being validated for quantification of low phosphonate levels therein (LOQs of 0.08-0.15 mg/kg fresh weight). Thereby, grapevines were shown to take up well detectable amounts of phosphonate through the roots, although the total amount found in berries was significantly lower when applying a 0.54 % (w/v) phosphonate solution to the roots (6 mg/kg) than after foliar spray application (38 mg/kg). Furthermore, the determination of the ratios of phosphonate levels in leaves and those in stems allowed identifying whether the vines were sprayed with phosphonate or took up phosphonate through the roots, e.g., from contaminated groundwater. We also present data from open-field vineyards to validate the results obtained with container vines. Besides soil-borne phosphonate, we also found phosphonate residues in enological additives and processing aids, also contributing to potential phosphonate contaminations in the final wine product. In brief, our contribution will provide new insights into the origin of phosphonate in vines and derived wines originating from vineyards that had not been sprayed with phosphonate in the respective growing season.

IMPACT OF MUST NITROGEN DEFICIENCY ON WHITE WINE COMPOSITION DEPENDING ON GRAPE VARIETY

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Keywords: nitrogen deficiency, chemical markers, white wine, amino acid

Nitrogen (N) nutrition of the vineyard strongly influences the must and the wine compositions. Several chemical markers present in wine (i.e., proline, succinic acid, higher alcohols and phenolic compounds) have been proposed for the cultivar Chasselas, as indicators of N deficiency in the grape must at harvest [1]. Grape genetics potentially influences the impact of N deficiency on grape composition, as well as on the concentration of potential indicators in the wine. The goal of this study was to evaluate if the chemical markers found in Chasselas wine can be extended for other white wines to indicate N deficiency in the grape must.

This study was conducted on the vineyard of Agroscope in Changins (Switzerland) and focussed on four white grape varieties: Chardonnay, Sauvignon blanc, Gewürztraminer and Chasselas. Two treatments were set up (i.e, foliar N fertilisation at veraison and no fertilisation) for three years. Wine was produced for each treatment. The composition of the grapes was analysed at harvest and the potential indicators of N deficiency, mentioned above, were quantified in the wines. In addition, sensorial analysis of wines was carried out and highlighted the fact that wines from N-deficient must, regardless of grape variety, were less appreciated.

Nitrogen fertilisation significantly increased must N concentration (NH3 and amino acids (AA)) for all grape varieties, although the gain was related to the grape variety. Grape varieties influenced both the concentration and profile of AA in must. Nitrogen concentration in must was positively correlated with proline ($R^2 = 0.656$) and propan-1-ol ($R^2 = 0.579$) concentration in wine and negatively correlated with succinic acid, 2-phenyl-ethanol and catechin quantities in wines ($R^2 = 0.369$; 0.368 and 0.266 respectively). Grape variety affected the concentration of all N deficiency indicators in wine (p < 0.05).

These results confirm that the chemical markers, initially proposed for Chasselas, can be used for other white wines. However, the threshold of the markers in wine, indicating N deficiency in grape juice, must be determined for each grape variety separately.

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INFLUENCE OF CHITOSAN, ABSCISIC ACID AND BENZOTHIADIAZOLE TREATMENTS ON SAVVATIANO (*VITIS VINIFERA L.*) WINES VOLATILE COMPOSITION PROFILE

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I.SC.6

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Keywords: Savvatiano, Biostimulants, Volatile compounds, Aroma

In the last decades the use of bioestimulants in viticulture have been promoted as alternative to conventional pesticides. Moreover, as bioestimulants promote the biosynthesis of secondary metabolites in grape berries, several studies had investigated their influence on the accumulation of phenolic compounds (Monteiro et al., 2022). However, few studies, so far, are focused on the accumulation of the volatile compounds and their impact on the produced wines (Giménez-Bañón et al., 2022; Gomez- Plaza et al., 2012; Ruiz Garcia et al., 2014).

This study was conducted in a single vineyard of white autochthonous grapevine variety Savvatiano (*Vitis vinifera L.*) in Muses Valley (Askri, Viotia, Greece). Chitosan (CHT), Abscisic Acid (ABA) and Benzothiadiazole (BTH) were applied. The applications were performed at veraison stage, in a randomized complete block and grapes were harvested at their optimum technological maturity level. White wines vinification procedures were carried out (Miliordos et al., 2022) physiochemical parameters of must and wine, and wine aroma compounds were examined. Volatile compounds were analyzed using a gas chromatography coupled to a mass spectrophotometric detector (Miliordos et al. 2022). Results were statistically evaluated by analysis of variance (ANOVA at the $p \leq 0.05$ level) and principal component analysis (PCA). CHT treatment increased total terpenes, esters and monoterpenes concentration which may enhance the desirable aromas for Savvatiano wines. Moreover, ABA enhanced the concentration of total esters, while kept in lower levels higher alcohols than control wines related to unpleasant aromas. On the other hand, BTH kept in low levels monoterpenes and acetates, as well as concentration of acids (hexanoic acid, isobutyric, butyric, isovaleric) and alcohols were still in low levels compared to control wines and the CHT and ABA treated. Furthermore, these differences in the volatile compound levels could sensorially detected, by the sensory panel.

The application of biostimulants recorded promising results to enhance aroma profile of the produced white wines. More research on different Greek cultivars in different terroirs is needed in order to enhance our knowledge regarding the effect of biostimulants on grape and wine quality.

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AN AUTOMATIC CANOPY COOLING SYSTEM TO COPE WITH THE THERMAL-RADIATIVE STRESSES IN THE PIGNOLETTO WHITE GRAPE

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Keywords: Climate change, Precision irrigation, Sunburn damage, Phenolic maturity

In recent years characterized by hot dry summers, the implementation of innovative irrigation tools in the vineyard represents a crucial challenge to ensure optimal production and to avoid excess of water consumption. It is known that the grapevine reacts to multiple stresses - i.e., high temperatures and water shortage - through adaptive mechanisms that are detrimental to the yield. Furthermore, this condition is usually aggravated by high solar radiation, which could negatively affect the phenolic composition of the grapes. Therefore, a cooling system has been developed aiming to reduce bunches' sunburn damage. The system is composed of both a network of proximal sensors able to acquire the microclimatic data within the vineyard and an actuator that triggers the nebulizers when the air temperature threshold of 35 °C is exceeded. The system was evaluated at the experimental vineyard of University of Bologna during the 2022 season on Pignoletto, an Italian white grape cultivar. Three treatments were evaluated: non-defoliated control (C), vines subjected to defoliation of the basal leaves (DI) and vines subject to the same defoliation and sprayed with nebulized water (FOG) in order to verify the effects on yield attributes, berry necrosis and secondary metabolites such as flavonols, responsible for white wine browning. The application of nebulized water in the cluster zone was able to reduce the temperatures of the berries compared to C and DI. Furthermore, the vines subjected to nebulized water showed to be more productive than the C and DI vines without affecting any technological maturity parameters. In conclusion, the cooling system of the fruiting area seems to be an excellent device for reducing the negative effect of multiple summer stresses on grapes with regards in sunburn damage and grape composition.

EFFECTS OF DIFFERENT PRUNING TYPES ON CHARENTE UGNI BLANC GRAPE AND WINE QUALITY

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Keywords: Short and High pruning, Chemical analyses, Methyl salicylate, Wine tasting

Since the use of sodium arsenite was banned in 2001, Grapevine Trunk Diseases (GTDs) have become even more widespread increasing (1). To avoid pathogen entry, pruning, an age-old practice, is increasingly coming to the fore. As the vine is a liana (2), any excessive woody proliferation has to be stopped. This can preserve grapevine life, provided it does not damage the diaphragm.

Our trial concerned two Ugni blanc parcels planted in 2006 and 2015, in vineyards managed by JAS HENNESSY & CO, at Juillac-le-Coq and Saint-Preuil in Charente. The parcels were set out in double Guyot-Poussard. Starting in 2018, two different types of pruning quality were used in both parcels. Short pruning damaged the diaphragm, whereas high pruning ensured a desiccation cone to keep the diaphragm safe. The aim of our work was to measure over three years the different impacts of these two types of pruning on grape and wine quality.

From 2020 to 2022, weight and quality of grapes were analysed at harvest. Microvinification was then carried out. On both musts and wines, several chemical analyses were performed: (i) amino acids, to determine the nitrogen status of future wine quality; (ii) ester and higher alcohol aroma wine markers; (iii) methyl salicylate, a specific GTDs plant marker. Triangular wine tasting was carried out on six-monthold wines.

Amino acid results tended to differ with pruning quality. This was not the case for the ester and higher alcohol results, which only showed differences between the vintages. The methyl salicylate level was low in wines, thereby confirming the findings of Xavier Poitou (3) for the Ugni blanc cultivar. Although the wine tasting analyses did detect differences between short and high pruning for the young parcel, it was more difficult to do so for the older one.

The present study confirms the interest of applying high pruning on a long-term basis. It can have an indirect effect on the plant's physiological functioning, keeping the vines safer and preserving the grape quality.

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IMPACT OF HARVEST DATE ON THE FINE MOLECULAR COMPOSITION OF MUST AND BORDEAUX RED WINE (VAR. MERLOT, CABERNET SAUVIGNON). FOCUS ON ACIDITY AND SENSORY IMPACT AFTER FIVE YEARS OF AGING.

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Keywords: Climate Change, Premature aging, acidity, taste

Climate change has brought several impacts that are becoming increasingly intense during the last few years and put at risk the quality of the berries or even the plant's sustainability. Such extreme climatic events impact the composition of the wine while modulating its quality and the consumer preferences (Tempère et al., 2019). The three most important changes that take place in the must are: 1) decrease acidity, 2) increase of the concentration of sugar, hence increase of alcohol in the wine, and 3) modification of the sensory balance and the development for example of cooked fruit aromas. These nuances are also associated with the "premature aging" phenomenon, mostly during bottle aging. In the context of this work, impact molecular markers [] were evaluated in Bordeaux red wines after 5 years of aging under controlled conditions.

The main goal of this study was to examine the importance of the link between the must acidity at

Numerous results shed light on the significant impact of the maturity level as well as the variety on the molecular markers after 5 years of aging. This gives us some valuable information in order to better understand the role of harvest date and the acidity level in the must on wine evolution during aging but also to better adapt Bordeaux terroirs and grape varieties to future changes.

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ELEMENT

Wine chemistry, wine components with physiological effects

CHANGES IN CU FRACTIONS AND RIBOFLAVIN IN WHITE WINES DURING SHORT-TERM LIGHT EXPOSURE: IMPACTS OF OXYGEN AND BOTTLE COLOUR

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Keywords: Riboflavin, Cu fractions, Wine bottle colour, Photo-degradation

Copper in white wine can be associated with Cu(II) organic acids (Cu fraction I), Cu(I) thiol species (Cu fraction II), and Cu sulfides (Cu fraction III). The first two fractions are associated with the repression of reductive aromas in white wine, but these fractions gradually decrease in concentration during the normal bottle aging of wine. Although exposure of white wine to fluorescent light is known to induce the accumulation of volatile sulfur compounds, causing *light-struck* aroma, the influence on the loss of protective Cu fractions is uncertain. Riboflavin is known to be a critical initiator of photochemical reactions in wine, but the rate of its decay under short-term light exposure in different coloured bottles and for wine of different oxygen concentrations is not well understood. This study aims to gain insights into the rate of change in Cu fractions and riboflavin concentrations during the exposure of white wine with different oxygen concentrations and/or different bottle colours to light over a period of days to months.

A Chardonnay wine with an addition of 0.5 mg/L riboflavin, 0.3 mg/L Cu and different concentrations of oxygen (10 and 0.5 mg/L) was exposed to fluorescent light at 20°C in Flint coloured bottles. The wine was also investigated using other coloured bottles (Arctic blue, French green, Antique green and Amber) with the minimum oxygen concentration. The Cu fractions were quantified using colorimetry and riboflavin concentrations measured by ultrahigh-performance liquid chromatography. The results showed that for wine in Flint bottles with low oxygen, light exposure accelerated the decrease in Cu fraction I and II, with the change in Cu fraction I being most pronounced (i.e., a 10-fold decrease in 24 hours). In contrast, high oxygen concentrations resulted in no light-induced decrease in Cu fractions I or II. Riboflavin concentrations became depleted after only 20 hours of irradiation under high oxygen concentrations, while 0.07 mg/L remained in the wine with low oxygen. The darker coloured wine bottles slowed the changes observed for Cu fractions and riboflavin from a minimum period of hours (i.e., for Flint bottles) to a maximum period of months (i.e., for Amber bottles). Although light is known to induce light-struck aroma in wine, this study has demonstrated it can also accelerate the removal of protective fractions of Cu and this has implications for the general reductive development of wine.

CHARACTERIZATION AND IDENTIFICATION OF YEAST BIOACTIVE PEPTIDES RELEASED DURING FERMENTATION AND AUTOLYSIS IN MODEL WINE

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Keywords: Wine, Peptides, Yeast, Autolysis

Aging wine on lees is a consolidated practice during which some yeast components (e.g., polysaccharides, proteins, peptides) are released and solubilized in wine thus, affecting its stability and quality. Apart from the widely studied mannoproteins, the role of other yeast components in modulating wine characteristics is still scarce. Wine peptides have been studied for their contribution to taste, antioxidant, and antihypertensive potentials. However, the peptides detected in wine can be influenced by the interaction between yeasts and grape components. Therefore, to study the actual contribution of yeasts to the presence of wine peptides, the concentration and profile of peptides released by yeasts during and after fermentation was studied in model conditions.

A synthetic must, prepared replacing amino acids with NH₄Cl as the sole nitrogen source, was inoculated with an oenological *Saccharomyces cerevisiae* strain. The resulting synthetic wine was sampled weekly over the first month, and monthly in the following five months. After centrifugation, each sample was ultrafiltered (3 kDa MWCO), and the peptides on the filtrate were quantified and separated by RP-HPLC. The peptides present in the 7 (end of fermentation) – and 120-day samples, were characterized by LC-MS/MS, thus determining their sequence and the putative origin. Moreover, their potential bioactivity was studied in silico using the BIOPEP Database.

Results showed that the total concentration of peptides increased during the first two weeks before plateauing to $\simeq 0.91$ g/L. Nevertheless, the number of peptides (2263 at day 7; 1978 at day 120) and the amino acid sequence differed over time. Within the released peptides, in silico analysis revealed the presence of potential bioactive sequences in the samples taken at the end of fermentation and collected after 120 days of lees aging. The vast majority ($\simeq 95\%$) of the peptides showed a potential antihypertensive activity.

Results indicate that yeasts abundantly release different peptides during and after the alcoholic fermentation due to the presence of yeast cells. The high peptide concentration, variety, and bioactive potential reported here deserve further investigation to assess the role of this fraction on wine quality and, possibly, health effects.

IMPACT OF CLIMATIC CONDITIONS ON THE SEASONING QUALITY OF OAK WOOD FOR OENOLOGICAL USE (*QUERCUS PETRAEA*)

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Keywords: oak wood, fatty acids, unsaturated aldehydes, climate change

For coopers, seasoning and toasting are considered crucial steps in barrel making during which the oak wood develops specific organoleptic properties. Seasoning, carried out in the open air, allows reducing the moisture content of the staves to between 14 and 18% (compared to 70 to 90% after splitting) while modulating the intrinsic composition of the oak wood. Toasting consists of applying different degrees of heat to a barrel for a specific period of time. As the temperature increases, oak wood produces a wide range of chemical compounds through thermal degradation of its intrinsic composition. Many studies have been conducted to identify the key aroma compounds in oak wood, and in a recent work we reported the identification of two new unsaturated aldehydes responsible for the "puff pastry" and "metallic" nuances present in toasted oak wood aroma: (2E,4E,6Z)-nonatrienal (I) and trans-4,5-epoxy-(E)-2-decenal (II).1 In foods, these aldehydes are derived from the oxidative degradation of linolenic and linoleic acids, respectively. This degradation is promoted by heat, light and metal ions. However, no data are available on the presence of fatty acids in oak wood for oenological use (Quercus petraea). In this context, this work aimed to study the distribution of fatty acids in oak wood by focusing on the seasoning process taking into account the impact of climatic conditions. To do so, we studied in parallel the evolution and distribution of unsaturated aldehydes and fatty acids in seven oak wood staves during the seasoning process (0, 12, 18 and 36 months) depending on the location (Merpins, Châlon-en-Champagne and Beaumes-de-Venise). They were selected for their climatic diversity (average temperature and rainfall). Based on this experimental protocol, 84 samples were analyzed. The study of unsaturated aldehydes was carried out by GC-NCI-MS (NH₃) analysis, while the study of fatty acids required the development of a quantification method by GC-TOF MS analysis after liquid-liquid extraction and derivatisation. The results show a significant impact of climatic conditions on the distribution of unsaturated aldehydes and fatty acids. For example, the highest levels of unsaturated aldehydes (1.5 ng/g wood (I) and 13.2 ng/g wood (II)) were detected in oak wood seasoned in Merpins and Beaumes-de-Venise (southern France) compared to that seasoned in Châlon-en-Champagne (northern France). Conversely, linolenic acid was detected at a higher level in seasoned oak wood in Châlon-en-Champagne (9.5 µg/g wood). It is likely that "warm" climates lead to a degradation of fatty acids in favor of the formation of unsaturated aldehydes. These new results underline the potential effect of global warming on the quality and sensory identity of oak wood and barrels. To go further, these samples were also toasted. The impact on the aroma of red wine will be discussed.

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III.SC.4 NEUROPROTECTIVE AND ANTI-INFLAMMATORY PROPERTIES OF HYDROXYTY-ROSOL: A PROMISING BIOACTIVE COMPONENT OF WINE

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Keywords: hydroxytyrosol, alpha-synuclein, wine, neuroprotection

Hydroxytyrosol (HT) is a phenolic compound present in olives, virgin olive oil and wine. HT has attracted great scientific interest due to its biological activities which have been related with the ortho-dihydroxy conformation in the aromatic ring. In white and red wines, HT has been detected at concentrations ranging from 0.28 to 9.6 mg/L and its occurrence has been closely related with yeast metabolism of aromatic amino acids by Ehrlich pathway during alcoholic fermentation. One of the most promising properties of this compound is the neuroprotective activity against pathological mechanisms related with neurodegenerative disorders including Alzheimer's and Parkinson's disease. Alpha-synuclein (asyn), is a 140 amino acid protein abundant in the brain. In Parkinson's disease, insoluble forms of this protein accumulate forming inclusions termed Lewy bodies which unravel different molecular events that finally cause the death of dopaminergic neurons. In order to evaluate the capacity of HT to inhibit asyn fibril formation and to study the effect of this compound against asyn induced toxicity and inflammation, several techniques have been used including fluorescence spectroscopy, transmission electronic microscopy, RT-PCR, western blot and immunohistochemistry. Our results demonstrate that HT (at micromolar levels, 25-50 μM) presents a strong inhibitory effect preventing not only αsyn aggregation but also exercising a destabilising effect by disaggregating asyn fibrils. Moreover, HT is able to counteract asyn-induced toxicity totally reverting the death of neuronal cells (PC12 cell line). Additionally, HT can reduce inflammation induced by asyn fibrils in microglial cells (BV2 cell line). Indeed, a reduction of mRNA levels of TNF- α , iNOS, IL-1 β , IL-6 and CXCL10 was observed after the co-treatment of BV2 with HT and αsyn fibrils. Our results also demonstrated that the molecular mechanisms involved in this effect are related with the modulation of mitogen activated protein kinases (MAPKs) and the generation of reactive oxygen species through nicotinamide adenine dinucleotide phosphate (NADPH) oxidase. To sum up, our data support the use of HT to prevent neurotoxicity and inflammation associated with Parkinson's disease.

NEW INSIGHTS INTO THE FATE OF MARKERS INVOLVED IN FRESH MUSHROOM OFF-FLAVOURS DURING ALCOHOLIC FERMENTATION

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Keywords: fresh mushroom off-flavor, alcoholic fermentation, 1-hydroxyoctan-3-one, glycosides

The fresh mushroom off-flavour (FMOff) has been appearing in wines since the 2000s. Some C8 compounds such as 1-octen-3-one, 1-octen-3-ol, 1-hydroxyoctan-3-one, 3-octanol and others are involved in this specific off-flavour [1-3]. At the same time, glycosidic precursors of some FMOff compounds have been identified in musts contaminated by Crustomyces subabruptus [4], highlighting the role of aroma precursors in this specific taint. However, the fate of these volatile molecules and glycosidic fractions during fermentation is not well known. The aim of this work was to study the effects of alcoholic fermentation by Saccharomyces cerevisae (Levuline CHP® strain) on FMOff glycosidic precursors and volatile compounds present in healthy and contaminated Pinot noir musts, using Crustomyces subabruptus as fungal infection. The volatile compounds of FMOff and the glycosidic fractions of the initial musts and the resulting wines were analysed by GC-MS. The analysis of glycosidic precursors revealed for the first time the presence of 1-hydroxyoctan-3-one glycosides only in the contaminated musts. In addition, the Levuline CHP® strain was able to release a 1-hydroxyoctan-3-one glycosidic fraction during alcoholic fermentation. For the volatile FMOff compounds, the effect of fermentation was both compound and matrix dependent. Indeed, both 1-octen-3-one and 3-octanone showed fluctuating evolution depending on the initial matrix. The 3-octanol was systematically produced during alcoholic fermentation whatever the initial matrix with concentrations reaching up to 0.35 µg/L and 0.58 µg/L under healthy and contaminated conditions respectively. Finally, 1-hydroxyoctan-3-one reached an identical threshold concentration (around 150 µg/L) whatever the type of matrix and regardless of its initial level. Interestingly, this compound plays a dual role as substrate and product for the yeast highlighting a potential metabolic node in the FMOff biogenesis.

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III.SC.6 NOVEL BENZENETHIOLS WITH PHENOLS CAUSE ASHY, SMOKE FLAVOR PERCEPTION IN RED WINES

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Keywords: smoke, guaiacol, sensory, benzenethiol

Smoke impacts on wines are becoming a worldwide problem; the size and severity of wildfires increasing due to influences from changing climates.¹ For over a century, wines have been known to have a unique issue of absorbing chemical compounds derived from wildfire smoke wherein the flavor of the subsequent wine becomes ashy, rubbery, campfire-like, and smoky.² The economic impacts of a smoke-impacted wine can last for years depending on the grape varietal, costing Oregon and Washington states in the United States over a billion dollars from the 2020 wildfires, as an example.³ While years of research have indicated elevated concentrations of smoke-related compounds, such as guaiacol and syringol, in wines after smoke events, unfortunately, replicating the sensory experience using smoke-associated phenols has not had much success.⁴ In our study, we found elevated concentrations of benzenethiols in smoke-impacted wines. 13C-labelled smoke data suggests phenol conversion into benzenethiol. A follow-up sensory experiment showed that the ashy aftertaste of smoke-impacted wines can be replicated by adding elevated amounts of benzenethiols and phenols to a wine that was previously not influenced by smoke. The novel targets could lead to better fining methods to remove the unwanted benzenethiols to achieve the flavors and aromas desired by winemakers and consumers.

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OENOLOGICAL TANNINS FOR PREVENTING THE LIGHT-STRUCK TASTE IN WHITE AND ROSÉ WINES

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Keywords: Riboflavin, Light exposure, Wine fault, Storage

The light exposure of wine can be detrimental as a relevant loss of aromas takes place [1] and light-induced reactions can occur. The latter involves riboflavin (RF), a photosensitive compound, that is fully reduced by acquiring two electrons. When the electron-donor is methionine, the light-struck taste (LST) can appear leading to cooked cabbage, onion and garlic odours-like [2]. The use of oenological tannins can limit the appearance of LST in both model wine [3] and white wine [4]. This research aimed to evaluate the impact of certain oenological tannins, selected in a previous study as the most effective against LST [5], in both white and rosé wines.

Six white wines and two rosé wines (5 still and 3 sparkling wines) produced in different vintages, were added with grape seed, tea and tara tannins (40 mg/L) at bottling or disgorgement. The wines were stored in the dark until the light exposure that was carried out under controlled condition [3] at bottling, and after 4 and 9 months of storage. Tannin-free wine samples were considered at each sampling point as control. The total flavonoids (FLVs), the color index (at 420 nm or 520 nm) and the content of RF were determined. The sensory analysis was also performed.

As expected, the addition of oenological tannins led to an increase of FLVs reaching the highest concentrations in the presence of tara tannins followed by tea tannins; a comparable content of FLVs was observed in control wines and those added with grape seed tannin. Only slight color index changes were found in the wines added with tannins. The light exposure did not affect neither FLVs nor the color index. In the samples stored in the dark, the content of RF ranged from about 50 μ g/L (in 2 white sparkling wine samples) up to 130 μ g/L that decreased when the wines were kept under light. The perception of bitterness and astringency was evident in particular with tara and grape seed tannins getting stronger especially in the 2 low-RF wine samples after the light exposure. A decrease of the overall aromatic profile was evidenced after the light exposure and LST was less perceived in the presence of tea tannins that seemed to limit aroma loss.

This study evidenced the impact of the wine on the light-induced fault that can have different wine-dependent facets and it seems of higher intensity in younger wines. Among the oenological tannins tasted, tea tannin was the most effective against LST and, in some cases, also in limiting the aroma decay.

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III.SC.8 OPTIMIZING THE IDENTIFICATION OF NEW THIOLS AT TRACE LEVEL IN AGED RED WINES USING NEW OAK WOOD FUNCTIONALISATION STRATEGY

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Keywords: Red wines, Thiol compounds, Meaty aroma, Oak wood functionalisation strategy

During bottle aging, many thiol compounds are involved in the expression of bouquet of great aged red wines according to the quality of the closure.^{1,2} Identifying thiol compounds in red wines is a challenging task due several drawbacks including, the complexity of the matrix, the low concentration of these impact compounds and the amount of wine needed.^{3,4}

This work aims to develop a new strategy based on the functionalisation of oak wood organic extracts with H_2S , to produce new thiols, in order to mimic what can happen in red wine during bottle aging. Following this approach and through sensory analysis experiments, we demonstrated that the vanilla-like aroma of fresh oak wood was transformed into intense "meaty" nuances similar to those found in old but non oxidized red wines.5 Functionalized samples were analysed by gas chromatography coupled with a pulsed flame photometric detector (GC-PFPD) and olfactometry (GC-O) to optimize the reaction conditions. Analysis of functionalized oak wood organic extracts by GC-O and GC-PFPD led us to detect six OZ reminiscent of "meaty" nuances and associated with sulphur compounds. One of them was characterized by preparative multi-dimensional gas chromatography coupled with olfactometry and time of flight mass spectrometry (Prep-MDGC-O-TOF MS) and identified as 2-methoxybenzenethiol.

This thiol was also identified in red wines following extraction by SPE, separation and detection by means of GC-MS/MS (SRM mode). The validation of the quantification method was carried out before its use to study its distribution in wines, young and old from different appellations and according to the OTR (determined by coulometry) of the closure. We show that its concentration can reach the odour detection threshold determined at 607 ng/L. Following the same strategy, five other thiols reminiscent of "meaty" nuances, including 2,5-dimethylfuran-3-thiol, 5-methyl-2-furfurylthiol, o-toluenethiol, 2,6-dimethylbenzenethiol and 2,6-dimethoxybenzenethiol were also identified for the first time in red wines. Their sensory impact will also be discussed.

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PHOTOCHEMICAL DEGRADATION OF TRYPTOPHAN IN MODEL WINE: IMPACT OF HEAVY METALS AND OXYGEN ON 2-AMINOACETOPHENONE FORMATION

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III.SC.9

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Keywords: 2-aminoacetophenone, iron, oxygen, riboflavin

The wine industry worldwide faces more and more challenges due to climate change, such as increased dryness in some areas, water stress, sunburn and early harvesting during hot summer temperatures¹. One of the resulting problems for the wine quality might be a higher prevalence of the untypical aging off-flavor (ATA)². A substance, which Rapp and Versini made responsible for ATA, is the 2-aminoacetophenone (2-AAP)³. 2-AAP in wine causes a naphthalene, wet towels, wet wool, acacia flower or just a soapy note⁴. The formation of the substance occurs via the degradation of tryptophan and the tryptophan metabolite indole-3-acetic acid. The formation of 2-AAP is promoted by abiotic stress factors such as drought, low nitrogen content and high temperature, and by microorganisms via riboflavin, known as a photosensitizer5. In this study, the influence of other abiotic factors, namely oxygen and heavy metals, on the light-induced degradation of tryptophan to 2-AAP was investigated. Model wine with 0.53 µmol/l riboflavin was treated with UV-C light to stimulate tryptophan degradation. A linear increase in the intensity of UV-C light exposure caused a linear increase of 2-AAP. Increasing oxygen in the model wine supported the production of 2-AAP verifying that tryptophan degradation via riboflavin follows an oxidative pathway. Indeed, 2-AAP production decreased by 81 % when oxygen was reduced from saturation to anoxic conditions. It was also found that the presence of heavy metals led to a significant reduction of 2-AAP: 0.1 mmol/l Fe²+ decreased 2-AAP by 63 %, and 0.1 mmol/l Cu²+ decreased 2-AAP by 32 %. This observation can be explained by the Fenton reaction which requires Fe²+ and/or Cu²+ to produce – in this case – acetaldehyde from ethanol. It is suggested that the Fenton reaction acts as a competitive reaction to the photosensitized production of 2-AAP. As a lateral observation, the model wine in this study turned yellow after being UV-C radiated. The LC-MS signal suggested the substance lumichrome; its signal increased with the more yellow color of the model wine. Accordingly, riboflavin could not only act as a photosensitizer but also degrade itself after exposure to light.

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PINKING PHENOMENA ON WHITE WINES: RELATION BETWEEN PINKING SUS-CEPTIBILITY INDEX (PSI) AND WINE ANTHOCYANINS CONTENT

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Keywords: White wines, Pinking, PSI, Monomeric anthocyanins

Pinking is the emergence of pink tones in white wines exclusively produced from white grape varieties, known as pinking phenomena for many years. Pinking is essentially appeared when white wines are produced under reducing conditions [1,2,3]. Pinking usually occurs after bottling and storage of white wines, but its appearance has also been described after alcoholic fermentation or even as soon as the grape must is extracted [4]. Therefore, the purpose of this work was to investigate the existence of anthocyanins in white wines made from different white grape varieties and grown locations and critically evaluate the most common method used for predicting pinking appearance in white wines: the Pinking Susceptibility Index (PSI). Anthocyanins were concentrated by SPE [1]. Also, the products formed by hydrogen peroxide oxidation of the same wines were isolated using this method. The correlation between the PSI and the whole visible spectra was studied by multivariate statistical methods, PCA and PLS analysis, to evaluate the spectral regions in the visible spectra most important to the measured PSI. No correlation between anthocyanins concentration and the Pinking Susceptibility Index (PSI) was observed contrarily to the colour of wines exposed to oxygen (r = 0.871, p < 0.00005) [5]. The oxidation of wines with hydrogen peroxide resulted in the formation of various compounds. PSI was correlated with compounds absorbing in the 400–480 nm region, probably more related to the browning than the pinking phenomenon. The lack of correlation between the PSI and anthocyanins concentration in white wines can be due to the different chemical compositions of white wines that yield various compounds after oxidation that might not be related to the natural wine pinking phenomenon.

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Enological Practices and Processes

BORDEAUX RED WINES WITHOUT ADDED SULFITES SPECIFICITIES: COMPOSI-TIONAL AND SENSORY APPROACHES TOWARDS HIGHLIGHTING AND EXPLAI-NING THEIR SPECIFIC FRUITINESS AND COOLNESS

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Keywords: Wines without added sulfites, Methyl salicylate, Carbonyl compounds, Sensory analysis

With the development of naturality expectations, wines produced without any addition of sulfur dioxide (SO_2) become very popular for consumers and such wines are increasingly present on the market. Recent studies also showed that Bordeaux red wines without added SO_2 could be differentiated from a sensory point of view from similar wines produced with SO_2^1 . Thus, the aim of the current study was to characterize from a sensory point of view, specific aromas of wines without added SO_2 and to identify compounds involved.

First, sensory profile were established for wines produced from the same merlot grapes with or without SO_2 addition throughout all winemaking process². This led to demonstrate that wine without added SO_2 was perceived differently than the wine with added SO_2 with a specific fruity aroma and a higher coolness. Moreover, to validate that presence of free SO_2 was not only at the origin of these differences, triangle tests were also performed from these wines.

In a second step, targeted analyses were performed on compounds known to be impacted by SO₂ and/ or with specific impact on fruity aroma. Thus, acetaldehyde, diacetyl, and methyl salicylate, previously identified by a sensory target approach³, were studied. These quantification approaches allowed to observe that wines without added SO₂ presented free acetaldehyde, higher concentrations in methyl salicylate and lower concentrations of free and total diacetyl. Based on these results, sensory characterization of these compounds in wines without added SO₂, and considering the impact of SO₂ were done using sensory profile. This allowed to show that presence of methyl salicylate, acetaldehyde and/or free SO₂ modified perception of coolness, whereas diacetyl and methyl salicylate modified fruity aroma.

Finally, experimental wine produced from the same grapes respectively with and without SO_2 addition were supplemented, according to their compositional specificities, with SO_2 and diacetyl for the wine without added SO_2 and with methyl salicylate for that one with added SO_2 . These two supplemented wines were compared by triangle test and were perceived differently, thus underlined that differences between red wine produced with and without added SO_2 from Bordeaux involve other compounds than those considered here and suggests that further investigations are needed.

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IV.SC.2 ANALYZING THE ROLE OF ELEMENTAL SULFUR IN GRAPE JUICE ON THE DEVE-LOPMENT OF POLYFUNCTIONAL MERCAPTANS IN SAUVIGNON BLANC WINES

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Keywords: Sauvignon blanc, polyfunctional mercaptans, elemental sulfur, varietal thiols

Sauvignon blanc is characterized by distinctive aromas, both fruity and herbaceous. The "green" character has been attributed to the methoxypyrazines, while the "fruity" character is associated with polyfunctional mercaptans. Polyfunctional mercaptans are of great significance due to their high impact on wines and associated low perception thresholds.

Elemental sulfur (S^o) is widely used to protect grapevines from powdery mildew. Machine harvesting can enhance the formation of C6-compounds from grape lipids, leading to desirable polyfunctional mer-captan formation in wines.

A missing piece of information in most past studies on the formation of polyfunctional thiols has been the concentration of elemental sulfur in grapes. In this research, we aimed first to develop an easy and applicable method for a winery setting to analyse elemental sulfur concentration in grape juice samples. With this method in place, trials were then established to examine the link between elemental sulfur in the juice and 3MH/3MHA formation in wines. The trials were undertaken during three consecutive harvests in New Zealand in 2020, 2021, and 2022.

The study developed a sulfide sensor to measure elemental sulfur levels in grape juice samples and investigated the correlation between S^o and polyfunctional mercaptan concentration in resulting wines. We reduced S^o to sulfide using dithiothreitol in acidic conditions and used an ion-selective electrode to measure sulfide concentrations. GC-MS was used to compare thiol concentration in wine with juice S^o levels from 2020 and 2021 samples. The investigation was expanded in 2022 by manually applying S^o to grapes at various intervals prior to harvesting and analyzing the relationship between residual S^o levels in juice and polyfunctional mercaptans in resulting wines.

The study established a dependable method based on ion-selective analysis and produced accurate calibration curves. The reduction process was found to be effective and the apparatus performed well with both standard and juice samples. Additionally, the results from the 2020 and 2021 trials revealed a correlation between increased juice elemental sulfur and a higher formation of 3MH/3MHA, supporting the theory that S^o contributes to the formation of 3MH in wine. This correlation was further confirmed in the 2022 trial, which saw a substantial increase in 3MH/3MHA in wines resulting from the manual application of S^o to the grapes through late spraying in the field.

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IV.SC.3 ANTIOXIDANT CAPACITY OF INACTIVATED *NON-SACCHAROMYCES* YEASTS

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Keywords: Yeast derivatives, Antioxidant, Wine stability, Non-Saccharomyces

The importance of the *non-Saccharomyces* yeasts (NSY) in winemaking has been extensively reviewed in the past for their aromatic or bioprotective capacity while, recently their antioxidant/antiradical potential has emerged under winemaking conditions. In the literature the antioxidant potential of NSY was solely explored through their capacity to improve glutathione (GSH) content during alcoholic fermentation [1], while more and more studies pointed out the activity of the non-glutathione soluble fraction released by yeasts [2].

Our study proposed to combine untargeted UHPLC-Q-ToF MS based metabolomic analysis with DPPH antiradical activity [3] to explore the antioxidant capacity of compounds released by inactivated *non-Saccharomyces* yeast (INSY) in wine like model solution. In our experimental plan, 3 INSY species were compared to one inactivated *Saccharomyces cerevisiae* yeast (ISY) selected for its high antioxidant capacity [4]. In that way, both the species and the production process were evaluated for their impact on the metabolic fingerprint and the antioxidant capacity. Then, unsupervised analysis has been used to extract ions correlated with the antioxidant capacity of the INSY.

Our results show that, all the INSY can accumulate GSH during the specific production process with yields ranging from +170% to +360% compared to the corresponding classical production process. Among the tested INSYs, one presenting equivalent antioxidant capacity to the control ISY while was 4 times less concentrated in GSH (4.73+/-0.09 mg/g against 20.95+/-0.34 mg/g, respectively). The principal component analysis of the 3511 ions detected by UHPLC-Q-ToF MS clearly grouped INSY by species, independently of the production process. 73 specific ions presenting strong and significant spearman correlation (rho < -0.6, p-value < 0.05) with the DPPH scores, clustered the most antioxidant INSY and the control *Saccharomyces* in different groups, indicating that the antioxidant capacity of these two products should be driven by different pools of compounds.

These results are very valuable for future research perspectives while they point out that, first, GSH alone is not relevant to explain the antioxidant capacity of INSY soluble fraction and other more reactive compounds must be considered. Second, they support the fact that INSY antioxidant capacity is essentially driven by a specie specific metabolism and opens an avenue for the selection new species with great enological potential.

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BIOPROTECTION BY ADDING NON-SACCHAROMYCES YEASTS : ADVANCED RESEARCH ON THIS PROMISING ALTERNATIVE TO SO₂

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Keywords: Bioprotection, Non-Saccharomyces, SO₂ alternative

Sulphur dioxide has been used for many years for its antimicrobial, antioxidant and antioxydasic properties in winemaking but nowadays, it is a source of controversy. Indeed, consumers are more attentive to the naturalness of their foods and beverages and the legislation is changing to reduce the total SO₂ levels allowed in wines. To limit and replace the doses of sulphur dioxide applied, winemakers can now use bioprotection consisting in live yeast addition as alternative, seems to be promising. This process, lightly used in from the food industry, allows to colonize the environment and limit the development or even eliminate undesirable microorganisms without altering the sensory properties of the product. Recent research took the advantage of the availability of commercial non-Saccharomyces yeast to evaluate their bioprotective potential in oenology. From 2017 to 2021, different experiments were conducted using a mixture of two species (Torulaspora delbrueckii and Metschnikowia pulcherrima) as bioprotection applied at 50 mg/L directly on grapes or musts on different varieties and ripening stages. Our data showed that the bioprotection was successfully implanted in the medium, whith a lower colonization for over-ripened harvests. By using 18S metabarcoding analysis in grape must, we showed that fungal communities such as Hanseniaspora, Aspergillus or Botrytis were significantly less abundant when bioprotection was applied instead of SO₂. Furthermore, bioprotection added in the must rapidly consumed dissolved O₂ and had a negative impact on the strict aerobic acetic acid bacteria by limiting the development of these spoilage microorganisms. Experiments carried out in white must showed that bioprotection also limits the oxidation phenomena: the concentrations of glutathione were significantly higher in bioprotected white musts and final wines. Finally, bioprotection used in red winemaking presents a chemical signature, characterized by fatty acid ethyl esters, increasing the perception of fruitiness in young red wines, but to a lesser extend compared to the same yeast strains in mixed fermentation with S. cerevisiae. After bottling, the bioprotected wines were not sensorially different from wines without SO₂ addition but were different from classical sulphited wines. This research confirm the antimicrobial and a partial protection from oxidation by bioprotection in winemaking and its capacity to preserve sensory properties of wines.

DEVELOPMENT OF DISTILLATION SENSORS FOR SPIRIT BEVERAGES PRODUCTION MONITORING BASED ON IMPEDANCE SPECTROSCOPY MEASUREMENT AND PARTIAL LEAST SQUARES REGRESSION (PLS-R)

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Keywords: Spirit beverages, acetaldehyde, ethanol, impedance spectroscopy

During spirit beverages production, the distillate is divided in three parts: the head, the heart, and the tail. Acetaldehyde and ethanol are two key markers which allow the correct separation of distillate. Being toxic, the elimination of the head part, which contains high concentration of acetaldehyde, is crucial to guarantee the consumer's health and security. Plus, the tail should be separated from the heart based on ethanol concentration. Nowadays, online or in-line sensors for acetaldehyde monitoring during distillation do not exist and the online sensors for alcohol monitoring, based on density measurement, remain expensive for producers. In this work, we demonstrate the development of distillation monitoring sensors based on electrical impedance spectroscopy (EIS) measurements¹⁻³, combined with PLS-R (partial least squares regression) modeling. Four types of sensors are proposed and tested with wine-based distillates. Using PLS-R, the best correlations were found for one electrode, named "SpotsSym". With an R2 up to 89.9% for acetaldehyde concentration prediction and an R2 up to 86.8% for ethanol, the obtained results indicate the promising potential of the proposed approach. To our knowledge, this is the first report of sensors capable of simultaneously measuring ethanol and acetaldehyde concentrations. Furthermore, these sensors offer the advantages of being low-cost and non-destructive. Based on these results, the development of an in-line distillation monitoring system is possible in a near future, providing a promising tool for spirit beverages producers. Regarding the enology part, according to the preliminary results obtained by our research team, applications of our approach can also be developed for wine fermentations monitoring.

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IMPACT OF THE WINES' QUALITY ON THE WINE DISTILLATES' ORGANOLEPTIC PROFILE

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Keywords: wine spirit, volatil compounds, destillation, organoleptic quality

Brandy de Jerez (BJ) is a spirit drink made exclusively from spirits and wine distillates and is characterized by the use of casks for aging that previously contained Sherries. The quality and sensory complexity of BJ depend on the raw materials and some factors: grape variety, conditions during processing the wine and its distillation, as well as the aging in the cask. Therefore, the original compounds of the grapes from which it comes are of great interest (1 y 2) being in most cases the Airén variety. Their relationship with the quality of the musts and the wines obtained from them has been studied (3) and varies each year of harvest depending on the weather conditions (4). It is also influenced by specific viticultural techniques and ripeness (5). The organoleptic profile and physicochemical characteristics of wine distillates depend on the wine used to produce them, as the distilled product retains a flavor and aroma characteristic of the raw material used. In the Marco de Jerez area, the grape juice (must) is obtained under conditions that can contribute with herbaceous compounds and tannins that are not desirable for wine quality, and facilitates the drainage of the must during pressing, improving the extraction yield. The pressure applied plays an important role and, depending on the level applied, the following are obtained: "primera yema" (PY)(lower pressure) "segunda yema"(SY), -average pressure-, and finally "mosto prensa" (MP), (higher presurre). PY, SY and MP have different profiles and qualities and, therefore, will directly influence the quality of the distillates. Traditionally, the wines distilled for the production of Brandy de Jerez usually come from other D.O.'s and are characterized by poor quality. For this reason, it is interesting to fully characterize four different wines of the three qualities described (PY, SY and MP). The classic oenological parameters, polyphenol, organic acid profiles and volatile compounds are determined. After the complete characterization, all wine qualities will be distilled under the same conditions to determine if these distinguishing characteristics are transferred to the distillates obtained. A better knowledge of the raw materials will allow to improve the elaboration and manufacturing of products from wine distillates and to develop new products, being of great industrial interest. From a scientific point of view, it is of great interest as it is the first study to evaluate the influence of the press on the organoleptic quality of the distillates.

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INOCULATION OF THE SELECTED *METSCHNIKOWIA PULCHERRIMA* MP1 AS A BIOPROTECTIVE ALTERNATIVE TO SULFITES TO PREVENT BROWNING OF WHITE GRAPE MUST

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Keywords: Metschnikowia pulcherrima, Browning, SO₂ alternative, Bioprotection

Enzymatic browning (BE) of must is caused by polyphenol oxidases (PPOs), tyrosinase and laccase. Both PPOs can oxidize diphenols such as hydroxycinnamic acids (HA) to quinones, which can later polymerize to form melanins [1], which are responsible of BE in white wines and of oxidasic haze in red wines. SO_2 is the main tool used to protect must from BE thanks to its capacity to inhibit PPOs [2]. However, the current trend in winemaking is to reduce and even eliminate this unfriendly additive. Among the different possible alternatives for protecting must against BE, the inoculation with a selected *Metschnikowia pulcherrima* MP1 is without any doubt one of the most promising ones.

For that purpose, white grapes were harvested, pressed and diluted 5 times with a model grape must synthetic buffer at pH = 3.50 and supplemented or not with 20 mg/L of SO₂, 2 UA/mL of laccase activity and 250 mg/L of the selected *M. pulcherrima* MP1 (Level2 InitiaTM, Lallemand Inc, Montreal, Canada). Immediately, the samples were saturated with O₂ and its concentration was noninvasively monitored overtime by luminescence (Nomasense TM O₂ Trace Oxygen Analyzer by Nomacorc S.A., Thimister Clermont, Belgium) [3]. Once oxygen consumption attained an asymptotic behavior the samples were used for color analysis [4] and for HPLC analysis of HA [5].

As expected, in the absence of SO_2 , the must actively consumed O_2 and HA, and it turned intensely brown whereas in the presence of SO_2 , the O_2 consumption rate (OCR) was significant lower, the HA concentration was maintained at significant higher levels and the yellow color intensity remained at low values. In presence of laccase, OCR and browning intensity were even higher than in control conditions and the supplementation with SO_2 reduced both parameters but not as much as in the control must.

Inoculation with the selected *M. pulcherrima* MP1 increased significantly OCR and protected the must from BE since the final yellow color was significantly lower and the HA concentration significantly higher than in control conditions although this protection was not so effective as that of SO₂. It seems therefore that selected *M. pulcherrima* MP1 consumes O₂ very effectively making that some of the initially dissolved O₂ is not consumed by PPOs. In the presence of laccase, the supplementation with MP also protected the must from browning but not so efficiently.

This data confirms that the use of the selected *M*. *pulcherrima* MP 1 can be an interesting tool for reducing the dose of SO₂ without affecting seriously its final color quality.

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INTENSE PULSED LIGHT FOR VINEYARD WASTEWATER: A PROMISING NEW PROCESS OF DEGRADATION FOR PESTICIDES

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Keywords: Photo-degradation, phytosanitary products, metabolites, wastewater

The use of pesticides for vine growing is responsible for generating an important volume of wastewater. In 2009, 13 processes were authorized for wastewater treatment but they are expensive and the toxicological impact of the secondary metabolites that are formed is not clearly established. Recently photodecomposition processes have been studied and proved an effectiveness to degrade pesticides and to modify their structures (Maheswari *et al.*, 2010, Lassale et al., 2014). In this field, Pulsed Light (PL) seems to be an interesting and efficient process (Baranda et al., 2017). Therefore, the aim of this work was to investigate the PL technology as a new process for the degradation of pesticides.

The degradation by PL with a LPBox (Sanodev) of 20 pesticides widely used in viticulture was studied by HPLC-MS/MS. Firstly, untargeted analyses were performed in order to observe secondary metabolites formed during PL treatments. This study allowed to detect 118 metabolites and 53 hypotheses of structures were proposed using m/z, isotopic patterns of the molecules containing halogens as well as results previously obtained in the literature. Then, a quantitative method was built for the 20 studied pesticides and for the compounds previously identified as secondary metabolites. Two transitions per compound were used following fragmentation experiments. The developed MRM method allows absolute quantification of the parent molecules and relative quantification of 87 major secondary metabolites. In order to further study the degradation ability of LPBox on pesticides, 7 pesticides were selected because of their rapid degradation with PL. An optimization was made to identify the number of pulses needed to degrade the 7 pesticides from their LC50 (Daphnia Magna) to a concentration lower than their limit of quantification (LOQ). These experiments also demonstrate that it is possible to relatively quantify secondary metabolites of pesticides after PL treatment. Real wastewater samples were also treated by PL showing effective degradation of pesticides.

In conclusion, our results proved that PL has an effective impact on all pesticides treated although the fluence needed is molecule-dependent. An optimization in terms of fluence showed that it was possible to degrade pesticides from a toxic concentration to a concentration below the LOQ.

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MOVING FROM SULFITES TO BIOPROTECTION: WHICH IMPACT ON CHARDONNAY WINE?

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Keywords: metabolomic, sensory, integrative approach, alternative

Over the last few years, several tools have been developed to reduce the quantity of sulfites used during winemaking, including bioprotection. Although its effectiveness in preventing the development of spoilage microorganisms has been proven, few data are available on the impact of sulfite substitution by bioprotection on the final product. The objective of this study was therefore to characterize Chardonnay wines with the addition of sulfite or bioprotection in the pre-fermentation stage. The effects of both treatments on resulting matrices was evaluated at several scales: analysis of classical oenological parameters, antioxidant capacity, phenolic compounds, non-volatile metabolome and sensory profile. This integrative approach was used for the first time in the context of bioprotection on white wine. The analysis of classical oenological parameters did not reveal any differences between the two treatments. However, the use of sulfites in the pre-fermentation stage seemed to induce a higher antioxidant capacity than bioprotection in wine. This result was confirmed by the decrease in the concentration of some phenolic compounds in the bioprotected wines. UHPLC-q-ToF-MS analysis of finished wines revealed specific footprints reflecting the impact of each treatment. As a result, 618 biomarkers were associated to sulfite treatment, mainly represented by CHON compounds, which could correspond to peptides. Moreover, bioprotection treatment was characterized by 364 biomarkers, including predominantly lipids. These highlighted biomarkers could be associated with various metabolic pathways such as amino acid biosynthesis and cofactors biosynthesis. These important differences in metabolite composition observed between the wines could be explained by the presence or the absence of sulfites, known for their effects on yeast metabolism and wine compounds. In contrast to metabolomic analysis, a very small difference was perceived between the two treatment from a sensory point of view. Thus, this study revealed substantial changes in wines regarding their composition, without impacting their sensory profile. This integrated approach has provided new knowledge on the impact of sulfite substitution by bioprotection on Chardonnay wines.

NEW PLANT BIOPOLYMERS FOR THE COLLOIDAL STABILITY OF THE COLORING MATTER OF RED WINES

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Keywords: Plant exudate, Coloring matter, Colloidal stability

The color as well as the "clarity" of red wines are ones of the qualities required by the consumers. Red wines must have colloidal stability from its bottling to its consumption. The supplementation of red wines with additives, and especially Acacia *senegal* gum, contributes to its organoleptic properties such as the colloidal stabilization of the coloring matter. In a global perspective of limitation of additives in the field of enology, one of the objectives 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.

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, polydispersity and intrinsic viscosity) 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.

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, one of them 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. 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. This increased efficiency towards the colloidal stability of the coloring is correlated to the intrinsic biochemical and structural properties of this exudate. This natural exudate could therefore be of interest for its use in enology.

IV.SC.11 PAIRING WINE AND STOPPER: AN OLD ISSUE WITH NEW ACHIEVEMENTS

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Keywords: wine storage, stoppers, volatile profile, phenolic profile

The sensory characteristics of wine are a topic studied by several researchers over time, but it continues to be a current and challenging subject. These characteristics are fundamental for the consumer acceptability, which has increasingly aroused their interest to modulate them in line with current market trends and innovation demands. The wine physical-chemical and sensory properties depend on a wide set of factors: they begin to be designed in the vineyard and are later constructed during the various stages of winemaking. Afterwards, the wine is placed in bottles and stored or commercialized. During the storage of bottled wine several physical-chemical changes may occur, modulated by the position of the bottle, type of closure, temperature, and storage time, which impact the oxygen entrance ratio. In fact, the permeability of the stoppers to oxygen is considered one of the most important properties that influences wine sensorial properties during post-bottling (1,2). In the present study, red and white table wines stored in a horizontal position for 17.5 (white wines) and 35 months (red wines), using natural cork stoppers, different types of microagglomerated cork stoppers and a synthetic one, were characterized. To achieve a holistic view of the changes that may have occurred during bottling, a set of analysis were implemented, namely, determination of volatile components by comprehensive gas chromatography-mass spectrometry with time of flight analyser (GC×GC-ToFMS), determination of phenolic profile by ultra-high-performance liquid chromatography, coupled with tandem mass spectrometry (UHPLC-DAD-MSⁿ), sensorial analysis performed by a trained panel, and also determination of colour, acidity (total and volatile), SO₂ (free and total), and pH. The strategy used in this study provides new chemical data that allow evaluating the effect of the stopper among different type of wines. Physical-chemical and sensory analysis unveiled that the type of stopper modulates the characteristics of the wine, and its selection may be used as an oenological tool in the construction of the wine identity.

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SUB-CRITICAL WATER: AN ORIGINAL PROCESS TO EXTRACT ANTIOXIDANTS COMPOUNDS OF WINE LEES

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Keywords: Wine lees, antioxidant, subcritical water, response surface methodology

Wine lees are quantitatively the second most important wine by-product after grape stems and marc [1]. In order to recycle, distilleries recovered ethanol and tartaric acid contained in wine lees but yeast biomass is often unused. It has already been demonstrated that this yeast biomass could be upcycled to produce yeast extracts of interest for wine chemical stabilization [2]. In addition, it is well known that lees, during aging, release compounds that preserve wine from oxidation. Currently, very few studies have focused on the characterization and valorisation of the antioxidant component of lees. Although the role of glutathione has been demonstrated [3], recent studies have shown that S- and N- containing compounds are the main contributors to the antioxidant metabolome of wine [4]. Thus, the valorisation of wine lees to obtain compounds with antioxidant capacity seems to be of great interest for the wine industry.

In order to obtain extracts with antioxidant properties from white wine lees, we studied the interest of subcritical water as a green extraction process. The extraction conditions (temperature, extraction duration and stirring speed) were optimized by Response Surface Methodology (RSM) to maximize the antioxidant properties of the obtained extracts. The composition of the soluble fraction such as total phenolic content, protein, SH- compounds and glutathione was determined by spectrophotometry and LC-MS methods. Global antioxidant activity of extracts was determined by DPPH (radical-scavenging power), FRAP (Ferric reducing antioxidant potential) and OCR (Oxygen Consumption Rate) in model wine solution.

Results show that temperature is the key parameter for obtaining extracts with high antioxidant activity. Interestingly, we observe that the antioxidant potential does not seem to be related to the presence of a single molecule but rather to the presence of a pool of reducing compounds.

To conclude, subcritical water is a promising eco-sustainable process to obtain antioxidant compounds from wine lees. Such extracts could be used for a targeted application in oenology as well as in other sectors (food, cosmetics, pharmaceuticals).

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Sensory properties : psychophysics-cognitive psychology, experimental economy, connexions with neurosciences

SENSORY CHARACTERIZATION OF COGNAC EAUX-DE-VIE AGED IN BARRELS REPRESENTING DIFFERENT TOASTING PROCESS

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Keywords: Cognac, Sensory analysis, Barrel toasting

Cognac is an outstanding french wine spirit appreciated around the world and produced exclusively in the Nouvelle-Aquitaine region, and more precisely in the Cognac area. According to AOC regulations (Appellation D'origine Controlée), the spirit required at least 2 years of continuous ageing in oak barrels to be granted the title of Cognac. The oak wood will import color, structure and organoleptic complexity. The different steps during barrel-making process, such as seasoning and toasting, influence the above quality attributes in both wines and spirits. Barrel toasting is probably the most important step in barrel manufacturing, as it influences oak wood chemical composition, which is then likely to migrate into the wine and spirits during ageing, affecting their organoleptic properties. From a sensory point of view, no studies have been conducted focusing on the influence of barrel toasting on Cognac eaux-de-vie. Therefore, the aim of our study is to perform a sensory characterization of Cognac eaux-de-vie aged for 12 months in barrels representing different toasting levels. Eight eaux-de-vie aged in barrels with 8 different toastings were studied. The 8 toastings represented 4 different temperatures (low, medium, medium plus and high) and two toasting lengths for each temperature (one so-called "normal" and the other "slow"). Sensory analysis was carried out on these eaux-de-vie through several tests. First, a sorting test showed the differences between the samples and then training was carried out on previously chosen descriptors in order to build a sensory profile and perform a ranking test. The study was realised at an alcohol level of 40 % (v/v), which is the alcohol level of a commercial Cognac. The results showed that the eaux-de-vie are strongly impacted by the toasting of the barrel during the first year of ageing and it is possible to differenciate them according to the toasting used. Light barrel toasting results in significantly different eaux-de-vie from those aged in high-toast barrels. The latter tends to result in high quality eaux-de-vie. This study is a first step in the characterisation of Cognac eaux-de-vie aged in barrels made with different toastings and can lead to practical recommendations in order to obtain the ideal toasting temperatures that give the best olfactory and gustatory profiles in eaux-de-vie.

WINE SWIRLING: A FIRST STEP TOWARDS THE UNLOCKING OF THE WINE'S TASTER GESTURE

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Keywords: Wine swirling, Champagne, Diode Laser Sensor, CO₂

Right after the pouring of wine in a glass, a myriad of volatile organic compounds, including ethanol, overwhelm the glass headspace, thus causing the so-called wine's bouquet [1]. Otherwise, it is worth noting that during wine tasting, most people automatically swirl their glass to enhance the release of aromas in the glass headspace [1]. About a decade ago, Swiss researchers revealed the complex fluid mechanics underlying wine swirling [2]. However, despite mechanically repeated throughout wine tasting, the consequences of glass swirling on the chemical space found in the headspace of wine glasses are still barely known.

A preliminary study was thus conducted to characterize the dynamic parameters of the wine's taster gesture. From a kinematic point of view, wine swirling, on a flat support, follows an orbital motion described by its radius of gyration and its angular speed. A video processing program was developed to decipher the basic statistical parameters of this orbital motion done by a panel of 85 participants swirling INAO glasses filled with increasing levels of a water/ethanol mixture. Based on these statistical data, a homemade 3D-printed orbital shaking device was designed to replicate a standardized and repeatable glass swirling motion. Actually, In champagne and sparkling wine tasting, from the service of wine into the glass, gas-phase CO_2 was found to massively invade the glass headspace [3,4]. Therefore, the idea has emerged that gas-phase CO_2 could be considered as an ideal tracker to better understand the consequences of wine swirling on the chemical headspace inhaled by wine tasters. A spectrometer initially developed to monitor gas-phase CO_2 under static conditions was thus upgraded for the monitoring of gas-phase CO_2 in the headspace of champagne glasses automatically swirled by the 3D-printed orbital shaking device [5]. The first datasets recorded thanks to this setup show a sudden drop in the CO_2 concentration in the glass headspace, probably triggered by the liquid wave traveling along the glass wall following the action of swirling the glass.

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HYBRID GRAPEVINE CV BACO BLANC, BETWEEN TRADITION AND MODERNISM: FOCUS ON ENDOGENOUS EUGENOL AS RESISTANCE FACTOR TO *BOTRYTIS CINEREA*

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Keywords: ontogeny, inducibility, resistant hybrid vine, phenylpropanoids

The well-known antifungal and antibiotic molecule, eugenol, is widely spread in various plants including clove, basil and bay. It is also abundant in the hybrid grapevine cultivar (cv) Baco blanc (Vitis vinifera x Vitis riparia x Vitis labrusca), created by François Baco (19th century) in the Armagnac region. This study confirmed this cv as highly resistant to Botrytis cinerea by comparing fruit rot incidence and severity with two Vitis vinifera cultivars: Folle Blanche and Ugni Blanc. We have demonstrated the efficiency of eugenol in vitro, by further investigating the effect of small concentrations of eugenol, 3 to 4 ppm (corresponding to IC10), on B. cinerea. By comparing the two major modes of action (direct or volatile antibiosis), the vapour inhibiting effect of eugenol was more powerful. In the skin of Baco blanc berry, the total eugenol concentration reached a maximum at veraison, i.e. 1118 to 1478 µg/kg. Leaf removal in the bunch zone induced a significant increase of 32 % in eugenol at veraison, which was also associated with a significant decrease in B. cinerea infection in the vineyard. Eugenol, as a natural endogenous molecule of Baco blanc, is therefore an inducible compound. Very interestingly, in terms of fruit ontogenic resistance, a significant negative correlation was established between the technological maturity of berries and the total eugenol content in the berry skin. This correlation was observed on 3 plots and confirmed over several years (2021 and 2022): it therefore appears to be intrinsic to the biology of cv Baco blanc. Moreover, the temporal study of two forms of eugenol tends to hypothesise the effectiveness against B. cinerea of precursor forms of eugenol. Such bound forms are structures which are currently being researched in our laboratory. For all these reasons, eugenol appears to be a biochemical marker of ontogenic resistance in Baco blanc and presumably an important resistance factor in this old cv of renewed interest.

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I.P.2 IMPACT OF GRAPE-ASSOCIATED MOLDS IN FRESH MUSHROOM AROMA PRODUCTION

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Keywords: Mycobiot, growth modeling, volatile organic compounds (VOCs)

Mycobiota encountered from vine to wine is a complex and diversified ecosystem that may impact grape quality at harvest and the sensorial properties of wines, thus leading to off-flavors [1–3]. Among known off-flavors in wine, fresh mushroom aroma (FMA) has been linked to some mold species, naturally present on grapes, producing specific volatile organic compounds (VOC) [4–5]. The most well-known are 1-octen-3-ol and 1-octen-3-one, although many other VOC are likely involved. To better understand the FMA defect, biotic and abiotic factors impacting growth kinetics and VOC production of selected fungal species in must media and on grapes were studied.

In total, 18 fungal isolates, belonging to 7 genera and 9 species, were selected based on their off-odor production profiles on malt medium among 685 isolates from our working collection. Growth rates were measured using solid synthetic must (MS) and real must (MR) and compared to those obtained in liquid must by laser nephelometry. Sensorial analysis and VOC profiles (GC-MS) were also determined for the same isolates, individually or in co-cultures with two FMA producing *Botrytis* isolates, after growth on must and grapes.

Among the generated physiological data, optimal growth temperatures were 27–28°C, 26–30°C, 21–22°C for *Botrytis* spp., *Penicillium crocicola* and *P. citreonigrum*, respectively, depending on the isolate. Fastest growth rates were observed for *B. cinerea* and *P. crocicola*, while *Cladosporium* subtilissimum and P. *bre-vicompactum* isolates were slowest. For VOC profiles, *P. crocicola*, *P. bialowiezense* and *Clonostachys* ro-sea produced known FMA compounds (1-octen-3-one and 1-octen-3-ol) at higher levels when co-ino-culated with *Botrytis* spp. on grapes. For must trials, a species effect on VOC profiles was clearly observed (92 VOC identified). To confirm these findings, further co-inoculation studies were performed on two grape varieties (Meunier and Pinot noir) and, so far, sensorial analyses showed similar trends. Overall, this study provides novel knowledge about changes in fungal growth kinetics and VOC profiles in musts and on grapes. These results provide new insights for the wine making to better understand how FMA off-flavors are generated by molds.

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FOLIAR APPLICATION OF METHYL JASMONATE AND METHYL JASMONATE PLUS UREA: INFLUENCE ON PHENOLIC, AROMATIC AND NITROGEN COMPOSITION OF TEMPRANILLO WINES

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Keywords: elicitor, nitrogen fertilizer, quality, Vitis vinifera

Phenolic, volatile and nitrogen compounds are key to wine quality. On one hand, phenolic compounds are related to wine color, mouthfeel properties, ageing potential. and are associated with beneficial health properties. On the other hand, wine aroma is influenced by hundreds of volatile compounds. Fermentative aromas represent, quantitatively, the wine aroma, and among these volatile compounds, esters, higher alcohols and acids are mainly responsible for the fermentation *bouquet*. Finally, nitrogen compounds affect the development of alcoholic fermentation and the formation of flavour metabolites. Different approaches have been studied to improve the wine quality. Foliar application of elicitors and nitrogen compounds to vineyard has been studied to palliate the effects of climate change in grape composition, and therefore, to enhance grape and wine quality. Methyl jasmonate (MeJ) is an elicitor able to trigger a response of defense in plants, that induces the production of secondary metabolites. Urea is a nitrogen fertilizer widespread employed due to its small molecular size, higher water solubility, and low cost. MeJ and urea (Ur) have been studied separately as a foliar application in vines. Describing an enhance of volatile, phenolic and nitrogen compounds in grapes, although their effect in wines sometimes is less evident.

In this trial, three treatments were carried out as foliar application: Control, MeJ and MeJ+Ur, during two growing seasons (2019 and 2020) in Tempranillo vineyard. The analysis of phenolic and nitrogen compounds were carried out by HPLC-DAD [1, 2]. Volatile compounds were determined by SPME-GC-MS [3].

The effect of foliar treatments was season dependent. In 2019, MeJ and MeJ+Ur wines were characterized by a higher content of total acylated anthocyanins, but a lower content of total esters, alcohols and acids than control wines. MeJ+Ur wines presented a higher total amino acids content than control and MeJ wines. However, in 2020, MeJ+Ur treatment increased the total content of flavonols, flavanols, hydroxycinnamic acids, stilbenes and total amino acids when compared with control. MeJ wines presented a low content of esters and acids, whereas MeJ+Ur did not show differences with control. Overall, the synergic effect of MeJ+Ur foliar treatment was greater than the effect of MeJ application in order to improve the wine chemical composition

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I.P.4 INVESTIGATING TERROIR TYPICITY: A COMPREHENSIVE STUDY BASED ON THE AROMATIC AND SENSORIAL PROFILES OF RED WINES FROM CORBIÈRES APPELLATION

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Keywords: : red-blended-wine , molecular marker , Aroma compound , Sensorial attribute

Volatile compounds play a significant role on the organoleptic properties defining wines quality. This particular role was exploited in several studies with the aim to differentiate wines from a more or less extensive production area, according to their sensory profile ^[1], as well as their chemical composition ^[2,3] (Di Paola-Naranjo et *al.*, 2011; Kustos et *al.*, 2020). Indeed, since aroma compounds development in grapes depends primarily on the environmental conditions of the vines and grapes (soil and climate), it is conceivable that these parameters craft the aromatic signature of the wine produced, in relation to its origin (Van Leeuwen et al., 2020). In this work, a general study on the aromatic and sensorial profile of wines produced in five sub-regions of the Corbières denomination, a renowned red grape varieties viticultural region in South France, was reported. The objective of this study was to identify the aroma compounds and sensorial descriptors enabling a discrimination of the five sub-regions, and to evaluate their link with the soil and climate characteristics of the geographical areas. The analyses were carried out on two vintages (2018 and 2019) on wines produced from a blend of the four main varieties (Syrah, Grenache, Carignan and Mourvèdre). Aroma compounds were analyzed by HS-SPME-GC-MS in full scan mode and 44 compounds had significantly different concentrations among the zones. Several chemical families of compounds were highlighted as being more significantly present in wines of certain regions. The significant presence of those varietal (e.g. linalool, C13-norisoprenoids) or fermentative aromas (higher alcohols and ethyl and acetate esters) in a particular geographical area could be linked to soil features, climate vintage conditions and topographical traits (sunlight exposition, altitude, etc.). Sixteen sensorial descriptors were assessed and wines were compared by Quantitative Descriptive Analysis (QDA) profile method. Descriptors that appeared significant were linked to some aromatic compounds identified (e.g. β -damascenone and cooked red fruits) as well as related between each other (e.g. humus and amylic). In a process of subdivision of the denomination, this study allowed a first chemical and sensorial characterization of these terroirs, proposing valuable elements in the definition of the typicity of wines.

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THE EFFECT OF COPPER ON THE PRODUCTION OF VARIETAL THIOLS DURING THE ALCOHOLIC FERMENTATION OF COLOMBARD AND GROS MANSENG GRAPE JUICES

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Keywords: 3-sulfanylhexan-1-ol, copper, alcoholic fermentation, yeast

Nowadays, the rapid growth of vineyards with organic practices and the use of copper as the only fungicide against downy mildew raises again the question of the effect of copper on varietal thiols in wine, especially 3-sulfanylhexan-1-ol (3SH) and its acetate (3SHA). A few decades ago, several works indicated that the use of copper in the vineyard had a negative effect on the content of varietal thiols in Sauvignon blanc wines [1, 2]. However, these studies only considered the concentration of the reduced form (RSH) of varietal thiols, without quantifying the oxidised ones. For this purpose, we proposed to monitor both reduced and oxidised forms of varietal thiols in wine under copper stress during alcoholic fermentation to have a more complete picture of the biological and chemical mechanisms. In the present work, Colombard and Gros Manseng grape juices were fermented under different copper levels (from 0.2 to 3.88 mg/L) to mimic the consequences of organic practices on grape and must. The consumption of thiol precursors and the release of varietal thiols (both free and oxidised forms of 3SH and 3SHA) were monitored by LC-MS/MS according to previously published methods [3, 4]. It was found that the highest copper content (3.6 and 3.88 mg/L for Colombard and Gros Manseng, respectively) significantly increased yeast consumption of precursors (by 9.0 and 7.6% for Colombard and Gros Manseng, respectively). Surprisingly, this higher consumption of precursors was not associated to higher thiol concentrations. Indeed, for both varieties, the content of free thiols in the wine decreased significantly (by 84 and 47% for Colombard and Gros Manseng, respectively) with the increase of copper in the starting must, as already described in the literature [1, 2]. However, the sum "reduced+oxidized" forms of 3SH produced during fermentation was constant for the Colombard must regardless of the copper conditions, which means that the effect of copper was only oxidative for this variety. In Gros Manseng, on the other hand, the sum "reduced+oxidized" forms of 3SH increased with the copper content, up to 90%. This last result suggests that copper probably modifies the regulation of the production pathways of varietal thiols and has also a key role of oxidation. These results complement our knowledge on the effect of copper during thiol-oriented fermentation and the importance of considering both "reduced+oxidized" forms to distinguish chemical from biological effects.

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THE EFFECT OF PRE-FERMENTATIVE GLYPHOSATE ADDITION ON THE METABOLITE PROFILE OF WINE

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Keywords: Glyphosate, Viticultural Management, Wine Metabolites, Fermentation

The synthetic herbicide glyphosate has been used extensively in viticulture over many decades to combat weeds. Despite this, the possible influence of residual glyphosate on both the alcoholic fermentation of grape juice and the subsequent metabolite profile of wines has not been investigated. In this study, Pinot noir juice supplemented with different concentrations of glyphosate ($0 \ \mu g \ L-1$, $10 \ \mu g \ L-1$ and $1000 \ \mu g \ L-1$) was fermented with commercial *Saccharomyces cerevisiae* yeast strains. Using a combination of analytical methods, 80 metabolites were quantified in the resulting wines. When the pre-fermentative grape juice was spiked with glyphosate at $1000 \ \mu g \ L-1$, the concentrations of 21 of these were found to have decreased significantly in the finished wines. This study has shown for the first time that the presence of glyphosate during alcoholic fermentation by yeast influences the resulting wine metabolite profile.

WHICH TERROIR-RELATED FACTORS INFLUENCE THE MOST VOLATILE COMPOUND PRODUCTION IN COGNAC BASE WINE?

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Keywords: Aroma compounds, Grape composition, Base wine for Cognac distillation, Ugni blanc

Cognac is a famous spirit produced in southwest France in the region of the eponymous town from wines mainly from Vitis vinifera cv. Ugni blanc. This variety gives very acidic and poorly aromatic base wines for distillation which are produced according to a very specific procedure. Grapes are picked at low sugar concentrations ranging 13-21 °Brix and musts with high turbidity (>500 NTU) are fermented without sulphite addition [1]. Fermentative aromas, as esters and higher alcohols, are currently the main quality markers considered in Cognac spirits. Hence it is important to better understand the effect of potential influential factors on the production of these compounds as well as on their precursors in berries. This communication deals with the study of the impact of various terroir components among maturity, vine rootstocks, water and nitrogen status that can influence grape and corresponding wine composition. All experiments used Ugni blanc grapes and were conducted in commercial vineyards in the Cognac region as well as in the GreffAdapt plot (13 rootstocks selected) [2]. Fermentations were performed at laboratory scale in triplicate similar to Cognac base wine elaboration under harvest-like conditions and standardized conditions, where sugars and YAN were all corrected to the same values [3]. Berry composition at harvest, including detailed amino acid profile, and wine fermentative aromas, such as higher alcohols and esters, were determined. All the parameters tested here could be ranked from the most influential to the least on ester concentrations. Under harvest-like conditions, nitrogen status was found to be the most influential followed by maturity level and finally water status, which was the least impactful parameter despite a very warm and dry 2022 grape-growing season. Higher alcohol acetates were about twice higher in the high nitrogen-status vines (+ 30 mg/L of YAN) compared to the control. Under standardized conditions, maturity was found the most impactful although the initial differences in must sugars and nitrogenous compounds were smoothed, and nitrogen status was the least. Indeed, fatty acid ethyl esters differed considerably depending on maturity and their concentrations were the lowest when grapes were picked around 13-15 °Brix compared to the other two more advanced maturities. These findings highlight the importance of maturity as a key parameter for growers to take into consideration for Cognac production.

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OENOLOGICAL AND SUSTAINABILITY POTENTIAL OF WINES PRODUCED FROM DISEASE RESISTANT GRAPE CULTIVARS (PIWI WINES)

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I.P.8

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Keywords: PIWI, winemaking, social sustainability, ecological transition

The strategy for sustainability in the wine sector of the EU refers to a set of practices and principles that aim to minimize the negative impact of wine production on the environment, social and economic sustainability. Sustainable wine production involves a range of practices that are designed to reduce waste, conserve resources, and promote the well-being of workers and communities.

- 1. Vineyard management: Sustainable vineyard management involves practices that minimize the use of chemicals and pesticides, conserve water, and promote soil health
- 2. Energy efficiency: Wineries can reduce their carbon footprint by implementing energy-efficient practices, such as using renewable energy sources, investing in energy-efficient equipment, and improving insulation.
- 3. Water conservation: Water is a critical resource in wine production, and sustainable wineries seek to minimize water use through measures like drip irrigation, rainwater harvesting, and recycling wastewater.
- 4. Packaging and shipping: Sustainable wineries aim to reduce the environmental impact of their packaging and shipping practices by using recycled materials, minimizing packaging waste, and reducing transportation emissions.
- 5. Social responsibility: Sustainable wineries also prioritize social responsibility by treating workers fairly, supporting local communities, and promoting diversity and inclusion.

One of the proposed approaches is to expand the use of disease resistant hybrid grape cultivars (DRHGC) ('PIWI' grapes), and to introduce new DRHGCs, which have the potential to assist with the implementation of the European Green Deal 2050 and the EU 'Farm to Fork' strategy. DRHGCs have thus been very recently permitted for PDO wines, leading to a completely new perspective in the production of wines with protected appellation ("Regulation (EU) 2021/2117," 2021). DRHGCs are of interest since they allow for much fewer treatments in the vineyard and thus can limit the indirect negative consequences of such treatments: improved job security due to less labor in the fields; less soil compaction in the vineyard; positive impacts on responsible tourism and on neighbouring activities, particularly in the context of (perurban viticulture. However, the characteristics of DRHGCs wines are different, which makes it necessary to take measures and make changes in winemaking technology to maintain high quality. The winemaker must account for high titratable acidity, malic acid, pH, protein, polysaccharide levels and low condensed tannin levels. This can leave them vulnerable to microbial spoilage and would lower the astringency of DRHGC wines. DRHGCs often have problems due to too high yeast assimilable nitrogen leading to excessively hot fermentations. An interdisciplinary analysis is being carried on in South Tyrol where PIWI wines are cultivated, with the aim to produce a case test on different target groups: producers, retailers and buyers, hospitality workers, and consumers regarding both the environmental advantages and the particularities of wines made from DRHGCs (PIWI wines).

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PESTICIDE RESIDUES IN THE VINEYARD ENVIRONMENTS: VINE LEAVES, GRAPE BERRIES, WINES, HONEYBEES AND ASIAN HORNETS

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Keywords: analysis, pesticide contamination, QuEChERS, LC-MS/MS

Synthetic pesticides are widely used in viticulture to ensure steady harvest quality and quantity. Fungicides are primarily used to control grapevine diseases but insecticides and herbicides are likewise used. Pesticide residues in viticultural areas currently represent a strong societal concern, but may also affect different trophic chains in such areas. In this project we wish to analyse honeybees collected from hives placed in different vineyards, their natural predator (the invasive hornet Vespa velutina), as well as the honey, grape berries, and wines produced. In order to screen the different pesticides found in our study areas, it was first necessary to optimize the extraction procedure. Pesticide residues in plant matrices such as leaf or grape berry are regularly monitored at the ISVV using QuEChERS (Quick, Easy, Cheap, Effective, Rugged and Safe) extraction followed by LC-MS/MS analysis. A QuEChERS method was adapted based on previously published work in order to analyse honeybees and single hornets, for which the quantity of samples is limited. The method was improved using a zirconium-based sorbent for d-SPE, which is used to reduce the matrix effect in lipidic commodities. The performance of this developed method was evaluated for 42 pesticide residues. A significant matrix effect was however noted for some molecules, thus procedural calibration was used to quantify pesticide residues in real samples. Methodological developments and pesticide residue quantification results in various matrices will be presented.

I.P.10 ACCUMULATION OF GRAPE METABOLITES IS DIFFERENTLY IMPACTED BY WATER DEFICIT AT THE BERRY AND PLANT LEVELS IN NEW FUNGUS DISEASE-TOLERANT GENOTYPES

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Keywords: water deficit, primary metabolism, anthocyanins, thiol precursors

The use of new fungus disease-tolerant varieties is a promising long-term solution to better manage chemical input in viticulture, but unfortunately little is known regarding these new hybrids fruit development and metabolites accumulation in front of abiotic stresses such as water deficit (WD). Thus, prior to the adoption of such varieties by the wine industry in Mediterranean regions, there is a need to consider their suitability to WD.

This study aimed to characterize, from 2019 to 2021, 6 new fungus disease-tolerant varieties selected by INRAE (Floreal, G5 and 3159B for white grapes and Artaban, 3176N and G14 for red grapes) in comparison to *V. vinifera* Syrah. A gradient of WD was applied and followed by weekly measures of predawn water

potentials. Grape development was non-destructively monitored to determine the arrest of berry phloem unloading, moment at which all grapes were harvested, as way to objectify the sampling date at a precise physiological landmark. Primary metabolites (glucose, fructose, tartrate, malate and yeast assimilable nitrogen) and main cations (K⁺, Mg²⁺, Ca²⁺, Na⁺, NH₄⁺) were assessed by HPLC and enzymatic methods. Secondary metabolites as anthocyanins and thiol precursors were assessed by HPLC-UV and LC-MS/ MS, respectively.

Genotype was the main factor explaining the variations in metabolites and cation concentration in berries at the ripe stage. At the phloem unloading arrest, primary metabolites and main cation concentrations were the lowest in G14 and the highest in Floreal and Syrah. Regarding secondary metabolites, all genotypes showed higher values than the *V. vinifera* Syrah. Yet, the red hybrid 3176N emerged as the richest genotype in both anthocyanins and total thiol precursors, reaching values of 1609 mg/L and 539 µg/kg respectively. Despite the low contribution of WD to metabolite concentrations, it consistently reduced the total accumulation of primary and secondary metabolites per berry and per plant, with different intensities depending on the genotype. Our results show that WD can ultimately reduce the production of metabolites per unit of fruit and per plant without significantly improving the concentration of compounds of interest in the grape.

AROMA AND SENSORY CHARACTERIZATION OF XINOMAVRO RED WINES FROM DIFFERENT GREEK PROTECTED DESIGNATIONS OF ORIGIN, EFFECT OF TERROIR CHARACTERISTICS

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Keywords: GC-MS, sensory analysis, terroir, typicity

The quality of wines has often been associated with their geographical area of production. The aim of this work was to characterize Protected Designation of Origin (PDO) Xinomavro red wines from different geographical areas of Amyndeon and Naoussa in *Northern Greece*, elaborated with variables that contribute to their differentiation, such as soil characteristics, altitude, monthly average temperature and rainfall.

Xinomavro fruit parcels from different vineyards within the two PDO zones (5 PDO Naoussa and 6 PDO Amyndeon) were vinified following a standard winemaking process. A total of 25 aroma compounds were quantified using gas chromatography-mass spectrometry (GC-MS) with simultaneous full scan and selected ion monitoring for data recording, and odor activity values (OAVs) were determined. A trained panel evaluated the wines using sensory descriptive analysis, rating a total of 13 aroma attributes.

According to the quantitative data, a complex aroma profile rich in higher alcohols, ethyl esters, acetate esters and fatty acids, with a contribution of terpenes and volatile phenols was recorded. Statistical data analysis techniques, ANOVA and Principal Component Analysis, showed the structure of the experimental data and the significant differences for each compound in the different wines. PDO Amynteon wines presented higher concentrations in 1-hexanol and higher intensity of green bell pepper attribute, while PDO Naoussa wines were higher in ethyl octanoate, ethyl 2-methylbutyrate and eugenol, with higher scores in berry fruit and spices attributes. Terroir and meso-climate characteristics correlated well with the data obtained and helped identify how typical aromas could be an expression of terroir.

This study provides an approach to the chemo-sensory fingerprinting of Xinomavro PDO wines. It will be further used to advance the understanding of how impact aroma compounds and sensory characteristics shape terroir expression and how this could be manipulated by viticultural and winemaking practices, under current and future climatic conditions.

CLIMATE CHANGE EFFECT ON POLYPHENOLS OF GRIGNOLINO GRAPES (*VITIS VI-NIFERA L*.) IN HILLY ENVIRONMENT

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Keywords: meteorological conditions, vineyards age, vineyard aspect, grape polyphenols

Current changes of ecoclimatic indicators may cause significant variation in grapevine phenology and grape ripening. Climate change modifies several abiotic factors (e.g. temperature, sunlight radiation, wa-ter availability) during the grapevine growth cycle, having a direct impact on the phenological stages of the grapevine, modulating the metabolic profile of berries and activating the synthesis and accumula-tion of diverse compounds in the skin of berries, with consequences on the composition of the grapes.

The influence exerted by different meteorological conditions, during three consecutive years (2020-2022) on secondary metabolites such as the polyphenolic profile of Grignolino grapes was investigated. The samples were collected from three vineyards characterized by different microclimatic conditions mainly related to the vineyard aspect and to a different age of the plants.

Significant variations were observed in temperature and rainfall patterns among the different vintage years. The response of grape secondary metabolism to abiotic stress, with particular emphasis on polyphenolic profile of the berries was evaluated using spectrophotometry and HPLC.

The results highlighted significant differences among the vintages for each vineyard as to the berry weight and the contents of the several classes of polyphenols. 2022 Vintage distinguished by a series of extreme conditions in terms of high temperature and low rainfall, showed low berry weight and skin contents of almost all polyphenols, especially for the southwest exposed vineyards. The seeds, on the other hand, exhibited higher amounts of polyphenols, possibly due to their greater extractability. Berry weight being equal, fresh conditions during the green phase until veraison, followed by dry and hot pre harvest period privileged phenolics mainly in the berries of the southeast vineyard. Younger vineyard showed more result variability through the years.

The intensity and mainly the timing of meteorological fluctuations affected the final content in phenolics of Grignolino grapes. In hilly environment, conditions of limited water availability and high temperature, that lately are characterizing the second part of berry development, seem to support the grape quality of Grignolino, a cultivar of medium-late ripening, by limiting the differences on bunch ripening, allowing a greater accumulation of secondary metabolites, but maintaining at the same time an optimum balance sugar/acidity.

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DOES LIGNIN AN ACCEPTABLE MARKER OF GRAPESEED MATURATION AND QUALITY?

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Keywords: Grapeseed, maturation, biochemistry

Usually the winemaker consider polyphenols from the grape berry as an actor of the wine quality. There are frequently consider as a marker of grape maturity. It is commonly known that winemaker consider tannins and anthocyanins as main polyphenol actors for winemaking practices and wine quality. Here we will focus on the characterisation of lignins in grape seeds. Previous studies suggest that the seed is lignified [1], which could explain the change in colour of the seed when it reaches maturity and thus provide a reliable indicator for describing the maturity stage in the seed. Furthermore, lignin is contained in the outer envelope of the seeds [2] and would constitute a mesh that affects the extractability of tannins and therefore the quality of the wine obtained following the winemaking process [3], since these are mainly responsible for the astringency and bitterness in the wine. In order to provide initial answers on the establishment of lignin in the seed, as well as on the quantity and type of lignin found in the seed, a preliminary two-stage study was conducted.

In a first stage, the lignin biosynthesis pathway was studied using qPCR approach with a focus on key genes of the lignin pathway (PAL, 2 isoforms of COMT, CCOAMT, F5H and 2 isoforms of CAD).The analysis of level of transcripts show a differential regulation and timing of transcripts accumulation depending of the stage of maturity and the vintage studied.

In a second step, it was undertaken to identify and quantify the different lignin monomers present in the grapeseed. For this purpose, an extraction of lignins was carried out with an ethanol:toluene, ethanol, water sequence on seed powder. In order to determine the lignin content after extraction, an acetyl bromide procedure was performed as well as a thioacydolysis protocol to cleave the β -O-4 bonds of the lignin polymer and release the different lignin monomers G from guaiacyl, S from syringyl and H from p-hydroxyphenyl. Their identification and quantification was undertaken by HPLC-MS.

This first work on lignin determination in grapeseed give a solid baseline to go further in the comprehensive way to transfer of oenological molecules from grapeseed to must and wine almost in the context of climate change.

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EFFECTS OF BIODYNAMIC VINEYARD MANAGEMENT ON GRAPE RIPENING MECHANISMS.

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Keywords: Biodynamics, viticulture, grapes, maturity

Biodynamic agriculture, founded in 1924 by Rudolph Steiner, is a form of organic agriculture. Through a holistic approach, biodynamic agriculture seeks to preserve the diversity of agriculture and the existing interactions between the mineral world and the different components of the organic world. Biodynamic grape production involves the use of composts, herbal teas and mineral preparations such as 500, 501 and CBMT.

Several scientific studies have provided evidence on the effects of biodynamic farming on the soil, the plant and the wine. Numerous empirical opinions of wine growers support the existence of differences brought by such a management.

The objective of our study is to build a scientific experiment to validate this knowledge and opinion by providing understanding of the biological behavior of the plant and the grape, and, finally, of the differences observed on the wine.

Our trial aims at evaluating the effects of a biodynamic management on the mechanisms of grape ripening. It is conducted on 8 parcels of the Lafite Rothschild vineyard, 4 of which have been receiving biodynamic preparations since 2017 and 4 not.

The parameters of technological (sugars, TA, malic acid, tartaric acid, pH), phenolic (glories method), and textural maturity (Penetrometry, Aw) of the berries from veraison onwards were monitored in the 8 plots. The content of polysaccharides and pectin was analyzed during the ripening period on the grape skins. Once harvested, the berries were tasted by a trained panel.

Depending on the parameters, differences were observed and seemed to confirm the empirical vision of biodynamic practitioners.

Further analyses will have to be carried out to confirm these observations and evaluate the mechanisms involved.

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EVALUATION OF A SEAWEED EXTRACT OF *RUGULOPTERYX OKAMURAE* AGAINST *ERYSIPHE NECATOR* IN GRAPEVINE

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Keywords: sustainability, Fungicides, seaweed, circular economy

Powdery mildew, caused by *Erysiphe necator*, is a widespread disease that causes high economical losses in viticulture. The main strategy to control the disease is the recurrent application of sulphur based phytochemical compounds. However, in order to reduce their accumulation in the environment and promote the sustainability of the sector, the European Commission has applied restrictions to the number of pesticide treatments and the maximum quantity of fungicides to be applied in viticulture. Seaweeds, in particular macroalgae, are marine resources rich in sulphated polysaccharides with bio-protective potential for the plant, representing an environmentally-friendly alternative approach for sustainable wine production.

In the current work we investigated the antifungal efficacy of the brown invasive macroalgae *Rugulop-teryx okamurae* against *Erysiphe necator*. *Rugulopteryx* was collected from Algeciras coast (South Spain) and an aqueous extract was developed using a water /ethanol extraction protocol. A foliar spraying (6 gr/L) of *Rugulopteryx okamurae* extract was applied to Tempranillo and Cabernet Sauvignon plants grown in a green-house facility and the antifungal activity of the extract was tested by monitoring disease symptoms after fungi infection. Results showed that while the incidence of powdery mildew was similar in treated than in control plants (water treated), the disease severity was 1.7 fold lower for treated plants in comparison to controls. Further research by exploring grapevine resistance/defence mechanisms is necessary to explain this extract 's mode of action.

Evidencing the efficacy of *Rugulopteryx okamurae* as a biostimulant/fungicide is a finding of major importance, as it would be a first step towards its inclusion in a circular scheme, reducing its accumulation on the coast and at the same time benefiting the wine sector.

EXPLORING RED WINE TYPICITY OF CORBIÈRES: EVALUATION OF THE DEGREE OF IMPACT OF VINIFICATION PROCESS ON THE CHEMICAL COMPOSITION AND ORGANOLEPTIC PROPERTIES OF WINES FROM DIFFERENT TERROIR

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Keywords: red-blended-wine, vinification process, aroma compounds, polyphenols

It is important nowadays for wine producers to create a product that is an expression of their terroir, a concept including the interaction between a place (topography, climate, soil), the people (tradition, winemaking and viticultural practices) and the resulting product (grape varieties, wines)^[1]. Nonetheless, wine's typicity linked to those terroirs must be easily recognizable by consumers thanks to distinctive sensory characters and composition ^[2]. Among the compounds of interest, aromatic compounds and polyphenols play an important role in the quality of red wines, by impacting on the odour, color and astringency. To explore the influence of terroir factors, including climate, soil and human practices, on the chemical and sensory profile of wines, red wines from five terroirs of the Corbières appellation were subjected to a general study approach. The analyses were done on two vintages (2018 and 2019) and wines were characterized by a blend of the four main varieties (Syrah, Grenache, Carignan and Mourvèdre). A comprehensive analysis approach was adopted for the study of wines' aromatic, color and astringency profiles. Volatile compounds were evaluated by HS-SPME-GC-MS, while wine pigments and derived pigments were assessed through spectrophotometric measurements. Moreover, wines were compared through a Quantitative Descriptive Analysis (QDA) sensorial profile method. The aim was to identify the "molecular markers" that could characterise the different wines and to assess whether these markers were related to each other and explained by their area of origin. In this study winemaking parameters were also considered and multifactorial analyses were performed to link these data to the chemical and/or sensory profiles. Results found for the Color Intensity (CI), the Total Polyphenol Index (IPT) and the nuance could be explained by the percentage of blending, color extraction techniques and ageing time. Differences in the aroma profile were mainly attributed to some fermentative and certain varietal aromas. Sensorial descriptors that appeared significant were related between each other, as well as to some aromatic and polyphenolic features highlighted (red fruits, IPT and astringency). This study could allow a first analytical characterization of five terroirs, proposing valuable elements in the definition of the typicity of wines.

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FLAVONOID POTENTIAL OF MINORITY RED GRAPE VARIETIES

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Keywords: Flavonoid profile, minority grapes, HPLC-DAD-MSn and HPLC-MS-MRM, Chemotaxonomic markers

The alteration in the rainfall pattern and the increase in the temperatures associated to global climate change are already affecting wine production in many viticultural regions all around the world (1). In fact, grapes are nowadays ripening earlier from a technological point of view than in the past, but they are not necessarily mature from a phenolic point of view. Consequently, the wines made from these grapes can be unbalanced or show high alcohol content. Dramatic shifts in viticultural areas are current-ly being projected for the future (2). However, these wine-growing areas have the potentiality to stay in place if they shift from the "international" varieties to autochthonous varieties, usually better adapted to the local climate of the growing area (3). In the Spanish "Castilla y León" region, an important number of minority *Vitis vinifera* L varieties have been identified and conserved in a germplasm bank. It is therefore interesting to study their potential to make quality wines. For this purpose, the present study aimed at determining by HPLC-DAD-MSn (4) and HPLC-MS-MRM (5) the anthocyanin, flavonol and flavanol composition of some of them (Mandón de Zamadueñas, Mandón de Arribes, Gajo Arroba, Tinto Jeromo, Bruñal, Merenzao, Estaladiña and Cenicienta) coming from different parts of Castilla y León.

Quantitative differences were observed in the total anthocyanin contents and in the proportions of individual pigments. Malvidin derivatives prevailed over the rest of the anthocyanins in all cases, but in Merenzao and Estaladiña grapes, the proportion of the latter ones were greater than in the other varieties. Varieties also differed in the *p*-coumaroyl/acetyl derivatives ratio and in the proportion of caffeoyl derivatives. Flavonol total content and profile also changed among varieties, with myricetin and quercetin derivatives being the most abundant ones. Flavanol profile, which has been reported to be less useful for chemotaxonomic purposes than anthocyanin and flavonol profiles, was also different even for varieties coming from the same part of the Castilla y León region, highlighting again the existence of varietal differences in flavonoid composition. The knowledge of the flavonoid composition of these red grape minority varieties will be helpful for enologists to adapt the winemaking process to exploit the potential of each variety and to obtain quality wines from the natural resources of the region.

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FREE TERPENE RESPONSE OF 'MOSCATO BIANCO' VARIETY TO GRAPE COLD STORAGE

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Keywords: grape cold storage, aroma, terpenes, Muscat varieties

Temperature control is crucial in wine production, starting from grape harvest to the bottled wine storage. Climate change and global warming affect the timing of grape ripening, and harvesting is often done during hot summer days, influencing berry integrity, secondary metabolites potential, enzyme and oxidation phenomena, and even fermentation kinetics. To curb this phenomenon, pre-fermentative cold storage can help preserve the grapes and possibly increase the concentration of key secondary metabolites.

In this study, the effect of grape pre-fermentative cold storage was assessed on the 'Moscato bianco' white grape cultivar, known for its varietal terpenes (65% of free terpenes represented by linalool and its derivatives) and widely used in Piedmont (Italy) to produce Asti DOCG wines. The study involved two experiments: a 12 h short-term storage under fresh (15 °C) and sunny outdoor (peak of 43 °C) conditions, and a medium-term storage under five different temperatures (5, 7, 12, 17, 19 °C) and durations (12, 24, 54, 84, 96 h), according to a Central Composite Design then evaluated using response-surface methodology (RSM). Berry skin break force mechanical property and juice physiochemical parameters were analyzed, as well as juice free terpene compounds using GC-MS.

In the short-term trial, after 4 and 8 h of storage the cooled sample showed a higher concentration of linalool, but at the end of the storage (12 h, when external temperature dropped to 25–20 °C after sunset), an opposite situation was found, possibly indicating a higher terpene solubilization in their thermoto-lerance defense role.

The medium-storage experiment indicated that the sum of the 13 detected terpenes in grape juice significantly decreased progressively after 75 h of storage, particularly in samples stored at the highest temperature tested (19 °C). However, the RSM model indicates that storage times shorter than 50 h contributed to higher terpenes, as well as the increase in storage temperature. The berry skin break force was not affected significantly by the treatments.

In conclusion, grape cold storage may offer several advantages in winemaking, but further studies are needed on this variety for assessing the best storage temperature and length conditions, as well as for the comparison between free and glycosidically-bound terpenes in juice and in the resulting wine.

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IMPACT OF NEW BIO STIMULANTS ON GRAPE SECONDARY METABOLITES UNDER CLIMATE CHANGE CONDITIONS

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Keywords: aroma precursors, polyphenols, Vermentino, Sangiovese

In a context of climate change and excessive use of agrochemical products, sustainable approaches for environmental and human health such as the use of bio stimulants in viticulture represent a potential option, against abiotic and biotic threats. Bio stimulants are organic compounds, microbes, or a combination of both, that stimulate plant's vital processes, allowing high yields and good quality products. In vines, may trigger an innate immune response leading to the synthesis of secondary metabolites, key compounds for the organoleptic properties of grapes and wines.

During this research the prospect of foliar application of bio stimulants to improve the aromatic and polyphenolic potential of the grapes was investigated in two consecutive years, characterized by hot and dry summers. Two different products, prepared with specific fractions of inactivated yeasts, were compared, and applied in different points during veraison with two- or three-time application protocol. The experiment involved two cultivars cultivated in Tuscany, a white (Vermentino) and a red one (Sangiovese). Quali-quantitative determination of the aromatic composition of the grapes was carried out using GC-MS, whereas polyphenols in skins and seeds were analyzed by spectrophotometry and HPLC methods.

The bio stimulants did not affect the vine yield, but higher berry weight and reduced sugar contents were noted at harvest in the grapes from treated with respect to the control vines. All treatments enhanced polyphenolic potential in berry skins of red grapes, whereas modifications on anthocyanins percentages and reduction of flavonols were also observed, suggesting a protective effect of the treatments against solar radiation stress. Moreover, grapes from treated vines differ significantly for the lower content of polyphenolic compounds in their seeds. As regards aroma precursors, three-time application triggered significantly higher contents for almost all chemical classes of aromatic precursors for Vermentino whereas all treatments enhanced the accumulation of terpenoids and benzenoids in the berries of Sangiovese.

Bio stimulants thus, improved the qualitative parameters of the grapes, but their effect was different based on the frequency and the timing of the application, the chemical class of the compounds and the cultivar examined. Further future investigation is necessary to optimize bio stimulant application to contrast stress conditions and improve grape quality.

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I.P.20 NEAR INFRARED SPECTROSCOPY FOR THE ESTIMATION OF TEMPRANILLO BLANCO VOLATILE COMPOSITION ALONG GRAPE MATURATION

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Keywords: grape aromatic composition, NIR spectroscopy, non-destructive, TF-SPME

Grape volatile compounds are mainly responsible for wine aroma, so it is important to know the varietal aromatic composition throughout ripening process. Currently, there are no tools that allow measuring the aromatic composition of grapes, in intact berries and periodically, throughout ripening, in the vineyard or in the winery. For this reason, this work evaluated the use of near infrared spectroscopy (NIR) to estimate the aromatic composition and total soluble solids (TSS) of Tempranillo Blanco berries during ripening. For this purpose, NIR spectra (1100–2100 nm) were acquired from 240 samples of intact berries, collected at different dates, from veraison to overripening. From these same samples, the concentration of volatile compounds was analyzed using Thin Film–Solid Phase Microextraction–Gas Chromatography–Mass Spectrometry (TF–SPME–GC–MS), and the TSS were quantified by refractometry. Calibration, cross–validation and prediction models were built from spectral data using modified partial least squares regression (MPLS). Determination coefficients of cross–validation (R²CV) above 0.5 were obtained for all volatile compounds, their families, and TSS. These findings support that NIRS can be successfully use to estimate the aromatic composition as well as the TSS of intact Tempranillo Blanco berries in a non–destructive, fast, and contactless form, allowing simultaneous determination of technological and aromatic grape maturities.

PHENOLICS DYNAMICS OF BERRIES FROM *VITIS VINIFERA* CV SYRAH GRAFTED ON TWO CONTRASTING ROOTSTOCKS UNDER COMBINED SALINITY AND WATER STRESSORS AND ITS EFFECT ON WINE QUALITY

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Keywords: phenolics, rootstocks, combined stress, wineberry quality

Wine regions are getting warmer as average temperatures continue raising affecting grape growth, berry composition and wine production. Berry quality was evaluated in plants of Vitis vinifera cv Syrah grafted on two rootstocks, Paulsen (PL1103) and SO4, and grown under two salinity concentrations (LS:0.7dS/m and HS:2.5dSm⁻¹) in combination with two irrigation regimes (HW:133% and CW:100%), being the seasonal water application 483mm (control, 100%). Spectrophotometer measurements from berry skin during veraison and harvest stages and from "young" wine samples, were indicative of the stressors effect and the mediation of the rootstocks. At veraison (i) total phenolics content were high under LSHW (0.7dSm⁻¹ and high water conditions) for SO4 and PL1103. (ii) Tannins were higher in SO4 under LSHW and in PL1103 under HSCW (2.5dSm⁻¹ and control water conditions). (iii) Higher carotenoids were found at HSCW for both rootstocks. At harvest: (i) total phenolics content decreased dramatically from veraison to harvest stage under high salinity in both rootstocks. Phenolic content decreased by 34% in SO4 and 32% in PL1103. Under LS (0.7dSm-1) total phenolics content decreased by 29% in both rootstocks. (ii) Tannins in SO4 were higher under LSCW (0.7dSm-1 and control water conditions) while in PL1103 were higher under HSHW (2.5dSm-1 and high water conditions). (iii) Carotenoids highly accumulated under HSHW in both rootstocks. In young wine samples: (i) total phenolics content was higher in wines made from berries under HSCW in SO4 while in PL1103 was higher under LSCW treatment. (ii) Tannin content was higher in the wine made from berries under HSHW from SO4 and with berries from PL1103 grafts under LSCW. (iii) For carotenoids the highest content was found in wines made with SO4 under LSCW and with PL1103under HSCW. In conclusion, our results show a clear mediating effect of the rootstock on Syrah berry metabolism and wine quality. This data should be considered when planning the use of reclaimed water in irrigation strategies or when growing plants in saline soils. Moreover, graft tolerance and mediating effects on berry metabolism might not be consistent, requiring a compromise between yield and quality.

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I.P.22 REGULATION OF CENTRAL METABOLISM IN THE LEAVES OF A GRAPE VINES VA-RIETAL COLLECTION ON A TEMPERATURE CLINE

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Keywords: Grape (Vitis vinifera), high temperature, metabolite, transcript profiling

Grape (Vitis vinifera) is one of the world's oldest agricultural fruit crops, grown for wine, table grape, raisin, and other products. One of the factors that can cause a reduction in the grape growing area is temperature rise due to climate change. Elevated temperature causes changes in grapevine phenology and fruit chemical composition. Previous studies showed that grape varieties respond differently to a temperature shift of 1.5°C; few varieties had difficulties in the fruit development or could not reach the desired Brix level. In this study, six grapevine varieties (Syrah, Petit Syrah, Petit Verdot, Tempranillo, Sangiovese, and Pinot Noir), grown in Ramat Negev (30°58'43.4"N 34°42'31.6"E, 300 m asl and 79.4 mm rainfall) experimental vineyard showing different sugar accumulation patterns between temperature regimes were studied during a heatwave event. The physiological activities of these varieties were measured at three different times (7am, 12pm and 6 pm) during the heatwave. GC-MS based metabolite profiling and targeted transcript analysis were used to study the central metabolism in leaves in response to increasing temperature from morning to evening. Results showed that Pinot Noir had higher rates of transpiration, stomatal conductance and photosynthetic assimilation compared to Syrah. The metabolite profiling analysis revealed that the metabolic activity was generally higher in the morning for all varieties, decreasing during noon and evening. This research provides valuable insights into the impact of global warming on grapevine metabolism and the potential implications for wine production.

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SHIRAZ FLAVONOID EXTRACTABILITY IMPACTED BY HIGH AND EXTREME HIGH TEMPERATURES

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Keywords: Extractability, High temperature, Flavonoids, Tannins

Climate change is leading to an increase in average temperature and in the severity and occurrence of heatwaves, and is already disrupting grapevine phenology. In Australia, with the evolution of the weather of grape growing regions that are already warm and hot, berry composition including flavo-noids, for which biosynthesis depends on bunch microclimate, are expected to be impacted [1]. These compounds, such as anthocyanins and tannins, contribute substantially to grape and wine quality. The goal of this research was to determine how flavonoid extraction is impacted when bunches are exposed to high (>35 °C) and extreme high (>45 °C) temperatures during berry development and maturity. The sole effect of temperature was investigated on well-irrigated potted Shiraz grapevines grown in a glasshouse, where either the whole vine or bunches-only were heated using fans. For both experiments, berries were sampled at harvest, peeled, ground and total flavonoids were extracted using 60% acetone [2]. Two additional assays evaluated the potential temperature impact on subsequent wine composition using wine-like extraction (15% ethanol) [3] or micro-scale winemaking. Detailed tannin composition was primarily determined by LC-MS/MS after phloroglucinolysis [2], with complementary total tannin concentration (methyl cellulose precipitable assay). Secondary metabolites such as phenolic acid and anthocyanins were also analyzed.

The present work showed that short spells of high temperature may not impact on skin and seed tannin extractability when assessed on visually undamaged berries by harvest. Indeed, while total skin tannin concentrations, extracted with 60% acetone, were clearly reduced by a rise of temperature around véraison, skin extractable tannin (15% ethanol) and seed tannin concentrations were not impacted. In damaged berries at harvest, skin tannins were dramatically reduced while seed tannins were mostly preserved. Wine quality, made with a mix of heat-damaged and undamaged berries, was significantly reduced when about 20% (by mass) of the berries were visually damaged and necrotic, corresponding to about 50% of damaged berries (in number). Maintaining wine quality under a changing climate with more frequent extreme events leading to heat stress and/or water stress is challenging. However, this study showed that the impact of heatwaves in the vineyard may be compensated by a better extraction during winemaking and require further investigations at winery scales.

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REDUCING NITROGEN FERTILIZATION ALTERS PHENOLIC PROFILES OF *VITIS VINIFERA L.* CV. CABERNET GERNISCHT WINE OF YANTAI, CHINA

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Keywords: Cabernet Gernischt, Vitis vinifera, Nitrogen, Phenolic composition

Nitrogen (N) fertilizer is important for grape growth and the quality of wine. It is essential to address the mismatch between N application and wine composition. Cabernet Gernischt (Vitis vinifera L.), as one of the main wine-grape cultivars in China, was introduced to Yantai wine region in 1892. This grape cultivar is traditionally used for quality dry red wine with fruit, spices aroma, ruby red and full-bodied wines. In order to regulate vine growth and improve grape and wine quality, Cabernet Gernischt grapevines were subjected to decreased levels of N treatments, compared to normal N supply treatment, during grape growing seasons of 2019 and 2020. Three N application treatments were imposed from leaf development to verasion: The normal N treatment corresponding to the control (N2), reducing N application by half treatment and no N application treatment corresponding to N1 and N0, respectively. Individual phenolics were determined by UHPLC-MS/MS. The result showed that reducing N had significantly decreased shoot pruning weight and yield, but the effect on fruit ripening was depending on season. N reduction treatment significantly improved wine phenolic parameters including total phenolic, tannnins and anthocyanins, and enhanced most of individual anthocyanins, and some non-anthocyanin phenolics especially stibenes including piceatannol, trans-resveratrol and polydatin, regardless of season. The overall results highlighted the importance of reducing N application during grape growing season in modifying wine phenolic profiles.

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EVALUATION OF THE OENOLOGICAL POTENTIAL OF NEW RESISTANT VARIETIES MEETING TYPICAL BORDEAUX CHARACTERISTICS

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Keywords: varietal potential, microvinification, resistance, typicity

Varietal innovation is a major lever for meeting the challenges of the agro-ecological transition of vineyards and their adaptation to climate change. To date, selection work has already begun in the Bordeaux region through the Newvine project. The aim of this project is to create new vine varieties with resistance to mildew and powdery mildew, adapted to the climatic conditions of the Bordeaux region and enabling the production of wines that are in line with consumer tastes and the expected typicity of Bordeaux wines.

Crosses were made by INRAE Colmar and IFV (UMT Genovigne) between varieties carrying 2 genes of resistance to mildew and 2 to powdery mildew, and Petit Verdot or Cabernet Franc grape varieties. 168 genotypes resulting from these crosses were planted on a plot in the Bordeaux vineyard with 5 vine plants per genotype. This system allowed to study the monitoring of certain agronomic aptitudes for a period of 3 vintages: resistance to mildew and powdery mildiou, sensitivity to other bio-aggressors, phenology, bearing, production, maturity.

In this study, the varietal oenological potential of 168 clones (grapes, wines) has been evaluated during the 2022 vintage by integrating 1) the study of the grape ripening characteristics and the composition of musts at harvest 2) by adapting the winemaking modalities for the part of new varietal creations which have been selected for vinification 3) by assessing the sensory quality and analytical composition of the wines made from 68 varieties.

The winemaking conditions were adapted to allow the fermentative monitoring of a large number of samples (12 white clones and 56 red clones) and the production of wine according to traditional Bordeaux methods.

All of these wines have been chemically analysed and then subjected to a sensory analysis by an expert panel. To assess their oenological potential, a jury of experts rated each wine sensory characteristic according to its quality, its typicality and characterized the presence of any faults. The results show a great variability between the varieties in terms of ripening profile, grape and wine composition and the diversity of sensory profiles of the wines (some atypical or marked by alterations, others presenting typicity close to the expected profiles).

This project will capitalize on the information necessary for the selection of varieties that meet the objectives and the implementation of devices to acquire the data necessary for the registration and classification of these varieties.

I.P.26 INFLUENCE OF GRAPE RIPENESS ON MACROMOLECULES EXTRACTABILITY FROM GRAPE SKIN TISSUES AND GRAPE SEEDS DURING WINEMAKING

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Keywords: ripeness, grape, extractability, wine, stability

A consequence of climate change is the modification of grape harvest quality and physico-chemical parameters of the obtained wine: increase in alcoholic degree, decrease in pH, and modification of the extractability of macromolecules, which leads to problems of microbiological, tartaric, colour and colloidal stability. In order to respond to these problems, the winemaking processes must be anticipated and adapted with a better knowledge of macromolecule extractability in grapes and their evolution, according to the grape variety, vintage and winemaking process. The purpose of this study was to understand 1) how the harvest date can influence the extractability of macromolecules, polysaccharides and phenolic compounds, which are responsible for wine stability 2) how to adapt the winemaking process to the harvest date in order to optimise wine quality

In this study, the extraction of grape seeds and grape skins macromolecules in wine has been investigated during the 2020 and the 2021 vintage at different grape maturity stages. At each maturity stage, three winemaking modalities have been produced: (i) a control modality (whole berry), (ii) a grape seeds modality made of exclusively with seed and (iii) a grape skins modality made of exclusively with skins. The evolution of skin, seed and wine macromolecules content (such as polysaccharides and phenolic compounds) has been followed during the winemaking, from alcoholic fermentation to post-fermentative maceration.

The findings have shown that grape berry ripeness influences the extractability levels, types and kinetics of macromolecules.

ASSESSMENT OF GRAPE QUALITY THROUGH THE MONITORING OF PHENOLIC RIPENESS AND THE APPLICATION OF A NEW RAPID METHOD BASED ON RAMAN SPECTROSCOPY

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Keywords: Grape quality, Phenolic ripeness, Anthocyanins, Red wines

The chemical composition of grape berries at harvest is one of the key aspects influencing wine quality and depends mainly on the ripeness level of grapes. Climate change affects this trait, unbalancing technological and phenolic ripeness, and this further raises the need for a fast determination of the grape maturity in order to quickly and efficiently determine the optimal time for harvesting. To this end, the characterization of variety-specific ripening curves and the development of new and rapid methods for determining grape ripeness are of key importance.

As part of this ongoing project, 35 vineyards (26 cv. Nebbiolo, 9 cv. Barbera) from Langhe, Roero, and Monferrato terroirs (Piemonte, Italy) were monitored during two consecutive vintages (2021–2022). The Nebbiolo vineyards were further classified, based on historical data, into ripening classes according to the harvest period estimation (early, medium, and late Nebbiolo). To study the evolution of grape ripening, four grape samples were taken from each vineyard during the ripening period (mid-August - late September), and grape quality assessment was performed by means of parameters commonly used in wine industry: juice technological maturity and phenolic ripeness parameters (total and extractable anthocyanins-EA%, share of tannins from seeds-Mp%). Preliminary results showed differences among cultivars and ripening classes, with a strong influence of the climatic conditions of the vintage, being both hot vintages with a strong water deficit (and decrease in berry weights and anthocyanin accumulation) for the 2022 vintage.

To have a more in-depth insight into the phenolic changes of the grapes during ripening, total extractions of the skins and seeds phenolics were carried out to better characterize the composition of Nebbiolo and Barbera berries. Lastly, this data was used to train a new approach based on Raman spectroscopy (RS), in an attempt to develop a method for the rapid determination of berry quality. At each sampling point, the acquisition of the grape Raman spectra was carried out in parallel with the other chemical analyses, developing a prediction model by correlating technological and phenolic ripening parameters with RS results.

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WHAT'S FUTURE FOR SANTORINI'S VITICULTURE IN THE CONTEXT OF CLIMATE CHANGE

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Keywords: Assyrtiko, Kouloura, Climate change, Greece

The own-rooted vineyard of Santorini is a unique case of vineyard worldwide that is been cultivated for thousands of years. On the island's volcanic soil, the vines are still cultivated with traditional techniques, which are adapted to the specific and extreme weather conditions that prevail on it. While climate change is a reality in the Mediterranean region, will Santorini vineyard endure its impact? The study of the traditional training systems, techniques and vine density, as well as the application of sustainable solutions (cover crops and use of kaolin etc.) revealed sustainable methods for the adaptation of the local viticulture to new climatic phenomena that tend to be more and more frequent in the region due to climate change. The purpose of this study aims at presenting the current situation, highlighting on the status of traditional viticulture of the island and providing tools for viticulturists in order to adapt to these new conditions with a sustainable manner.

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Grape and wine microorganisms : diversity and adaptation

II.P.1 CONVOLUTIONAL NEURAL NETWORK TO PREDICT GENETIC GROUP AND SUL-FUR TOLERANCE OF *BRETTANOMYCES BRUXELLENSIS*

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Keywords: Brettanomyces bruxellensis, deep learning, cell morphology, genetic groups

The spoilage yeast *Brettanomyces bruxellensis* presents many strain dependent characteristics, particularly sulfur tolerance (1). Climate change and the evolution of oenological practices are at the origin of matrices with low levels of sulfur dioxide and higher pH. These parameters favor the development of this yeast and lead to serious financial losses for winemakers (2). Thus, it is essential to discriminate *B. bruxellensis* isolates at the strain level in order to predict their stress resistance capacities. Few predictive tools are available to reveal intraspecific diversity within *B. bruxellensis* species; also, they require expertise and can be expensive. In this study, to make analysis even faster, we further investigated the correlation between genetic groups previously described (3) and cell polymorphism using the analysis of optical microscopy images via deep learning. A Convolutional Neural Network (CNN) was trained and allowed the discrimination of *B. bruxellensis* isolates in 4 of the 6 genetic groups (GG), with an accuracy of 96.6% (4). Future works will have to be done for the no tested genetic groups. But already these results confirm the possibility to develop a tool allowing to determine the tolerance of a contaminant, in a short time, in order to help wine industry professionals to choose the appropriate corrective measure.

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INVESTIGATION OF MALIC ACID METABOLIC PATHWAYS DURING ALCOHOLIC FERMENTATION USING GC-MS, LC-MS, AND NMR DERIVED ¹³C-LABELED DATA

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II.P.2

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Keywords: 13C-labeling, malic acid, central carbon metabolism

Malic acid has a strong impact on wine pH and the contribution of fermenting yeasts to modulate its concentration has been intensively investigated in the past. Recent advances in yeast genetics have shed light on the unexpected property of some strains to produce large amounts of malic acid ("acidic strains") while most of the wine starters consume it during the alcoholic fermentation. Being a key metabolite of the central carbohydrate metabolism, malic acid participates to TCA and glyoxylate cycles as well as neoglucogenesis. Although present at important concentrations in grape juice, the metabolic fate of malic acid has been poorly investigated. In this work, we used ¹³C-labeled malic acid to understand the main routes of its consumption and its de novo production. Two strains selected for their opposed malic acid metabolism were compared by combining several analytical chemistry techniques. The isotopic enrichment of intracellular amino acids was measured by GC-MS, the relative quantification of intracellular and extracellular labeled compounds was achieved by 2D-NMR, and the absolute quantification of labeled and unlabeled extracellular organic acids was achieved by LC-MS/MS. Although, both strains consume most of the malic acid provided, the "acidic strain" produces de novo malic acid during the second part of the alcoholic fermentation. In addition, ¹³C-filiation analyses provided evidence that most of the TCA is fed by glycolytic pyruvate and/or by cytosolic acetyl-CoA. Our results also confirmed that malic acid may be a secondary source of TCA cycle during alcoholic fermentation especially in high malic acid consuming strains that has an efficient malo-ethanolic fermentation. Finally, ¹³C-labeled compounds belonging to amino acids, alcoholic fermentation and neoglucogenesis pathways were identified, highlighting the pleiotropic position of malic acid in both catabolic and anabolic routes.

II.P.3 INSIGHTS ON THE ROLE OF GENES ON AROMA FORMATION OF WINES

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Keywords: gene expression, Saccharomyces cerevisiae, yeast metabolism, volatile profile

Yeast secondary metabolism is a complex network of biochemical pathways and the genetic profile of the yeast carrying out the alcoholic fermentation is obviously important in the formation of the metabolites conferring specific odors to wine. The aim of the present research was to investigate the relative expression of genes involved in flavor compound production in eight different *Saccharomyces cerevisiae* strains.

Two commercial yeast strains Sc1 (*S.cerevisiae* x *S.bayanus*) and Sc2 (*S.cerevisiae*) and six indigenous *S. cerevisiae* strains (Sc3, Sc4, Sc5, Sc6, Sc7, Sc8) isolated during spontaneous fermentations were inoculated in Assyrtiko and Vidiano grape must. The fermentation kinetics, content of organic acids and glycerol production was monitored daily throughout the experiment. Transcript profiling of yeast genes involved in aroma formation and volatile composition of the must/wine was performed through real-time RT-PCR and SPE/GC–MS respectively, at four different time points of the fermentations. Specifically, a total of 28 volatile compounds were determined and the relative expression levels of 46 genes coding for acetyl-CoA synthetases, amino acid permeases, transaminases, reductases, decarboxylases, alcohol and aldehyde dehydrogenases, alcohol acetyltransferases, acyltransferases, esterases and glycosidases were recorded.

The relative expression levels of the genes implicated in amino acids, higher alcohols, ethyl esters, and terpenes metabolism, such as *EEB1*, *EHT1*, *EXG1*, *ARO8*, *ARO9*, *PDC5*, *PDC6*, *ADH6*, *ADH3*, *AAD14*, *AAD16*, were higher at the first three time points studied, since the most active period of aroma compound accumulation appears to occur in earlier fermentation stages. Additionally, the transcriptome data revealed substantial changes in expression patterns of genes between the different strains tested. In terms of the volatile characterization of the wines, the concentration levels of total esters and total alcohols appeared to be clearly distinct between the wines, which confirms that the production of volatile compounds is strain depended. Remarkable differences in the gene expression levels were observed when comparing the different strains which resulted in different aroma profiles.

This study enhances our understanding on yeast aroma metabolism-related gene expression and regulation. This knowledge can be a tool to modulate aroma production and orient the fermentation process towards a desirable wine aromatic profile.

INCREASING PINOT NOIR COLOUR DENSITY THROUGH SEQUENTIAL INOCULATION OF FLOCCULENT COMMERCIAL WINE YEAST SPECIES

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II.P.4

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Keywords: Anthocyanins, Colour density, Pinot noir, Sequential inoculation

Vitis vinifera L. cv. Pinot noir can be challenging to manage in the winery as its thin skins require careful handling to ensure sufficient extraction of wine colour to promote colour stability during ageing.¹ Literature has shown that fermentation with flocculent yeasts can increase red wine colour density.² As consumers prefer greater colour density in red wines,³ the development of tools to increase colour density would be useful for the wine industry. This research explored the impact of interspecies sequential inoculation and co-flocculation of commercial yeast on Pinot noir wine colour. Six commercial non-Saccharomyces yeast species and two commercial Saccharomyces cerevisiae strains were assayed based on their sedimentation rates in synthetic grape must, both individually and in combination, to determine flocculation ability. The most flocculent S. cerevisiae and non-Saccharomyces spp. yeast pairings, RC212 + BIODIVA and VL3 + BIODIVA, were used in a 20 L-scale Pinot noir winemaking trial. Ultraviolet-visible spectrophotometric measurements of wine colour parameters, and sensory evaluation of wine appearance, found that mixed species fermentations produced wines with greater colour density. Total and monomeric anthocyanin concentrations were lower in sequentially-inoculated wines, despite being the main source of young red wine colour. Pigmentation assays indicated a higher adsorption of anthocyanins by BIODIVA than S. cerevisiae, suggesting that greater amounts of cell wall mannoproteins in flocculent yeast may scavenge anthocyanins during fermentation, allowing for their subsequent release from the lees and potential for enhanced formation of copigments. Findings from this research have wide application in the industry to increase red wine colour intensity, particular in thinskinned red grape varieties.

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II.P.5 MOUSY OFF-FLAVOURS IN WINES: UNVEILING THE MICROORGANISMS BEHIND IT

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Keywords: Mousy off-flavor, Brettanomyces bruxellensis, Lactic acid bacteria, Wine

Taints and off-flavours are one of the major concerns in the wine industry and even if the issues provoked by them are harmless, they can still have a negative impact on the quality or on the visual perception of the consumer. Nowadays, the frequency of occurrence of mousy off-flavours in wines has increased.

The reasons behind this could be the significant decrease in sulphur dioxide addition during processing, the increase in pH or even the trend for spontaneous fermentation in wine. This off-flavour is associated with Brettanomyces bruxellensis or some lactic acid bacteria metabolisms. Three N-heterocyclic compounds (APY, ETHP, ATHP) have been described as involved in mousiness perception. Thus far, no study addressed the variability in that N-heterocycles production according to microorganism strains from different species. Twenty-five wines presenting mousy off-flavour were analysed. In total, 252 bacteria with 90.5 % of Oenococcus oeni and 101 yeast strains with 53.5 % of Saccharomyces cerevisiae were isolated and identified. Even if B. bruxellensis have been isolated during this study, it has been shown that in most mousy wines, it wes not found. Their capacity to produce mousy compounds was investigated using Stir Bar Sorptive Extraction-Gas Chromatography-Mass Spectrometry (SBSE-GC-MS) in a standardised N-heterocycle assay medium (NHAM). While four and three species of yeast and bacteria, respectively, were isolated from mousy wines, only three species of microorganisms were associated with N-heterocycles production: B. bruxellensis, Lentilactobacillus hilgardii and Oenococcus oeni. The screening was then extended to collection strains for these three species to improve their genetic representativity. Our results show that the levels and the ratios of the three N-heterocycles present huge variations according to the species but all the tested strains were able to produce mousiness in the NHAM

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UNRAVELLING THE ROLE OF LACTIC ACID BACTERIA ON SPARKLING WINE ELABORATION THROUGH METABOLOMICS APPROACH

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Keywords: sparkling wine, malolactic fermentation, Xinomavro, bacteria

Xinomavro is a red grape variety from Northern Greece (Protected Designation of Origin), known for the nice acidities, perfectly appropriate for sparkling wine production (Rosé and Blanc de Noir). The elaboration of sparkling wine requires technical as well as scientific skills. Although the impact of the yeast strains and their metabolites on the final product quality is well documented, the action of bacteria still remains unknown.

The present work focuses (i) on the population diversity of lactic acid bacteria isolated from sparkling wines and (ii) on the technological effect of the species during sparkling wine elaboration. Bacterial strains were typed by multiple loci VNTR analysis (MLVA) based on five tandem repeats loci and 3 different strains were chosen as starters for the sparkling wine production.

Xinomavro base wine was treated according to the winery production protocol and second fermentation was realised in the bottle under 6 different inoculation schemes.1) addition of *S. cerevisiae* (Lalvin DV10) 2) addition of *S. cerevisiae* (Lalvin DV10) and Lysozyme (40g/hL) 3) addition of *S. cerevisiae* (Lalvin DV10) and *O. oeni* Greek strain (UNIWA collection) 4) addition of *S. cerevisiae* (Lalvin DV10) and *O. oeni* French strain (CRBO collection) 5) addition of *S. cerevisiae* (Lalvin DV10) and *O. oeni* Commercial strain 6) addition of Schizo saccharomyces pombe strain (NRRL collection). Twelve months after the second fermentation in the bottle, oenological parameters were determined according to the OIV protocols, the volatile compounds produced were measured by GC/MS, and the metabolomic fingerprint analysis were acquired by an UPLC-HDMS-QTof-MS instrument. Finally, all produced wines were evaluated by quantitative descriptive sensorial analysis.

Malolactic fermentations were realized in all cases except the condition n°2 where lysozyme was added. Forty compounds were quantified and separated according to their chemical classes (monoterpenes, norisoprenoids, aldehydes, alcohols, esters, acids, and ketones) while statistical analysis showed the presence of three groups of sparkling wines according to the inoculation scheme. The untargeted metabolomic approach clearly discriminated the action of bacteria and revealed intra species variability at strain level. This is the first time that highlights the role of lactic acid bacteria and precisely of the species of *O*. *oeni* to sparkling wine elaboration.

A NEW SPECIFIC LINEAGE OF *OENOCOCCUS OENI* IN COGNAC APPELLATION WINES

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Keywords: Oenococcus oeni, Malolactic fermentation, Cognac, Biodiversity

Oenococcus oeni is the main lactic acid bacteria (LAB) species which conducts the malolactic fermentation (MLF) in wine. During MLF, *O. oeni* converts malic acid into lactic acid, which modulates wine aroma composition leading to better balanced organoleptic properties. *O. oeni* is a highly specialized species only detected in environments containing alcohol such as wine, cider or kombucha. Genome analysis of more than 240 strains showed that they form at least 4 main phylogenetic lineages and several sublineages, which are associated with different beverages or types of wines. Distilled wines produced in Cognac appellation of origin undergo MLF. Given the lack of knowledge of LAB present in distillation wines, the control of MLF and the further storage of wines is a difficult task. Therefore, the aim of this work is to analyze the biodiversity of *O. oeni* strains naturally occurring in cognac distilled wines and to determine if they confer a particular quality to the spirit after distillation.

559 samples of wines were collected before, during and after MLF from 24 wineries located in almost all the regions of Cognac appellation during 4 vintages from 2019 to 2022. The samples were processed to isolate single colonies of LAB, which were typed at the species and strain levels by MLVA (Multiple Loci of Variable Number of Tandem Repeats Analysis). About 5000 colonies of *O. oeni* isolates were obtained and assigned to 688 different strains. The most abundant strains in each winery were further analyzed at the genomic level. A total of 49 draft genomes were produced by Illumina MiSeq. The distances between these 49 genomes and 240 other publicly available *O. oeni* genomes were calculated using ANI (Average Nucleotide Identity) and used to reconstruct a phylogenetic tree. The tree showed that 34 of the 49 strains grouped together in a new phylogenetic lineage and contain only stains isolated during MLF in the wineries, which suggests that this lineage is specific and predominant in all the cognac wines. Moreover, the same strains were often found in the same wineries during consecutive vintages. The results suggest that the strains of this genetic lineage share specific genetic properties conferring them a better adaptation to cognac wines, and may in addition confer specific aromatic characteristics to cognac wines during MLF.

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ALCOHOLIC FERMENTATION DRIVES THE SELECTION OF OENOCOCCUS OENI STRAINS IN WINE

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Keywords: Oenococcus oeni, Malolactic fermentation, Population dynamics, phylogroups

Oenococcus oeni is the predominant lactic acid bacteria species in wine and cider, where it performs the malolactic fermentation (MLF) (Lonvaud-Funel, 1999). The *O. oeni* strains analyzed to date form four major genetic lineages named phylogroups A, B, C and D (Lorentzen et al., 2019). Most of the strains iso-lated from wine, cider, or kombucha belong to phylogroups A, B+C, and D, respectively, although B and C strains were also detected in wine (Campbell-Sills et al., 2015; Coton et al., 2017; Lorentzen et al., 2019; Sternes and Borneman, 2016). This study was performed to better understand the distribution of the phylogroups in wine and cider. Their population dynamics were determined by qPCR all through wine and cider productions, and the behavior of the strains was analyzed in synthetic wines and ciders. Phylogroups A, B and C were all represented in grape must and throughout the alcoholic fermentation, but on the transition to MLF, only phylogroup A remained at high levels in all wine productions. In the case of cider, phylogroups A, B and C were detected in stable levels during the process. When they were tested in synthetic wine and cider, all phylogroups performed MLF, but with different survival rates depending on the ethanol content. In this sense, ethanol and fermentation kinetics are the main agent that drives the selection of phylogroup A strains in wine, while B and C strains dominates in cider containing less ethanol.

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AROMATIC AND FERMENTATIVE PERFORMANCES OF *HANSENIASPORA VINEAE* IN DIFFERENT SEQUENTIAL INOCULATION PROTOCOLS WITH *SACCHAROMYCES CEREVISIAE* FOR WHITE WINEMAKING

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II.P.9

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Keywords: Hanseniaspora vineae, non-Saccharomyces, sequential inoculation, wine aroma

Hanseniaspora vineae (Hv) is a fermenting non-Saccharomyces yeast that compared to Saccharomyces cerevisiae (Sc) present some peculiar features on its metabolism that make it attractive for its use in wine production. Among them, it has been reported a faster yeast lysis and release of polysaccharides, as well as increased ß-glucosidase activity. Hv also produces distinctive aroma compounds, including elevated levels of fermentative compounds such as ß-phenylethyl acetate and norisoprenoids like safranal. However, it is known for its high nutritional requirements, resulting in prolonged and sluggish fermentations, even when complemented with Sc strain and nutrients. The study aims to assess the impact of progressive inoculation of Sc yeast during white wine fermentation at different stages: 24 h, 48 h, 72 h, 100 h, and 200 h after the initial inoculation of Hv yeast. The latter time point corresponds to the halfway of the fermentation process. The concentration of some yeast-derived aroma compounds was evaluated in wines by GC-MS/MS (2-phenylethyl alcohol, ß-phenylethyl acetate, isoamyl acetate and ethyl hexanoate) as indicators of the metabolic response of yeasts during fermentation. The 200 h protocol took an average 13 extra days to complete alcoholic fermentation compared to the pure Sc. The difference decreased as the moment of sequential inoculation neared, with a difference of -4 days for the 24 h protocol. Regarding volatile compounds, the production of isolamyl acetate and ethyl hexanoate were higher in Sc wines respect to any Hv wines (up to 2.5-fold), for which it was found no significant differences between them. However, every Hv protocol was richer in ß-phenylethyl acetate. Interestingly, the 24 hour protocol produced the highest concentration (~11-fold than Sc) while the 200 h protocol showed the lowest (-8-fold), demonstrating a downward trend with respect to the time of Sc inoculation. Conversely, 2-phenylethanol concentration was higher in the 200 h protocol and it showed a positive correlation with reduced inoculation time. Results confirm the ability of Hv to change the aroma features of wines, increasing the rose-like scents that characterise ß-phenylethyl acetate. By reducing the delay in Sc inoculation, the performance of Hv became better aligned with industrial standards while also maintaining an increased production of ß-phenylethyl acetate.

II.P.10 DEVELOPMENT OF BIOPROSPECTING TOOLS FOR OENOLOGICAL APPLICATIONS

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Keywords: yeast, bioprospection, wine

Wine production is a complex biochemical process that involves a heterogeneous microbiota consisting of different microorganisms such as yeasts, bacteria, and filamentous fungi. Among these microorganisms, yeasts play a predominant role in the chemistry of wine, as they actively participate in alcoholic fermentation, a biochemical process that transforms the sugars in grapes into ethanol and carbon dioxide while producing additional by-products. The quality of the final product is greatly influenced by the microbiota present in the grape berry, and the demand for indigenous yeast starters adapted to specific grape must and reflecting the biodiversity of a particular region is increasing. This supports the concept that indigenous yeast strains can be associated with a "terroir".

While some non-*Saccharomyces* species have been found to affect the chemical composition of wine, their low fermentation ability limits their usefulness, as they are unable to fully metabolize the sugars in the grape juice and produce only small amounts of ethanol. However, non-*Saccharomyces* strains have several oenological properties that are fundamental for the organoleptic properties of wine. As a result, the use of mixed non-*Saccharomyces/Saccharomyces* fermentation can be a valid alternative to spontaneous fermentation, as it can mimic natural biodiversity and increase the organoleptic properties of wine while minimizing microbial alternations.

The objectives of this work were to prospect and precisely identify genetically yeasts (more than 300 strains) of interest for the production of fermented beverages using an innovative protocol in several Swiss vineyards, establish a methodology to phenotypically characterize the isolated yeasts, and develop a procedure to assist winegrowers in their use of mixed *saccharomyces* and non-*saccharomyces* yeasts.

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II.P.11 ENRICHMENT OF THE OENOLOGICAL MALDI-TOF/MS PROTEIN SPECTRA DATABASE FOR RELIABLE OENOLOGICAL YEAST AND BACTERIA IDENTIFICATION

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Keywords: Yeast, bacteria, MALDI-TOF/MS, identification, database

The Matrix Assisted Laser Desorption/Ionization–Time-Of-Flight Mass Spectrometry (MALDI-TOF MS) technology is commonly used in food and medical sector to identify yeast or bacteria species isolated from a nutritive culture media. Since a decade, brewery and oenology industries have been attracted to this method which combines fast analysis times, reliability and low cost of analysis. Briefly, this method is based on the comparison of the MALDI-TOF/MS protein spectra of an isolated colony of yeast or bacteria with those contain in a manufacturer's reference protein spectra database. Initiated in 2015, the creation of the first oenological mass spectra database has proved to be essential for increase quality of species identification. Indeed some yeast or bacteria of interest in oenological environment are less representative or absent from manufacturer's reference spectra database such as some species of *Pichia* or *Starmerella* genus for yeast and *Acetobacter* species for bacteria. Moreover, many study demonstrated that yeast and bacteria isolated from wine related environment have some particular genetic and phenotypic characteristics and commonly belong to separated subgroups within the species. These wine strains specificity make essential to create an oenological dedicated MALDI-TOF/MS spectra database with wine related environment isolated yeast and bacteria strains to obtain successful identification by MALDI-TOF/MS.

The oenological mass spectra database contains today more than 200 yeast and bacteria species, corresponding to 40 different oenological yeast species and 28 distinct species of acetic and lactic acid bacteria, mainly provide by the Biological Resources Centre CRBO (ISVV). The database has been implemented and successfully used in several studies related to yeast and bacteria species diversity analysis as well as the impact on the winemaking process (pre-fermentery stages without SO₂, use of chitosan). This oenological mass spectra database is extensible and constantly implemented to meet the needs or future challenges of the wine industry. Finally, this innovative method of MALDI-TOF/MS, completed with oenological mass spectra database, allows quick and cheap implantation validation of grape juice bioprotection preparation composed of oenological yeast species.

EXPLORING THE METABOLIC AND PHENOTYPIC DIVERSITY OF INDIGENOUS YEASTS ISOLATED FROM GREEK WINE

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Keywords: indigenous yeast, Greek terroir, phenotyping

Climate change leads to even more hostile and stressful for the wine microorganism conditions and consequently issues with fermentation rate progression and off-character formation are frequently observed. The objective of the current research was to classify a great collection of yeast isolates from Greek wines based on their technological properties with oenological interest. Towards this direction, fourteen spontaneously fermented wines from different regions of Greece were collected for further yeast typing. The yeast isolates were subjected in molecular analyses and identification at species level. Random Amplified Polymorphic DNA (RAPD) genomic fingerprinting with the oligo-nucleotide primer M13 was used, combined with Matrix Assisted Laser Desorption Ionization-Time of Flight Mass Spectrometry (MALDI-TOF MS) technique. All yeast isolates were scrutinized for their sensitivity to killer toxin, production of metabolites such as acetic acid and H_2S , enzymatic activity of β -glucosidase and resistance to different concentrations of the antimicrobial agents; SO₂. Qualitative data were statistically treated by homogeneity of variances, one sample Kolmogorov-Smirnov and off between-subjects effects tests. According to our results, among the 190 isolates, S. cerevisiae was the most dominant species (83,5%) while some less common non-Saccharomyces species such as Trigonopsis californica, Priceomyces carsonii, Zygo saccharomyces bailii, Brettanomyces bruxellensis and Pichia manshurica were identified in minor abundancies. Moreover, based on phenotypic typing, the majority of isolates were neutral to killer toxin test and exhibited low acetic acid production. Additionally, statistically significant differences were observed between the different levels of H₂S production in terms of sample origin and yeast species. Finally, Hierarchical Cluster Analysis revealed the presence of four yeast groups based on phenotypic fingerprinting. This study proposed a fast preselection of wine autochthonous yeast with oenological potential using a simple phenotypic-based methodology.

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II.P.13 FERMENTATION POTENTIAL OF INDIGENOUS NON-*SACCHAROMYCES* YEASTS ISOLATED FROM MARAŠTINA GRAPES OF CROATIAN VINEYARDS

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Keywords: non-Saccharomyces, monoculture fermentation, FTIR, yeast cell counting

indigenous non-Saccharomyces yeast for use in wine production has increased in The interest in recent years because they contribute to the complex character of the wine. The aim of this work was to investigate the fermentation products of ten indigenous strains selected from a collection of native yeasts established at the Institute for Adriatic Crops and Karst Reclamation in 2021, previously isolated from Croatian Maraština grapes, belonging to Hypopichia pseudoburtonii, Metschnikowia pulcherrima, Metschnikowia sinensis, Metschnikowia chrysoperlae, Lachancea thermotolerans, Pichia kluyveri, Hanseniaspora uvarum, Hanseniaspora guillermondii, Hanseniaspora pseudoguillermondii, and Starmerella apicola species, and compare it with commercial non-Saccharomyces and Saccharomyces strains. The Maraština sterile grape juice was inoculated with yeast isolates at a concentration of 10⁶ cells/mL in a laboratory flask. The fermentation process was monitored by psycho-chemical parameters and yeast cell counting on WL agar plates. Samples were analyzed by infrared spectroscopy with Fourier transformation (FTIR). Residual sugar after alcoholic fermentation was between 2.3 and 6.8 g/L for all species. M. chrysoperlae was yeast first finished fermentation after 20 days. Production of volatile acidity was similar for all indigenous yeasts (0.55-0.68 g/L) except H. pseudoguillermonondii which produced 0.87 g/L of volatile acidity and the lowest level of ethanol (11.5 % vol). On the other side, M. sinensis produced wines with the highest level of ethanol (12.7 % vol) and with low concentrations of malic acid. Fermentation with H. pseudoburtonii showed the highest level of lactic acid, 0.67 g/L. The obtained results allow the selection of yeasts for further research in the selection of potential starter cultures for creating a wine with regional character.

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FLOW CYTOMETRY, A POWERFUL AND SUSTAINABLE METHOD WITH MULTIPLE APPLICATIONS IN ENOLOGY

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Keywords: Brettanomyces bruxellensis, Flow cytometry, Specific quantification, Sustainable analysis

Flow cytometry (FCM) is a powerful technique allowing the detection, characterization and quantification of microbial populations in different fields of application (medical environment, food industry, enology, etc.). Depending on the fluorescent markers and specific probes used, FCM provides information on the physiological state of the cell and allows the quantification of a microorganism of interest within a mixed population. For 15 years, the enological sector has shown growing interest in this technique, which is now used to determine the populations present (of interest or spoilage) and the physiological state of microorganisms at the different stages of winemaking.

By studying the other quantification methods now routinely available in enology and comparing them to our FCM method, we show that alternative methods only provide a partial, and sometimes erroneous, view of the microbial populations. Coupled with the use of different markers (vitality markers and probe specific to *B. bruxellensis*), FCM allows the precise and specific quantification of cells and provides information on their physiological state. In addition, it is the only method that provides a comprehensive view of the present populations, all this in a short time and at a controlled cost.

These advantages make it a method of choice for multiple technical applications in wine microbiology such as the prevention of spoilage during aging, the control of fermentation activity or the quality control of barrel cleaning protocols.

Finally, aside from its technical interest, FCM responds to a major challenge for the wine industry: the commitment to sustainable development. Based on recent work evaluating the environmental impact of analysis techniques, we show through an original comparative study of *B. bruxellensis* quantification methods that FCM is a more sustainable technique than its alternatives usually used in the sector.

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FUNGAL CHITOSAN IS AN EFFICIENT ALTERNATIVE TO SULPHITES IN SPECIFIC WINEMAKING SITUATIONS

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Keywords: Antiseptic, Spoilage, Chitosan, Sulfites

The most common method to prevent or eliminate microbes in wine is sulfur dioxide (SO₂) addition. However, as risk of acute allergy exists, the European Union has classified SO_2 as one of the 14 priority food allergens (EU Regulation N°1169/2011, Annex II). The legal dose admitted in both conventional and organic farming will be downgraded probably in the near future, according to consumer's expectations. In addition, sulfur dioxide addition does not always prevent microbial spoilage, because of the emergence of tolerant/resistant strains. Winemakers thus need alternate and efficient antiseptic methods to reduce total SO₂ content in wines. The resolutions of the 7th general assembly of the International Organization of Vine and Wine (OIV/OENO 338A/2009) and the European Union (EC 53/2011) authorized the addition of fungal chitosan to reduce spoilage microorganism populations especially Brettanomyces bruxellensis. Chitosan is a partially acetylated polysaccharide of glucosamine. It is positively charged at wine pH, which allows it to interact with the microorganisms and particles present in the wine. With the trend in oenology of limiting SO₂, more and more questions arise as to the impact of fungal chitosan on other microorganisms from grapes and wine-related environment. It was shown recently that most species were affected, at least transiently, by chitosan treatment (Miot-Sertier et al. 2022). However, a high variability prevails within most species and sensitive, intermediate and tolerant strains can be observed, as well as different efficiencies depending on the wine chemical parameters or the winemaking stage when the treatment is performed.

In order to have a clear opinion on the usefulness of a chitosan treatment, we have carried out tests in various situations in which sulphites were not enough to protect the wine (presence of tolerant strains in particular). Though chitosan does not solve all the microbial spoilage issues, this study reveals that chitosane can be an interesting alternative to sulphites in certain situations. Furthermore, when the antiseptic effect is clear it seems durable and hence, wines are protected for microbial spoilage over long periods.

The study also shows that structural differences among fungal chitosans impact their efficiency. The organoleptic consequences of the treatment are also evaluated on red and white wines.

IMPACT OF ABIOTIC AND BIOTIC FACTORS ON BIOADHESION PROPERTIES OF BRETTANOMYCES BRUXELLENSIS

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Keywords: Brettanomyces bruxellensis, Wine, Spoilage, Bioadhesion

Brettanomyces bruxellensis is an ubiquitous yeast associated with different fermentation media such as beer and kombucha, where its presence is beneficial to bring an aromatic typicity. However, it is a main spoilage yeast in wines, in which it produces volatile phenols responsible for organoleptic deviations causing significant economic losses (Chatonnet et al., 1992). Cellar and winery equipment's are considered as the first source of contamination, during fermentation and wine ageing process (Connel et al., 2002). Indeed, it is possible to find B. bruxellensis in the air, on walls and floors of the cellars, on small materials, vats and barrels. Furthermore, specific strains are recurrently isolated in wines of certain wineries, thus showing the ability of the species to be resident in the cellar for long periods (Cibrario et al., 2019). Bioadhesion phenomena and biofilm formation are protective mechanisms that could explain the persistence of *B. bruxellensis* in the winery and recurrent wine contaminations. A subset of 17 *B.* bruxellensis strains, representative of the species genetic diversity and showing contrasting bioadhesion phenotypes, were selected to study the impact of pH and ethanol concentration on electronegativity (Zeta potential), hydrophobic character (MATS) and bioadhesion on stainless steel by confocal microscopy. The experimental design consisted in multi-strains and multi-species bioadhesions in order to observe potential interactions. Our results show that pH and ethanol concentrations do not impact the phenotypes but that the strains and genetic groups are the main factors explaining the variance sugges ting the role of genetic mechanisms on bioadhesion properties. Regarding multispecies bioadhesion, a decrease in the bioadhesion of *B. bruxellensis* is observed in association with lactic acid and acetic acid bacteria. Multi-strains bioadhesion of B. bruxellensis show that the most bioadhesive strain is present in higher proportions during the first stages of the bioadhesive process comparing with other strains. This study provides new insights into the impact of environmental factors on *B. bruxellensis* lifestyles as bioadhesion in response to stressful environments, with major consequences on surface colonization in food industry and wine spoilage.

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II.P.17 MICROFLUIDIC PLATFORM FOR SORTING YEAST CELLS ACCORDING TO THEIR MORPHOLOGY.

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Keywords: breeding, yeast spores, hybridization, microfluidics

In this work we briefly present a microfluidic device aiming to sort yeast cells according to their morphology. The technology is based upon microfluidic chips made out of Polydimethylsiloxane and glass using soft lithography processes and replica molding. The microfluidic device was used for encapsulating single yeast cells in liquid droplets containing growth medium. Liquid droplet containing yeast cells were sorted using a real time imaging and decision-making process. The technology used is based upon a home-made platform (labview based) for comparison of cells with recorded templates (1). Sorting is achieved by triggering electric fields on chip to actuate the hit droplets in the selection channels. Budding cells, ascospores, zygotes are differentiated at rates of ~30 drops/sec. From freshly mated strains, newly formed F1-zygotes can be sorted, while single ascus can be sorted from sporulating strains. Around 300 positive droplets with the desired event can be collected in a fraction tube using a single chip in one hour. This microfluidic platform constitutes an original device for achieving critical operation of yeast breeding at a Hight ought level.

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NEW INSIGHTS INTO THE EFFECT OF *TORULASPORA DELBRUECKII/SACCHA-ROMYCES CEREVISIAE* INOCULATION STRATEGY ON MALOLACTIC FERMENTA-TION PERFORMANCE

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Keywords: Wine microorganisms, Alcoholic fermentation, Malolactic fermentation, Inoculation strategy

Winemaking is influenced by micro-organisms, which are largely responsible for the quality of the product. In this context, Non-Saccharomyces and Saccharomyces species are of great importance not only because it influences the development of alcoholic fermentation (AF) but also on the achievement of malolactic fermentation (MLF). Among these yeasts, Torulaspora delbrueckii allows in sequential inoculation with strains of S. cerevisiae shorter MLF realizations [5]. Little information is available on the temporal effect of the presence of *T. delbrueckii* on (i) the evolution of AF and (ii) the MLF performance. Therefore, the objective of this study is to evaluate the effect of sequential time (2, 4 and 6 days) of T. delbrueckii/ S. cerevisiae on the achievement of MLF by two strains of Oenococcus oeni. AF and the following MLF were performed in a synthetic must supplemented with linoleic acid and b-sitosterol. The results showed that differences were observed in the duration of the AF as for example co-inoculated AF lasted less time, even compared to the control, while sequential AF were prolonged in time. Regarding the abundance of the species in co-inoculation S. cerevisiae dominated the fermentation process from the middle to the end as previously described in literature [2,3]. In sequential fermentations, T. delbrueckii represented a higher percentage at the end, 40-30% of the total population. In relation to the differences between sequential conditions it seems that during the fermentation with 4 days of T. delbruekii contact the population was higher than 2 and 6 days. As for the supplementation with lipids to the synthetic must we could observe that yeast viability increased, acetic acid decreased and AF and MLF performance improved.

Regarding MLF T. delbrueckii improved the total time of the process comparing with *S. cerevisiae* as described in literature [1,4]. However, in the co-inoculated wines MLF had a longer duration. Regarding sequential wines, in the 4-day contact condition with *T. delbruekii* the MLF was shortened to two days, with the two *O. oeni* strains, so this seemed to be the best strategy combination.

Overall, these findings highlight the importance of considering both the inoculation strategy and the specific strains used to a better understanding of the complex interactions between these species in the fermentation process.

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II.P.19 OENOLOGICAL POTENTIAL OF AUTOCHTHONOUS SACCHAROMYCES CEREVISIAE STRAINS AND THEIR EFFECT ON THE PRODUCTION OF TYPICAL SAVATIANO WINES

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Keywords: yeast selection, technological properties, sensory evaluation, terroir wine

Due to the global demand for terroir wines, the winemaking industry has focused attention on exploiting the local yeast microflora of each wine growing region to express the regional character and enhance the sensory profile of wines such as varietal typicity and aroma complexity. The objective of the present study was to isolate and compare the indigenous strains of *Saccharomyces cerevisiae* present in different vineyards in the Mesogeia – Attiki wine region (Greece), evaluate their impact on chemical composition and sensory profile of Savatiano wines and select the most suitable ones for winemaking process.

Yeast populations were collected from spontaneous alcoholic fermentation of Savatiano musts. The yeast isolates were tested for basic oenological parameters including sulphur dioxide and ethanol tolerance as well as H₂S production. Four *S.cerevisiae* strains were selected for microvinification in order to assess their technological properties and sensorial characteristics. The fermentation kinetics was monitored throughout the experiment, while the content of organic acids and glycerol production have been controlled daily using HPLC analysis.

Our study revealed that the indigenous *S. cerevisiae* strains are able to metabolize all sugars, produce a satisfactory amount of ethanol and contribute to a distinct sensory profile. Although, different growth rates and metabolic differences between strains were observed. The overall evaluation of the data highlights the potential of the indigenous *S. cerevisiae* strains to provide promising results in wine industry.

PHENOTYPIC DIVERSITY AND BIO-PROTECTION CAPABILITY OF *METSCHNIKO-WIA* SP. IN OENOLOGY

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Keywords: wine fermentation, bioprotection, Metschnikowia, diversity

Nowadays, the trend is to reduce the use of chemical inputs in the food sector, including in oenology. One of the inputs widely used in the wine making process are sulfites, for its several properties: antimicrobial and antioxidiant. This use isn't without consequences on consumer's health and environment, it can lead for example to allergic reactions and pollution. To limit the addition of chemical inputs, microbial alternatives are used. It consists to inoculate in grape must, a micro-organism able to inhibit the growth of the negative indigenous flora during the phase before the fermentation and to guarantee the sensory qualities of wines. One of the specie the most used for its bio-protection capacity is the genus *Metschnikowia*. This project aims to study the phenotypical diversity in wine fermentative conditions and the diversity of bio-protector character of *Metschnikowia* sp.

To study the phenotypical diversity, 16 species of *Metschnikowia* within 50 strains have been selected depending their localization, their origins, and the species. These strains are used to ferment grape synthetical must, and the products of carbon central metabolism are analyzed by HPLC and the production of volatile molecules by GC-MS. In parallel, these strains are put in co-culture, with an acetic bacterium (Gluconobacter oxydans), known to lead to acetic souring, in commercial grape juice to study the interaction between yeast at 106cell/mL, and bacteria at 103cell/mL. Their growth is followed at day 0, 1, 2 and 7 through drop test on selective medium.

Similar to the fermentation aspects, the volatile profiles of the different strains were quite different, which corroborated the diversity of the *Metschnikowia* yeasts. The results of the drop test show an effect of bio-protection from the species *Metschnikowia* on the growth of *G. oxydans*. This work can underline both the potential of *Metschnikowia* yeast strains for inhibiting spoilage wine microorganisms and increasing aroma compounds.

II.P.21 PROFILING OF LIPIDS IN WINES FROM MONOCULTURE FERMENTATION WITH INDIGENOUS *METSCHNIKOWIA* YEAST SPECIES

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Keywords: lipids, Metschnikowia species, indigenous yeast, Maraština wine

Lipids are a diverse group of organic compounds essential for living systems. They are vital compounds for yeast which makes them an important modulator of yeast metabolism in alcoholic fermentation. This study presents a comprehensive lipidome analysis of wine samples from the Vitis vinifera L., Maraština. The fermentation trails were set up in monoculture with different indigenous yeast strains selected from a collection of native yeasts established at the Institute for Adriatic Crops and Karst Reclamation in 2021, previously isolated from Croatian Maraština grapes: Metschnikowia pulcherrima, Metshnikowia sinensis/shanxiensis, and Metschnikowia chyrsoperlae. Commercial yeast M. pulcherrima FLAVIA served to control fermentation. Regarding the good sensitivity, specificity, and dynamic range of the UHPLC-MS/MS method, different classes of lipids were identified and quantified: free saturated fatty acids (6), free unsaturated fatty acids (5), triterpenoid (1), glycerophospholipid (1), glycerolipid (1), and free fatty acid esters (6). Methyl stearate was the only compound that is identified and quantified in wine but not found in must. The most abundant lipid compound in the Maraština musts and all experimental Maraština wines was palmitic acid (C16:0). Fermentation trials with M. chyrsoperlae yeast strain showed the highest concentrations of glycerolipid, triterpenoid, and free fatty acids. Mystric, linoleic acid, and glycerophospholipid had the highest concentrations in the fermentation with the M. sinensis/ shanxiensis yeast strain, whereas *M. pulcherima* dominated in the production of ethyl free fatty esters.

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PROTEOMIC STUDY OF THE USE OF MANNOPROTEINS BY *OENOCOCCUS OENI* TO IMPROVE MALOLACTIC FERMENTATION

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Keywords: mannoproteins, Oenococcus oeni, malolactic fermentation, proteomics

Malolactic fermentation (MLF) is a desired process to decrease acidity in wine. This fermentation, carried out mostly by *Oenococcus oeni*, is sometimes challenging due to the wine stress factors affecting this lactic acid bacterium. Wine is a harsh environment for microbial survival due to the presence of ethanol and the low pH, and with limited nutrients that compromise *O. oeni* development. This may result in slow or stuck fermentations. After the alcoholic fermentation the nutrients that remain in the medium, mainly released by yeast, can be used in a beneficial way by *O. oeni* during MLF. Among them, mannoproteins stand out, being the main component of the yeast cell wall. These polysaccharides are released in different amounts during the winemaking process in alcoholic fermentation and aging on the lees. It has been described that the mannoproteins released by yeasts can activate the development MLF due to detoxification but little is known about the possible metabolization of mannoproteins by *O. oeni*.

The aim of this work was to evaluate the changes in the proteome of *O. oeni* PSU-1 due to the presence of mannoproteins. The addition of 2 g/L of a purified extract of mannoproteins resulted in the decrease of the duration of MLF in wine synthetic medium. This could be correlated to the decrease in mannoprotein content after MLF. Proteomic analysis of *O. oeni* cells allowed the identification a total of 956 proteins. From these, 59 showed significant differences in abundance due to mannoprotein presence. On one side, the functional category of carbohydrate transport and metabolism was the most affected by mannoprotein addition and represented 25% of the proteins showing an increased abundance with respect to the control condition. Remarkably, one protein with increased abundance was a permease of the phosphotransferase system (PTS). Mannose, which can be liberated from mannoproteins as a result of *O. oeni* mannosidase activity, has been described as a PTS substrate, and could be implicated in *O. oeni* growth stimulation [1, 2]. On the other side, amino acid transport and metabolism, together with translation, were the functional categories that showed a higher number of proteins with decreased abundance in comparison to the control condition. In conclusion, *O. oeni* PSU-1 proteome was modified due to mannoprotein addition, indicating the metabolic use of these compounds that resulted in a stimulatory effect on MLF.

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II.P.23 RED WINE AGING WITHOUT SO₂: WHAT IMPACT ON MICROBIAL COMMUNITY?

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Keywords: wine without SO₂, microbial communities, MALDI-TOF MS, malolactic fermentation

Nowadays, the use of food preservatives is controversial, SO_2 being no exception. Microbial communities have been particularly studied during the prefermentary and fermentation stages in a context of without added SO_2 . However, microbial risks associated with SO_2 reduction or absence, particularly during the wine aging process, have so far been little studied. The microbiological control of wine aging is a key issue for winemakers wishing to produce wines without added SO_2 . The aim of the present study is to evaluate the impact of different wine aging strategies according to the addition or not of SO_2 on the microbiological population levels and diversity.

In 2021 and 2022, microbial community were monitored on merlot red wines during the wine aging process with different SO_2 management and no SO_2 addition. An experimental design (30L) was set up in triplicate and samples were collected from vatting to bottling to perform microbial analysis: population levels were monitored by plating on agar selective media for cultivable yeasts, acetic and lactic acid bacteria. From a subset of colonies obtained on solid medium, identifications at species level were made using the MALDI-TOF MS combining with a homemade database created by the laboratory.

In 2021, our results showed that without SO₂, significant higher population levels of yeast and bacteria comparing with the sulphiting wines were present during the wine aging process. As expected, the higher species diversity was found at vatting. During the winemaking process, different species of lactic acid bacteria (10), acetic acid bacteria (3) and yeasts (8) were identified. Surprisingly, the effectiveness of SO₂ addition at the end of MLF on the lactic acid bacteria showed contrasting results considering initial SO₂ addition or not at vatting: population levels were significantly lower when SO₂ was added only after malolactic fermentation. Our results regarding the impact of SO₂ nanagement during the winemaking process could provide opportunities for winemakers to reduce SO₂ levels. Furthermore, for the first time, microbial communities have been monitored throughout the winemaking process, in a reduced or without added SO₂ context.

THE IMPACT OF NON-*SACCHAROMYCES* YEASTS ON THE WHITE WINE QUALITY

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II.P.24

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Keywords: non-Saccharomyces yeasts, volatile thiols, esters, sensory properties

Selected strains of non-Saccharomyces yeasts showed a positive effect on sensory characteristics and aromatic complexity of wine. A sequential microbial culture of non-Saccharomyces and S. cerevisiae species is usually inoculated due to poorer fermentability of non-Saccharomyces species. The aim of the study was to investigate the role of non-Saccharomyces yeasts in the production of white wines. We evaluated how individual combinations of sequential inoculations of non-Saccharomyces and S. cerevisiae species affect the aromatic compounds (volatile thiols and esters) and sensory characteristics of the wines. Sauvignon Blanc and Istrian Malvasia musts were inoculated sequentially with different species of non-Saccharomyces yeasts (Pichia kluyveri FrootZen (Chr. Hansen Holding A/S), Kluyveromyces dobzhanskii Re19L, Pichia guilliermondii ZIM624, Starmerella orientalis 126, Torulaspora delbrueckii IVV7, Lachancea thermotolerans BLF LT7 (Laffort),) and yeasts of the Saccharomyces genus (S. cerevisiae Zymaflore X5 (Laffort), S. uvarum NO608/1, S. cerevisiae Ca39). Fermentation kinetics and reducing sugars content were monitored gravimetrically during alcoholic fermentation. After completion of alcoholic fermentation, physicochemical analyses were performed, and the content of volatile thiols and esters was determined by GC-MS and the content of hydroxycinnamates by HPLC-DAD. We also performed sensory analysis using intensity ranking test. The mixed yeast cultures showed differences in fermentation kinetics, in the ability to release thiols, and in the synthesis of esters during alcoholic fermentation. According to the sensory evaluation, the best evaluated wine was produced with the yeast K. dobzhanskii Re19L in the vinification of Sauvignon Blanc and with T. delbrueckii IVV7 in the Istrian Malvasia variety.

II.P.25 WINE FERMENTATION METABOLITES PRODUCED BY TWO *TORULASPORA DEL-BRUECKII* STRAINS ISOLATED FROM OKANAGAN VALLEY, BC, CANADA VI-NEYARDS

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Keywords: Non-Saccharomyces yeast, Wine aroma, Fermentation metabolites, Volatile compounds

Wine aroma is influenced by various factors, from agricultural practices in the vineyard to the enological choices made by winemakers throughout the vinification process. Spontaneous fermentations have a characteristically deeper complexity of aromas when compared to fermentations that have been inoculated with *Saccharomyces (S.) cerevisiae* because of the diversity of microflora naturally present on grape skins. Non-*Saccharomyces* yeast are being extensively studied for their ability to positively contribute to wine aroma and flavour. These yeasts are known to liberate more bound volatile compounds present in grape must than *S. cerevisiae* through the enzymatic action of β -glucosidases and β -lyases¹. *Torulaspora (T.) delbrueckii* is known to stand out among these nonconventional yeasts by increasing the content of esters, terpenes and thiols in wine fermentations, together with low production of acetic acid, hydrogen sulfide and acetaldehyde.

In a previous study using Pinot Noir grapes from the Okanagan Valley (BC, Canada)², a collection of non-*Saccharomyces* yeasts was isolated from late-stage spontaneous lab fermentations. These yeasts were screened for their fermentative performance based on residual sugar, ethanol concentration and production of non-volatiles such as glycerol and acetic acid in single fermentations using Chardonnay juice. From these, two strains of *T. delbrueckii* were selected for further analysis. The aim of the present work was to examine oenological traits such as ethanol, sulfite, and copper sulfate resistance for the two *T. delbrueckii* Okanagan Valley strains, the *T. delbrueckii* reference strain CBS1146, the commercial *T. delbrueckii* strain Zymaflore Alpha and a control *S. cerevisiae* strain. These five strains were also used to perform single yeast fermentations in Muscat juice. Non-volatile compounds were quantified by HPLC/RID and analyzed by ANOVA with no significant differences in residual sugars, ethanol and glycerol production, while CBS1146 displayed lower acetic acid than the other 4 strains. Volatiles such as terpenes, primary alcohols and esters were also semi-quantified by SPME-GC/MS, followed by Partial Least Squares-Discriminant Analysis. Differences were observed among the strains in aroma compounds including limonene, γ -terpinene, α -terpineol, ocimene, phenylethyl alcohol and 2-phenethyl acetate. This work will add to developing research on *T. delbrueckii* from the perspective of BC and Canadian wines.

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WINE LEES AS A SOURCE OF NITROGEN FOR *OENOCOCCUS OENI* TO IMPROVE MALOLACTIC FERMENTATION PERFORMANCE

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Keywords: wine lees, malolactic fermentation, Oenococcus oeni, nitrogen compounds

Malolactic fermentation (MLF) is a desired process in red and acidic white wines, after alcoholic fermentation (AF), carried out by the lactic acid bacterium (LAB) *Oenococcus oeni*. The advantages are an increase of pH, microbiological stabilization and organoleptic improvement of the final wine. However, the presence of stress factors such as ethanol, low pH, high total SO₂, lack of nutrients and presence of inhibitors, could affect the successful completion of MLF [1]. Changes in amino acid composition and deficiencies in peptides after AF, showed that MLF can be delayed, signaling its importance for bacterial growth and L-malic acid degradation during MLF [2].

Wine lees accumulated after fermentation, contain approximately 25% of dried matter, that consists of 25 to 35% tartrate salts, 35 to 45% microorganisms (predominantly yeasts) and 30 to 40% organic residues [3]. During vinification, through yeast autolysis there is a release of nitrogen compounds that could be beneficial for LAB [4]. The monitorization of nitrogen compounds during MLF and aging on lees in red wine with *O.oeni* has revealed the breaking down of peptides and rise in free amino acid concentration, supporting the idea of proteolytic activity [5].

The aim of the present work was to observe the effect of the addition of different wine lees, produced in the vintage 2022 in the cellar (Mas dels Frares, Tarragona, Spain) by different vinification processes, on MLF. Protein concentration, primary amino nitrogen, free amino acids and ammonia were determined in wine lees coming from red and white wine with different inoculation strategies and fermenting temperatures. The ones presenting greater differences in nitrogen compounds composition were selected for the addition in synthetic wine with pH 3.5, ethanol 12% (v/v) and low nitrogen content. MLFs were carried out at 20 °C with two different strains of *O.oeni* showing differences in MLF performance. Changes in nitrogen compounds during MLF were evaluated. Under most of the conditions, the lees addition (1 g/L) produced a reduction in the MLF duration in comparison with the control condition. This effect is bacteria strain and lees dependent. Overall, it was confirmed that the addition of wine lees could be beneficial. This effect could be linked to the proteins and amino acids input.

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II.P.27 EVALUATION OF INDIGENOUS *SACCHAROMYCES CEREVISIAE* ISOLATES FOR THEIR POTENTIAL USE AS FERMENTATION STARTERS IN ASSYRTIKO WINE

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Keywords: Indigenous yeast, S. cerevisiae, Strain variability, Assyrtiko wine

Assyrtiko is a rare ancient grape variety that constitutes one of the most popular in Greece. The objective of the current research was to evaluate indigenous Saccharomyces cerevisiae isolates as fermentation starters and also test the possible strain impact on volatile profile of Assyrtiko wine. 163 S. cerevisiae isolates, which were previously selected from spontaneous alcoholic fermentation, were identified at strain level by interdelta-PCR genomic fingerprinting. Yeasts strains were examined for their fermentative capacity in laboratory scale fermentation on pasteurized Assyrtiko grape must. Daily glucose and fructose consumption was monitored and at the final point free sorting task was conducted to categorize the samples according to their organoleptic profile. The most performant strains were selected and subsequently subjected in a second laboratory scale fermentation. Oenological properties such as, titratable acidity, glucose/fructose consumption, total acidity, volatile acidity, pH, L-malic acid, yeast assimilable nitrogen, free and total SO₂ as well as sensory characteristics were determined. Finally, two wines with different aromatic profiles were subjected in Gas Chromatography- Olfactometry- Mass Spectrometry (GC-O MS) analysis. The molecular typing revealed the presence of 20 different S. cerevisiae strains from which 65% indicated high fermentative capacity. Hierarchical Cluster Analysis (HCA) based on sensory evaluation results clearly discriminated the produced wines and led to the selection of 4 strains. After the second pilot fermentation all selected strains resulted in dry wines with desirable technological and organoleptic characteristics. Additionally, statistically significant differences were noticed regarding the perception of tropical fruits and acidity while according to the results of GC-O MS analysis both samples revealed similar aromatic profiles. To the best of our knowledge, this is the first assay that explores the yeast strain effect on the aromatic profile of Assyrtiko variety by means of GC-O MS analysis.

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STATISTICAL COMPARISON OF GROWTH PARAMETERS OF NINE BIOPROTECTION STRAINS IMPLEMENTED ON ARTIFICIALLY CONTAMINATED SYNTHETIC MUST

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Keywords: bioprotection, pre-fermentation maceration, non-Saccharomyces yeasts

In recent years, consumer demand for products without chemical additives increased, becoming a priority for the wine sector. SO_2 is widely used for its multiple properties including antiseptics, antioxidants and antioxidasics and the strategy of bioprotection in winemaking represents now an alternative to this chemical additive. In oenology, results have highlighted the interest of bioprotection to limit the development of microorganisms like *Hanseniaspora uvarum* and thus reduce the doses of sulphite. Indeed, this species is considered because of its acetic acid and methyl butyl acetate production, the latter can cover the varietal character of wines.

Previous data sled to the development of a bioprotection concept based more specifically on the principle of early yeasting (pre-fermentation phases). In recent years, non-Saccharomyces yeasts have seen a resurgence of interest. Today, some of these strains are commercialized, such as Torulaspora delbrueckii, Metschnikowia pulcherrima or Lachancea thermotolerans. The antimicrobial effect may be linked to interaction mechanisms between microorganisms, including competition for nutrients and the production of inhibitory compounds. Therefore, the mechanisms involved, their mode of action and their effectiveness according to the technical constraints of winemaking process, more particularly at pre-fermentation stage conducted at low temperature, have yet to be explored. The experiments presented here involve 9 bioprotection strains (3 T. delbrueckii, 3 Metschnikowia sp. and 3 L. thermotolerans) grown on synthetic must artificially contaminated at different population levels (5.104 or 5.106 UFC/mL) with a mix of Hanseniaspora sp. strains. The growth parameters of all the strains characterized at 20°C were compared to those obtained at 12°C. This comparison underlined a negative effect on some bioprotectants, independently of the genus and species tested. Growth monitoring in coculture at 12°C showed that when the rate of contamination of the medium is higher than the rate of inoculation of bioprotectants, none of the bioprotection strains reach a biomass equivalent to that of the Hanseniaspora mix. Nevertheless, for lower contamination, two strains of M. pulcherrima and M. fructicola had a negative impact on the development of Hanseniaspora. The analysis of the amino acid consumption of the different strains tested could explain the different behaviors observed.

IMPACT OF *METSCHNIKOWIA PULCHERRIMA* DURING FERMENTATION ON AROMATIC PROFILE OF VIDAL BLANC ICEWINE

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Keywords: *Metschnikowia pulcherrima*, mixed culture fermentation, Vidal blanc icewine, volatile aroma compounds

Non-Saccharomyces yeasts not only increase microbial diversity during wine fermentation, but also have a positive effect on improving wine aroma. Among these non-Saccharomyces yeast species, Metschnikowia pulcherrima is often studied and used in winemaking in recent years, but its application in icewine has been rarely reported. In this study, indigenous M. pulcherrima strains and Saccharomyces cerevisiae strains (commercial and indigenous strains) were sequentially inoculated for icewine fermentations; meanwhile, pure S. cerevisiae fermentations were used as the control; indigenous strains used above were screened from spontaneous fermentations of Vidal blanc icewine. The aim was to study the effect of *M. pulcherrima* on the aroma complexity of icewine, which is of great significance to the application of *M. pulcherrima* in icewine production. The results showed that *M. pulcherrima* was completely replaced by S. cerevisiae at the middle and later fermentative stages in mixed culture fermentations. Compared with the icewine fermented with pure S. cerevisiae, mixed culture fermented icewines contained lower concentrations acetic acid and ethanol, and higher concentrations glycerol and succinic acid. The inoculation of M. pulcherrima greatly impacted the levels of several important volatile compounds, and more ethyl esters (such as ethyl caprylate, ethyl hexanoate, ethyl heptanoate, eta.), 2,4-hexadienoic acid, decanal, 1-octanol, and trans-rose oxide were produced, and the pleasant fruity and flowery characteristic was intensified. Moreover, the relevance of strain-specificity within *M. pulcherrima* to aroma compound differences was shown.

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IMPACT OF MINERAL AND ORGANIC NITROGEN ADDITION ON ALCOHOLIC FER-MENTATION WITH S. CEREVISIAE

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Keywords: alcoholic fermentation, nitrogen addition, organic/mineral nitrogen, S. cerevisiae

During alcoholic fermentation, nitrogen is one of essential nutrient for yeast as it plays a key role in sugar transport and biosynthesis of and wine aromatic compounds (thiols, esters, higher alcohols). The main issue of a lack in yeast assimilable nitrogen (YAN) in winemaking is sluggish or stuck fermentations promoting the growth of alteration species and leads to economic losses. Currently, grape musts are often characterized by low YAN concentration and an increase of sugars concentration due to global warming, making alcoholic fermentations even more difficult. YAN depletion can be corrected by addition of inorganic (ammonia) or organic (yeast derivatives products) nitrogen during alcoholic fermentation.

The aim of this work was to study the impact of the timing and the nature of nitrogen addition (mineral, organic or mixed) on alcoholic fermentation. First, 16 commercial strains were inoculated in Sauvignon blanc grape must deficient in YAN (110 mgN/L) and with reducing sugars concentration adjusted to 240 g/L (potential alcohol content of 14.3 %vol.). Fermentation kinetics of strains were then classified in 3 groups: stuck, sluggish or complete alcoholic fermentations. New experiments were carried on in the same grape must supplemented in YAN with ammonium (mineral) or yeast derivatives products (100% organic or mixed 30% organic - 70% mineral) to get 200 mgN/L. YAN additions were made at the beginning of alcoholic fermentation (single addition) or in two additions (50% at the beginning + 50% at the middle of alcoholic fermentation).

Our results showed that supplementing YAN twice with the mixed yeast derivative allowed complete alcoholic fermentations with reduced durations for all strains that initially showed stuck and sluggish fermentations.



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Wine chemistry, wine components with physiological effects

A NEW TOOL TO QUANTIFY COMPOUNDS POTENTIALLY INVOLVED IN THE FRUITY AROMA OF RED WINES. DEVELOPMENT AND APPLICATION TO THE STU-DY OF THE FRUITY CHARACTER OF RED WINES MADE FROM VARIOUS GRAPE VARIETIES

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Keywords: aroma compounds, GC-MS, fruity aroma, red wine

A wide range of olfactory descriptors ranging from fresh and jammy fruit notes to cooked and oxidized fruit notes could describe the fruity aroma of red wines ^[1]. The fruity character of a wine is mainly related to the grape variety selected, to the terroir and the vinification process applied for its conception. In white wines, some volatile compounds confer directly their aroma to the wine while the question of "key" compound is more complex in red wines. According to many studies performed over the past decades, some fruity ethyl esters are directly involved in the fruity perception of red wines while others, present at subthreshold concentrations, participate indirectly to the fruity expression via perceptive interactions ^[2]. However, a few non-fruity aroma compounds not belonging to ester family are known to contribute to the fruity notes via synergic effects while 1,8-cineole (a monoterpenoid) is involved in the blackcurrant aroma of particular red wines ^[3, 4].

This study intends to explore the fruitiness of red wines produced from different grape varieties. An analytical method was developed and optimized using liquid-liquid extraction and gas chromatography coupled to mass spectrometry (GC/MS) to determine the concentrations of aroma compounds potentially involved in the fruity aroma of red wines. The aim of this method was to reduce sample preparation and analysis time, as this tool requires a single sample preparation and a single injection to quantify 43 aromatic compounds including 19 esters, 13 monoterpenes, 5 C13-norisoprenoids and 1 C6-aldehyde and 5 C6-alcohols. A total of 37 volatile compounds were detected and quantified in commercial single-varietal red wines from the 2018 vintage made from grape-varieties planted around the Mediterranean (Greece, Cyprus, Spain, Portugal and France).

A generation of olfactory descriptors was coupled to instrumental analyses to investigate their fruity aromas. Samples were selected by experts according to their qualitative fruity aromas marked by "fresh red- and black-berry fruit" and "red- and black-berry jammy fruit" notes. Differences were observed regarding the variations in concentrations of several aroma compounds. Some variations are partially correlated to the olfactory descriptors cited by experts.

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BIOSORPTION OF UNDESIRABLE COMPONENTS FROM WINE BY YEAST-DERIVED PRODUCTS

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Keywords: 4-ethylphenol, biosorption, yeast cell walls, competitive adsorption

4-Ethylphenol (EP) in wine is associated with organoleptic defects such as barn and horse sweat odors. The origin of EP is the bioconversion reaction of p-coumaric acid (CA), naturally present in grapes and grape musts by contaminating yeasts of the genus *Brettanomyces bruxellensis*.

Yeast cell walls (YCW) have shown adsorption capacities for different compounds. They could be applied to wines in order to adsorb either CA and/or EP and thus reduce the organoleptic defects caused by the contaminating yeasts.

In this work, we selected four YCW derived from two different yeast genera (*Brettanomyces/Dekkera* and *Saccharomyces*) prepared by two processes, autolysis or with a high-pressure homogenizer (HPH). We investigated the effects of both genus and treatment on the capacity of adsorption of CA and EP. The operating parameters affecting adsorption, such as contact time, sorbent dosage, and initial CA and EP concentration, were studied to evaluate their influence on the adsorption capacity. The competition between the two adsorbates on the sorption sites was also investigated.

The adsorbed amounts of CA and EP by the YCW increased as the concentration of the adsorbent increased, regardless of their initial concentration. This might be explained by the increase in active vacant sorption sites and surface area available for the adsorption of CA and EP.

At equilibrium, the specific adsorption capacity of YCW increased when the initial concentration of adsorbate increased. The resistance to mass transfer of the adsorbate between liquid and solid phases is overcome by the driving force, which is determined by the initial concentration of the adsorbate. As the adsorbate's initial concentration increased, the concentration gradient's driving force increased, which explains the increase in adsorption.

The specific adsorption capacity decreased when the two adsorbates were together in the medium. First, the adsorption capacity of EP is higher than CA, indicating that EP has specific sites on the YCW that are different from CA. Second, the adsorption capacity is higher when the adsorbate is alone in the medium, implying competition between the two adsorbates. There are probably some common sites for CA and EP on YCW.

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III.P.3 CHEMICAL DRIVERS OF POSITIVE REDUCTION IN NEW ZEALAND CHARDONNAY WINES

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Keywords: Chardonnay, Flint, Mineral, Volatile Sulfur Compounds

According to winemakers, wine experts and sommeliers, aromas of wet stone, mineral, struck match and flint in white wines styles, such as those produced from Vitis vinifera L. cv. Chardonnay, are considered to be hallmarks of positive reduction.¹² In recent years, the production of Chardonnay styles defined by aroma characteristics related to positive reduction has become more desirable among wine experts and consumers. The chemical basis of positive reduction is thought to originate from the concentration of specific volatile sulfur compounds (VSCs), including methanethiol (MeSH) imparting mineral and chalk notes,³ and benzenemethanethiol (BMT) responsible for struck match and flint.¹⁴ However, the role of other aroma compounds, including esters, higher alcohols, and other VSCs, and their contribution to the sensory perception of positive reduction in New Zealand (NZ) Chardonnay wines has not been fully investigated. We selected 12 commercial NZ Chardonnay wines to represent a range of styles from low to high intensities of mineral and flint. Wine aroma profiles were analysed using headspace solid phase microextraction (HS-SPME) coupled with gas chromatography-mass spectrometry (GC-MS). Quantitative descriptive analysis (QDA) was performed on the same wines using a trained panel. Wines varied greatly in both their chemical and sensorial characteristics. Multivariate analysis showed that there were several key VSCs found to be explanatory variables driving the perception of attributes related to positive reduction in the NZ Chardonnay wines. These results will be presented in the context of winemaking techniques that can be applied by the industry to achieve Chardonnay styles with positive reduction, if desired by the winemaker.

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EFFECTS OF HYDROXYTYROSOL ON THE CHEMICAL PROFILE AND SENSORY ATTRIBUTES OF A RED TUSCAN WINE

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Keywords: Projective mapping, CATA, polyphenolic profile, hydroxytyrosol

The chemical profile and sensory attributes were studied in Borrigiano IGT Toscana wine (Italy), a blend of Sangiovese 85% and Cabernet Sauvignon 15% grapes harvested in September 2020, where 2-(3,4-dihydroxyphenyl)ethanol (hydroxytyrosol, HT, [1]) was added to a 750-ml wine bottle in 3 different amounts (30, 60, 120 mg) and compared with the control (no HT addition). The study aimed to evaluate whether Polyphenol-HT1®, a high purity HT (>99%) produced by Nova Mentis using biotechnology, could be used as a supplement to sulfites and how it would impact the sensory and chemical profile of this wine [2]. Each sample was prepared in triplicate. The chemical profile and sensory analysis were studied every three months (T1, T3 and T6) for a total of six months of storage. HT stability and evolution of sensory attributes were also investigated. The oenological parameters (such as free and total SO₂, residual sugars, organic acids) were evaluated with multiparametric wine analyser, the dissolved oxygen was measured according to OIV protocols, and HPLC-DAD was used to evaluate the phenolic profile [3]. To explore the effects of HT addition, Multiple Factor Analysis (MFA) was applied. The Projective Mapping sensory protocol [4], combined with CATA (check-all-that-apply) method, were chosen to achieve a rapid categorization and characterization of Borrigiano wine using an internal panel of fourteen assessors (aged 25-40 years old). Procrustean Multiple Factor Analysis (pMFA) and CLUSTATIS methods [5] were used to manage the sensory data. Evaluators were asked to rank wine samples according to their preferences and a frequency table was constructed. The HT addition (at different concentrations) and storage time influenced the chemical profiles and sensory attributes. After six months of storage, free sulfur dioxide remained higher in wines with the highest HT content. On the contrary, the dissolved oxygen was higher in the control wines, and was negatively correlated with the HT content. Acetic acid, which is the most important quality parameter of wine, was higher in the control wine samples. The assessors preferred the samples with the highest amount of HT; in fact, this wine gained first position for a greater number of times in the ranking constructed by the panel. The samples with the highest amount of HT had the lowest values of astringency, the highest level of vegetal, red fruit, dried fruit and wood aroma and red fruit flavour.

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EXPLORING THE ROLE OF TRANSITION METAL IONS IN THE EVOLUTION OF ESTERS COMPOSITION OF YOUNG WHITE WINE DURING AGEING

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III.P.5

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Keywords: esters, transition metal ions, hydrolysis, radical scavengers

Young white wines are typically released to the market a few months after harvest, to be consumed within a year, when their fresh fruity aromas are still dominant and appealing to modern consumers. Esters, particularly higher alcohol acetates (HAAs) and ethyl esters of fatty acids (EEFAs), play a central role in the fruity expression of young white wines [1]. However, these esters are known to undergo significant hydrolysis during the first months of aging [1, 2]. Therefore, understanding the factors that affect the hydrolysis of esters is crucial for wine producers. Although the hydrolysis of esters in wine matrices has been extensively studied for decades [3], the role of transition metal ions on the fate of esters in wines is still poorly documented.

This study aimed to explore the influence of Fe, Mn and Cu on the evolution of the ester composition of young white wines after 8-weeks of artificial ageing at 30 °C under different conditions. Young white wines were spiked with different mixtures of Fe, Mn and Cu, to reach final concentrations of 5 mg/L, 4 mg/L and 1 mg/L of metal ions, respectively. Wines were then aged in 20 mL SPME vials, full and half-full (oxidative conditions). The presence of gallic acid was also tested in interaction with metal ions added.

The presence of the Fe, Mn, and Cu mixture, described, above significantly increased the hydrolysis of HAAs and EEFAs in two different wine samples, with an 18% and 25% drop in HAAs and a 12% and 15% drop in EEFAs, respectively, compared to the same wine samples without the addition of metal ions. The oxidative aging did not affect this trend, except for EEFAs with long carbon chains (C10 and C12), which showed a decrease in concentration when the vial was half-full in comparison to full vial.

In contrast, the presence of gallic acid at 50 mg/L limited the effect of the metal ion mixture on esters hydrolysis. Each metal ion was also tested individually. Fe alone or in association with Cu had the same impact as the mixture of the three metal ions. Surprisingly, esters hydrolysis was significantly boosted with the addition of Cu and Mn alone or in mixture, but also when Fe was mixed with Mn. The addition of Mn alone had the strongest impact with a drop of 40% and 30% of HAAs and EEFAs concentration, respectively.

This work opens new research perspectives on how transition metal ions can shape the evolution of wine esters and, more broadly, the aromatic composition of wine.

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HYDROXYTYROSOL PRODUCTION BY DIFFERENT YEAST STRAINS: *SACCHA-ROMYCES* AND *NON-SACCHAROMYCES* AND THE RELATION WITH THE NITROGEN CONSUMPTION

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Keywords: hydroxytyrosol, Ehrlich pathway, yeast, tyrosol

Hydroxytyrosol (HT) is a phenolic compound with extensive bioactive properties. It is present in olives, olive oil and wines. Its occurrence in wines is partly due to yeast synthetise tyrosol from tyrosine by the Ehrlich pathway, which is subsequently hydroxylated to .

The aim of the present work is to study how different yeast strains can influence in the HT production and, how the different nitrogen consumption of each strain can interfere the production of bioactive compounds.

A total of six strains were evaluated for the production of HT, four of them were *Saccharomyces* and two of them were *non-Saccharomyces*. The *Saccharomyces* ones were Red Fruit, QA23, Uvaferm and Lalvin Rhone, and the *non-Saccharomyces* were *Torulaspora delbrueckii* and *Metschnikowia pulcherrima*.

In order to know the nitrogen consumption of each yeast, the nitrogen content in the extracellular media was measured at the early days of the fermentation.

The alcoholic fermentation was performed in synthetic must prepared according to the instructions of Riou et al., 1997. Fermentation was carried out in sextuplicate for each strain. lasting 10 days each. A total of 360 samples were collected. The growth of yeast, the weight of the flask, density, and the Baume grade of the must were recorded daily to monitor the fermentation.

Prior to the analyses of the compounds, a cleaning step was performed using a Solid Phase Extraction (SPE). The protocol for the SPE was optimized following the instructions of AOAC, 20212. All the compounds of the Erlich pathway (tyrosine, hydroxyphenylacetic acid, tyrosol, hydroxyphenylacetaldehyde acid, hydroxyphenylpyruvic acid and hydroxytyrosol) were evaluated thanks to a validated method of UHPLC-HRMS. The analysis was carried out in a Waters Acquity UHPLC (Milford, Massachusetts, USA) coupled to a Waters Xevo TQ (Milford, Massachusetts, USA) triple quadrupole mass spectrometer. The MassLynx MS software was used. The column used was an Acquity UPLC BEH C18. The chromatographic conditions consisted of two mobile phases, water with 0.2% acetic acid (A) and acetonitrile (B), with a gradient elution programmed.

The obtained results show that the *Saccharomyces* strains have a higher production of HT than *non-Saccharomyces*. Significant differences were observed between strains for the production of HT. The highest production was in day 5 for Uvaferm, reaching a concentration of 4 ng/mL. A different nitrogen consumption was observed for each yeast.

IDENTIFYING POTENTIAL CHEMICAL MARKERS RESPONSIBLE FOR THE PERMISSIVENESS OF BORDEAUX RED WINES AGAINST *BRETTANOMYCES BRUXELLENSIS* USING UNTARGETED METABOLOMICS

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Keywords: Untargeted metabolomics, Brettanomyces bruxellensis, UHPLC-UV-HRMS, Wine

All along the red winemaking process, many microorganisms develop in wine, some being beneficial and essential, others being feared spoilers. One of the most feared microbial enemy of wine all around the world is *Brettanomyces bruxellensis*. Indeed, in red wines, this yeast produces volatile phenols, molecules associated with a flavor described as "horse sweat", "burnt plastic" or "leather". To produce significant and detectable concentrations of these undesired molecules, the yeasts should first grow and become numerous enough. Even if the genetic group of the strain present and the cellar temperature may modulate the yeast growth rate¹ and thus the risk of spoilage, the main factor seems to be the wines themselves, some being much more permissive to *B. bruxellensis* development than others. However, common parameters such as pH, alcohol or sugars composition² cannot fully explain the permissiveness differences observed between the wines studied.

The present study aims at identifying (if any) the chemical markers specifically present in permissive wines and absent from resistant ones or conversely. To achieve this goal, the metabolite profiles of red wines coming from different châteaux in Bordeaux area and displaying different permissiveness was examined. The chemical composition was studied using targeted and untargeted metabolic profiling by UHPLC-UV-HRMS and ¹H-NMR. Meanwhile, the wines were inoculated with selected *Brettanomyces* strains; the microbial growth kinetics were studied and used to classify the wines into distinct groups. With the help of unsupervised statistical analyses, these results were combined in order to draw correlations between the chemical markers and the wine permissiveness.

The nature and the origin of the chemical markers identified is discussed and additional assays are currently performed to confirm the incidence of each marker on the risk of *Brettanomyces* development.

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III.P.8 MODELLING THE AGEING POTENTIAL OF SYRAH RED WINES BY ACCELERATED AGEING TESTS: INFLUENCE OF ANTIOXIDANT ASSAYS AND PHENOLIC COMPOSITION.

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Keywords: Red wine, Ageing capacity, Oxygen, Polyphenols

Red wine ageing is an important step in the red wine evolution and impacts its chemical and sensory characteristics through many chemicals and physico-chemical reactions. The kinetics of these evolutions depend on the wine studied and influence the wine ageing potential. Generally, high quality red wines require a longer period of bottle ageing before consumption¹. The ageing potential is an important parameter for wine quality and is related to the capacity of a wine to undergo oxidation over time². Phenolic compounds which are ones of the main substrates for oxidation can then potentially modulate ageing potential³.

The aim of this study was to assess the influence of phenolic composition and antioxidant properties on the ageing capacity of 14 Syrah red wines. This ageing capacity was measured by accelerated ageing tests (AATs) recently developed in our laboratory (thermal test at 60°C, enzymatic test with laccase and chemical test with H_2O_2)4. Different parameters were measured such as anthocyanin and flavanol contents, spectrophotometric antioxidant assays, voltammetric behaviour, colour parameters and free SO_2 levels. Statistical analyses were performed to model the results of the ATTs from the initial phenolic composition and antioxidant properties of Syrah red wines.

High correlations were obtained between the initial phenolic composition and the antioxidant properties of red wines. The results showed significant differences between the three studied ATTs, revealing specific mechanisms for each accelerated ageing condition. The Partial least squares (PLS) regression models results, based on measured parameters, had overall very good accuracy and involved different explaining variables for each test. The models have excellent predictive capacities with correlation coefficients (r²) between 0.89 et 0.98.

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NEW INSIGHTS INTO VOLATILE SULPHUR COMPOUNDS SCALPING ON MICROAGGLOMERATED WINE CLOSURES

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Keywords: volatile sulphur compounds, wine closure, scalping, sorption

The evolution of wine during bottle ageing has been of great interest to ensure consistent quality over time. While the role of wine closures on the amount of oxygen is well-known [1], closures could also play other roles such as the scalping phenomenon of flavour compounds. Flavour scalping has been described as the sorption of flavour compounds by the packaging material, which could result in losses of flavour intensity. It has been reported in the literature that volatile sulphur compounds (VSC) can be scalped on wine closures depending on the type of closure (traditional and agglomerated cork, screw-cap, synthetic [2]). However, no studies have been carried out to correlate the permeability of agglomerated closures with the scalping effect. In this study, we studied the evolution of the scalping effect of 7 VSC on 4 micro-agglomerated closures in model and Shiraz wines.

In practice, each closure was fully immerged in 20 mL of wine containing a precise concentration of 7 VSC (13.3 µmol/L) and maintained at room temperature under stirring (300 rpm) for 7 days. Finally, the residual concentration of VSC in the wines was monitored after 1 h, 6 h, 3 days and 7 days by HS-SPME-GC-MS/MS. All experiments were performed in triplicate and a control experiment without closure allowed us to evaluate the losses of VSC due to chemical mechanisms in both matrices.

In general, the concentrations of VSC decreased in all experiments from 1 h to 7 days, regardless of the wine matrix, indicating a possible flavour scalping. In fact, the residual concentrations of VSC after 7 days of monitoring were significantly lower in wines containing a closure ($2.1\pm0.5 \mu$ mol/L) than in the control experiment (6.3 μ mol/L). VSC were rapidly degraded after 6 h of contact of both wine matrices with closures, but no significant effect of the type of closure was observed. Consequently, it seems that the permeability of the agglomerated closure could not affect the scalping of VSC.

From a quantitative point of view, a simple material balance allowed us to characterize the fate of VSC during simulated ageing conditions:

- Synthetic wine: 27±3 % of sorption on the whole closure/ 60% of chemical mechanisms/ 13±3% of residual VSC.

- Shiraz wine: 22±1% of sorption on total closure/ 61% of chemical mechanisms/ 17±1% of residual VSC.

Finally, if we only consider the size of the closure mirror, the VSC scalping in the bottles is negligible and corresponds to 4% maximum of the initial concentration.

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III.P.10 PREVALENCE OF OAK-RELATED AROMA COMPOUNDS IN PREMIUM WINES

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Keywords: oak barrel, wine, aroma compounds, quantitation

Barrel fermentation and barrel-ageing of wine are commonly utilised practices in premium wine production. The wine aroma compounds related to barrel contact are varied and can enhance a range of wine aromas and flavours, such as 'struck flint', 'caramel', 'red berry', 'toasty' and 'nutty', as well as conventional oaky characters such as 'vanilla', 'spice', 'smoky' and 'coconut'. A survey of commercially produced premium Shiraz, Cabernet Sauvignon, Pinot Noir and Chardonnay wines was conducted, assessing the prevalence of compounds that have been proposed as barrel-ageing markers¹ including oak lactones, volatile phenols, furanones, aldehydes, thiazoles^{2,3}, phenylmethanethiol⁴ and 2-furylmethanethiol.⁵

Accurate quantitative data is necessary to help understand the compounds which might influence these aromas but their analysis is often not a trivial undertaking. The furanones, especially furaneol, are difficult to measure accurately in wine as they are very polar. Thus, an improved stable isotope dilution assay was developed using automated liquid–liquid microextraction and multidimensional–gas chromatography–mass spectrometry. Also, the quantification of aldehydes was simplified using automated headspace solid-phase microextraction and gas chromatography–tandem mass spectrometry with invial derivatisation. Thiazoles were quantified utilising gas chromatography–tandem mass spectrometry. Other targeted volatile compounds were quantified using previously published stable isotope dilution assay methods that are routinely used in-house. Wide concentration ranges were found for many of the targeted aroma compounds and this information will direct further detailed studies.

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SENSORY DEFINITION OF A TECHNICAL UNAVOIDABLE TRANSFER OF AROMA COMPOUNDS VIA SEALING IN A BOTTLING LINE IN ORDER TO PREVENT PROSE-CUTION DUE TO FRAUDULENT AROMATIZATION OF A SUBSEQUENTLY FILLED WINE

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Keywords: aroma migration, matrix dependent odor detection threshold, odor activity value, aromatized wines

In 2020, 12% of all bottled German wines were aromatized, which may increase further due to rising popularity of dealcoholized wines. As sealing polymers of a bottling line absorb aroma compounds and may release them into regular wines in the next filling¹, this unintentional carry-over bears the risk to violate the legal ban of any aromatization of regular wine. However, following EU legislation, German food control authorities accept a technical unavoidable transfer of aroma compounds, if this is of no sensory significance. In order to prevent any legal consequences of aroma migration, we propose how to define the requested absence of sensory significance and how to improve cleaning in respect to aroma compounds.

Using a novel direct analysis of sealing polymers revealed that cleaning of the filler removed only 11-62% of seven studied aroma compounds which are commonly used to aromatize wines, including γ -decalactone, α -ionone or eugenol¹. High temperature of 85 °C revealed the largest cleaning effect, while chemical additives such as caustic soda or ozone exhibited only minor efficacy². Complete removal of absorbed aroma compounds from sealing was not achieved, making a later release into subsequently bottled wines still possible.

Odor detection thresholds were determined separately in water, model wine and white wine for the monitored aroma compounds. Applying the odor activity concept, we could show that migration of aroma compounds into the subsequently bottled wines were of no sensory relevance³.

Studying aroma migration in two industry scale bottling lines we could confirm the uptake of marker compounds into sealing polymers during bottling mulled or aromatized wines. Despite ineffective cleaning, aroma compounds migrating back into the subsequently bottled non-aromatized regular wines were way below their sensory thresholds. Sensory evaluation by a 2-out-of-5-test of the wine before and after bottling indeed revealed no significant difference.

In conclusion, despite migration of aroma compounds into sealing of a bottling line, cleaning and dilution effects in the subsequently filled wine prevented any aroma carry-over of sensory relevance. Thus, the analytical determination of "illegal" added aroma traces in a regular wine due to this technically una-voidable transfer, would not lead to legal prosecution. This legal evaluation could be a show case, how to apply the de-minimis concept to assess traces of pesticides or other contaminants into wine.

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III.P.12 THE INFLUENCE OF COMMERCIAL SACCHAROMYCES CEREVISIAE ON THE POLY-SACCHARIDES AND OTHER CHEMICAL PROFILES OF NEW ZEALAND PINOT NOIR WINES

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Keywords: Yeast, New Zealand Pinot noir, Polysaccharides, Chemical profile

Wine polysaccharides (PS) play an important role in balancing mouthfeel and stability of wine and even influence aroma volatility. Despite this, there is limited research into the effect of winemaking additives on the polysaccharide profile and other macromolecules of New Zealand (NZ) Pinot noir wine. In this study the influence of a selection of commercial S. cerevisiae strains on the chemical profile, including polysaccharides, of New Zealand Pinot noir (PN) wine was investigated. Research scale PN fermentations using five strains of commercially available S. cerevisiae (Lalvin EC1118 and RC212, Levuline BRG YSEO, Viallate Ferm R71 and R82) were undertaken. PS were qualified and quantified using HPLC-RID. Wine produced using Vialatte Ferm R71 had higher PS content than the uninoculated control, primarily for the high and medium molecular weight PS. Wine colour and tannins were spectrophotometrically analysed. R82 wines were found to have statistically higher colour density and lower hue than EC1118, R71 and control wines. Furthermore, R82 wines had statistically lower concentrations of tannins than BRG YSEO wines. The aroma profiles were examined using SPME-GC-MS and it was found that R82 wines had statistically higher concentrations of several aromatic esters and alcohols compounds than all other wines, including ethyl isobutyrate, ethyl octanoate, ethyl hexanoate and butanol. Varietal thiols and thiol precursors were measured using LC-MS/MS. There was no statistical difference between then concentration of 3SH in the wines, but some differences in concentrations of varietal precursors, Cys-3SH and GSH-3SH, were measured.

THE ODORIFEROUS VOLATILE CHEMICALS BEHIND THE OXIDATIVE AROMA DEGRADATION OF SPANISH RED WINES

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Keywords: Chemosensory analysis, Gas chromatography-olfactometry (GC-O), Oxidative aging, Wine's longevity

It is a well-established fact that premature oxidation is noxious for wine aromatic quality and longevity. Although some oxidation-related aroma molecules have been previously identified, there are not works carrying out systematic research about the changes in the profiles of odour-active volatiles during wine oxidation.

Different types of wines in terms of region, grape variety, oak aging and price were subjected to an oxidative aging procedure, sensory analysis, gas-chromatography olfactometry (GC-O) and quantitative analysis. Sensory notes such as dried fruit, cooked vegetables or liquorice-alcohol were oxidation-related. The GCO analysis of the samples with highest oxidation notes, revealed highest levels of four odour zones, which were identified in a dual system GC-O/FID-GC-O/MS as 1,1-diethoxyethane (liquor, strawberry, sweet), 2,4,5-trimethyl-1,3-dioxolane (fruity, solvent), 3-methylbutanal (solvent, yeasty) and methional (boiled potato, cooked vegetables).

The two aldehydes were quantified by gas chromatography-mass spectrometry (GC-MS). together with isobutanal, 2-methylbutanal and phenylacetaldehyde. All them were already present in significant amounts before oxidation. However, as they were forming odourless reversible adducts with SO₂ (α -hy-droxyalkylsulphonates)1 they were initially non-odour active. However, as free SO₂ disappeared during oxidation² they become odour-active in oxidized samples. Additional quantities were formed during oxidation, most likely by the reaction of wine dicarbonyls with the amino acid precursors. This additional formation was particularly relevant for 2-methylbutanal, followed by methional and isobutanal, while for phenylacetaldehyde and 3-methylbutanal, quantities formed were smaller than those originally present. These results confirm that both, pre-existent levels of Strecker aldehydes and the ability to form them during oxidation, are relevant in wine stability.

Acetals were determined by L–L microextraction followed by GC–MS. Results revealed that during oxidation there is a clear increment on the levels of acetals formed from the condensation of acetaldehyde with ethanol, 2,3-butanediol and glycerol; leading to 1,1-diethoxyethane, 2,4,5-trimethyl-1,3-dioxolane and several heterocyclic acetals, respectively. Levels formed were high enough to be odour-active. This suggests that the formation of acetals is an essential part of the sensory changes noted during wine oxidation.

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III.P.14 VOLATILE AND GLYCOSYLATED MARKERS OF SMOKE IMPACT: LEVELS AND PATTERNS OBSERVED IN 2020 WINES FROM THE UNITED STATES WEST COAST

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Keywords: smoke impact, volatile phenols, glycosylated phenols, wine

Smoke impact in wines is caused by a wide range of volatile phenols found in wildfire smoke. These compounds are absorbed and accumulate in berries, where they may also become glycosylated. Both volatile and glycosylated forms eventually end up in wine where they can cause off-flavors, described as "smoky", "bacon", "campfire" and "ashtray", often long-lasting and lingering on the palate. In cases of large wildfire events, economic losses for all wine industry actors can be devastating.

In order to assess smoke impact, a selection of volatile and glycosylated smoke-derived phenols is proposed, mainly based on research from Australia (1, 2). It includes the volatile phenols guaiacol, 4-methylguaiacol, ortho-, meta- and para- cresol, phenol, syringol, and 4-methylsyringol, as well as their glycosylated forms guaiacol rutinoside, 4-methylguaiacol rutinoside, cresol rutinoside, phenol rutinoside, syringol gentiobioside, and 4-methylsyringol gentiobioside. The accurate and reproducible measurement of these markers is now possible worldwide, due to the commercial availability of standards and isotopic analogues.

The 2020 vintage has been particularly affected by wildfires all over the western part of the United States, giving us an opportunity to collect extensive data for this suite of markers in wines from smoke-exposed grapes.

In the large majority of cases, levels of both volatile and glycosylated markers in wines appeared closely related to the intensity of vineyard's exposure to smoke. This confirmed the relevance of these markers in the western United States.

In some cases, however, volatile markers were relatively low, sometimes barely indicating any exposure to smoke, while glycosylated markers were high. This suggested very efficient glycosylation mechanisms in grapes and vines exposed to smoke.

We also observed opposite patterns, meaning high levels of volatile markers in combination with low levels of glycosylated markers. This may be the consequence of impaired glycosylation pathways in the plants, possibly related to a severe heat wave experienced in mid-August 2020.

These observations confirm that measuring both volatile and glycosylated markers is advisable in order to identify wines from smoke exposed grapes.

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YEAST DERIVATIVE PRODUCTS: CHARACTERIZATION AND IMPACT ON RIBOFLAVIN RELEASE DURING THE ALCOHOLIC FERMENTATION

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Keywords: Light-struck taste, Flavins, Gluthathione, Must

Light-struck taste (LST) is a wine fault that can occur in white and sparkling wines when exposed to light. This defect is mainly associated to the formation of methanethiol and dimethyl disulfide due to light-induced reactions involving riboflavin (RF) and methionine [1]. The presence of RF in wine is mainly due to the metabolism of yeast [2] which fermenting activity can be favoured by using yeast derivative products (YDPs) as nutrients. Nonetheless, a previous study showed the addition of YDPs before the alcoholic fermentation (AF) led to higher concentrations of RF in wines [3]. Due to the widespread use of YDPs in the winemaking process, this study aimed to understand the possible relation between the content of RF in wine and the YDP adopted as nutrient for AF.

The experimental plan included (i) the characterization of selected YDPs and (ii) their addition as nutrient in microvinification trials using must as medium. Fourteen commercial YDPs of different nature (inactivated yeasts [IYs], mannoproteins [MPs], yeast autolysates [YAs], yeast extract [YEs] and yeast hulls [YHs]) were chosen. They were characterized in terms of flavins (RF, FAD and FMN), amino acid profile and sulfur-containing compounds (e.g. reduced glutathione [GSH], cysteine, cell wall cysteine and adsorbed cysteine). The characterized IYs, YEs and YHs were used for fermentation trials, carried out in Chardonnay must with 4 *Saccharomyces cerevisiae* strains. Flavins were quantified before and after the alcoholic fermentation.

Both YEs and one YA were the richest in RF which increase up to 20 µg/L was estimated considering an addition in must of 40 g/hL. These YDPs showed also the highest concentration of amino acids (up to 300 mg/L). However, among the flavins, FMN was the major one in most of the analyzed YDPs. With regards to GSH, this tripeptide was found at the highest concentration in the same YA (13.2 mg/g). The addition of YDPs caused a variation in RF released during AF depending on both the yeast strain and YDP nature. The addition of YEs caused an RF increase in must of about 15–20 µg/L, in accordance to RF content found in these products. For one of the yeast strains investigated, RF fatherly increased up to 30 µg/L during AF.

These results evidence the impact of YDPs on RF content indicating that the selection of nutrients combined with the choice of fermenting yeast strain should be considered for preventing the risk of LST appearance.

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III.P.16 INVESTIGATION OF FILM COATINGS AS A PROTECTIVE LAYER IN REDUCING THE ABSORPTION OF SMOKE PHENOLS INTO PINOT NOIR GRAPES

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Keywords: smoke, remediation, film, phenol

Wine grapes exposed to wildfire smoke have resulted in wines with burnt and ashy sensory characteristics¹, that are undesirable qualities in wine. In extreme wildfire events, this can lead to total loss of grape crop. Currently there are no effective solutions in the market to prevent the uptake of smoke compounds into grapes. In this study, previously developed innovative film coatings were tested to analyze their effectiveness in reducing smoke phenol absorption². Four different cellulose nanofiber-based film types were investigated. The film types varied in their chitosan and/or β -cyclodextrin composition. Film coatings were applied at veraison in vineyards in the Rogue Valley and Willamette Valley. The Rogue Valley experienced two smoke events during the season from wildfires in California. The grapes from Willamette Valley experience heavy smoke exposure using designed smoke cages. At harvest, half of the grapes were washed to remove the films. This was to determine if smoke phenols were blocked or bound to the film coatings. Further analysis of the interaction of smoke phenols with film coatings was done by observing any volatile phenol diffusion through the film using a custom-made polytetrafluoroethylene apparatus. Free and bound smoke phenols in grape juice were analyzed using GCMS and smoke glycosides using LCMS³. Results show some of the film coatings were effective in reducing the amount of smoke compounds absorbed into the grapes, primarily guaiacol, 4-methyl guaiacol, syringol and 4-methyl syringol. The cresol compounds were not greatly impacted. The collective results of this study show promise for film coatings as an effective preventative technique for grape smoke exposure. Optimization of the film coating formulation will lead to the reduction in smoke sensory characteristics in wine and ultimately diminish the loss of product.

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UNTARGETED METABOLOMICS ANALYSES TO IDENTIFY A NEW SWEET COMPOUND RELEASED DURING POST-FERMENTATION MACERATION OF WINE.

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III.P.17

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Keywords: Untargeted metabolomic analysis, Taste, Sweetness, Mass spectrometry

The gustatory balance of dry wines is centered on three flavors, sourness, bitterness and sweetness. Even if certain compounds were already identified as contributing to sweetness, some taste modifications remain largely unexplained^{1,2}. Some empirical observations combined with sensory analyzes have shown that an increase of wine sweetness occurs during post-fermentation maceration³. This step is a key stage of red winemaking during which the juice is left in contact with the marc, that contains the solid parts of the grape (seeds, skins and sometimes stems). This work aimed to identify a new taste-active compound that contributes to this gain of sweetness.

Recent developments have highlighted the interested of untargeted metabolomic analysis for oenology⁴⁵. Using similar tools, an original approach has been developed here to discover new sweet molecules released during post-fermentation maceration. In this context, different samples were taken from eight Bordeaux wineries over three vintages. These samples, coming from a total of 240 vats, were collected at two distinct stages, giving rise to two modalities: at the end of alcoholic fermentation and just before running-off the vat, that is before and after post-fermentation maceration. The analyses were assayed using liquid chromatography-high resolution mass spectrometry (UHPLC-Q-Exactive Plus, Orbitrap analyzer). Data processing was carried out using the MzMine 2 software followed by a differential analysis and statistical study executed with the R software to obtain a list of ions showing a strong increase during maceration. The MS² spectral data, obtained by fragmentation of molecules, provided information for their identification.

One of these ions was selected and considered for a targeted purification by various separative techniques (SPE, CPC and HPLC-preparative). Its structural elucidation by NMR allowed to identify this compound for the first time in wine. Furthermore, sensory analysis revealed its pronounced sweet taste.

This study proposes new tools to investigate taste-active compounds in wine. More generally, the results bring new insights to understand the chemical origin of wine taste and open promising perspectives for practical applications.

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III.P.18 AGEING BOTTLED WINES SUBMERGED IN SEA: DOES IT IMPACT WINE COMPOSITION?

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Keywords: ageing, red wine, volatiles

Aging wines is a common practice in oenology, which in recent years has undergone some innovations. Currently, we are witnessing the practice of aging bottled wine in depth, immersed in the sea or in reservoirs, for variable periods of time, but so far, little is known about the impact of aging in depth on the physicochemical properties, of wines.

The objective of this work was to evaluate the impact of this practice on the physicochemical characteristics, in particular to verify changes in the volatile composition of wines bottled and subsequently immersed in depth. A red wine from Cabernet Sauvignon was bottled and a set of bottles were submerged from July to February (2020), another set of bottles were submerged from February to September (2020) and another set was kept in the wine cellar. Bottles from each set were analyzed (in triplicate) in July 2021.

Wines basic parameters were analyzed according to OIV methods (1). Phenolic compounds and color were determined by UV-VIS spectrophotometry (2,3,4). Volatile compounds were determined by HS-SPME coupled with gas chromatography with time-of-flight mass spectrometry (GC/TOFMS) detection (5).

Regarding the color, the intensity and tone did not change significantly with aging in depth. There was a decrease in the content of total flavonoids and non-anthocyanic flavonoids and an increase in the content of total polyphenols, free anthocyanins and total anthocyanins.

Regarding volatile compounds 60 were identified and 26 shows a significantly difference among the 3 set of bottles when an analysis of variance was performed. A canonical discriminant analysis, performed only with variables that were significantly different, allows to discriminate the wines regarding the volatile compounds. In spite of being a preliminary study, results pointing out to a difference in wine characteristics as a result of maintaining bottles submerged in sea.

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III.P.19 AROMA ASSESSMENT OF COMMERCIAL *SFORZATO DI VALTELLINA* WINES BY INSTRUMENTAL AND SENSORY METHODOLOGIES

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Keywords: volatile compounds, sensory analysis, Sforzato di Valtellina, red wine

Sforzato di Valtellina DOCG is a special dry red wine produced from partially dehydrated Nebbiolo winegrapes growing in the Rhaetian Alps valley of Valtellina (Lombardy, Italy). Valtellina terraced vineyards are located at an altitude of 350–800 m according to 'heroic' viticulture on steep slopes. The harvested grape bunches are naturally dehydrated indoors, where a slow and continuous withering occurs (about 20% w/w of weight loss), until at least 1st December when the grapes reach the desired sugar content and can be processed following a normal winemaking with maceration. The wines can be marketed only after 20 months of aging, of which at least 12 in wood. Despite the increasing economic importance, few studies have been published on *Sforzato di Valtellina* wine and to our knowledge none on volatile organic compounds (VOCs).

In this study, VOCs of *Sforzato di Valtellina* wine were determined by HS–SPME–GC–MS. Sensory analysis was also performed by QDA and CATA methodologies with the aim of establishing correlations between VOCs content and perceived aroma intensity and descriptors. Thirty-two wines were analyzed in 2021 from two consecutive vintages (17 wines of 2016 and 15 wines of 2017), representing wineries producing 90% by volume of this denomination.

In addition to fermentative VOCs (mainly ethyl esters and some acids), terpenes and norisoprenoids were found in contents that could potentially contribute to the aroma of *Sforzato di Valtellina* wines. The 2016 wines were significantly richer in total VOCs than 2017 ones, particularly regarding total norisoprenoids and fermentative esters. This result was confirmed also in sensory analysis, with 2016 wines having higher aroma intensity than 2017 ones. Despite the variability found among the wines analyzed for each vintage, some differences were reported in the contents of single compounds: vitispirane and TDN (norisoprenoids), β -pinene and linalool (terpenes), diethyl malate, ethyl hexanoate, ethyl octanoate and methyl octanoate (esters), and hexanoic and octanoic acids (volatile acids). During aging, some compounds can be formed through chemical reactions depending on wine composition and storage conditions. These reactions include ester hydrolysis and formation (associated to fruity-related descriptors), hydrolysis of non-volatile glycoside precursors and chemical rearrangements of norisoprenoids and monoterpenes promoting balsamic-type descriptors, associated here mainly to vitispirane and β -pinene.

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III.P.20 ASSESSMENT OF 'DOLCETTO' GRAPES AND WINES FROM DIFFERENT AREAS OF OVADA DOCG

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Keywords: Dolcetto, phenolic content, autochthonous varieties, red wines

Dolcetto (*Vitis vinifera* L.) is one of the traditionally cultivated varieties in Piedmont (north-east Italy). Dolcetto wines have long been associated with local consumption and they are little known internationally. In particular, the Ovada area (south-east Piedmont), even if it represents a small share of the regional PDO Dolcetto production, is one of the oldest and vocated territory, giving wine also suitable for aging. In this study, the basic composition and phenolic content of Dolcetto grapes for Ovada DOCG wines have been investigated in three different vintages (2020–2022), as well as the main aspects of the derived commercial and experimental wines (basic parameters, phenolics, volatile compounds, sensory properties).

Grapes from fifteen vineyards, belonging to three Ovada DOCG areas, were harvested at the same grape soluble solids content (about 13.0–13.5% v/v potential alcohol) and were evaluated in terms of basic traits, phenolic ripeness, and skins and seeds phenolic composition. The commercial wines produced from these vineyards were analyzed for 2020 and 2021 vintages. Among them, representative vineyards were also selected for experimental standardized winemaking to establish correlations between grapes and wines results.

The results showed different acidic content at harvest, with higher values for area 3 that resulted also in lower sugar content, and differences among the vintages studied according to the weather conditions (2020 was wetter than 2021 and 2022). The phenolic ripeness parameters changed moderately among the three areas, even though the cell maturity index (EA%) and the seed maturity index (Mp%) reported no significant differences, with a higher vintage effect. The berry skins phenolic composition differed among areas, being the lower values of total polyphenols, total flavonoids, and total anthocyanins observed in area 1. Significant differences for polyphenols were found depending also on the vintage. Moreover, the seasonal conditions affected the berry weight, increasing the seeds polyphenols ratio on the total content in the drier years (2021-2022) although with different extent depending on the area. The results on experimental wines could be useful to assess if they correspond to those predicted from grape analysis, helping winemakers in improving vinification protocols according to the desired wine style.

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CHARACTERISTIC EXTRACTION OF THE PHENOL COMPOUNDS IN KOSHU (*VITIS VINIFERA* CV.) WINE DURING THE MACERATION

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Keywords: Koshu, phenol, proanthocyanidin, maceration

Koshu is one of the indigenous grape variety that has been grown in Japan for more than one thousand years. Recent research showed that it has 70% of *Vitis vinifera* genes. In 2010, the Koshu variety was included in 'International List of Vine and Varieties and their Synonyms' managed by the 'International Organisation of Vine and Wine' and has further fueled its popularity in Japan. It is the most cultivated variety for winemaking in Japan.

Koshu berries have light purple skins. The variety is mainly used to produce white wines such as an aromatic wine and a wine produced by sur lie method although various styles are produced. Whereas it is also used to produce orange wine which is fermented with their seed and skin. Previous study showed that β -damascenone could contribute to the fruity aroma in this style of wine. However, there is a few knowledge about the behavior of extraction of the phenol compounds during the fermentation and maceration, even though they are important compounds for the taste balance of such style. Furthermore, wines produced by Koshu grapes are sometimes described as bitter. Thus, the object of this study is to reveal the character of Koshu wine which is produced by the maceration at the point of view of phenol compounds.

In this study, Koshu was compared with other *V. vinifera* varieties, Sauvignon blanc (SB) and Merlot (MN). Each fruit tissues were separeted in pulp, seed and skin, and soaked separately in model wine solution (12% ethanol, tartaric acid 3000 mg/L, pH 3.2) during 14 days for study its capacity of extraction of phenol compounds at same alcohols degrees as wine. The results showed that proanthocyanidin was extracted by SB and MN seed and its content incresase during the sorking. On the other hand, proanthocyanidin was not extracted by Koshu seed, despite the presence of proanthocyanidin. Furthermore, these 3 varieties were fermented with seed and skin during 28 days for study the behavior of extraction of the phenol compounds during the fermentation and maceration in Koshu wine. The result showed that the content of proanthocyanidin and total phenols in Koshu wine were dramatically decreased during maceration. In contrast, those of SB and MN wine were stable or increased during maceration.

These new findings on the unique characteristics of proanthocyanidin in Koshu grape could might be showed the diversity of character in *Vitis*. And these insights are expected to contribute to the control of the taste of wine.

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CHARACTERIZATION AND ANTIBACTERIAL ACTIVITY OF A POLYPHENOLIC EXTRACT OBTAINED BY GREEN SUPERCRITICAL CO₂ EXTRACTION FROM RED GRAPE POMACE

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Keywords: polyphenolic extract, supercritical fluid, antioxidant, antibacterial

Upgrading wine industry solid wastes is considered as one of the main strategies to support the circular economy. Red grape pomaces constitute a rich source of polyphenols, which have been shown to possess antioxidant properties and to provide benefits for human and animal health. The objective of this work was to obtain and characterise polyphenolic extracts from red grape pomaces via green supercritical CO₂ extraction using ethanol as a co-solvent, and to evaluate their antibacterial activity against susceptible and multidrug-resistant *Escherichia coli* strains of animal intestinal origin.

Pomaces obtained from the vinification of red grapes of *Vitis vinifera* L. cv. Graciano were lyophilised and grinded. Effect of various values of ethanol concentration (10 – 70 %), extraction time (1 – 5 h), temperature (30 – 50°C), particle size (1.25 – 0.2 mm) and pressure (150 – 600 bar) were investigated. To-tal phenolic quantification and antioxidant activity were assessed by rapid in vitro spectrophotometric assays. Phenolic profiles were identified using ultra-high performance liquid chromatography coupled to a triple quadrupole/ion trap mass spectrometer and by GC-MS analysis. The antibacterial activity of the extracts was tested by the microtiter dilution assay against a collection of *E. coli* strains and minimal inhibitory concentration (MIC) values were determined.

Results showed that the extract obtained under the optimal conditions exhibited the highest value of antioxidant activity (3.79 mg Trolox equivalents/g) in the assay, and the highest antimicrobial activity (MIC value of 2 mg/mL) against all the studied antibiotic susceptible and resistant E. coli strains. Chemical analyses enabled the identification of 32 volatile compounds and phenolic compounds belonging to the groups of flavonols and hydroxybenzoic acids, and the contents of these phenolic compounds were positively correlated with the antibacterial activity.

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CHARACTERIZATION OF THE VOLATILE COMPOUNDS PROFILE OF COMMERCIAL GRAPPAS OBTAINED FROM THE POMACE OF AMARONE WINES

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III.P.23

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Keywords: Grappa, Amarone, volatile profile, GC-MS

Grappa is a traditional Italian alcoholic beverage, with an alcohol content generally between 40–60% vol., obtained from the distillation of grape pomace used for the production of wine. Grappa are often aged in wooden barrels. There are various types of grappa: young, aromatic, aged, extra-aged depending on whether the distillate comes from aromatic vines or is aged in wooden barrels for shorter or longer periods. There is also flavored grappa if herbs, fruit or roots are added. All this makes it an extremely heterogeneous product both from an organoleptic and compositional point of view. There is therefore the need to deepen the characterization of the different types in order to optimize the production processes and improve the products by enhancing their distinctive characteristics.

This work aims to characterize the volatile compounds profile of the Grappa obtained from the pomace of Amarone, a red wine from the province of Verona (northern Italy) produced from withered grapes of the Corvina and Corvinone varieties. For this study, the volatile profiles of 19 samples of Amarone grappa were compared with those of 7 grappas not obtained from Amarone pomace but from other vines. The aromatic profiles were obtained by SPE extraction of the volatile molecules followed by GC-MS analysis. A total of 62 compounds belonging to various chemical classes (alcohols, C6, terpenoids, sesquiterpeneoids, norisoprenoids, benzenoids, fatty acid esters) were identified and quantified in the samples. Through non-parametric statistical analysis (Kruskal Wallis) the compounds characterizing the Amarone grappas were identified, including: hexanoic acid, ethyl octanoate, 1,4-cineole, β -damascenone, β -ionone, TPB, 2,2 dihydrofarnesol and α -farnesol. Furthermore, the Amarone grappa was more characterized by compounds linked to the aging of the distillate in wood such as: syringaldehyde, vanillin, 2-methoxyphenol, whiskey lactone. In conclusion, these results allow us to better understand which compounds could be characterizing Amarone grappa, in order to study their behavior more thoroughly during the various production phases in order to manage the aromatic potential of these products.

III.P.24 DETERMINATION OF FREE AMINO ACIDS, AMINO ACID POTENTIAL AND PROTEASE ACTIVITY IN THE LEES AND STILL WINES OF CHAMPAGNE

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Keywords: Nitrogen, relesable nitrogen, lees, ageing

Prior to winemaking, organic or mineral nitrogen compound concentrations are usually measured in the vineyard and in grape musts. These indicators facilitate vine cultivation decisions, usually through yield or vigor. During vinification, yeast and bacteria metabolize nitrogen compounds in the musts in order to generate biomass. After fermentation, the microorganisms rerelease a part of this nitrogen as soluble compounds into the wines. Another part remains bound in the lees and can be lost during racking. The must's natural nitrogen quantities, additional supplements during fermentation, and lees contact management enhance the release of nitrogen compounds to the wines. During ageing these nitrogen compounds – primarily the amino acids – are implicated in the generation of odorous compounds such as heterocycles(1).

Yeast cellular autolysis facilitates the diffusion of proteins and peptides into the wine. Associated with protease activity, amino acids can be released during ageing. The organic nitrogen content of wines and lees post-fermentation is not yet fully understood. This study explored the quantities of free amino acids, potential amino acids, and protease activity in champagne still wines and their corresponding lees. To achieve this analysis, a new quantification method to detect protease activity was developed using fluorescein isothiocyanate bound to a casein substrate.

In addition, the current method used to quantify potential amino acids after acid hydrolysis was optimized and associated to the previously published HPLC/FLD method for quantifying free amino acids(2). The methods were also adapted for quantifying lees. This analytical toolbox allows the observation of nitrogen compound kinetics over time, and was subsequently applied to sixteen young wines and their corresponding lees. The results of this study highlight a high variability in amino acid content between wines and lees. This suggests huge differences between amino acids levels in wine and in lees. No direct correlation was observed between lees quantity and the concentration of free and potential amino acids, indicating the mechanism is more complex. After 5 months of wine ageing with lees contact, the wines with lower levels of amino acids showed higher protease activity. That result encourages us to continue studying yeast lees and their variable capacities to release amino acids into wine over time.

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III.P.25 EFFECTS OF INDUCED SUNBURN DAMAGES ON WHITE WINE PROPERTIES

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Keywords: climate change, sunburn, white wine, off-flavour

Climate change is a great challenge for the environment and affects the wine industry as well. Sunburn damage of sensitive grapes increase with severe heat periods. Besides significant loss of yield sunburn, modifies sensory properties of the wines and may cause climate-related off-flavours. To initiate sunburn in a controlled way, in 2021 sunburn was directly induced in the vineyard with the GrapeBurner device, exposing grapes of the varieties Riesling and Pinot Blanc with UV and IR radiation. This device was first assembled by Kai Müller of the university in Geisenheim and consists of a carriage with 6 UV/IR lamps. A 15 min irradiation was applied in early September at 60°Oe. Due to the colder season in 2021 the grapes were not harmed by previous sunburn damage. Nevertheless, we used non-defoliated grapes facing north of the vines to ensure that putative damage was only due to applied irradiation and not previous sun exposure. Three weeks after the treatment, a control and the irradiated grapes were harvested and directed to small scale winemaking following a standardised protocol. Sensory evaluation using descriptive analysis with a trained panel was complemented by aroma analysis using an established head space solid phase micro extraction GC-MS method.

For Riesling, the control had a sweeter and fruitier taste, and was reminiscent of ripe fruit, which correlated with a high content of ethyl esters. In contrast, wines from irradiated Riesling grapes had a smokier flavour and correlated with plant derived odorants such as vitispirane, linalool, β -damascenone, and 4-vinylguaiacol. Also the sensory panel described the wines from treated grapes as more astringent and bitter, as well as with a more intense yellow colour. These results were backed by Pinot Blanc trials, as the control was perceived more fruity due to enhanced ethyl esters. Vice versa, wines from the irradiated grapes were more smoky and astringent. In addition, 2-aminoacetophenone was enhanced, causing the atypical aging off-flavour.

These promising results stimulated further trials, in which the length of exposure was varied gradually, to determine a threshold from which irradiation is causing negative sensory and compositional changes. In parallel, mitigation strategies such as applying reflecting particles or adaptation to sun exposure by early defoliation will be examined in lieu of varying UV/IR radiation.

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EVALUATION OF INDIGENOUS CANADIAN YEAST STRAINS AS WINE STARTER CULTURES ON PILOT SCALE FERMENTATIONS

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Keywords: Indigenous strains, metabolites, volatile compounds, wine fermentation

The interactions between geographical and biotic factors, along with the winemaking process, influence the composition and sensorial characteristics of wine¹. In addition to the primary end products of alcoholic fermentation, many secondary metabolites contribute to wine flavor and aroma and their production depends predominantly on the yeast strain carrying out the fermentation. Commercially available strains of S. cerevisiae help improve the reproducibility and predictability of wine quality. However, most commercial wine strains available on the market have been isolated from Europe, are genetically similar, and may not be the ideal strain to reflect the terroir of Canadian vineyards². Instead, indigenous S. cerevisiae strains may enhance the typical sensory properties and characteristic profile of the wine region³. The Okanagan Valley is the major wine-producing region in British Columbia, Canada. The Measday lab has isolated S. cerevisiae indigenous strains from Okanagan Valley vineyards that are genetically distinct from commercial strains⁴. After evaluating the oenological characteristics of six indigenous strains isolated from Okanagan Crush Pad (OCP) winery in laboratory-scale fermentations, two were selected for pilot-scale winery fermentations to assess their potential as wine starter cultures. Fermentations with OCP088 and OCP125 yeast strains were carried out in triplicate 250L stainless steel barrels at OCP winery. Vin Gris (VG, Pinot Noir) and Pinot Gris (PG) varietals were chosen, the grapes were pressed, and the juice was settled to remove skins before inoculation. Major metabolites (organic acids, sugars, and ethanol) were quantified using HPLC-RID, sugar in both wines was mainly fructose, ranging between 16 g/L and 20 g/L, ABV of the finished product ranged between 10.8 and 11.3 %. Volatile compounds (terpenes, esters, ketones, and higher alcohols) were identified using SPME-GC/MS We identified the following number of volatile compounds in each fermentation: OCP125 PG (56), OCP088 PG (52), OCP125 VG (45), OCP088 VG (44). The majority of volatile compounds were esters, which are known for their contribution to wine quality. OCP 125 tended to produce more terpenes than OCP 088. Some of these compounds are responsible for honey and grapefruit-like aromas, which are atypical of these varietals, adding to the complexity of the final product.

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FOURIER TRANSFORM INFRARED SPECTROSCOPY IN MONITORING THE WINE PRODUCTION

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Keywords: FTIR spectroscopy, wine, quality control, authenticity assessment

The complexity of the wine matrix makes the monitoring of the winemaking process crucial. Fourier Transform Infrared Spectroscopy (FTIR) along with chemometrics is considered an effective analytical tool combining good accuracy, robustness, high sample throughput, and "green character". Portable and non-portable FTIR devices are already used by the wine industry for routine analysis. However, the analytical calibrations need to be enriched, and some others are still waiting to be thoroughly developed. For this reason, an extended literature review took place identifying gaps for further research meeting the needs of the modern wine industry (Thanasi et *al.*, 2022). The methodology that was followed was based on grouping the different studies according to the main sampling material used – 1) leaves, stems, and berries; 2) grape musts; 3) wines. For each sampling material the studies were categorized in terms of 1) main aim of the analysis; 2) type of sample; 3) sample preparation mode; 4) wavenumber range (/cm); 5) spectral pre-treatment; 6) statistical method.

The most important findings were: 1) the different sample preparation modes can influence the spectra; 2) a limited number of samples (less than 100 in most cases) was used and the validation took place with cross-validation tests; 3) the developed models were not applied to different grapevine cultivars, harvests, and types of wines; 4) many developed methods were focused on a specific oenological parameter or chemical compound or a specific stage of the winemaking process; 5) compounds with a concentration higher than 1 g/L are easier to be determined by FTIR; 6) the complexity of the wine matrix and the chemical similarity of the compounds under study makes the interpretation of the spectra very difficult due to several interferences.

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III.P.28 HOW OXYGEN CONSUMPTION INFLUENCES RED WINES VOLTAMMETRIC PROFILE

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Keywords: Red wine, Oxygen, Phenolic compounds, Voltammetry

Phenolic compounds play a central role in sensory characteristics of wine, such as colour, mouthfeel, flavour and determine its shelf life. Furthermore, the major non-enzymatic wine oxidation process is due to the catalytic oxidation of phenols in quinones. Due their importance, during the years have been developed different analytical methods to monitor the concentration of phenols in wine, such as Fo-lin-Ciocalteu method, spectrophotometric techniques and HPLC. These methods can also be used to follow some oxidation-related chemical transformations. However, these methods are complex, expensive and time consuming, thus not affordable for most small winery. Consequently, the development of several techniques that are faster, cheaper and user-friendly are currently of great interest. Among these, voltammetry has shown to be able to discriminate well wines according to their chemical composition, in particular in relationship to phenolic compounds.

Aims: Our work aimed to understand the effect of oxygenation on voltammetric signals of wines.

Material and Methods: DIfferent commercial red wines purchased from a local stores have been used in this study. The experimental protocol involved oxygenation of the wines in a 1 L bottle by hand shaking until the desired oxygen level was reached. Three level of oxygenation were chosen: 1.5 (TA), 5 (TB) and 7 mg/L O_2 (TC). Measures of oxygen have been carried out with a portable oximeter. The oxygenated wine was placed in 125 ml glass vials fitted with an oxygen sensor, filled without leaving any headspace and closed with sealing wax in order to not have oxygen exchanges with the outside.

Electrochimical measurements were performed with a with a potentiostat using disposable screenprinted sensors in a three-electrode arrangement. Total polyphenols, anthocyanins, free and total SO_2 measurements were carried out using a multiparametric analyser and the dedicated kit. For colour determination were measured the absorbances at wavelengths 420, 520 and 620.

The measures to determine the oxygen kinetic consumption were performed every 24 for hours. At the same time were carried out analysis with the multiparametric analyser and for colour determination.

Results: Red wine voltammograms were impacted by oxygenation, with several voltametric features showing variation in profile and peak intensity according to the level of oxygen consumption. Different signal treatments strategies were applied to highlight the regions of the voltammograms mostly affected by oxidation, in particular through the use of derivative voltammetry.

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IDENTIFICATION AND LEVELS OF PHENOLIC COMPOUNDS (TANINS, ANTHO-CYANS) IN RED VARIETAL WINES (PROKUPAC AND BLACK TAMJANIKA) FROM SERBIA

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Keywords: wine phenolics, Serbian red varietal wines, Prokupac, Black Tamjanika

The phenolic compounds of red wines represent a source of numerous benefits for human health, which is why they are a constant subject of scientific research. Winemaking in Serbia has a growing economic significance, with particularly autochthonous varieties included [1]. This research identifies and quantifies phenolic compounds of Serbian red varietal wines of Prokupac and Black Tamjanika varieties. Quantification of the level of phenolics has been conducted, including molecular tannins [(+)-catechin, (-)-epicatechin, procyanidin dimers B1, B2, B3, B4], molecular anthocyanins, and the mean degree of polymerization of tannins by HPLC by UV detection, total antioxidant capacity via spectrophotometric methods and chromatic characteristics via CIELAB. For research 21 Serbian red varietal wines were used - Prokupac and Black Tamjanika varieties, 16 and 5, respectively. Wine samples are from different Serbia winegrowing regions and different vintages, from 2015 to 2019. The aim of the research is an investigation of autochthonous Serbian red varietal wines with an emphasis on phenolic compounds. The results obtained are showing the diversity and important differences between the phenolic compounds of these two varieties.

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IMPACT OF CLIMATIC ZONES ON THE AROMATIC PROFILE OF CORVINA WINES IN THE VALPOLICELLA REGION

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Keywords: Corvina, huglin index, temperature increase, aroma profile

In Italy, in the past two decades, the rate of temperature increases (0.0369 °C per year) was slightly higher compared to the world average (0.0313 °C per year). It has also been indicated that the number and intensity of heat waves have increased considerably in the last decades. (IEA, 2022). Viticultural zones can be classified with climatic indexes. Huglin's index (HI) considers the temperature in a definite area and has been considered as reliable to evaluate the thermal suitability for winegrape production (Zhang et al., 2023).

In this scenario, understanding the relationship between climatic conditions existing in specific grape growing areas and the composition of the grapes and wines composition grown in that particular region is of major interest. The aim of this project is to investigate the aromatic profile of Corvina grapes and wines in the Valpolicella region and how it is impacted by the different climatic zones. Valpolicella is a wine-making region found in the north of Italy in the zona of Veneto, it is divided into three zones (Valpolicella Classica, Valpantena, and Orientale). All zones are subdivided into valleys and hilly areas which can range from 30 up to 500 m a.s.l., this variation in altitudes plays a role in the climatic conditions.

First, the climatic zones were studied in the region. Data from 24 weather stations across this region showed that there are 5 different HI climatic zones from temperate to too hot. Based on this information, grapes were obtained from 16 different vineyards from three different climatic zones (warm temperate, warm, and very warm), in order to carry out micro vinifications and grape macerations. Vinification was carried out in triplicate with 800 g of Corvina grape in bottles of 1 L *Saccharomyces cerevisiae* AWRI 796 (Experti Srl) and potassium metabisulphite was added, and fermentation was carried at 22 °C until it reached a concentration of ~1 g/L of glucose-fructose. In addition, grape macerates were also prepared to investigate the varietal compounds in the absence of yeast activity. Grape macerates were carried out in triplicate with 500 g of Corvina grapes in bottles of 1 L with ethanol (15% w/w), potassium metabisulphite, and dimethyl decarbonate at 22 °C for 15 days.

For the quantification of alcohols, esters, fatty acids, benzenoids, terpenes, and volatile sulfur compounds, a combined analytical strategy involving SPE and SPME extraction methods followed by GC-MS analysis was used. Enological parameters were measured using a Biosystems Y15 multiparametric analyzer. Results will contribute to developing tailored strategies for climate change management for Valpolicella wines.

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III.P.31 IMPACT OF RHIZOPUS AND *BOTRYTI*S ON WINE FOAMING PROPERTIES

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Keywords: wine foam, Rhizopus, yeast proteins, aspartic protease

A lot of work has been done on the impact of *Botrytis* on the foam of sparkling wines. This work often concerns wines produced in cool regions, where *Botrytis* is the dominant fungal pathogen. However, in southern countries such as Spain, in particularly hot years such as 2022, the majority fungal pathogen is sometimes Rhizopus. Like *Botrytis*, Rhizopus is a fungus that produces an aspartic protease. The aim of this study was therefore to determine the foaming properties of wines produced with a synthetic must contaminated by a Rhizopus or *Botrytis* culture. In order to confirm the identification of the fungal strain, the D1-D3 domains of the 28S rRNA gene were amplified and sequenced. BLAST search indicated 100% identity with a reference strain of *Rhizopus lyococcus* (CBS 320.35).

The complete experimental design presents 12 modalities (AF in triplicate, i.e. 36 bottles). The fungal isolates of *Botrytis cinerea* (*B. c.*) and *Rhizopus lyococcus* (*R. l.*) were cultured using a modified version of the method described by Gimenez et al. (2022). Alcoholic fermentations (AF) were performed in 500mL glass bottles from synthetic grape must supplemented or not with 50 mg/L of epicatechin. The yeast strain *S. cerevisiae* Lalvin EC1118 (Lallemand) was used for the AF process. To examine the impact of the pathogenic fungi, 10% (v/v) of *B.c.* or *R.l.* culture were added (separately) to the model grape juice. Furthermore, two different concentrations of L-malic acid were added to the fermentation media creating two sets of conditions : 2g/L of L-malic acid (pH=3.5) and 6 g/L of L-malic acid (pH=3). The results of the wines with fungus were compared to those of the control wines obtained without fungus.

The results of this study show that the presence of Rhizopus in the must significantly or highly significantly degrades the foamability and foam stability of the wines (foam measured with the KRUSS DFA100 equipment). The analysis of the protein composition by SDS-PAGE clearly shows a degradation of the yeast proteins by the fungal proteases of Rhizopus. Surprisingly, the *Botrytis* strain used did not affect the foam of the wines. These differences in proteolytic activity are confirmed by using BSA as a substrate: the Rhizopus culture degrades the 500 mg/L BSA in a few minutes, whereas the BSA degradation by the *Botrytis* culture remains considerably lower despite the longer culture of the fungus. Finally, the presence of epicatechin did not affect the wines' foaming properties.

INFLUENCES OF SO₂ ADDITION AND STORAGE CONDITIONS IN THE DETERMINATION OF MEAN DEGREE OF POLYMERIZATION OF PROANTHOCYANIDINS IN AGED RED WINES.

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Keywords: phloroglucinolysis, SO₂, proanthocyanidin mean degree of polymerization (mDP)

The structural diversity is one of the most remarkable characteristics of proanthocyanidins (PA). Indeed, PA in wines may vary in the B-ring and C-ring substitutes, the C-ring stereochemistry, the degree of polymerization (DP) and the linkage between the monomers. Knowing in detail the structural characteristics of the PA of a wine can help us to understand and modulate several sensorial characteristics of the wine, such as color, antioxidant properties, flavor, and mouthfeel properties. In the last years was discovered and confirmed the presence of sulfonated monomeric and oligomeric flavan-3-ols in wine [1], as well as was pointed out their importance in wine quality [1,2]. Aim of this work was to explore if and how the presence of the sulfonated PA can influence the wine PA profile and mDP, at different storage parameters. The sample set used included 5 single cultivar wines, four levels of SO₂ and two storage conditions, while all wines were analyzed by phloroglucinolysis reaction - UPLC-MS/MS recently published [3]. The results showed that after the phloroglucinolysis reaction the epicatechin sulfonate increased more than 30 times. The formation of the phloroglucinol adducts after the reaction is highly influenced for the storage conditions, and therebefore the mDP. The wines stored in cellar temperatures has the double of the concentrations of phloroglucinol adducts in comparison to the wines stored in room temperature. The inclusion of epicatechin sulfonate in the determination of mDP leads to lower values in all studied wines, highlighting the relevance of the sulfonated proanthocyanidins in the determination of this relevant parameter.

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INVESTIGATION INTO MOUSY OFF-FLAVOR IN WINE USING GAS CHROMATO-GRAPHY-MASS SPECTROMETRY WITH STIR BAR SORPTIVE EXTRACTION

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Keywords: mousiness, N-heterocycles, quantification, SBSE-GC-MS

Mousy off-flavor is one of the defects of microbial origin in wine. It is described as a particularly unpleasant defect reminiscent of rodent urine (a "dirty mouse cage"), and grilled foods such as popcorn, rice, crackers, and bread crust. Prior to the 2010s, mousiness was very uncommon but it has been becoming more frequent in recent years. It is often associated with an increase in pH as well as certain oenological practices, which tend to significantly decrease the use of sulfur dioxide.

Three major compounds, 2-acetyl-1-pyrroline (APY), 2-acetyltetrahydropyridine (ATHP) and 2-ethyltetrahydropyridine (ETHP), have been identified as responsible for mousiness in wines. A particularity of these compounds is that they are prone to tautomerism and can coexist in several forms. Moreover, the nitrogen atom in the heterocyclic ring can be protonated under specific conditions, i.e. with pH lower than pKa, inducing a positive charge and, as a consequence, an increase of polarity and a loss of volatility of the molecule.

To date quantification data reported in the literature are limited due to analytical issues related to the nature of these compounds. To fill the gap and later understand the parameters influencing mousiness, the objective of this study was to develop a simple and effective method to simultaneously determine trace levels of these three mousy N-heterocycles in wines. Therefore, a stir bar sorptive extraction (SBSE) followed by GC-MS analysis was developed (1).

Firstly, both previously reported tautomers of ATHP (2), 2-acetyl-1,4,5,6-tetrahydropyridine and 2-acetyl-3,4,5,6-tetrahydropyridine were identified, unlike to APY and ETHP. The extraction conditions were then optimized paying particular attention to the pH of the sample. The performance of the developed method was evaluated on white, rosé and red wines and the limits of detection and quantification of the method are lower than previously published concentrations in spoiled wine.

The method was then applied to provide quantitative data by analyzing 6 control wines and 68 wines produced without added sulfites. ETHP was detected in almost all wines produced with limited use of SO₂. ATHP was detected in almost all wines suspected of mousiness whereas APY was only detected in few cases. This method will provide a support for further studies aimed at understanding the phenomena that influence the occurrence of mousy off-flavor and the oenological parameters that modulate its expression.

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III.P.34 MAPPING OF GAS-PHASE CO₂ IN THE HEADSPACE OF CHAMPAGNE GLASSES BY USING AN INFRARED LASER SENSOR UNDER STATIC TASTING CONDITIONS

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Keywords: TDLAS, Champagne, CO₂, Effervescence

From the chemical angle, Champagne wines are complex hydro-alcoholic mixtures supersaturated with dissolved carbon dioxide (CO₂). During the pouring process and throughout the several minutes of tasting, the headspace of a champagne glass is progressively invaded by many chemical species, including gas-phase CO₂ in large majority. CO₂ bubbles nucleated in the glass and collapsing at the champagne surface act indeed as a continuous paternoster lift for aromas throughout champagne or sparkling wine tasting [1]. Nevertheless, inhaling a gas space with a concentration of gaseous CO₂ close to 30% and higher triggers a very unpleasant tingling sensation, the so-called "carbonic bite", which might completely perturb the perception of the wine's bouquet. Therefore, to enhance the champagne tasting experience, monitoring gas-phase CO₂ in the headspace of champagne glasses has become a topic of interest over the last dozen years [2–5].

Based on the Tunable Diode Laser Absorption Spectroscopy (TDLAS), a CO_2 -Diode Laser Sensor (CO_2 -DLS) with two distributed feedback (DFB) diode lasers emitting at 4986.0 and 3728.6 cm-1 was developed to allow the fine monitoring of gas-phase CO_2 over a large concentration range from 0.5% to 100%. Moreover, to perform the simultaneous spatial mapping of CO_2 along a multipoint array in the headspace of champagne glasses, two couples of galvanometric mirrors were combined with a couple of parabolic mirrors symmetrically positioned on either side of the glass headspace [4,5]. Thereby, the CO_2 -DLS shows a very high temporal resolution thus enabling an accurate monitoring and mapping of gas-phase CO_2 in the headspace of glasses.

Real-time monitoring of gas-phase CO_2 was thus performed with the CO_2 -DLS, under static tasting conditions, in the headspace of several types of champagne glasses showing distinct shapes and volume capacities (including the 21 cL INAO glass, a worldwide reference for sensory evaluation). Moreover, a brand-new glass recently proposed as a universal glass for the tasting of still and sparkling wines (the 45 cL ŒnoXpert) was also examined. A kind of CO_2 fingerprint, evolving in space and time, was unveiled for each glass type. After a strong increase of the gas-phase CO_2 concentration observed within the several seconds of the pouring step, a vertical stratification of CO_2 was unveiled in the headspace of glasses, with decreasing CO_2 concentrations while moving away from the champagne surface, and as time elapses.

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MAPPING THE CONCENTRATIONS OF GASEOUS ETHANOL IN THE HEADSPACE OF CHAMPAGNE GLASSES THROUGH INFRARED LASER ABSORPTION SPECTROSCOPY

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Keywords: Ethanol, Champagne, Interband Cascade Laser, Spectroscopy

Under standard wine tasting conditions, volatile organic compounds (VOCs) responsible for the wine's bouquet progressively invade the glass headspace above the wine surface. Most of wines being complex water/ethanol mixtures (with typically 10–15 % ethanol by volume), gaseous ethanol is therefore undoubtedly the most abundant VOC in the glass headspace [1]. Yet, gaseous ethanol is known to have a multimodal influence on wine's perception [2]. Of particular importance to flavor perception is the effect of ethanol on the release of aroma compounds into the headspace of the beverage [1]. Moreover, triggered by the presence of ethanol in wines, the Marangoni effect increases the exhaust of flavored molecules in the glass headspace [2]. In addition, ethanol is known to modify the orthonasal detection threshold of aromas (and especially the fruity aromas [2]), and it can also trigger the trigeminal system leading to tingling and/or warm sensation [2]. Monitoring gaseous ethanol, in space and time, in the headspace of wine glasses is therefore crucial to better understand the neuro-physicochemical mechanisms responsible for aroma release and flavour perception.

For this purpose, micro-gas chromatography was used in the past to simultaneously monitor gas-phase ethanol and CO₂ in the headspace of champagne glasses, but with a relatively poor temporal resolution leading to a one-minute data sampling interval [3], [4]. Since the last decade at GSMA (Groupe de Spec-trométrie Moléculaire et Atmosphérique), tunable diode laser absorption spectroscopy has shown to be a well-adapted method to accurately monitor gas-phase CO₂ in the headspace of glasses poured with champagne [5]. The tunability of semiconductor laser with current modulation provides CO₂ monitoring with a high temporal resolution of 42 measurements per seconds. Lastly, thanks to the recent interband cascade laser (ICL) technology, the CO₂ sensor was upgraded to monitor gaseous ethanol. This new quantum laser source, combined with previous technology developed for the monitoring of gas-phase CO₂, allowed us to simultaneously monitor gas-phase CO₂ and ethanol under standard still wine and sparkling wine tasting conditions. The first data sets obtained in the headspace of a glass poured with a standard brut-labelled Champagne wine are presented

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NEW TOOL FOR SIMULTANEOUS MEASUREMENT OF OXYGEN CONSUMPTION AND COLOUR MODIFICATIONS IN WINES

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Keywords: Oxygen consumption, colour, wine, kinetics

Measuring the effect of oxygen consumption on the colour of wines as the level of dissolved oxygen decreases over time is very useful to know how much oxygen a wine is able to consume without significantly altering its colour. The changes produced in wine after being exposed to high oxygen concentrations have been studied by different authors, but in all cases the wine has been analysed once the oxygen consumption process has been completed. This work presents the results obtained with the use of an equipment designed and made to measure simultaneously the level of dissolved oxygen and the spectrum of the wine, during the oxygen consumption process from saturation levels with air to very low levels, which indicate the total consumption of the dosed oxygen.

For this purpose, this equipment has been designed, built and prepared and has been validated with the measurement of red wines made from different grape varieties.

The equipment built has 2 mm quartz cuvettes for the measurement of the spectrum in the visible with a spectrophotometer and a sensor for the simultaneous measurement of dissolved oxygen with a luminescent measurement system, both measurements were carried out every 15 minutes during oxygen consumption. The tightness of the cuvettes during the process of measuring the kinetics of oxygen consumption was evaluated, as well as the reproducibility of the measurements of both parameters.

The results of this study show that the equipment designed and built is valid for monitoring the kinetics of oxygen consumption with the simultaneous measurement of the spectrum in the visible and dissolved oxygen. The tightness tests corroborated that all the cells used simultaneously are airtight, keeping their interior totally isolated from the exterior, showing a variability between cells of less than 10%. The results of the repeatability tests showed that the same wine measured simultaneously in the different cuvettes showed the same results both in the measurement of the consumption kinetics and in the measurement of the spectrum in the visible. The application of the system developed for the study of red wines allowed to know the characteristics of the consumption kinetics, obtaining that all red wines were initially able to take up the same amounts of oxygen (O_{max}), with values of 174 hPa. However, the wines made with Tempranillo grapes showed higher oxygen consumption ($\Delta O_{max_{min}}$), 115 hPa, and lower residual oxygen values (Omin), 59 hPa compared to than those made with the Garnacha grapes with 84 y 88 hPa of O_{\min} and $\Delta O\max_{\min}$, respectively. One of the main advantages of this equipment is the ability to record the changes produced in the spectrum as the wine consumes oxygen, thus, an increase in red tones (450 and 580 nm) was observed in all the wines studied. It was found that the wines made with the Garnacha grapes underwent increases in absorbance between 400 and 460 nm and between 610 and 670 nm as they consumed oxygen, indicating an increase in the compounds responsible for the purple and yellow hues. On the other hand, wines made with the Tempranillo grapes, as they consumed oxygen, showed a decrease in purple hues.

OPTIMIZATION OF EXTRACTION AND DEVELOPMENT OF AN LC-HRMS METHOD TO QUANTIFY GLUTATHIONE IN WHITE WINE LEES AND YEAST DERIVATIVES

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Keywords: Glutathione, wine lees, RSM design, quantification

Glutathione is a natural tripeptide composed of l-glutamate, l-cysteine and glycine, found in various foods and beverages. In particular, glutathione can be found in its reduced (GSH) or oxidized form (GSSG) in must, wine or yeasts¹. Numerous studies have highlighted the importance of GSH in wine quality and aging potential². During winemaking, especially during aging on lees, GSH helps prevent the harmful effects of oxidation on the aroma of the wine³. Nevertheless, the amounts of GSH/GSSG present in wine lees are often unknown and the choice of operating conditions (quantity of lees and aging time) remains empirical.

Based on these observations, the present work aims at proposing an optimized method to extract and quantify the potential of glutathione from white wine lees and yeast derivatives. In this context, several parameters, such as the type of solvent, the extraction time and the solid-liquid ratio, were investigated. For each matrix, the optimization study was carried out using the Box-Behnken Design (3-factor, 3-level) based on 33 factorial experiments. The results showed that the main factor influencing the extraction efficiency was the ethanol concentration. After development and validation of a liquid chromatography–high resolution mass spectrometry quantitation method, GSH and GSSG were assayed in various white wine lees and yeast derivatives in order to investigate the influence of oenological parameters on their content. The results showed differences in concentration depending on the grape varieties and strains used.

For the first time, this study focused on the use of design of experiments and analytical techniques to highlight the presence of glutathione in wine lees and yeast derivatives. Moreover, this research offers promising perspectives for a better understanding of lees antioxidant potential during wine aging. More generally, by-products such as lees can provide new natural products to the food industry, with safer and better antioxidant qualities against oxidative damage.

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OPTIMIZATION, VALIDATION AND APPLICATION OF THE EPR SPIN-TRAPPING TECHNIQUE TO THE DETECTION OF FREE RADICALS IN CHARDONNAY WINES

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Keywords: chardonnay, radical, wine oxidation, EPR

The aging potential of Burgundy chardonnay wines is considered as quality indicator. However, some of them exhibit higher oxidative sensitivity and premature oxidative aging symptoms, which are potentially induced by no-enzymatic oxidation such as Fenton-type reaction (Danilewicz, 2003). This chemical mechanism involves the action of transition metal, native phenolic compounds and oxygen which promote the generation of highly reactive oxygen species (ROS) such as hydroxyl radicals (OH) or 1-hydroxyethyl radicals (1-HER) from oxidation of ethanol. Such mechanism is involved in the radical oxidation occurring during bottle aging. According to Elias et al.,(2009a), the 1-HER is the most abundant radical in forced oxidation treated wines. Consequently, understanding its evolution kinetic in dry white wines is of great importance. Due to the reactivity (Danilewicz, 2003) and short half-life (10⁻⁶-10⁻⁹s) of ROS in aqueous solution (Pryor, 1986), their direct quantitation in wine is impossible. However, by means of spin-trapping technique, the radicals can form relatively stable adducts with spin-trap, and thus be monitored in real-time by electron paramagnetic resonance (EPR) (Elias et *al.*, 2009b).

This study aims to optimize and validate an EPR spin trapping method using POBN as spin trap, to monitor the formation kinetic of 1-HER in chardonnay white wine and investigate the impact of some enological parameters (pH, ethanol, acidity, sulfites) on their formation. 1-HERs were generated by Fenton reaction (Fe²+ and H₂O₂) in chardonnay wines. The relative amount of reactant was optimized. In addition, several strategies were developed to decrease the impact of bisulfite on the detection of EPR signal. Finally, the analytical method was validated in terms of repeatability and reproducibility and applied to many chardonnay wines. To some extent, this study provides new insights into radical behavior that may contribute to comprehensive understanding of the oxidative stability of chardonnay white wines.

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III.P.39 OTA DEGRADATION BY BACTERIAL LACCASES

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Keywords: Ochratoxin A reduction, lactic acid bacteria laccases, polyphenolic compounds, redox mediators

Laccases from lactic acid bacteria (LAB) are described as multicopper oxidase enzymes with copper union sites. Among their applications, phenolic compounds' oxidation and biogenic amines' degradation, have been described. Besides, the role of LAB in the toxicity reduction of ochratoxin A (OTA) has been reported (Fuchs et al., 2008; Luz et al., 2018). Fungal laccases, but not bacterial laccases, have been screened for OTA and mycotoxins' degradation (Loi et al., 2018). OTA is a mycotoxin produced by some fungal species, such as *Penicillium* and *Aspergillus* sp., which infect grape bunches used for winemaking. OTA degradation is paramount given that it has been described as human-health harmful according to EFSA.

The work aimed to evaluate the OTA degrading capacity of three heterologous LAB laccases expressed in E. coli. The experimental procedure consisted on testing bacterial laccases from *L. lactis*, *L. paracasei* and *P. parvulus* in acetate buffer pH 4 with or without CuSO4 and OTA in presence and absence of several concentrations of epicatequin and complete polyphenolic extracts from red and white wine as media-tors. Degradation of OTA was followed and quantified by analyzing samples with HPLC-QToF-MS.

According to the results, OTA degradation in the reaction buffer with copper was at least three times higher than without copper. In addition, 0.75 mM epicatequin was the optimum concentration to obtain the highest OTA degradation with *L. paracasei* laccase (78%). Then, *P. parvulus* and *L. lactis* laccases were tested at this concentration, averaging 70% degradation. Finally, mean values of 40% and 10% OTA degradation were revealed when using polyphenolic extracts from red and white wine, respectively, for the three laccases. The application of these LAB laccases on OTA degradation in real wine needs to be further explored.

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THE FLAVANOL PROFILE OF SKIN, SEED, WINES, AND POMACE ARE CHARACTE-RISTIC OF EACH TYPOLOGY AND CONTRIBUTES TO UNDERSTAND THE FLAVAN-3-OLS EXTRACTION DURING RED WINEMAKING

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Keywords: Tannins, Flavanols, Winemaking, Extraction

Wine flavanols are extracted from grape skin and seeds along red winemaking. Potentially, eight flavan-3-ol subunits may be present as monomers or as tannins constituents, being these catechin, epicathechin, gallocatechin, epigallocatechin end the gallates of the mentioned units. In this work the flavanol profiles of grape skins and seeds before (grapes) and after (pomace) red winemaking were studied together with the one in the corresponding wines. The trials were made over two vintages in Vitis vinifera cv. Tannat, Syrah and Marselan from Uruguay. A total of twenty wines were made under the same experimental conditions. The flavanol fractions were isolated from the samples using C18 solid phase extraction cartridges. A LC-MS system was used for analysis, composed by an HPLC couples to a mass spectroscopy system (triple-quadrupole ESI-MS/MS). Catechin and epicatechin registered the higher relative abundance in all typologies as expected. In the skins, the percentage of catechin was significantly higher than that of epicatechin while the opposite was observed in the seeds. In agreement with literature, the relative proportion of gallates was much higher in the seed than in the skins, while prodephinidins (PD%) exceeded 10% in skins and were found at traces levels in seeds. There were no differences among cultivars in the skins flavanol profile, but in the seeds, Marselan had a characteristic high proportion of catechin that almost matched that of epicatechin. Moreover, Tannat had higher proportion of epicatechin-gallate than catechin-gallate, while the opposite was registered in Syrah and Marselan. The seed-pomace flavan-3-ol monomers profile matched that of the seeds in the three-cultivar studied, showing that all compounds were extracted at the same rate along maceration. Nevertheless, the skin-pomace had a much higher proportion of epicatechin and of gallates that observed in skins, and a much lower of PD. These differences were of higher magnitude in the flavan-3-ol monomers profile, and in Marselan, which wines had a much higher contribution of flavanols from the seeds (observed as low PD% and high of gallates and epicatechin) than Tannat and Syrah. In Syrah and Tannat wines, the PD% was just slightly lower than in skins, while in Marselan they were much lower. Thus, skins adsorb flavanols released from seeds during maceration, while the trihydroxylated prodelphinidin monomers are the more easily extracted flavanols from skins.

III.P.41 THE POTENTIAL USE OF SOLUBLE POLYSACCHARIDES TO PREVENT THE OXIDATION OF ROSÉ WINES

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Keywords: rosé wine, polysaccharide, oxidation, phenolic compounds

Lately, rosé wine is rapidly increasing its popularity worldwide. Short-time macerations with the red skin of the grapes cause the partial extraction of anthocyanins, which are responsible for the pinki-sh-salmon hue of rosé wines. However, the low quantity of tannins (antioxidants) and richness in phenolic acids, which can be easily oxidized into yellowish pigments, tend to predispose rosé wines to an undesirable browning. Although the use of SO₂ for the prevention of oxidation is highly extended, this practice is expected to be reduced. Therefore, the search for alternative oenological adjuvants that prevent the oxidation and browning of rosé wines is highly desired.

Thus, the aim of this work is to assess the effect of the addition of soluble polysaccharides, issued from grape pomace on the oxidation process. To do this, rosé wines were made using grapes from V. vinifera cv Syrah and employing two different maceration times: short (S, 10 min) and long (L, 2 hours). Thus, two different wines were elaborated (SYS and SYL). Soluble polysaccharides were extracted, purified and characterized (by means of HPLC-DAD-MS and HPLC-RID) from white grape pomace and added to the rosé wines. Then, wines were submitted to an oxidation process by reaching oxygen saturation level in the solution. Wines' phenolic composition was studied before the oxidation process and then its evolution was monitored.

The extract of polysaccharides presented three main fractions: F1 (25%) with a MW of 104 kDa; F2 (13%) with a MW of 8 kDa and F3 (62%) with a MW of 2 kDa. The polysaccharide extract was analysed by HPLC-DAD-MS after acid hydrolysis and a chemical modification reaction, in order to obtain a derivative of the monosaccharide which could be detected by UV. The main constitutive monosaccharide units detected were: galacturonic acid (26.3%), arabinose (26.2%), galactose (16%), xylose (11.4%), glucose (9.0%), mannose (6.6%), rhamnose (3.2%) and glucuronic acid (1.3%).

Two antioxidant test (FRAP and ABTS) were performed on the polysaccharide extract for the purpose of measuring its potential use as an antioxidant. Phenolic composition was analysed by HPLC-DAD-MS during the duration of the study (60 days).

Results allowed us to assess the importance of polysaccharide addition to modify the ability of rosé wines to resist oxidation, evaluating the possible application of a natural polysaccharide obtained from wine's by-product as an oenological adjuvant.

UNRAVELING THE CHEMICAL MECHANISM OF MND FORMATION IN RED WINE DURING BOTTLE AGING : IDENTIFICATION OF A NEW GLUCOSYLATED HYDROXYKETONE PRO-PRECURSOR

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Keywords: cooked fruit aroma, 3-methyl-2,4-nonanedione, glucosylated precursors, identification

During bottle aging, the development of wine aroma through low and gradual oxygen exposure is often positive in red wines, but can be unfavorable in many cases, resulting in a rapid loss of fresh, fruity flavors. Prematurely aged wines are marked by intense prune and fig aromatic nuances that dominate the desirable bouquet achieved through aging (Pons et *al.*, 2013). This aromatic defect, in part, is caused by the presence of 3-methyl-2,4-nonanedione (MND). MND content was shown to be lower in nonoxidized red wines and higher in oxidized red wines, which systematically exceeds the odor detection threshold (62 ng/L). Concentrations up to 340 ng/L were evidenced in the most oxidized red wines as well as MND content up scaling was observed whatever the oxidation level. Very recently, we identified two new hydroxyketones (2-hydroxy-3-methylnonan-4-one) associated with MND distribution in aged red wines. We demonstrated that in red wine, their oxidation can produce MND (Peterson et al., 2020). To date, the origin of these precursors were not studied. During preliminary experiments, the presence in wine of a glycosylated form of this hydroxyketone was suggested by hydrolysis experiments. Based on the literature, we hypothesized the presence of a corresponding glucosylated precursor and developed a strategy for its organic multi-step synthesis. First, the MND hydroxylated precursor of MND was synthesized by aldolization (Crévisy et al., 2001). Then, based on literature, we optimized strategies for the O-glycosidation step. For this, the tetrabenzylated glucose was activated by imidation reaction (Chatterjee et al., 2018). Several deprotection methods for the glucoside moiety were then experimented. Finally, the use of palladium on carbon for the hydrogenolytic debenzylation lead to the target compound. A multi-step purification process (LC, HPLC) was carried out to reach sufficient purity. Glycosylated standard was characterized by Nuclear Magnetic Resonance (NMR) and by High Resolution Mass Spectrometry (HRMS) and then used to develop an LC-MS/MS for its identification in grapes and wines. The first analytical results lead to look deeper into the search for glucosylated compounds in various oenological samples (grapes, musts, red wines), affected or not by the nuances of "dried fruits".

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USE OF 13C CP/MAS NMR AND EPR SPECTROSCOPIC TECHNIQUES TO CHARACTERIZE MACROMOLECULAR CHANGES IN OAK WOOD (*QUERCUS PETRAEA*) DURING TOASTING

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Keywords: oak wood, thermal degradation, macromolecular components, aroma compounds

For coopers, toasting process is considered a crucial step in barrel production during which oak wood (Q. *petraea*) develops several aromatic nuances released to the wine during its maturation. Toasting consists of applying different degrees of heat to a barrel for a specific period. As the temperature increases, thermal degradation of oak wood structure produces a huge range of chemical compounds. Many studies have identified the main key aroma volatile compounds (whisky-lactone, furfural, eugenol, guaiacol, vanillin). However, detailed information on how the chemical structure of oak wood degrades with increasing toasting level is still lacking.

In this study, we characterised the structural changes of the main components (cellulose, hemicellulose and lignin) of oak wood staves subjected to heat treatments (non-toasted to 240 °C) by cross-polarisation/magic angle spinning nuclear magnetic resonance (CP/MAS NMR, 800 MHz) and electron paramagnetic resonance (EPR, X-band) spectroscopy techniques applied to solids. Furthermore, the results will be compared to those obtained by quantitative GC-MS (EI) analysis of oak wood volatile compounds generated by the same heat treatments. The ¹³C CP/MAS NMR data highlighted concomitant phenomena of demethoxylation and depolymerisation of syringyl units leading to the formation of guaiacyl units and the release of monomer units. EPR results revealed the same phenomena and led us to hypothesize that the nature of the radical formed evolved during toasting from syringyl to guaiacyl form. Our results are consistent with data on others wood species and confirm that 200 °C is a threshold temperature above which degradation of the macromolecular structure leads to the formation of radicals and volatile compounds. Overall, the study offers interesting perspectives for the application of spectroscopic techniques, particularly EPR spectroscopy, to monitor radical formation during barrel aging and, consequently, to assess the oxidative stability of wine.

VOLATILE AND GLYCOSYLATED MARKERS OF SMOKE IMPACT: EVOLUTION IN BOTTLED WINE

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Keywords: smoke impact, volatile phenols, glycosylated phenols

Smoke impact in wines is caused by a wide range of volatile phenols found in wildfire smoke. These compounds are absorbed and accumulate in berries, where they may also become glycosylated. Both volatile and glycosylated forms eventually end up in wine where they can cause off-flavors. The impact on wine aroma is mainly attributed to volatile phenols, while in-mouth hydrolysis of glycosylated forms may be responsible for long-lasting "ashy" aftertastes (1).

In order to assess smoke impact, a selection of volatile and glycosylated phenols is proposed, mainly based on research from Australia (2, 3). It includes the volatile phenols guaiacol, 4-methylguaiacol, or-tho-, meta- and para- cresol, phenol, syringol, and 4-methylsyringol, as well as their glycosylated forms guaiacol rutinoside, 4-methylguaiacol rutinoside, cresol rutinoside, phenol rutinoside, syringol gen-tiobioside, and 4-methylsyringol gentiobioside. The accurate and reproducible measurement of these compounds is now possible, due to the commercial availability of standards and isotopic analogues.

In this study, we investigated the stability of these markers in bottled wines from smoke-exposed grapes, during a two-year timeframe. Wines monitored were a Chenin Blanc and a Chardonnay (whites), a Grenache (rosé), two Cabernet Sauvignons, a Zinfandel and a Grenache (reds).

No significant increases in guaiacol were observed in the white and rosé wines. Slight increases (2-3 μ g/L) were observed in red wines, with the exception of the Pinot Noir (9 μ g/L). Non-significant to slight increases were observed for phenol, except in the Zinfandel (10 μ g/L). Large increases were observed for syringol in red wines only, especially in Cabernet Sauvignons (up to 60 μ g/L). No significant increases were observed for the other volatile phenols measured.

All measured glycosylated markers were stable, in all wines. Therefore, increases in volatile phenols, when they happened, were not explained by the hydrolysis of corresponding glycosylated forms measured.

The observed increases in guaiacol and syringol, as well as the stability of the glycosylated forms measured, are consistent with results from a previous study (4).

The stability of glycosylated markers makes them relevant in identifying wines from smoke exposed grapes, possibly for many years after bottling. A limitation is that some smoke impacted wines might show normal or even non-detectable levels of these glycosylated markers.

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VOLATILE COMPOSITION OF WINES USING A GC/TOFMS: HS-SPME VS MICRO LLE AS SAMPLE PREPARATION METHODOLOGY

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Keywords: red wine, volatiles, sample preparation, GC/TOFMS

Wine aroma analysis can be done by sensorial or instrumental analysis, the latter involving several methodologies based on olfactometric detection, electronic noses or gas chromatography. Gas Chromatography has been widely used for the study of the volatile composition of wines and depending on the detection system coupled to the chromatographic system, quantification and identification of individual compounds can be achieved.

Prior to the chromatography, a sample preparation step is almost always required, but unfortunately there is no extraction procedure that can aid in the detection of the wide range of volatile compounds that exists in a wine sample. Wine volatile profile is characterized to have thousands of compounds with varying chemical properties, like molecular weight, structure, polarity and molecular structures. Moreover, they exist in a wide range of concentration, which, sometimes implies that a pre-concentration step is also required, if the ones existing in very low concentrations are of interest. As far as sample preparation methods for the analysis of wine aroma concerns, one can found thousands of bibliographic references, but the most used ones are probably the liquid-liquid extraction (LLE) and the solid-phase microextraction (SPME). Extensive reviews on the different sample preparation methods that has been used for wine analysis, along with each one advantages and drawbacks, has already received researcher's attention (Costa Freitas et al, 2012)

In light of the above, this work intents to discuss the use of two different sample preparation methods to quantify and identify volatile compounds in wines.

Two sample preparation methods were compared: a micro liquid-liquid extraction with 500mL of dichloromethane (based on Vilanova et al, 2010) and a HS-SPME (based on Pereira et al 2021). Chromatographic method was the same for both sample preparation method.

The number of compounds identified by HS-SPME was higher than the ones identified by micro-LLE. 26 compounds were identified in wines by both sample preparation methods. Since the majority of compounds identified by each sample preparation methodologies are different, choose to do one or another, or even both, should be taken into consideration when the goal is to go deep on volatile composition of wines.

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VOLATILE, PHENOLIC AND COLORIMETRIC CHARACTERIZATION OF THREE DIFFERENT LAMBRUSCO APPELLATIONS

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Keywords: Sparkling red wines, Charmat, Champenoise, Varietal characterization

Lambrusco is a commercially successful sparkling red and rosé wine. With 13.06 million litres sold in 2021 was the second best-selling Italian wine after Chianti. According to National Catalogue of Vine Varieties there are thirteen Lambrusco Varieties with which to date are produced seven PDO wines. Among these, "Lambrusco Salamino di Santa Croce", "Lambrusco Grasparossa di Castelvetro" and "Lambrusco di Sorbara" are the only ones that can be considered mono-varietal appellations, all located in Modena area. The PDOs contemplate the possibility of producing wines by secondary fermentation either in tank (Charmat method), or in bottle (Classico method). Sur lie is a third method commonly employed for Lambrusco, similar to the Classico method, from which differs for the absence of disgorgement.

The present study has two aims: we intended to provide for the first time a detailed characterization of the volatile chemical and phenolic composition and the colorimetric parameters of the three mono-varietal Lambrusco PDOs "Lambrusco Salamino di Santa Croce", "Lambrusco Grasparossa di Castelvetro" and "Lambrusco di Sorbara" and we wanted to investigate the influence of the three production methods considered, Charmat, Classico and Sur lie on relevant aroma compounds.

Volatile composition was investigated thanks to GC-MS coupled with different extraction techniques (SPE and SPME). Total polyphenols and anthocyanins were evaluated with specific enzymatic assays, tannins with methyl cellulose precipitable assay. CIElab parameters were studied with a reflectance co-lorimeter.

We found that the three types wine were differentiated by many volatile compounds. Lambrusco Grasparossa showed higher content of cyclic terpenes and sulphur compounds, Salamino higher content of linalool and 1,4-cineole, while Sorbara showed quite high levels of β -myrcene, 1,8-cineole, TDN, vitispirane and *cis*-3-hexenol. Fermentation-derived compounds showed a wide intra-varietal variability.

The type of secondary fermentation method (Charmat, Classico or Sur lie) can impact significantly Lambrusco volatile composition, highlighting the importance of various complex phenomena including aging period, adsorption of volatile compounds on yeast lees as well as formation of volatile compounds from amino acids.

With regard to the content of total polyphenols, tannins and anthocyanins, Lambrusco di Sorbara was characterised by a lower content than Grasparossa and Salamino, which was also reflected in the colorimetric parameters.

HOLISTIC APPROXIMATION OF THE INFLUENCE OF *SACCHAROMYCES* STRAINS ON WINE AROMA PRECURSORS

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Keywords: Saccharomyces, Glycosidic aroma precursors, Metabolomics, Wine varietal aroma

Wine varietal aroma is the result of a mixture of compounds formed or liberated from specific grape-aroma precursors. Their liberation/formation from their specific precursors can occur spontaneously by acid catalyzed rearrangements or hydrolysis or by the action of the yeast enzymatic activities. The influence of yeast during fermentation on the production of these volatile compounds has been widely studied however, the effect of this influence during aging is not fully understood. In order to evaluate these processes several indirect strategies have been used to study aroma precursors although they are not useful to understand the chemistry of the process. Therefore, the deep development of liquid chromatograph-mass spectrometers (LC-MS) during the last years has promoted some direct analysis of aroma precursors to identify them.

The objective of the present work is to study the influence of yeast on the aromatic precursors of wine and how that modulates the wine aroma during aging and its longevity. For that, four different yeasts (three S. cerevisae strains, Lalvin QA23[™], Lalvin Sauvy[™] and Lalvin Rhône 2056®, and S. kudriavzevii CR89D1) were selected attending to their different abilities to modulate aroma compounds. A must obtained combining 6 different grape varieties was fermented with the 4 strains and wines were aged under anoxia during 12, 24 and 96 hours at 75°C. After this process volatile compounds of young and aged wines were analyzed by gas chromatography mass spectrometry (GC-MS) and in parallel, the aromatic precursor fraction of must and young wines was characterized using UPLC-QTOF-MS untargeted analysis.

The targeted approach revealed remarkable differences in levels of vinylphenols, some terpenes, polyfunctional mercaptans, esters and some lactones. However, the concentration of norisoprenoid aroma compounds was not influenced by yeast. As it was expected, the metabolomic study revealed notable changes on young wines respect to the grape must, although the effect of yeast on putative glycosidic aroma precursors was marginal. These compounds were more influenced during aging, which supports the relevance of aging for producing varietal aroma derived from glycosidic precursors. This study has also made it possible the putative identification of some glycosidic precursors, which have to be studied to evaluate their relevance on the wine varietal aroma.

EFFECT OF FERMENTATION TEMPERATURE GRADIENT AND SKIN CONTACT ON ESTER AND THIOL PRODUCTION AND TROPICAL FRUIT PERCEPTION IN CHAR-DONNAY WINES

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Keywords: fermentation gradient, skin-contact, CATA, sensory analysis

Wines with tropical fruit aromas have become increasingly more available¹². With increased availability of different wine styles, it has become important to understand the compounds that cause the fruity aromas in wine. Previous work using micro fermentations showed that fermentation temperature gradients and time on skins resulted in an increase in thiol and ester compounds post fermentation and these compounds are known to cause tropical fruit aroma in wines³. This work aimed to scale up these fermentations/operations to determine if the desired aromas could still be achieved and if there is a perceivable difference in tropical fruit aromas, liking, and emotional response in the wines at the consumer level. Four treatments were tested at varying fermentation temperature gradients and skin contact times: control fermentation at 13°C with no skin contact (SCOFGO), fermentation at 13°C with 18 hours of skin contact (SC1FG0), fermentation temperature gradient by time (20°C for 4 days then reduced to 13°C) with no skin contact (SCOFG1), fermentation temperature gradient by time with 18 hours of skin contact (SC1FG1). A change in winemaking scale did not alter the pH, residual sugar, or alcohol of the wines. Chemical analysis and descriptive sensory analysis were conducted to determine the alterations on the composition and aroma profiles of these wines. Check-all-that-apply (CATA) showed different prominent aromas for each wine treatment, with pome fruit, stone fruit, pineapple, honeysuckle, honey, and passionfruit being the most perceived aromas. Descriptive analysis (DA) showed that SC1FG0 was significantly different from both SCOFG1 and SC1FG1. SC1FG0 presented the most tropical fruit aromas, SC1FG1 presented more stone fruit, and SC0FG1 presented more honey and lemon/lime. Understanding the causes of tropical fruit aromas in wine and processes that alter these compounds is necessary to ensure winemakers can achieved tropical fruit quality consistently.

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CHARACTERIZATION OF THE AROMA PROFILE OF COMMERCIAL PROSECCO SPARKLING WINES

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Keywords: Prosecco, sparkling wine, volatile compounds, SPME; GC-MS

The typicality of a wine, as well as its aromatic identity, are attributes that are highly sought after and requested by the current market. It is therefore of considerable technological interest to investigate the aromatic aspects of specific wines and to identify the odorous substances involved. In this thesis work, the characterization of the aromatic composition of Prosecco wines available on the market with a price range between 7 and 13 euros was carried out. These wines came from three different areas of origin such as Valdobbiadene, Asolo and Treviso.

To obtain a general view, the wines were subjected to basic chemical analyzes, in addition to the analysis of the aromatic profile by gas chromatography coupled with mass spectrometry (GC-MS) and various extraction techniques, Solid Phase Extraction (SPE) and headspace solid phase micro-extraction (HS-SPME). A total of 73 volatile molecules were analyzed. The respective OAVs have been calculated for their impact on the aroma of Prosecco wine. The resulting molecules with the greatest impact were ethyl hexanoate, isoamyl acetate and beta-damascenone mainly from fruity notes. Also important is the molecule of ethyl cinnamate which gives floral notes. Subsequently, the possible effects of subzones in wines were investigated. For what the Kruskal Wallis test was used and from this 16 compounds were identified that differ between the different areas of origin of the wines. The resulting compounds found in samples characterizing the areas of origin of the samples were: isoamyl alcohol, octanoic acid, limone-ne, 3-carene, α -pinepinene, α -phellandrene, p-cymene, rose oxide, TPB, carbon disulfide, diethyl diulfide, dimethyl disulfide, diethyl disulfide, α -pinene, α -myrcene and ethyl thioacete. These molecules are major norisoprenoids, esters and sulfur compounds. Finally, the wines were evaluated from a sensorial point of view by mean of a sorting task analysis. The clustering in three groups of the wine samples was observed, partially attributable to the areas of origin.

AGEING REVEALS THE TERROIR OF AGED RED BORDEAUX WINES REGARDLESS OF THE VINTAGES! TARGETED APPROACH USING ODOROUS COMPOUNDS LE-VELS INCLUDING TERPENES AND C¹³ NORISOPRENOIDS

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Keywords: Wine identity, Aroma compounds, Terroir, Ageing

The chemistry of wine is notably complex and is modified by ageing of the bottles. The composition of wines is the result of vine production (under the influence of vintage, climate and soils); yeast production (under the influence of juice composition and fermentation management); lactic bacteria production (under the influence of young wine composition and malolactic fermentation management); and of the ageing process either in vats, barrels or bottles or both. The composition is linked to the quality perceived by consumers but also to their origin, sometimes associated to the "terroir" concept.

Even if the chemical identity of a wine is shaped by a large variety of factors (soil, climate, varieties, microbiology, ageing process), we know now that the terroir and the maturation plays a key role in the sensorial and chemical identity of wines even after ageing (1-4). The aims of that study was to explore the links between terroir, ageing or vintages and the chemical composition. A targeted approach have been tested. It involves the quantification of molecular markers such as esters, terpenes, norisoprenoids or sulphur compounds. It have been applied to a large set of wines composed by 80 samples produced by 7 wineries during a selection of vintages between 1990 to 2007. The statistical analysis of the results permits to highlight similar compositions between wines produced in the same winery despite the variation of berry composition due to the vintage, the variations dues to technical choices and to ageing time. In the current study, the whole volatile composition is essential to the uniqueness of the wines because there are no compounds that are exclusively involved in discrimination of estate. This shows the complex effect of the grape and wine matrix on achieving a typical product. Overall, in the aromatic matrix, there is an existence of a hierarchy in the importance of compounds that permits the unicity of Bordeaux estate. Hence, three families of compounds (terpenes, norisoprenoids and esters) which made it possible to discriminate between the seven Bordeaux estates studied and are therefore influenced by the composition of the grapes. These include TDN, vitispirane, β -damascenone, terpinen-1-ol, α -terpinene, methyl salicylate, cis-linalooxide, ethyl esters of fatty acids (C_4C_2, C_6C_2, C_8C_2) and many others. It's interesting to note that even after years of bottle ageing, the imprint of the grape is still visible. The personality of each estate through its specific terroir is therefore an indispensable element for the aromatic singularity of each great wine.

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IDENTIFICATION OF NEW RESVERATROL DERIVATIVES FORMED IN RED WINE AND THEIR BIOLOGICAL PROPERTIES

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Keywords: Stilbene, resveratrol, oxidative coupling, isomerisation

Stilbenes are natural bioactive polyphenols produced by grapevine. Recently, we have reviewed the natural presence of these compounds in wines [1]. This study showed that the resveratrol and its glycoside, the piceid, are the most abundant stilbenes in wines. Resveratrol is a well-known stilbene with a wide range of biological activities. Due to its specific structure, resveratrol can be oxidized in wines to form various derivatives including oligomers [2]. In this study, we investigate the resveratrol and piceid transformation in wines.

First, the transformations of resveratrol and piceid by oxidative coupling in presence of metals and by photo-oxidation under light exposure were investigated in model solutions. Structural elucidation of oxidative products was obtained by NMR. Secondly, the formation in wines of these compounds was monitored by liquid chromatography coupled with accurate mass spectrometry. The main results will be presented and discussed. Finally, the biological properties of these compounds were evaluated on cell line models. The results will be presented and compared with those obtained with resveratrol.

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METHYL SALICYLATE: A TRENDY COMPOUND MARKER OF ZELEN, A UNIQUE SLOVENIAN VARIETY

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Keywords: Zelen, typicality, methyl salicylate, 4-vinylguaiacol

The wine market interest for autochthonous varieties, particularly from less known wine regions, has significantly raised in the past few years. In that context, Slovenia, a small country from central Europe with a long winemaking tradition, is getting more and more attention, particularly through its range of unique regional varieties. Among them, Zelen, meaning "green" in Slovene, can only be found in the Vipava valley region, located on the western side of the country, near the border with Italy. When they are young, Zelen wines display very singular aromas reminiscent of rosemary, sage and white fruit. Despite its uniqueness, Zelen wine aromatic typicality is poorly documented in the literature. The goal of this study was to highlight some potential aromatic markers specific to Zelen in comparison to other international and regional varieties grown in Slovenia.

A first batch of 28 white wines from different Slovenian wine regions including 8 Zelen wines, were analysed for their contents in volatile thiols by GC/MS/MS, terpenoids, and untargeted screening by HS/ SPME-GC/MS. Thereafter a second batch of 67 wines from Vipava valley including 25 Zelen wines, were analysed for their contents in methyl salicylate and volatile phenols by HS/SPME-GC/MS.

The first batch of analyses showed that Zelen had lower content in volatile thiols and higher concentration in some monoterpenols such as linalool in comparison to other varieties. Nevertheless, two compounds identified with the untargeted analysis seemed to be particularly important in Zelen wines aromatic profile: methyl salicylate and 4-vinylguaiacol. The second batch of analysis confirmed this trend with the average concentration of methyl salicylate at 14 µg/L and 3 µg/L in Zelen and other wines respectively. The highest concentration was measured at 38 µg/L in one Zelen wine, which corresponds to the sensory threshold measured in neutral white wines [1]. Methyl salicylate has recently gained some attention as it was found that this compound could contribute to the Italian Verdicchio and Lugana wines aromatic profile [1,2] and to some Bordeaux red wines made under specific conditions [3,4]. Zelen wines also displayed higher concentrations of 4-vinylguaiacol with 30% of Zelen wines having concentrations above the perception threshold reported for white wines [5]. Preliminary sensory investigations suggested that both compounds could potentially contribute to Zelen aromatic typicality.

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PROGRESS OF STUDIES OF LEES ORIGINATING FROM THE FIRST ALCOHOLIC FERMENTATION OF CHAMPAGNE WINES

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Keywords: Champagne, Lees, Fermentation, Aroma

Champagne wines are produced via a two-step process: the first is an initial alcoholic fermentation of grape must that produces a still base wine, followed by a second fermentation in bottle – *the prise de mousse* – that produces the effervescence. This appellation produces non-vintage sparkling wines composed of still base wines assembled from different vintages, varieties, and regions. These base wines, or "reserve wines," are typically conserved on their fine lies and used to compensate for quality variance between vintages (1). Continuously blending small amounts of these reserve wines into newer ones also facilitates preserving the producer's "house style." Some of these wines therefore possess the potential to age and maintain their organoleptic quality for even decades. Despite this potential, while the prise de mousse and final product have been extensively studied, far less research has been realised on the Champagne base wines and lies.

The aim of our ongoing study is to apply a multi-disciplinary strategy to study Champagne base wine lies. First, lies production was standardized on a laboratory scale at differing volumes (125 mL, 1 L, 5 L) for both synthetic solutions modelled on Champagne grape musts, and actual musts originating from the region. Confocal microscopy was then utilised to observe yeasts cells present in the wines and lees, as well as any enzymatic activity, creating a visual reference of autolytic dynamics over a one-year period. Simultaneously, these solutions were analysed for volatile odorous compounds and their precursors, including dimethyl sulphide (DMS,) monoterpenes, and heterocycles. Particular attention was given to amino acid concentrations, as previous studies show the importance of lees and amino acid content on ageing potential of reserve wines(2). *Initial results show a dynamic evolution of volatile compounds in the early stages of aging, highlighting the potential contribution of lies to the longevity of still base wines.* In addition, chemical analyses revealed unexpected data concerning the production of DMS in wines. This tandem approach allowed a preliminary analysis of Champagne reserve wine kinetics and the corresponding release of volatile compounds during the initial stages of lees aging.

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UNEXPECTED PRODUCTION OF DMS POTENTIAL DURING ALCOOLIC FERMENTA-TION FROM MODEL CHAMPAGNE-LIKE MUSTS

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Keywords: dimethyl sulfide, fermentation

The overall quality of aged wines is in part due to the development of complex aromas over a long period (1.) The apparition of this aromatic complexity depends on multiple chemical reactions that include the liberation of odorous compounds from non-odorous precursors. One example of this phenomenon is found in dimethyl sulphide (DMS) which, with its characteristic odor truffle, is a known contributor to the bouquet of premium aged wine bouquet (1). DMS supposedly accumulates during the ten first years of ageing thanks to the hydrolysis of its precursor dimethylsulfoniopropionate (DMSp.) DMSp is a possible secondary by-product from the degradation of S-methylmethionine (SMM), an amino acid identified in grapes (2), which can be metabolized by yeast during alcoholic fermentation. As a consequence, the totality of DMSp is not released into a young wine (3). Previous studies show that DMS and DMSp are effective as quality indicators for Champagne wines.

However, in beer, dimethyl sulphide (DMS) is either the result of the reduction of dimethylsulfoxide (DMSO) or the hydrolysis of DMSp, and is also linked with the fermentative process (4). Our current question: is the DMS present in wines liberated exclusively from DMSp of vegetal origin – i.e., produced by the vines – or do yeast likewise contribute DMSp during fermentation?

That question is particularly important in the case of Champagne wines because of the double fermentation required for its production. As part of an ongoing study of these Champagne base wines, lies production using *Saccharomyces cerevisiae* in both grape must and model solutions were standardized at a laboratory level. Modalities omitting DMSp and DMS in the original solution allowed us to monitor the appearance of DMSp during and post-fermentation. While the yeast in these modalities did not initially produce DMS, concentrations of DMSp rose from the onset of fermentation. Further analysis showed this onset coincided with a dramatic drop in methionine concentrations in the fermenting must. While the precise correlation is still being determined, these initial results showed DMSp can originate in both the vineyard and from yeast activity during fermentation, and implies that it may be possible to improve aging quality production using oenological techniques.

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III.P.55 RED WINE AGING THROUGH ¹H-NMR METABOLOMICS

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Keywords: wine aging, NMR metabolomics, oak barrels, fingerprinting

Premium red wines are often aged in oak barrel. This widespread winemaking process is used, among others, to provide roundness and complexity to the wine. The study of wine evolution during barrel aging is crucial to better ensure control of wine quality.

¹H-NMR has already been proved to be an efficient tool to monitor winemaking process [1]. Indeed, it is a non-destructive technique, it requires a small amount of sample and a short time of analysis, yet it provides clues about several chemical families. The aim of the present study is to investigate the evolution of wine during aging in oak barrels with NMR-based metabolomics.

Red wines, produced in an estate of Bordeaux region, were kept in oak barrels from three different manufacturers. They were firstly sampled after one month of aging. They were then resampled after twelve months of storage in oak barrels within the estate cellar. The evolution of wine constituents during aging was measured by ¹H-NMR-based metabolomics. NMR spectra were submitted to targeted and untargeted approaches.

Data were then statistically processed through multivariate statistical analysis such as principal component analysis (PCA), and orthogonal projections to latent structures discriminant analysis (OPLS-DA). It was used to better watch the distribution of metabolic variance, and to sharpen the separation between observations groups. The results of supervised models were validated using cross permutation tests and ANOVA. Statistical significances were then assessed for the potential discriminant compounds thanks to analysis of variance (ANOVA) or t-test. Based on this analysis, wine maturation effect was monitored, and discriminant metabolites were identified.

Regarding aging effect, wines analyzed after one month of aging exhibit higher contents of amino acids, catechin and epicatechin, acetoin and choline. On another side, wines analyzed after twelve months of aging present higher contents of acetic acid, ethyl lactate, arabinose, and glucose.

As it concerns barrel origins, samples showed higher heterogeneity after one month than after twelve months. However, significant differences were observed between wines depending on the barrel manufacturers.

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NEW METHOD FOR THE QUANTIFICATION OF CONDENSED TANNINS AND OTHER WINE PHENOLIC COMPOUNDS USING THE AUTOMATED BIOSYSTEMS SPICA ANALIZER

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Keywords: Tannins, Polyphenolic profile, Automated methods, Wine

Wine phenolic compounds are important secondary metabolites in enology due to their antioxidant and nutraceutical properties, and their role in the development of color, taste, and protection of wine from oxidation and spoilage. Tannins are valuable phenolic compounds that contribute significantly to these wine properties, especially in mouthfeel characteristics; however, tannin determination remains a significant challenge, with manual and time-consuming methods or complex methodologies. The purpose of this study is to propose a novel method for quantifying condensed tannins in finished wine products. This method aims to provide an accurate approximation of condensed tannin levels, similar to the widely used precipitation assay that involves the polysaccharide polymer methyl cellulose. The new tannin approximation is based on the strong correlation observed between Total Polyphenol Index (TPI) and methyl cellulose precipitable (MCP) tannin assay as both are determined at 280 nm, and using the epicatechin calibration is possible to obtain a value that is equivalent to condensed tannins determined by MCP tannin assay. Thus far, the results have shown a strong correlation between this new method and MCP tannin assay, with an r2 value of 0.83 and a sample size of 60. The present study has included wines from diverse geographical locations and varying ages. In addition to quantifying condensed tannins, the study also aimed to explore potential correlations that may explain differences found in the wines by analysing other polyphenolic parameters. These parameters included catechins determined via the DMACA reaction, anthocyanins based on their structural transformation arising from a change in pH, total polyphenols measured using the Folin-Ciocalteu reaction, TPI, and the chromatic features of wines at 420, 520 and 620 nm. Moreover, the study incorporated analyses of pH, total acidity, and ethanol content to obtain a comprehensive understanding of the wines' chemical composition. Noteworthy, the study is expanding the sampling to consider more matrices within the winemaking process.

All of the aforementioned parameters, including the condensed tannin, were determined automatically using the BioSystems SPICA® analyzer. The technical simplicity of automated methods for phenolic evaluation, will lead to enhanced efficiency, robustness, and accuracy. Furthermore, these automated methods may facilitate greater field applications, leading to increased profitability and an opportunity to improve wine quality.

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UNCOVERING THE ROLE OF BERRY MATURITY STAGE AND GRAPE GENOTYPE ON WINE CHARACTERISTICS: INSIGHTS FROM CHEMICAL CHARACTERISTICS AND VOLATILE COMPOUNDS ANALYSIS

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Keywords: Volatiles compounds, SPME-GC-MS, Composition parameters, Grape maturity

In a climate change context and aiming for sustainable, high-quality Bordeaux wine production, this project examines the impact of grape maturity levels in various cultivars chosen for their adaptability, genetic diversity, and potential to enhance wine quality. The study explores the effects on wine composition and quality through sensory and molecular methods. We studied eight 14-year-old Vitis vinifera cv. grape varieties from the same area (VITADAPT plots 1 and 5): Cabernet Franc, Cabernet Sauvignon, Carmenère, Castets, Cot, Merlot, Petit Verdot, and Touriga Nacional. We examined three berry maturity stages from the 2022 vintage: mid-veraison (MV), mid-maturity (MM), 7 days before maturity (M-7), at maturity (M), and 10 days post-maturity (M+10). Classical composition parameters were monitored during maturation. Fine volatile compounds, including lactones, furanones, norisoprenoids, and carbonyls as ripening and over-ripening markers, were quantified in grapes and wines using SPME-GC-MS, while thiols were analyzed in wines by SPE-GC-MS/MS. For example, according to the maturity stages, a significant increase in alcohol content was observed, which varied depending on the grape genotype. The highest concentrations were found in Petit Verdot (13.78 g/L in M-7), Cabernet Sauvignon, Merlot, and Petit Verdot (15.21, 15.30, and 15.75 g/L in M) and Merlot (16.68 g/L in M+10). These values were directly related to the higher sugar concentrations found in their must during the evaluated periods. Total acidity and pH levels vary among cultivars and are also influenced by different maturation stages. Some cultivars show more significant changes over time, while others display more modest fluctuations. As expected, the pH values and total acidity in wines from different cultivars were inversely related. Concerning the analyzed volatile compounds, surprisingly, Petit Verdot exhibited the highest concentrations of γ-nonalactone, followed by Cabernet Sauvignon and Cot, at all maturity stages including M-7 (6.39, 3.90, 3.61 µg/L), M (20.98, 8.98, 6.05 µg/L), and M+10 (13.93, 12.40, 8.48 µg/L), respectively. Overall, this study offers a new method to assess varieties' sensitivity to overripening and vital insights into the impact of berry maturity stage and cultivar on wine physicochemical traits and volatile compound profiles. These findings can be a foundation for future research aiming to predict or model wine's chemical and sensory properties.

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Enological Practices and Processes

ALCOHOLIC FERMENTATION AND COLOR OF ROSÉ WINES: INVESTIGATIONS ON THE MECHANISMS RESPONSIBLE FOR SUCH DIVERSITY

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Keywords: wine, alcoholic fermentation, rosé wine color, polyphenols

Color is one of the key elements for the marketing of rosé wines due to their packaging in transparent bottles. Their broad color range is due to the presence of pigments belonging to phenolic compounds extracted from grapes or formed during the wine-making process. However, the mechanisms responsible for such diversity are poorly understood. The few investigations performed on rosé wines showed that their phenolic composition is highly variable, close to that of red wines for the darkest rosés but very different for light ones [1]. Moreover, large variations in the extent of color loss taking place during fermentation have been reported but the mechanisms involved and causes of such variability are unknown. The hypothesis of this work was that the color and composition of light and darker rosé wines are driven by different mechanisms occurring during alcoholic fermentation, depending on the initial must composition. To test this hypothesis, three different Vitis vinifera grape varieties commonly used for the elaboration of rosé wines in French Provence area were selected for their different color potential: Grenache, Syrah, and Cinsault. The reactions and adsorption on yeast lees of phenolic compounds and their role in color and composition changes during alcoholic fermentation of rosé musts were investigated using UV-visible spectrophotometry, ultra-high performance liquid chromatography coupled to triple quadrupole mass spectrometry, and high performance size-exclusion chromatography coupled to UV-visible spectrophotometry.

Targeted mass spectrometry analysis exhibited large varietal differences in must and wine compositions, with higher proportions of hydroxycinnamic acids in Cinsault and Grenache whereas higher concentrations of anthocyanins and flavanols were found in Syrah. Syrah must color was mainly due to anthocyanins which were partly converted to derived pigments through reactions with yeast metabolites, resulting in a limited color drop during alcoholic fermentation. UV-visible spectrophotometry and size exclusion chromatography data indicated that Grenache and Cinsault musts contained oligomeric pigments derived from hydroxycinnamic acids and flavanols which were mostly lost during fermentation due to adsorption on lees. This work highlighted the impact of must composition, reflecting varietal characteristics, on changes occurring during fermentation and consequently wine color.

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CHARACTERIZATION OF ENOLOGICAL OAK TANNIN EXTRACTS BY MULTI-ANA-LYTICAL METHODS APPROACH

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Keywords: Oak extract, Ellagitanins, oses and polyols, HRMS

Oak tannin extracts are commonly used to improve wine properties. The main polyphenols found in oak wood extracts are ellagitannins¹ that release ellagic acid upon hydrolysis and comprise numerous structures². Moreover, oak tannin extracts contain other compounds giving a complex mixture. Consequently, the official OIV method based on gravimetric analysis of the tannin fraction adsorbed on polyvinylpolypyrrolidone is not sufficient to describe their composition and highlight their chemical diversity.

Eight commercial oak tannins were characterized by a combination of analytical approaches, Polyphenols were analyzed using the official OIV method, UV spectrophotometry, UPLC-UV-MS analysis before and after acidic methanolysis, and HPLC-SEC-UV. Neutral sugars and polyols were determined as alditol acetates by GC-FID analysis, before and after hydrolysis. Protein content was estimated by the Kjeldahl method. Finally, samples were compared by a non-targeted metabolomic approach based on UHPLC-HRMS/MS.

Gravimetric analysis, absorbance values at 280 nm, and the quantities of ellagic acid released by methanolysis revealed some differences between samples, indicating variations in their tannin composition. This was confirmed by HPLC-SEC-UV analysis evidencing differences in tannin size distribution, particularly in larger polymer content.

All samples contained significant quantities of sugars, and in particular xylose, mostly found in the linked form, and of quercitol, a polyol marker of oak origin. These compounds contributed to up to 25% of the whole extract composition, the proportions of free and combined sugars and polyols also showing large variations between tannins. The protein content was very low, generally representing less than 1% of the mass.

Non targeted UPLC-HRMS analysis detected major ellagitannins such as vescalagin, castalagin, and roburins A-E, but also a large number of derivatives as well as other molecules such as lignans and quercotriterponosides, and highlighted large differences between samples. Tannin extracts also contained aldehydes (HMF, furfural, syringaldehyde, sinapaldehyde, vanillin) in variable quantities.

This work demonstrates the variability in the composition of commercial oak tannin extracts, likely to impact their properties, and emphasizes the need for detailed multi-method characterization in the frame of quality control and selection of tannins for specific applications.

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EFFECTS OF WINEMAKING FACTORS AND AGEING ON THE POLYPHENOLIC AND COLORIMETRIC PROFILES IN RED WINES PRONE TO COLOUR INSTABILITY

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Keywords: Colour instability, Grape freezing, Chemical profile, Colorimetry

The effects of (A) grape freezing, and (B) malolactic fermentation, have been evaluated on the chemical and colorimetric profiles of red wines from Schiava grossa cv. grapes, thus prone to colour instability. The aim was to observe if specific variables (e.g. grape freezing) could improve the extraction and stability of pigments. The samples were studied from musts up to twelve months in bottle. The study was conducted with independent parallel micro-vinifications (12 = 4 theses x 3 replicates) under strictly-controlled conditions. The measured parameters included: 1) sugars, organic acids and %ABV (measured by specific enzymatic methods or by OIV reference methods), 2) dissolved oxygen (measured according to OIV protocols), 3) semi-quantitative determination of pigments, profile of non-anthocyanidin phenols, and profile of condensed tannins (LC-QqQ/MS [1]), spectrophotometric indexes (Hue and Intensity), colorimetric indexes (CIELab parameters), and the volatile profiles (GCxGC-ToF/MS [2]). A striking relation among the abundances of four anthocyanidin monoglucosides (peonidin-3-glu, malvidin-3-glu, petunidin-3-glu, and cyanidin-3-glu) has been observed in the musts from frozen grapes, but not in wines from frozen or non-frozen grapes. Cyclic procyanidins showed neither significant differences in concentration in must and wine due to any specific applied factor, nor due to specific treatments (such as with bentonites), proving again their applicability as markers for the grape variety in wine [3]. A substantial drop in peonidin-3-glu over the vinification (the main anthocyanin in Schiava cv. grapes) was studied in relation to the applied study factors. Grape freezing increased the extraction of peonidin-3-glu in the must, though the rate of its subsequent loss was faster than in wines from non-frozen grapes. Nonetheless, peonidin-3-glucoside was still more concentrated in the wines from frozen grapes than in wines from non-frozen grapes up to wine bottling. The wines made from frozen grapes and without malolactic fermentation had the highest colorimetric parameters a* (green \rightarrow red), Δ E* (difference in colour), C* (chromaticity), and ΔH^* (difference in tone) colorimetric parameters. b* (blue \rightarrow yellow) was highest in wines from frozen grapes, but regardless of the application or not of the malolactic fermentation.

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HOW DOES ULTRASOUND TREATMENT AFFECT THE AGEING PROFILE OF AN ITALIAN RED WINE?

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Keywords: Ultrasound, Wine ageing, Chemical profile, Sensory analysis

Many wine styles require moderate or extended ageing to ensure optimal consumer experience. However, few consumers have the interest or ability to age wine themselves, and holding wine in optimal conditions for extended periods is expensive for producers. A study was conducted on the use of ultrasound energy on wine, with particular reference to its impact on sensory and chemical profiles. The OIV has authorised the use of ultrasound for processing crushed grapes (must) in Resolution OENO 616-2019, but not yet for finished wine¹². Nonetheless, the method is considered to have potential for optimising wine ageing^{3,4}. Ultrasound treatment was carried out on sealed bottles of Buttafuoco red wine using an ultrasonic cleaning bath with 600 W power at 40 kHz. Both short (5 min) and long (30 min) treatments were conducted twice weekly. Four break points were defined at 3, 6, 9, and 12 months, when chemical and sensory analyses were conducted. For profiling of wines, GC×GC-MS, LC-MS, CIELab, spectrophotometry, and multiparametric analyses were undertaken. For sensory analysis, the Triangle Test was undertaken at T3, and Qualitative Descriptive Analyses at T6, T9, and T12. Results have shown clear differentiation between the treatments in chemical composition, due to the duration of the treatment applied via ultrasound. This has also influenced basic parameters such as tartaric acid and sulfur dioxide levels. The overall pattern is complicated as non-linear effects were observed for specific species in relation to long and short treatments. Some compounds displayed a decrease for the short treatment with respect to the control (no treatment), but then showed an increase at long treatments with respect to the short treatments. In addition, the chemical compositions of all wines were also influenced by ageing over the time period. For example, acetic acid decreased with ageing but did not differ between treatments. Colour was also affected by ageing but not by treatment. The sensory results have not shown clear trends based on treatments, with the short treatments appearing to be somewhat distinctive, but with the long and control treatments clustering. Sensory results were also clearly influenced by ageing. It is suggested that ultrasound treatment has a potential application for accelerated ageing of commercial wines ahead of release to market. However, further study is recommended to gauge consumer preferences regarding the extent of treatment applied.

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IMPACT OF FINING WITH K-CARRAGEENAN, BENTONITE, AND CHITOSAN ON PROTEIN STABILITY AND MACROMOLECULAR COMPOUNDS OF ALBARIÑO WHITE WINE PRODUCED WITH AND WITHOUT PRE-FERMENTATIVE SKIN MA-CERATION

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Keywords: Fining, Polysaccharides, Protein stability, Macromolecular composition

Pre-fermentative skin maceration is a technique used in white wine production to enhance varietal aroma, but it can increase protein concentration, leading to protein instability and haze formation [1]. To prevent protein instability, wine producers typically use fining agents such as bentonite, before wine bottling, which can negatively impact sensory characteristics and produce waste [2,3]. The aim of this study was to understand the impact of alternative techniques such as the application of polysaccharides (k-carrageenan and chitosan) on protein stability and on the wine macromolecular composition. The results showed that k-Carrageenan reduced the content of pathogenesis-related proteins (thaumatin-like proteins and chitinases), and consequently the protein instability in Albariño wines obtained with and without pre-fermentative skin maceration, and it was more efficient than sodium and calcium bentonites. Fungal chitosan was unable to heat stabilise both wines, and pathogenesis-related protein levels remained unchanged. Besides the impact on the protein content and wine protein instability, the use of k-carrageenan, chitosan, sodium, and calcium bentonites also differently impacted the wine polysaccharide content. Fungal chitosan decreased the wine polysaccharide content by 60%. Sodium and calcium bentonite also decreased the levels of wine polysaccharides although to a lower extent (16% to 59%) [4]. k-Carrageenan did not affect the wine polysaccharide composition. In conclusion, the results indicate that k-carrageenan is a suitable solution for white wine protein stabilisation, having a more desirable impact on the wine macromolecular fraction than the other fining agents tested, reducing the levels of the wine pathogenesis-related proteins without impacting polysaccharide composition.

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IV.P.6 IMPACT OF MANNOPROTEIN N-GLYCOSYL PHOSPHORYLATION AND BRANCHING ON WINE POLYPHENOL INTERACTIONS WITH YEAST CELL WALLS

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Keywords: *Saccharomyces cerevisiae* cell walls, Mannosyl phosphorylation, Mannan branching, Wine polyphenols adsorption

Yeast cell walls (CWs) may adsorb wine components with a significant impact on wine quality. When dealing with red wines, this adsorption is mainly related to physicochemical interactions between wine polyphenols and cell wall mannoproteins. However, mannoproteins are a heterogeneous family of complex peptidoglycans including long and highly branched N-linked oligosaccharides and short linear O-linked oligosaccharides, resulting in a huge structural diversity. Furthermore, the presence of mannosyl phosphate groups confers a net negative charge to the cell surface. The structural features in mannoproteins that promote their interactions with polyphenols and adsorption specificity are not clearly established yet. This work aimed to study the impact of mannosyl phosphorylation and mannan backbone branching on polyphenol adsorption by yeast cell walls.

Saccharomyces cerevisiae BY4742 Wild-type and mnn4 and mnn2 mutants (involved in N-glycosyl phosphorylation and mannan backbone branching, respectively) were obtained from EUROSCARF. Cell walls were purified and characterized in terms of total nitrogen, neutral sugar, and global charges. Their interactions with a red wine polyphenolic pool were studied in a wine-like solution by means of adsorption isotherms. Polyphenols were analyzed by means of UV-visible spectrophotometry and High-Performance Size-Exclusion Chromatography.

High molecular weight tannins and derived pigments were preferentially adsorbed whatever CW types, however, their affinity was significantly lower for mutants as compared to the wild-type. The *mnn4* and *mnn2* mutations induced changes in the mannose/protein ratio and a decrease of the CW net charge at wine pH. Both mutations induced a decrease in polyphenol affinity as well as a decrease in CW biosorption capacity, however, the latter was much more pronounced for the *mnn4* mutant (N-glycosyl phosphorylation).

This work evidenced the key role of mannosyl phosphorylation on yeast CW functionality regarding wine polyphenol adsorption.

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IV.P.7 INSIGHT THE IMPACT OF GRAPE PRESSING ON MUST COMPOSITION

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Keywords: White grape, Must extraction, Sparkling wine, Phenols

The pre-fermentative steps play a relevant role for the characteristics of white wine [1]. In particular, the grape pressing can affect the chemical composition and sensory profile and its optimized management leads to the desired extraction of aromas and their precursors, and phenols resulting in a balanced wine [2-4]. These aspects are important especially for must addressed to the sparkling wine as appropriate extraction of phenols is expected being dependent to grape composition, as well. To the best of our knowledge, a gap exists regarding grape composition – pressing conditions – must composition. To fulfill this gap and support the wine industry, this research aimed to clarify the impact of grape pressing based on both grape and must composition.

Chardonnay (7 samples) and Pinot blanc (2 samples) grapes were collected in vintage 2022 from different vineyards in Franciacorta area (Lombardy, Italy). These grapes were used to produce musts under an industrial scale following the pressing conditions adopted by wineries. Must samples were obtained at different extraction yields (e.g. running juice, 20, 30 [first fraction], 40, 50 [second fraction], 60 and 70 [third fraction] % must yields). The chemical parameters, turbidity units (NTU), color index (ABS 420 nm), total phenol index (TPI), polyphenol oxidase (PPO) activity and antioxidant capacity (AC) were assessed in both grape and must samples.

A decreasing trend of readily assimilable nitrogen and titratable acidity was found in must samples with higher extraction yields, while the opposite was observed for pH, NTU, color index, TPI and AC with a different extend dependent from the grape varieties and pressing conditions. Considering the first fraction must, a high variability in phenol extraction was found, from 16% to about 35%. Such a difference could be attributable to the different pressing conditions adopted as comparable levels of TPI were detected in grapes used (1.7-2.2 g/L, RDS=10%). The PPO activity seemed to be unaffected by the increased must extraction yield. Grape variety was influential on phenol content for the same must yield being higher for Pinot blanc probably due to its thinner skin in comparison to Chardonnay.

This study suggests the phenol-related indexes should be considered in addition to the chemical parameters for the accurate management of the pressing step; it also has been clarifying the relation existing between the composition of grape and must.

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METHYL SALICYLATE, A COMPOUND INVOLVED IN BORDEAUX RED WINES PRO-DUCED WITHOUT SULFITES ADDITION

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IV.P.8

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Keywords: Wines without added sulfites, Methyl salicylate, Sensory analysis, GC-O

Sulfur dioxide (SO₂) is the most commonly used additive during winemaking to protect wine from oxidation and from microorganisms. Thus, since the 18th century, SO₂ was almost systematically present in wines. Recently, wines produced without any addition of SO₂ during all the winemaking process including bottling became more and more popular for consumers. A recent study dedicated to sensory characterization of Bordeaux red wines produced without added SO₂, revealed that such wines were perceived differently from similar wines produced with using SO₂ and were characterized by specific fruity aromas and coolness1,2. The aim of this study was therefore to progress in wines without added SO₂ specificities characterization, focusing on compounds involved in their particular aroma.

To identify these compounds, a sensory targeted approach using semi-preparative HPLC3 followed by GC-O and GC-MS characterization was applied. For that, the same wines than those used for previous sensory characterization were studied. These wines were produced in 2017 from same merlot grape batches, according to a standard winemaking process, with or without SO₂ addition. First of all, wine aroma extracts were fractionated by semi-preparative HPLC to identify fractions perceived differently between wines. After comparing the fractions of the wines with or without sulfites, three consecutive fractions have been selected for their olfactive difference between the wines. These fractions were then analyzed by GC-O and GC-MS. Methyl salicylate was identified as responsible for sensory differences observed between these fractions. This compound was quantified4 in a large set of commercial red wines. Methyl salicylate was present at higher concentrations in the wines without added SO₂ ranging from 6 to 105 μ g/L whereas, in the wines with added SO₂, its concentration was below 10 μ g/L. Sensory threshold of methyl salicylate was determined in red wines at 62.3 μ g/L and one-quarter of the wines without sulfites studied, presented a concentration higher than this threshold.

Finally, methyl salicylate qualitative sensory impact was characterized in wines without added SO₂ by sensory profile determinations. This was done after a descriptor generation procedure and an adapted training on natural references associated to generated descriptors. This revealed that methyl salicylate was at the origin of wine without added SO₂ coolness and modified fruity aroma perception of these wines.

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IV.P.9 OENOLOGICAL STRATEGIES FOR THE REMOVAL OF PINKING IN WHITE WINE

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Keywords: Wine treatments, PVI/PVP, Chitosan, Yeast derivatives

The pinking of in white wine is the turning of color from yellow to salmon hue. White wines obtained from certain grape varieties (e.g. Chardonnay, Sauvignon blanc, Riesling, Trebbiano di Lugana) showed to be susceptible to pinking [1] that has been evaluated by an assay providing the addition of hydrogen peroxide. Even if its appearance does not seem to affect the sensory properties [2], strategies are necessary for its removal. Nowadays, the treatment with polyvinylpolipirroline (PVPP) was reported to significantly decrease the pink color [3]. To assess other additives and co-adjuvants suitable for pinking removal, this study aimed to identify the wine treatment(s) most effective for achieving this purpose.

A white wine showing the pinking fault was added with several additives and co-adjuvants, including active charcoals (bleaching and deodorizing), bentonites, gelatine, PVPP, PVI/PVP, chitosan, potassium caseinate, kaolin, zeolite, silica, calcium phytate, oenological tannins (oak and grape skin), glutathione, ascorbic acid (without/with sulfur dioxide) yeast derivatives for a total of 23 removal assays. The wines were stored up to 26 days and their susceptibility to pinking was carried out at 4 sampling points (day 1, 5, 15 and 26) through the hydrogen peroxide test. The wine was considered susceptible to pinking (SP) when an increase of 5 mAU was observed at 500 nm [4]. Moreover, the pink color index at 500 nm (without hydrogen peroxide) was determined.

No change in the pink color index was found with the exception of potassium caseinate. Some of the tested additives and co-adjuvants were not effective in limiting SP, including active charcoals, bentonite, gelatine, kaolin, zeolite, silica, grape skin tannin, glutathione and ascorbic acid. For some of them, an increased SP was evidenced (e.g. kaolin, zeolite, grape skin tannin). The treatment with PVI/PVP strongly decreased the pinking susceptibility already after 1 day. In this condition, the wine was not SP anymore at day 15. For this sampling time, three of the yeast derivatives tested, chitosan, PVPP, potassium caseinate and oak tannins limited the pinking susceptibility. The addition of PVPP, the mainly used co-adjuvant, did not result the most relevant one to solve such significant color change. Further study will investigate the selected additives and co-adjuvants in other pink wines as well as in combination in order to identify the most promising treatment for the pinking removal.

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POTENTIAL DEACIDIFYING ROLE OF A COMMERCIAL CHITOSAN: IMPACT ON PH, TITRATABLE ACIDITY, AND ORGANIC ACIDS IN MODEL SOLUTIONS AND WHITE WINE

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Keywords: wine, chitosan, organic acids, adsorption

Chitin is the main structural component of a large number of organisms (i.e., mollusks, insects, crustaceans, fungi, algae), and marine invertebrates including crabs and shrimps. The main derivative of chitin is chitosan (CH), produced by N-deacetylation of chitin in alkaline solutions. Over the past decade, the OIV/OENO 338A/ 2009 resolution approved the addition of allergen-free fungoid CH to must and wine as an adjuvant for microbiological control, prevention of haziness, metals chelation and ochratoxins removal (European Commission. 2011). Despite several studies on application of CH in winemaking, there are still very limited and controversial data on its interaction with acidic components in wine (Colangelo et al., 2018; Castro Marin et al., 2021). Therefore, the aim of this work was to assess the effect of a fungoid CH on pH, titratable acidity, and organic acids content in white wine and wine model solutions. A powdered sample of CH was added to each solution from 0 to 2.0 g/L and maintained under stirring (150 rpm) for 3h at room temperature. Before and after treatment, samples were analyzed for pH, titratable acidity, and organic acids content. Based on preliminary results, the CH treatment influenced both pH and titratable acidity: pH increased from 3.16±0.02 to 3.30±0.02, while titratable acidity decreased from 5.25±0,05 g/L to 4.60±0.04 g/L as tartaric acid equivalents. In detail, reductions in tartaric acid by 5-15% and in malic acid by 7-11% were observed. At the highest dosage (2.0 g/L) the CH produced a greater removal of tartaric acid (up to 202 mg per g of CH) than of malic acid (up to 63.45 mg/g of CH). These outcomes highlighted the valuable role of an allergen-free CH-based adsorbent as an alternative adjuvant for deacidification of white wines.

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PRECISE AND SUSTAINABLE OENOLOGY THROUGH THE OPTIMIZED USE OF AD-JUVANTS: A BENTONITE-APPLIED MODEL OF STUDY TO EXPLOIT

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Keywords: Oenological practices, Precision oenology, Adjuvant optimization, Bentonite, Wine resilience

As wine resilience is the result of different variables, including the wine pH and the concentration of wine components, a detailed knowledge of the relationships between the adjuvant to attain stability and the oenological medium is fundamental for process optimization and to increase wine durability till the time of consumption.

This work merges our 10-years' studies¹ on bentonite along with information from the literature to design a study-model feasible to optimize the effects of adjuvants by maximizing the impact on targeted compounds, while minimizing the one on desirable wine components. The boosting was simply based on the frequently unintended uses of oenological adjuvants by winemakers based also on some lacking in the EU regulations, which produces jeopardized main and side-effects, as the ones by bentonite are emblematic.

Indeed, there is no EU regulated upper limit for the addition of bentonite during the winemaking process, but the International Oenological Codex establishes the properties of the oenological bentonites amending the three classes of Ca-, Na-, and Na-activated bentonite.

Our studies demonstrated that the from-bentonite enrichment in wine cations results from the clay Cation Exchange Capacity (CEC) and from the pH, ethanol content and ionic strength which also impact on the residual card-house clay structure that is an important property for deproteinization. Indeed, for the removal of hazing forming proteins (b-glucanases, thaumatin-like proteins, chitinases) clay properties as CEC, Swell Index (SI), and Specific Surface Area (SSA) as well as wine pH are more impacting than the bentonite dose.

Considering adjuvant side-effects, bentonite can remove phenolic compounds so to modify wine colour and astringency. About terpenic wines, double addition to must for clarifying and to wine for fining outlined less removal than to the solely wine treatment. Removal of aglycones by low doses and of glycosylated terpenols especially by Ca-bentonite were predicted by RSM. For the fermentative aroma compounds, adsorption intensity and capacity more depended on the characteristics of the bentonite (SSA and CEC) than on the properties of the substances: the main removal is an indirect effect of deproteinization, while a direct adsorption can be described by the Freundlich equation for only a few compounds.

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IV.P.12 S. CEREVISIAE AND O. ŒNI BIOFILMS FOR CONTINUOUS ALCOHOLIC AND MALOLACTIC FERMENTATIONS IN WINEMAKING

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Keywords: biofilms, continuous fermetnation, S. Cerevisiae, O. oeni

Biofilms are sessile microbial communities whose lifestyle confers specific properties. They can be defined as a structured community of bacterial cells enclosed in a self-produced polymeric matrix and adherent to a surface and considered as a method of immobilisation. Immobilised microorganisms offer many advantages for industrial processes in the production of alcoholic beverages and specially increasing cell densities for a better management of fermentation rates. Controlling the speed of alcoholic (AF) and malolactic (MLF) fermentations in wine can be an important challenge for the production of certain short rotation wines for entry-level market segments.

The objective of this work was to design a continuous winemaking process using yeasts and bacteria biofilms. In a first part we showed the possibility of inducing the adhesion and biofilm formation by *O. œni* and *S. cerevisiae* separately, in low nutriment medium, on different materials already used in the winery environment. Then the biofilm formation was implemented in a 250 ml continuous bioreactor system for both microorganisms. At the end of the biofilm formation step, quantities of attached biomass (CFU counts) were close for all materials and over 5 log (UFC/cm²) for *S. cerevisiae*, over 6.2 log (UFC/cm²) for O. œni.

For continuous fermentations the inoculated supports were used in a similar 250 ml bioreactor with 3 different modalities: alcoholic fermentation (AF) by *S. w* in grape must, or Malo-Latic Fermentation (MLF) by *O. oeni* in wine or, co-fermentation (simultaneous AF and MLF) with both species biofilms feeded with grape must. The progress of the continuous fermentations was analysed. Over periods of 3 to 4 weeks under a continuous regime with a 48h residence time, stable consumption rates of 4 g/l/h for glucose + fructose and 1,8 g/l/24h for L-malic acid were reached in co-fermentations.

This biofilm continuous reactor could be the first step towards perfectly controlled industrial winemaking processes.

WINE WITHOUT ADDED SO₂: OXYGEN IMPACT AND EVOLUTION ON THE POLYPHENOLIC COMPOSITION DURING RED WINE AGING

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Keywords: Oxygen, Evolution, Phenolic compounds, Wine without added sulphite

 SO_2 play a major role in the stability and wine during storage. Nowadays, the reduction of chemical input during red winemaking and especially the removing SO_2 is a growing expectation from the consumers. Winemaking without SO_2 is a big challenge for the winemakers since the lack of SO_2 affects directly the wine chemical evolution such as the phenolic compounds as well as its microbiological stability.

During the red wine aging, phenolic compounds such as anthocyanin, responsible of the red wine colour, and tannins, responsible of the wine mouthfeel organoleptic properties, evolved quickly from the winemaking process to aging [1]. A lot of new interaction and molecules occurred lead by oxygen [2] or the lack of SO₂ which induce wine properties changes [3]. Nowadays, the phenolic composition of the wine without added SO₂ have not been reported. The aims of this study is to characterise the impact of oxygen on the phenolic composition of the wine without added sulphites during ageing. The evolution of the polyphenolic matrix have been monitored in function of the oxygen consumption. For the experiment, the identical wine without sulphite have been divided in different 30 L stainless steel tank. An increase amount of oxygen have been introduce from 0 mg/L to 36 mg/L of oxygen. Oxygen consumption have been followed. After consumption, wine samples have been collected for chemical and sensory analyses, and the same amount of oxygen have been introduce again. In total, three different cycle have been followed and sampled. Different phenolic analysis have been performed. Anthocyanin's evolution have been followed from the monomeric anthocyanin to the polymerized pigments. Condensed tannins evolution have also be carry out as well as the crown procyanidins. A correlation between the oxygen amount and anthocyanin's evolution have been determined as well as the tannin's evolution. The research of specific phenolic markers from the wine without sulphite is on progress.

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2-YEARS STUDY ON COMPARISON BETWEEN THE VOLATILE CHEMICAL PROFILE OF TWO DIFFERENT BLENDS FOR THE ENHANCEMENT OF "VALPOLICELLA SUPE-RIORE"

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Keywords: Red wine, Valpolicella wine, Aroma, GC-Ms

Valpolicella is a famous wine producing region in the province of Verona owing its fame above all to the production of two Protected Designation of Origins (PDOs) withered wines: Amarone and Recioto. In recent years, however, the wineries have been interested in the enhancement and qualitative increase of another PDO, Valpolicella Superiore. All the Valpolicella PDOs wines are produced with a unique grape blend, mainly Corvina, Corvinone, Rondinella and a range of other minor varieties. From 2019 Valpolicella product regulation has changed the grape proportion of the blend allowing new composition parameters of wines. For this reason, studying the volatile chemical profiles to support wine makers in the effort to produce high quality wines represents a field of great interest. The study aimed to evaluate the volatile chemical and sensory composition of two different blends, one "traditional" (70% Corvina, 30% Rondinella) and an "experimental" (60% Corvinone, 20% Corvina, 20% Rondinella). The grapes were supplied by four wineries in Valpolicella, which provided both blends.Winemaking was performed under standardized conditions. Free volatile compounds as well as those obtained through hydrolysis of glycosidic precursors were analysed with gas chromatography mass spectrometry (GC-MS) coupled with SPE and SPME extractions. Fermentation kinetics were found to be influenced by the different composition of the blends. The wines of traditional blends were found to be richer in free terpenes, ethyl acetate, benzyl alcohol while the wines of experimental blends were found to be richer in esters, beta damascenone, methyl salicylate and 1-Pentanol.Furthermore, the wines of experimental blends were characterized by a higher content of anthocyanins in both vintages.In conclusion, this study has highlighted the potential of the different blends studied to produce wines with specific and different aromatic profiles.

ANTHOCYANINS EXTRACTION FROM GRAPE POMACE USING EUTECTIC SOLVENTS.

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Keywords: Extraction, malvidin-3-O-coumaroylglucoside, Eutectic solvents, Grape pomace

Grape pomace is one of the main by-products generated after pressing in winemaking. Emerging methods, such as ultrasound-assisted extraction with eutectic mixtures, have great potential due to their low toxicity, and high biodegradability. Choline chloride (ChCl) was used as a hydrogen bond acceptor and its corresponding hydrogen bond donor (malic acid, citric acid, and glycerol: urea). Components were heated at 80 °C and stirred until a clear liquid was obtained. Distilled water was added (30 % v/v). A solid-liquid ratio of 1 g pomace per 10 ml of eutectic solvent was used. Total anthocyanins were determined. Malvidin-3-O-coumaroylglucoside was quantified by HPLC. Metabisulfite bleaching and the viscosity of the extracts were also determined. The highest extraction was obtained for the methanol/ water system. The eutectic system that showed the highest extraction was the mixture of choline chloride, urea, and glycerol in a molar ratio of 1:1:1. Glycerol is classified as a polyol. It can modify the polarity of water so it can be used as a co-solvent in the extraction of polyphenols. In addition, it is considered a highly flexible molecule, capable of forming intra- and intermolecular hydrogen bonds [1]. The higher extraction of choline chloride glycerol and urea (molar ratio 1:1:1) could be due to the influence of the lower polarity of glycerol presenting a higher affinity, probably with malvidin-3-O-coumaroylglucoside, which is less polar than Mv-3-0-glc. In HPLC analysis, malvidin-3-0-coumaroylglucoside was the main anthocyanin identified in all extracts. In eutectic mixtures, viscosity is the property that limits the extraction process compared to conventional solvent extractions. The extract obtained with the choline chloride: urea: glycerol (1:2:2) system had the lowest viscosity values, while the rest of the extracts presented higher viscosities. Viscosity reflects how compact a molecular structure is. Therefore, it can be inferred that the systems with malic acid and citric acid with choline chloride in molar ratios 1:2 present a compact molecular structure with a minimum of holes, which results in less diffusion during the extraction process. The choline chloride: malic acid (1:1) system presented significant resistance to sulfite bleaching at pH 3.5, losing approximately 34 % of color. The choline chloride: urea: glycerol (1:1:1) system lost approximately 50 % of the color, presenting a lower resistance to discoloration.

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DETERMINATION OF MINERAL COMPOSITION IN CV. TERAN (*VITIS VINIFERA L.*) RED WINE AFFECTED BY PRE-FERMENTATIVE MASH COOLING, HEATING, SAI-GNÉE TECHNIQUE AND PROLONGED POST-FERMENTATIVE MACERATIONS

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Keywords: Teran grape variety, mineral composition, pre-fermentative mash treatment, prolonged maceration

This study aimed to determine mineral composition in red wine obtained from cv. Teran (Vitis vinifera L.), autochtonous Croatian grape variety. Six different vinification treatments, including the control treatment (7-day standard maceration), were performed to study the effects of: 48-hour pre-fermentative mash cooling (8 °C) followed by prolonged post-fermentative maceration of 13 days (C15), 28 days (C30), and saignée technique (juice runoff) proceeded with prolonged post-fermentative maceration of 13 days (CS15); and effect of 48-hour heating (50 °C) followed by prolonged post-fermentative maceration of 13 days (H15) and 28 days (H30) on macro- and microelements in wine. Respectively, macerations durated 15 and 30 days in total, including either pre-fermentative cooling or heating. Macro- (K, Ca, Mg, Na) and microelements (Al, Cu, Fe, Mn) were determined using the Optima DV 2000 inductively coupled plasma - optical emission spectrometer (Perkin Elmer, Shelton, Connecticut, USA) equipped with a Meinhard spray chamber, nebulizer, and peristaltic sample delivery system. The analysed elements were identified in line with ICP-OES using the PerkinElmer's WinLab 1.35 software and quantified by direct calibration method. One-way analysis of variance (ANOVA) and Fisher's least significance difference (LSD) test were used to compare mean values (p < 0.05). Statistics were performed using Statistica 10.0. software (Sta-Soft Inc. Tulsa, OK). The obtained results showed that the total content of macroelements in investigated wine ranged from 939.03 to 1038.57 mgL-¹. The total content of microelements ranged from 3.09 to 6.37 mgL-1, where was found that significantly the highest were treatments submitted to pre-fermentative heating (H15 and H30), despite duration of prolonged maceration. The most abundant minerals in investigated wine were potassium (K) among macroelements and iron (Fe) among microelements. The significantly highest concentration of iron (Fe) was found in the treatment equally affected with both pre-fermentative heating and prolonged post-fermentative maceration (H30). On the other hand, among the macroelements, the highest concentration of calcium (Ca) was found in treatments subjected to pre-fermentative heating (H15 and H30) regardless of maceration duration. Obtained results suggested that Teran red wine, affected with particular vinification processes considered as strong source of several micro- and macroelements.

IV.P.17 EFFECT OF DIFFERENT VITICULTURAL AND ENOLOGICAL PRACTICES ON THE PHENOLIC COMPOSITION OF RED WINES

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Keywords: red wine, phenolic compounds, earthenware vats, HPLC-DAD-MS

Global climate change is exerting a notable influence on viticulture sector and grape composition. The increase in temperature and the changes in rainfall pattern are causing a gap between phenolic and technological grape maturities [1]. As a result, the composition of grapes at harvest time and, consequently, that of wines are being affected, especially with regards to phenolic composition. Hence, wine quality is decreasing due to changes in the organoleptic properties, such as color and astringency, making necessary to implement new adaptive technologies in wineries to modulate these properties in order to improve wine quality.

The aim of this work is to study the effect of different viticultural practices, such as traditional cultivation, organic cultivation and the use of natural fertilizer on the phenolic composition of grapes. In addition, the effect on wine phenolic composition of using tanks made of different materials (stainless steel tanks, oak wood barrels/tanks or earthenware vats) at different stages of winemaking and aging was evaluated over three vintages. The detailed phenolic composition of grapes and wines was determined by HPLC-DAD-MS [2].

Results obtained showed that the use of natural fertilizer did not cause significant differences in the pigment composition of grapes. However, a combination of organic cultivation with natural fertilizer significantly increased the total content of pigments and flavanols when compared to traditional cultivation with no fertilization. Regarding wines, higher levels of total flavanols and anthocyanins were observed when alcoholic fermentation (AF) was carried out in stainless steel tanks than when wines were fermented in earthenware vats. In the first ones (AF in stainless steel tanks), the type of container (oak barrels or earthenware) employed for the subsequent malolactic fermentation (MLF) did not have a significant influence in their phenolic composition. However, higher levels of phenolic compounds were observed in wines with AF made in stainless steel tanks and MLF in earthenware vats than in wines in which both fermentation processes occurred in earthenware vats. The obtained results showed that the type of tank as well as the stage at which it is used might have a significant influence on the phenolic composition of the wines. This could allow envisaging the most adequate tanks for each step of winemaking and aging in order to obtain wines with an adequate phenolic composition.

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IV.P.18 EFFECT OF FUMARIC ACID ON SPONTANEOUS FERMENTATION IN GRAPE MUST

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Keywords: Fumaric acid, Alcoholic fermentation, Malolactic fermentation, Spontaneous fermentation

Malolactic fermentation (MLF)¹, the decarboxylation of L-malic acid into L-lactic acid, is performed by lactic acid bacteria (LAB). MLF has a deacidifying effect that may compromise freshness or microbiological stability in wines² and can be inhibited by fumaric acid [E297] (FA). In wine, can be added at a maximum allowable dose of 0.6 g/L³. Its inhibition with FA is being studied as an alternative strategy to minimize added doses of SO₂⁴. In addition, wine yeasts are capable of metabolizing and storing small amounts of FA and during alcoholic fermentation (AF). Our aim was to study the effect of FA addition in natural grape must without SO₂ on alcoholic and malolactic fermentation. AF was performed on Muscat of Alexandria grape must without SO₂ under two different conditions. i) Grape must 1 without FA, pH 3.49 and ii) Grape must 2 with 0.6 g/L of FA, pH 3.39; both had an L-malic acid concentration of 1.44 g/L. AF was developed at 20°C and spontaneously, monitored by must density determination. The evolution of L-malic acid and FA was monitored enzymatically⁵ and plate counts were performed for Saccharomyces, non-Saccharomyces and LAB populations. In both grape musts, no significant differences were observed in the development of AF. In grape must 1 MLF was performed during AF and produced a lactic bite. A progressive decrease in FA was observed in grape must 2 during AF, reaching 0.087 g/L at the end. From the wine obtained from grape must 2, two conditions were prepared i) a wine uncorrected with FA with a concentration of 0.087 g/L and ii) a wine with FA correction to 0.6 g/L. MLF was tried to take place at a temperature of 20°C under two new conditions, i) spontaneous and ii) with inoculation of O. oeni VP41 (Lallemand S.A.). MLF was monitored following the evolution of L-malic acid and LAB populations by plate count. MLF was not performed in all conditions, except for wines without FA correction inoculated with LAB. In conclusion, the addition of FA in must at pH 3.5 without SO₂ with low initial LAB populations may be an effective strategy to prevent MLF during AF in conditions of absence of SO₂. However, FA supplementation in the grape juice will not inhibit the subsequent development of the MFL in the wine, since a large part of this acid is metabolized by the yeasts, being necessary supplementing with FA again to ensure the non-development of malolactic fermentation in the case of high LAB populations.

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EFFECT OF MANNOPROTEIN-RICH EXTRACTS FROM WINE LEES ON PHENOLIC COMPOSITION AND COLOUR OF RED WINE

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Keywords: lees, mannoprotein, colour wine, phenolic compounds

In 2022, wine production was estimated at around 260 million hl. This high production rate implies to generate a large amount of by-products, which include grape pomace, grape stalks and wine lees. It is estimated that processing 100 tons of grapes leads to ~ 22 tons of by-products from which ~ 6 tons are lees [1]. Wine lees are a sludge-looking material mostly made of dead and living yeast cells, yeast debris and other particles that precipitate at the bottom of wine tanks after alcoholic fermentation. Unlike grape pomace or grape stalks, few strategies have been proposed for the recovery and valorisation of wine less [2]. Nevertheless, this by-product could become a source of interesting compounds, such as mannoprotein rich extracts (MRE). Therefore, the aim of this work was to obtain MRE from different lees, to characterize them, and to evaluate their effect on wine colour and on the phenolic composition of red wines.

Red, rosé and white wines were used as sources of lees, which were collected after the alcoholic fermentation with different Saccharomyces cerevisiae commercial varieties. The extraction of MRE was performed by physical extraction (autoclave) followed by a purification with ethanol. The protein and polysaccharidic moieties of the purified extracts were characterized by SDS-PAGE, Lowry method, HR-SEC-RID and HPLC-DAD-MS. The obtained MRE were added to a red wine (Vitis vinifera L. cv Tempranillo) and the changes in the phenolic composition and colour were analysed by HPLC-DAD-MS and triestimulus colorimetry, respectively, before and after the stabilization of the wine (involving cold treatment). Results obtained showed that the extraction yield of MRE was efficient (~ 40 mg/g wet lees) for all types of lees assayed, which supports the valorisation of wine lees as a sustainable source of MRE. Interestingly, MRE presented important structural and compositional differences, both in the protein content and in the polysaccharidic profile, although the source of lees, namely red, white and rosé wines, was not the main factor determining these differences, but the winemaking techniques or the S. cerevisiae strain employed. Furthermore, the addition of the MRE to red wine had an effect on the stabilization of wine colour and its phenolic content that rely mainly on the saccharidic characteristics of each MRE. These results pointed out that MRE from wine less could be a potential tool to improve the colloidal stability of wine phenolic compounds.

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EFFECT OF MICRO-OXYGENATION IN COLOR OF WINES MADE WITH TOASTED VINE-SHOOTS

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Keywords: color, fixed micro-oxygenation, SEGs, winemaking techniques implementation

The use of toasted vine-shoots (*SEGs*) as an enological tool is a new practice that seeks to improve wines, differentiating them and encouraging sustainable wine production. The micro-oxygenation (MOX) technique is normally combined with alternative oak products with the aim to simulate the oxygen transmission rate that takes place during the traditional barrel aging. Such new use for *SEGs* implies a reduction in color due to the absorption by the wood of the responsible compounds, therefore, given the known effect that MOX has shown to have on the modification of wine color, its use together with the *SEGs* could result in an interesting implementation with the aim to obtain final wines with more stable color over time.

To achieve this, Tempranillo wines were in contact with their own *SEGs* and with those from Cabernet Sauvignon variety in two different doses (D1 and D2). *SEGs* were added at the end of malolactic fermentation and two fixed doses of micro-oxygenation (low, LMOX; and high, HMOX) were considered during the entire period of *SEGs* contact. At the end of the *SEGs*-MOX treatments, wines were bottled and stored at temperature and humidity-controlled conditions for 6 months. Wines were characterized in terms of visible spectra, CIELab and individual anthocyanin compounds (HPLC-DAD) to study the color evolution at bottling time and after 3 and 6 months in the bottle.

The results showed that at the end of the treatments, wines micro-oxygenated with the lower dosage (L-MOX) received 6.24 ± 0.87 mg/L per month while those from higher dosage (H-MOX) received 11.91 ± 0.71 mg/L per month. The spectral information showed that in general there was a decrease in the color of *SEGs*-MOX wines with respect to the control, being more pronounced at bottle time. This reduction was greater when the higher *SEGs* dose were used, but MOX doses considered did not seem to have a differentiating effect. Specific, only in wines with Cabernet Sauvignon *SEGs* and D1 the H-MOX produced less color loss; however, for Tempranillo *SEGs*, the highest dose (D2) combined with L-MOX showed the least color reduction. This reduction in color was observed during the bottle time, being less pronounced after 6 months. The greatest reductions were observed for the red tones (A520) and to a lesser extent for the blue ones (A620). The anthocyanin pormenorized analysis revealed the same behavior, being malvidin-3-O-glucoside the one that presented the greatest decrease. These first results could indicate that MOX would have to establish it based on *SEGs* variety and dosage.

EFFECT OF OXIDATION ON LOW MOLECULAR WEIGHT PHENOLIC FRACTION, SA-LIVARY PROTEINS PRECIPITATION AND ASTRINGENCY SUBQUALITIES OF RED WINES

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Keywords: oxidation, astringency, subquality, greenness

Changes in the low molecular weight phenolic fraction, obtained by liquid-liquid microextraction technique, were studied after controlled oxidation of two typologies of Sangiovese wines (Brunello di Montalcino and Chianti Classico) belonging to two vintages (2017 and 2018). The fractions were characterized by LC-MS and quantified by HPLC. The most abundant extracted compounds were the phenolic acids. The effect of oxidation, vintage, and wine typology was stated by a three-ways ANOVA. Gallic and syringic acids significantly increased after oxidation while (-)-epicatechin decreased the most. The interaction and precipitation with salivary proteins, mechanism at the basis of astringency, was carried out at three phenolic concentrations (3.0–5.0–7.5 g/L). The amount of salivary proteins and phenolic compounds were in turn analysed by HPLC. Only the caftaric, cis- and trans-coutaric acids, and procyanidin dimer B7 showed a significant precipitation with salivary proteins. The oxidated wine fractions showed a high interaction and more precipitation with salivary proteins than the not oxidated ones. However, the high precipitation of the low molecular weight phenolics was not correlated with the sensory astringency of wines. Control wines were characterized by unripe astringency felt with acidity, described as greenness subquality, due to the high content of (+)-catechin, (-)-epicatechin, caftaric acid and myricetin-3-O-glucoronide in the fractions. The decrease of such phenolic compounds by oxidation was correlated with the corresponding increase of silkiness and velvety sensation in the oxidated wines.

EFFECTS OF LEAF REMOVAL AT DIFFERENT BUNCHES PHENOLOGICAL STAGES ON FREE AND GLYCOCONJUGATE AROMAS OF SKINS AND PULPS OF TWO ITA-LIAN RED GRAPES

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Keywords: defoliation, secondary metabolites, aromas

Canopy-management practices are applied in viticulture to improve berries composition and quality, having a great impact on primary and secondary grape metabolism. Among these techniques, cluster zone leaf removal (defoliation) is widely used to manage air circulation, temperature and light radiation of grape bunches and close environment. Since volatiles are quantitatively and qualitatively influenced by the degree of fruit ripeness, the level of solar exposure, and the thermal environment in which grapes ripen, leaf removal has been shown to affect volatile composition of grape berries [1].

The aim of this research was to assess the impact of vine defoliation on free and glycosylated VOCs (Volatile Organic Compounds) of two Italian red grapes: Nebbiolo (neutral) and Aleatico (semi-aromatic). Defoliation was performed at fruit set phenological stage for Aleatico grapevines, and for Nebbiolo also at berries touch.

Solid Phase Extraction/Gas Chromatography–Mass Spectrometry (SPE/GC-MS) was carried out to analyse free and glycoconjugates VOCs isolated from skins and pulps as separate portions of the berries [2].

The results showed that defoliation had an almost negligible effect on free and glycosylated volatiles of Aleatico grapes, thus suggesting that defoliation at fruit set did not change the volatile composition of this grape variety. A different behaviour was observed for Nebbiolo grapes, on both free and bound VOCs, with a greater impact on the first. Indeed, all the 30 free VOCs identified were significantly (ANOVA; p<0.05) affected by defoliation and by the time at which it was carried out, with a greater influence on the skin components. Early defoliation at fruit set did not favour the accumulation of free VOCs in Nebbiolo skins, significantly reducing the content of several VOCs, such as n-butyl acetate, terpenes (α -terpineol, and nerol) and aldehydes (hexanal, and 2-hexanal). On the other hand, late defoliation performed at berries touch, reduced n-butyl acetate, but increased alcohols content (i.e., 3-methyl-1-butanol, 1-pentanol, 2-ethyl-1-hexanol, benzyl and phenylethyl alcohols), nerol, aldehydes (i.e., 2-hexanal), and vanillin.

Results suggest that the effects of defoliation at fruit set on the VOCs pattern is cultivar dependent and almost ineffective on Aleatico grapes. Moreover, early defoliation at fruit-set seems stressful for Nebbiolo grapes and its odorous and potentially secondary metabolites.

These results can be useful to improve canopy and winemaking precision practices.

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IV.P.23 ESTIMATING THE INITIAL OXYGEN RELEASE (IOR) OF CORK CLOSURES

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Keywords: Oxygen, Corks, Bottle, Wine

Many factors influence aging of bottled wine, oxygen transfer through the closure is included. The maximum uptake of wine before oxidation begins varies from 60 mg.L⁻¹ to 180 mg.L⁻¹ for white and red wines respectively [1].

The process of bottling may lead to considerable amounts of oxygen. The actual contribution of the transfer through the closure system becomes relevant at the bottle storage, but the amounts are small compared to prepacking operations [2] and to the total oxygen attained during filling.

When corks are inserted in bottlenecks, there is initial oxygen released (IOR) due to the compression exerted to accommodate them in the volume of the bottleneck. Then starts contribution of the transfer between the closure and the glass together through the cork.

The initial release of oxygen is significant compared to the transfer through the cork itself and has been reported around 60% to 70% of the total oxygen ingress in a bottle after the first month and around 90% to 97% after the second, either for corks tested under dry or under wine contact condition [3]. In a study designed for sparkling corks, inerting procedure allowed to reduce the initial oxygen release by around 1.5mg [4]. An identical procedure is used in the scope of oxygen transfer measurements as a preparation for the corks [5].

The current work aims at estimating the IOR of natural corks. Natural corks of a superior grade, 49 mm length and 24 mm diameter, ready for use were purged with nitrogen for 3 months to displace oxygen from cork cells. Then corks were inserted in bottles with controlled bottlenecks and oxygen ingress monitored using the non-invasive methodology [3] for two months. At 64 days, it was observed that corks submitted to the purging procedure released 1.4 mg of oxygen less. In a preliminary experiment purging for 1 month, the reduction of oxygen ingress was around 7% more which suggests that the additional months did not change much oxygen from the cork cells.

The ongoing project aims to consolidate the estimation of the IOR value and to be extended to micro agglomerated cork stoppers.

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IV.P.24 EVALUATING WINEMAKING APPLICATIONS OF ULTRAFILTRATION TECHNOLOGY

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Keywords: membranes, phenolics, proteins, wine

Ultrafiltration is a process that fractionates mixtures using semipermeable membranes, primarily on the basis of molecular weight. Depending on the nominal molecular weight cut-off (MWCO) specifications of the membrane, smaller molecules pass through the membrane into the 'permeate', while larger molecules are retained and concentrated in the 'retentate'. This study investigated applications of ultrafiltration technology for enhanced wine quality and profitability. The key objective was to establish to what extent ultrafiltration could be used to manage phenolic compounds (associated with astringency or bitterness) and proteins (associated with haze formation) in white wine. Nevertheless, ultrafiltration was also applied to red wine, despite the removal of anthocyanins and tannins (associated with colour and textural properties) being inherently detrimental to wine quality, so as to better characterise the chemical consequences of membrane filtration. The composition of permeate and retentate derived from pilot-scale fractionation of red and white wine using 10 and 20 kDa membranes, and different permeation rates (50, 80, 90, 95%) was investigated. The alcohol content and pH of permeate and retentate were not significantly different from that of the initial wine, but titratable acidity and macromolecules (proteins, polysaccharides and phenolic compounds, including anthocyanins for red wine) were progressively concentrated in the retentate, as a function of both membrane MWCO and the degree of permeation. Red wine permeates were stripped of much of their essential character, such that they were not considered commercially acceptable; whereas the removal of white wine phenolics demonstrated the potential for ultrafiltration to remediate oxidised or highly phenolic wines. Subsequent trials investigated the addition of retentate to (i) fermenting red grape must, (ii) dealcoholised wine, and (iii) permeate, as a potential strategies for enhancing wine colour stability, flavour intensity and/or mouthfeel properties. Whereas colour enhancements were not apparent, likely due to the inherent effects of dilution, differences in wine flavour and mouthfeel were perceived via sensory profiling using the Rate-All-That-Apply method. Findings will enable the wine industry to make informed decisions regarding the suitability of ultrafiltration technology as an innovative approach to improving wine quality and process efficiency, and therefore profitability.

EVIDENCE OF THE INTERACTION OF ULTRASOUND AND ASPERGILLOPEPSINS I ON UNSTABLE GRAPE PROTEINS

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Keywords: Ultrasound, Aspergillopepsins I, TLPs, Protein stability

Most of the effects of ultrasound (US) result from the collapse of bubbles due to cavitation. The shockwave produced is associated with shear forces, along with high localised temperatures and pressures. However, the high-speed stream, radical species formation, and heat generated during sonication may also affect the stability of some enzymes and proteins, depending on their chemical structure. Recently, Celotti et al. (2021) reported the effects of US on protein stability in wines. To investigate this further, the effect of temperature (40°C and 70°C; 60s), sonication (20 kHz and 100 % amplitude, for 20s and 60s, leading to the same temperatures as above, respectively), in combination with Aspergillopepsins I (AP-I) supplementation (100 µg/L), was studied on unstable protein concentration (TLPs and chitinases) using HPLC with an UV–Vis detector in a TLPs-supplemented model system and in an unstable white wine. In model wine, neither temperature nor sonication affected TLPs concentration, suggesting their unfolding reversibility. However, the presence of AP-I during US treatment reduced protein concentration, up to complete removal under the most powerful conditions. In wine, the temperature effect was enough to lower chitinase levels (~48% and ~54% reduction at 40°C and 70°C, respectively) but had an undetectable effect on TLPs level. US significantly reduced both protein families, being more effective on chitinases (52% and 69% reduction at 20 s and 60 s, respectively) than TLPs (~11%) with the most powerful treatment. Interestingly, US was more successful than heating on chitinase (32%) and TLPs (15%) removal at the most energetic conditions. The supplement of AP-I combined with heating or US further reduced protein concentration. For heat treatment, both proteins were affected at both temperature conditions (TLPs: ~25% and ~23%; chitinases: ~58% and ~46%), while AP-I combined with US only affected TLPs under the most energetic treatment (~18%). The study found that US can affect unstable grape proteins and has additional mechanisms beyond sonication-induced temperature increase. When combined with AP-I, it further reduces unstable proteins, and suggests interaction between the US and AP-I. Further investigation is required to determine if US treatment destabilises proteins through a mechanism distinct from temperature increase, considering other factors affecting protein stability in winemaking conditions.

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EVOLUTION OF CHEMICAL AND SENSORIAL PROFILE OF WINES ELABORATED WITH THEIR OWN TOASTED VINE-SHOOTS AND MICRO-OXYGENATION

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Keywords: differentiated wines, SEGs/MOX combination, winemaking technologies, wines quality

The positive contribution of toasted vine-shoots (*SEGs*, Shoot from vines – Enological – Granule) used in winemaking to the chemical and sensory profile of wines has been widely proven. However, the combination of this new enological tool with other winemaking technologies, such as micro-oxygenation (MOX), has not been studied so far. It is known that micro-oxygenation is used in wineries to stabilizes color, improves structure or combining with oak alternatives products to achieve a more effective aroma integration of wines. For that, its implementation in combination with *SEGs* could result in differentiated wines.

In this work, Tempranillo wines were in contact with their own *SEGs* in two different doses (D1 and D2), added at the end of malolactic fermentation and with two fixes dosages of micro-oxygenation (low, LMOX; and high, HMOX). At the end of the *SEGs*-MOX treatments, wines were bottled, and a sensory analysis was carried out over 6 months using a specific scorecard which included color, olfactory and taste descriptors. Also, along with the traditional olfactory and taste descriptors, a new one, named *SEGs*, was included to describe the specific impact of the vine-shoots. Besides, the phenolic and volatile compositions of wines were analyzed by HPLC-DAD and SBSE-GC/MS, respectively.

In the visual phase, the most significant factor was the time in bottle, being wines more violet at bottling and redder after 6 months. About the olfactory phase, the response was different depending on the *SEGs* dose, being wine elaborated with the highest doses of *SEGs* and MOX which showed the highest notes of toast, red fruits and nuts, and a very significant reduction of the herbaceous notes. This sensory profile was also maintained after bottle ageing, although the floral notes were slightly reduced, and the red and nuts notes increased. In the taste phase, panelists described wine elaborated with the highest micro-oxygenation dose (HMOX) and after 6 months in bottle with the most pronounced notes of *SEGs*, nuts and toast, independently of the *SEGs* dose used. On his part, tannins became less silky in all wines with time in bottle, regardless of the dose of *SEGs* and MOX used. Regarding the volatile compounds, bottle time was the most important factor for the differentiation, being wines from 3 months in bottle those that showed higher levels of aldehydes and norisoprenoids and, after 6 months, the concentration of alcohols, volatile phenols and esters was increased. As for phenolic compounds, a general decrease in the total content was observed with bottling, being trans-resveratrol the compound that remained practically constant.

EXTRACTIBLE COMPOUNDS FROM MICROAGGLOMERATED CORK STOPPERS TREATED WITH SUPERCRITICAL CO₂ AND NATURAL CORK STOPPERS USED FOR WINE BOTTLING

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Keywords: wine cork stoppers, polyphenols, suberic acid, sensory analysis

After bottling, the wine continues to evolve during storage. The choice of the stopper is an important factor in this evolution. In addition to the oxygen permeability of the closure, the migration of stopper compounds into the wine can also have an impact on the wine organoleptic properties. Many studies have shown that transfers of volatile compounds from the stoppers into the wine can happen depending on the type of closure used (1). Moreover, when cork-made stoppers are used, the migration of phenolic compounds from the stopper into the wine can also occur (2, 3). However, there are few studies on the migration of polyphenols from agglomerated corks treated with supercritical CO₂. Therefore, the present study aimed to quantify the polyphenols released by microagglomerated cork stoppers in hydroalcoholic solutions in comparison with those extracted from natural cork stoppers. Thus, extractions were performed on eight different batches of natural cork stoppers and eleven of microagglomerated cork stoppers treated with supercritical CO_2 . For this, six stoppers from each batch were immersed in 400 mL of 12% ethanol solution at 40°C for 10 days. The nineteen macerates were then analyzed by HPLC-DAD-ESI-QQQ to identify and quantify the extracted polyphenols and suberic acid. The microagglomerated corks released significantly fewer polyphenols (i.e., 25 times less). Regarding suberic acid, no differences were observed between both types of cork stoppers. Then, according to the groups obtained by a hierarchical ascending classification based on polyphenol composition, the macerates were pooled in equal volumes to reconstitute four new macerates of natural and/or microagglomerated cork stoppers. These four samples were then submitted to a panel of thirteen judges to perform a sensory profile with olfactory, taste, and mouthfeel descriptors. The results of this sensory profile showed that microagglomerated stoppers appeared to have the lowest overall impact on the olfactory and gustative perception of the hydroalcoholic solutions.

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FACTORS AFFECTING QUERCETIN SOLUBILITY IN SANGIOVESE RED WINE: FIRST RESULTS

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Keywords: Quercetin, solubility, wine, anthocyanins, mannoproteins

Quercetin (Q) is present in grape in form of glycosides and as aglycone. These compounds are extracted from grape skins during winemaking. In wines, following the hydrolysis reactions, the amount of quercetin aglycon can exceed its solubility value. Unfortunately, a threshold solubility concentration for quercetin in wine is not easy to determine because it depends on wine matrix (Gambuti et al., 2020).

This study is aimed at evaluating factors affecting the solubility of Q in red wine. The role of anthocyanins and mannoproteins (MPs) was evaluated. The role of anthocyanins in Q solubility was evaluated by adding known amount of grape derived anthocyanins to a model solution containing 30 mg/L of quercetin. Data showed that the solubility of Q increased by increasing the amount of grape derived anthocyanins in model solution until a complete dissolution of 30 mg/L of Q when 740,8903 ± 17,069 mg/l of anthocyanins were added. This is likely due to the π - π interactions between anthocyanins and Q determining the formation of stable copigmentation complexes in red wine (Whaterhouse 2016). In a further experiment the addition of two different mannoproteins to a model solution containing 30 mg/L of quercetin and grape derived anthocyanins was also tested in controlled conditions. A slight positive effect of MPs on quercetin solubility (until the twelve % of value detected in control samples) was observed. It is therefore likely that group of compounds tested are involved in Q colloidal stability.

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Angelita Gambuti1 • Luigi Picariello1 • Alessandra Rinaldi1,2 • Martino Forino1 • Giuseppe Blaiotta1 • Virginie Moine2 • Luigi Moio New insights into the formation of precipitates of quercetin in Sangiovese wines (2020)

FLAVANOL COMPOSITION OF VARIETAL AND BLEND WINES MADE BEFORE AND AFTER FERMENTATION FROM SYRAH, MARSELAN AND TANNAT

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Keywords: Polyphenols, Flavanols, Tannins, Wines

Background: The Flavan-3-ol extraction from grape skin and seed during red-winemaking and their retention into wines depend on many factors, some of which are modified in the winemaking of blend wines. Recent research shows that Marselan, have grapes with high proportion of skins with high concentrations of flavanols, but produces red-wines with low proportion of skin derived flavanols, differently to the observed in Syrah or Tannat. But the factors explaining these differences are not yet understood. Thus, the aim of this work was to evaluate if factors cited to affect tannin extraction and solubility, like solid to liquid ratio, anthocyanin concentration, seed to skin proportion, are behind the differences found in the flavanol composition of Syrah, Marselan and Tannat wines. Material and Methods: Over two vintages, 2019 and 2020, wines were made by the blending of grape-must before-fermentation (BFB) or of wines, after-fermentation (AFB), in proportion of 1/2-1/2 of Tannat-Marselan, Tannat-Syrah, Syrah-Marselan, and 1/3-1/3-1/3 of Tannat-Syrah-Marselan. The varietal wines (VW) were elaborated as well. All treatments were vinified by triplicate at experimental scale. Grape samples were taken before each winemaking. Macerations along 8 days were made in all cases. Spectrophotometric analysis were performed together with HPLC-ESI-Q-ToF determinations of flavan-3-ols. The wine to skin prodelphinidins quotient was used to estimate skin contribution to the wine flavanols. **Results:** In all cases, the flavanol structural composition of the grapes and of the varietal wines corresponded to the one expected for cultivar it belongs to. Thus, the results confirmed that under traditional red-winemaking, the flavanol composition of Syrah and Tannat wines mainly depends on the Skins while in Marselan mainly on seeds. The blend wines had a flavanol content and structural composition that closely matched the one that could be expected considering the composition of the varietal wines and the proportion of each cultivar in the blend. Therefore, there was also no significant effect of the time of blend (BFB vs AFB) on the flavanol concentration or composition of the wines. Conclusion: None of the factors that were modified in the winemaking of blend wines were behind the differences observed in the flavanol composition of the varietal wines of Syrah, Marselan and Tannat. Ongoing studies in Marselan may help to better understand the flavanol composition of wines.

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FUNCTIONALIZED MESOPOROUS SILICA IS A VIABLE ALTERNATIVE TO BENTONITE FOR WINE PROTEIN STABILIZATION

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Keywords: protein, stability, bentonite, wine

The presence of grape-derived heat unstable proteins can lead to haze formation in white wines [1], an instability prevented by removing these proteins by adding bentonite, a hydrated aluminum silicate that interacts electrostatically with wine proteins leading to their flocculation. Despite effective, using bentonite has several drawbacks as the costs associated with its use, the potential negative effects on wine quality, and its environmental impact, so that alternative solutions are needed.

This project aimed at evaluating the effectiveness of functionalized mesoporous silica (FMS), in removing heat unstable proteins from white musts and wines. FMS treatments were benchmarked against a commercial Na-Bentonite in a series of experiments conducted on heat unstable white musts and wines of different origin, vintage and variety, and on different scales (from few mL to 10 hL). The stabilizing properties of the fining agents were determined by analyzing the protein profiles of treated wines (by RP-HPLC), and by assessing protein stability via heat tests [3]. In addition, the treatments' impact on other wine parameters (e.g., organic acid profiles, metal content, macromolecules, lees formation, sensory analysis) were determined.

For each wine, the dose of bentonite and FMS needed to reach full protein stability was determined by fining rate trials. The amount of FMS needed to stabilize the wines was always in line with that of bentonite, with a small variability (±10%) attributable to differences in wine composition. FMS effectively removed both thaumatin-like proteins and chitinases in a dose dependent mode, without causing other modifications on wine composition in terms of organic acid profile, ethanol content, glycerol, volatile composition, and metal content that, on the other hand, was always modified by bentonite fining that always led to an increase in Fe and Al. The analysis by triangle test of two white wines (Sauvignon blanc and Traminer) stabilized with FMS or bentonite at similar addition rates revealed the lack of significant differences (total answers = 39, p = 0.5599 for S. blanc, p = 0.1184 for Traminer).

In general, FMS showed to effectively stabilized wines at addition rates similar to those of bentonite, without causing major compositional modification, nor detectable sensory impacts, and therefore they represent a good candidate to become a viable bentonite alternative.

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HAZE RISK ASSESSMENT OF MUSCAT MUSTS AND WINES : WHICH LABORATO-RY TEST ALLOWS A RELIABLE ESTIMATION OF THE HEATWAVE REALITY?

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Keywords: Haze risk, Muscat, wine, heat test

Wines made from Muscat d'Alexandria grapes exhibit a high haze risk. For this reason, they are systematically treated with bentonite, on the must and sometimes also on wine. In most oenological laboratories and in companies (trade, cooperatives, independent winegrowers), the test that is by far the most widely used, on a worldwide scale, remains the heat test at 80°C for 30 minutes to 2 hours (and sometimes up to 6 hours). The tannin test (sometimes coupled with a heat treatment) and the Bentotest are still used. In this study, we show that all these tests give much higher estimates of the haze risk than the risk assessed by a 24-48h treatment at 42°C, which represents a heat wave. For this purpose, we performed heat treatments ranging from 40 to 80°C in order to find out which test best reflects a heat wave episode. Each of these tests was carried out at different heating times (kinetic approach) and with wines presenting risks of protein breakage ranging from low to high. The results show that : 1) the test at 50°C for 1h (in a water bath) is by far the most correlated with the haze appearing when the wine is spent 24-48h at 42°C and 2) this test has a safety margin to choose the most adapted protein stabilisation treatment. Conversely, treatment at 80°C gives very high turbidities. The direct consequence of the 80°C-heat test is the use of too high doses of bentonite to eliminate a risk that is in fact poorly assessed. In this study dedicated to Muscat from Spain (Catalunya) wines, we show that it is possible, by means of a 1-hour heat test at 50°C carried out in the laboratory, to decide on the most appropriate treatment. In concrete terms, this translates into the reduction of bentonite doses, but also into the possibility of using oenological alternatives to this treatment.

IV.P.32 IMPACT OF ACIDIFICATION AT BOTTLING BY FUMARIC ACID ON RED WINE AFTER 2 YEARS

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Keywords: Wine acidification, Fumaric Acid, Red wine, Oenological parameters

Global warming is responsible for a lack of organic acid in grape berries, leading to wines with higher pH and lower titrable acidity. The chemical, microbiological and organoleptic equilibriums are impacted by this change of organic acid concentration. It is common practice to acidify the wine in order to prevent these imbalances that can lead to wine defects and early spoilage. Tartaric acid (TA) is most commonly used by winemaker for wine acidification purposes. Fumaric acid (FA), which is authorized by the OIV in its member states for the inhibition of malolactic fermentation, could also be used as a potential acidification candidate since it has a better acidifying power than tartaric acid. Thus, the objective of the present study is to investigate the impact of the addition of FA at bottling in comparison to TA on white and red wine's quality.

For this purpose, Cabernet Sauvignon wine was first split into two tanks, one of which was sulfited at 80 mg/L. The two batches, sulfite-free and sulfited, were then redivided into three batches, one control without any addition, one with TA addition at 2,5 g/L and one with FA addition at 2 g/L. The wine was then bottled and the following analysis were performed after 24 months. Classical oenological parameters (pH, titratable acidity), color parameters (color intensity, CIELAB), total phenolic compounds (IPT, Folin), as well as antioxidant capacities (CUPRAC, DPPH), total tannins, total anthocyanins and their composition (HPLC analysis) were also analyzed. Sensory analyses were also performed on the wines in order to assess the organoleptic impact of FA addition.

IN DEPTH CHARACTERIZATION OF OENOLOGICAL CHARACTERISTICS OF TWO LACHANCEA THERMOTOLERANS STARTER STRAINS

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Keywords: sequential inoculation, Lachancea thermotolerans, acidity, 2D gas chromatography

Non-Saccharomyces starter cultures became increasingly popular over the years because of their potential to produce more distinctive and unique wines. The major benefit of the use of Lachancea thermotolerans as a fermentation starter is its ability to produce relevant amounts of lactic acid and reduce alcoholic strength, making it valuable for mitigating negative impacts of climate change on grapes and wine quality. Besides, like any other non-Saccharomyces yeast, L. thermotolerans can significantly affect a whole range of other physico-chemical wine parameters. This study investigated the impact of two L. thermotolerans strains (LT2 and LT5) in fermentation of Malvazija istarska, a Croatian white grape cultivar that in some terroirs and growing seasons requires acidification and/or reduction of alcohol level. A strain of Saccharomyces cerevisiae (EC1118) was sequentially inoculated to finish LT fermentations, and as a monoculture control. Standard physico-chemical parameters were determined by the OIV methods. Organic acids, glycerol, and pathogenesis-related (PR) proteins were determined by HPLC-DAD. Targeted UPLC-MS/MS was performed to analyse phenolic composition, while total phenols were measured by UV/Vis spectrophotometry. Volatile aroma compounds were determined by untargeted metabolomics using GC×GC/TOF-MS complemented by GC-MS targeted analysis. Both L. thermotolerans starters increased total acidity, while the concentration of lactic acid increased from 0.08 g/L in control to 0.73 g/L in LT2 and 0.88 g/L in LT5 treatment wine. Significantly higher concentration of glycerol was determined in wines produced by LT2 strain. Phenol composition was affected without a uniform pattern, while total phenolic content was decreased by LT2 and increased by LT5 strain. Among PR proteins, only a single thaumatin-like protein was significantly reduced by both strains. The use of L. thermotolerans significantly modulated the volatile composition of wines and the most pronounced changes included increased linalool, ethyl lactate, ethyl isobutyrate, ethyl phenyl lactate, and diethyl succinate concentrations. Results from this study contribute to the overall knowledge and understanding of L. thermotolerans contribution to sequential fermentation, with the emphasis on its oenological potential to produce wines with improved acidity and complexity.

INFLUENCE OF THE NITROGEN / LIPIDS RATIO OF MUSTS ON THE REVELATION OF AROMATIC COMPOUNDS IN SAUVIGNON BLANC WINE

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Keywords: white must, nitrogen, lipids, esters

Production of volatile compounds by yeast is known to be modulated by must nitrogen. Nevertheless, various parameter of must quality have an impact on yeast fermentation. In this study we propose to evaluate the impact of nitrogen / lipids balance on a Sauvignon Blanc grape juice (Val de Loire).

Must was prepared from the same grapes at pilot scale. Three modalities were carried out: direct pressing, direct pressing with a pre-fermentation cold stabulation and pellicular maceration before pressing. Each juice had been clarified with and without pectolytic enzymes and spiked with different levels of grape solids and diammonium phosphate. The purpose of this experiment plan was to create four modalities with different nitrogen / lipids balances. These musts were fermented in laboratory normalized conditions. In addition of oenological analysis, free fatty acid and sterol were quantified in grape juices. After fermentation, varietal thiols, ethyl esters, higher alcohols and their acetates have been quantified.

Results showed that the nitrogen / lipids balance of grape must affect the concentration of aromatic compounds in wine, especially on the bioconversion of higher alcohols and 3-sulfanylhexanol into their acetates. Nitrogen supplementation was thus confirmed as having a positive effect on the yeast to ester acetates production. However, for the same level of nitrogen, lipids concentration may modulate ester acetates. More generally, a positive correlation has been observed between the nitrogen / lipids ratio and quantity of ester acetates in wine. Linear relation appeared between this nitrogen / lipids ratio and acetates / higher alcohols ratio.

Consequently, the nitrogen / lipids ratio seems to be a useful indicator for the winemaker to better control the desired aroma balance in white wines.

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INFLUENCE OF THE THICKNESS OF OAK ALTERNATIVES ON THE COMPOSITION AND QUALITY OF RED WINES

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Keywords: Oak alternatives, Thickness, wine composition, quality

Aging red wines in oak barrels is an expensive and laborious process that can only be applied to wines with a certain added value. For this reason, the use of oak alternatives coupled with micro-oxygenation has progressively increased over recent years, because it can reproduce the processes taking place in the barrels more economically and quickly [1]. Several studies have explored how oak alternatives [2-5] can contribute to wine composition and quality but little is known about the influence of their thickness. The aim of this research was therefore to study how different thickness of oak alternatives would influence the color, phenolic compounds and volatile composition of a red wine, especially with regard to the substances released by oak wood. For that purpose, a red wine was introduced in twelve 100-L plastic tanks with an oxygen permeability similar to oak barrels (Flexcube, Quilinox). Three tanks were supplemented with 2.5 g/L oak chips (between 7.5x3x1.5 and 20x13x3 mm), other three with 5 g/L of thin staves (7x47x960 mm), other three with 10 g/L of thick staves (17x47x960 mm) and finally the last three were maintained as controls. These dosages were chosen based on an equivalent oak impact intensity according to previous experiences.

All the wood alternatives were made of French oak (Q, petraea) with origin and characteristics as similar as possible. The wines were analyzed at 2, 6 and 12 months of aging in that conditions. Wines were also tasted by a trained panel at the end of aging time. The color intensity (CI), the Total Polyphenol Index (TPI) and the total tannins quantified by the methyl-cellulose precipitation method were significant higher in all wines supplemented with oak alternatives in respect to the controls, and it was observed that all these parameters increased as the thickness of the alternatives increased. In contrast, anthocyanins showed the opposite trend, being lower in concentration when the thickness of the alternatives was greater. Both trends, higher CI and lower anthocyanin concentration as the thickness of the oak alternatives increases, can be explained by the formation of polymeric pigments. In fact, the PVPP Index (% of combined anthocyanins) and the Ionization index (% of colored anthocyanins) augment as thickness increases. In general, all the volatile compounds coming from the wood (furans, vanillin, volatile phenols and whiskey-lactones) increased throughout the aging time and this increase was more important when the thickness of the alternatives was higher. Finally, the trained panel considered that color, aromatic intensity and complexity, sweetness, mouthfeel, structure and persistence of the wine improved significantly as the thickness of the alternatives increased. In addition, the panel preferred the wine aged with thick staves, followed in decreasing order by the wines aged with thin staves, oak chips and control. It can be concluded therefore that the thickness of the oak alternatives seems to have a clear influence on the composition and quality of the wines, the effect being significantly better when the thickness is higher.

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INFLUENCE OF WINEMAKING VARIABLES AND VINEYARD LOCATIONS ON CHE-MICAL AND SENSORY PROFILES OF SOUTH TYROLEAN PINOT BLANC

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Keywords: Pinot Blanc, pre-fermentative grape freezing, vineyard location, chemical profiles

Pinot Blanc, an important grape variety grown in some mountain areas of Northern Italy such as South Tyrol over the last decades, with its cultivation covering 10.3% of the total vineyards, has compatible climatic conditions (e.g. heat requirements) which are normally found in the geographical areas of the mountain viticulture [1,2,3,4]. Climatic changes are hastening the growth of this variety at higher elevations, particularly for the production of high quality wine. This report illustrates an example of a collaboration study where data were collected in a commercial winemaking setting to look into the factors that contribute to Pinot Blanc's typicity. The Control samples used a similar conventional vinification to compare three vineyards (Aldino, Montagna and Klaus). Four distinct winemaking techniques were examined for the vineyard "Aldino" taking into consideration characteristics like pre-fermentative grape freezing and co-inoculation with lactic acid bacteria. Musts before inoculation, young wines after one month and four month of aging and bottled wines at 0, 6 and 12 months of storage were investigated. The samples were analyzed by an offline HPLC-MS for the determination of the phenolic compounds and by HS-SPME-GCxGC-ToF/MS for determining the volatile profiles. The sensory analysis of the bottled wines was performed using Quantitative Descriptive Analysis (QDA ®) [5]. The profile of phenolic and volatile compounds of both musts and young wines were peculiar of each vineyards. For Aldino vineyard, the main differentiating factor for the musts and the young wines was the pre-fermentative grape freezing. No clear difference was observed in the phenolic and volatile profile as a function of co-inoculation with malolactic bacteria. For the bottled wines, specific sensory attributes contributed in the separation of the vineyards at all storage times. Furthermore, the overall quality judgement (OQJ) was significantly higher in all the wines at T12 storage time and for wines from Aldino. The sensory data were also combined with the chemical results to build multivariate models that exemplify how the components affect the wine's final quality. These built models as fingerprint databases could provide assistance to the winemakers during production and also render useful for authenticity purposes.

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MODULATION OF YEAST-DERIVED AROMA COMPOUNDS IN CHARDONNAY WINES USING ENCAPSULATED DIAMMONIUM PHOSPHATE TO CONTROL NU-TRIENT RELEASE

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Keywords: yeast nutrients, diammonium phosphate, aroma compounds, continuous supplementation

Yeast-derived aroma compounds are the result of different and complex biochemical pathways that mainly occur during alcoholic fermentation. Many of them are related -but not limited- to the availability of nutrients in the fermentation medium and linked to nitrogen metabolism and biomass produced. Besides, the metabolic phase of yeast also regulates the expression of many enzymes involved in the formation of aroma active compounds. The work investigates the overall effect of continuous supplementation of nutrients during alcoholic fermentation of a grape must on the volatile composition of wines. To this aim, diammonium phosphate was encapsulated mixed with a hydrophobic lipid matrix in two different supports designed to continuously release the salt for a final addition of 400 mg/L: a tablet-shaped support (Tb) of ~ 4 cm diameter and spherical microcapsules of ~0.2-1 mm diameter (Mc) obtained through spray cooling. The alcoholic fermentation was performed in triplicate at semi-industrial scale standardised conditions of turbidity (~100 NTU), yeast inoculum (200 mg/L) and fermentation temperature (19°C). Results were compared to those of wines fermented in absence of ammonium addition or supplemented with the same dose at the beginning of the alcoholic fermentation.

Among the metabolic compounds studied by GC-MS/MS, the production of acetate esters of higher alcohols was favoured by the Mc continuous ammonium release. This protocol almost doubled the total acetates formed in the untreated wines and increased ~33% and ~40% of those obtained with the one-shot supplementation and the Tb protocol respectively. Among alcohols, 2-phenylethanol and 2-methylbutanol were higher in the untreated wines and 1-propanol in the Mc protocol compared to others, even if the total amount of alcohols was not differentiated. Neither total fatty acids nor the corresponding ethyl esters were influenced by the nutrition protocol, even if some compounds were affected: ethyl hexanoate and ethyl octanoate were higher in the Mc protocol, differentiated from the Tb and one-shot protocols. Overall, nitrogen supplementation increased the total amount of esters in wines, being the Mc protocol the most performing, differentiated from the one-shot and Tb protocols that were statistically indistinguishable between them.

MONOSACCHARIDE COMPOSITION AND POLYSACCHARIDE FAMILIES OF LYO-PHILISED EXTRACTS OBTAINED FROM POMACES OF DIFFERENT WHITE GRAPE VARIETIES

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Keywords: white pomace, monosaccharides, grape polysaccharides, by-products

The recovery of bioactive compounds from grape and wine by-products is currently an important and necessary objective for sustainability. Grape pomace is one of the main by-products and is a rich source of some bioactive compounds such as polyphenols, polysaccharides, fatty acids, minerals and seed oil. Polysaccharides contained in the grape cell wall can be rhamnogalacturonans type II (RG-II), poly-saccharides rich in arabinose and galactose (PRAG), mannoproteins (MP), homogalacturonans (HG) and non pectic polysaccharides (NPP). The aim of this study was to evaluate the monosaccharide composition and polysaccharide families of extracts obtained from pomaces of different white grape varieties.

Twelve grape pomaces from 9 different white grape varieties of Castilla y León region were analysed after destemming and pressing the grapes. The polysaccharide extraction was carried out with the method previously developed by Canalejo et al. (2021). The lyophilised extracts were analysed by gas chromatography with mass spectrometry detector following the conditions developed by Guadalupe et al. (2012). An ANOVA and a principal component analysis (PCA) were carried out to determine the differences between grape varieties using the RStudio program.

Statistically significant differences were found between the white grape pomaces studied and even within the same grape variety. The Verdejo and Puesta en Cruz varieties showed the highest polysaccharide content due to the highest content in glucose and galacturonic acid. In addition, the Puesta en Cruz variety stood out for its higher rhamnose and galactose content. On the other hand, the Viura grape variety with highest maturity degree and the Sauvignon Rytos presented the lowest polysaccharide concentration. Considering the percentage of the different polysaccharide families, the PCA selected two components with an eigenvalue greater than 1, which explained 92.3% of the total variance. The Verdejo, Viura, Rufete Serrano and Soreli were more associated to the percentage of NPP, while the Malvasía, Puesta en Cruz and Sauvignon Blanc were more correlated with the HG and PRAGs.

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NEW TREATMENTS FOR TEMPRANILLO WINES BY USING CABERNET SAUVI-GNON VINE-SHOOTS AND MICRO-OXYGENATION

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Keywords: vine-shoots, micro-oxygenation, enological additive, bottle aging

Toasted vine-shoots as enological additive represents a promising topic due to their significant effect on wine profile. However, the use of this new enological tool with *SEGs* varieties different than wine and combined with others winemaking technologies, such as micro-oxygenation (MOX), has not been studied so far, despite this combination could result in wine with high chemical and organoleptic quality.

In this study, Tempranillo wines were in contact with Cabernet Sauvignon *SEGs* in two different doses (D1 and D2), added at the end of malolactic fermentation and with two fixed dosages of micro-oxygenation (low, LMOX; and high, HMOX). At the end of the *SEGs*-MOX treatments, wines were bottled, and a sensory analysis was carried out over 6 months using a specific scorecard which included color, olfactory and taste descriptors. Also, along with the traditional olfactory and taste descriptors, a new one, named *SEGs*, was included to describe the specific impact of the vine-shoots. Besides, the phenolic and volatile compositions of wines were analyzed by HPLC-DAD and SBSE-GC/MS, respectively.

In terms of sensory profile, wines were more purple at bottling, regardless of *SEGs* and MOX doses which decreased with bottle ageing, but the red color remained after 6 months in bottle. In the olfactory phase, wines were less herbaceous and showed more intense notes of nuts, toast, and red fruits after 6 months in bottle with both doses of *SEGs* and MOX. Finally, in the taste phase, panelists described the wines elaborated with D1 as more intense, highlighting the nuts, toast and vanilla notes after 6 months in bottle and with the HMOX. On his part, wines elaborated with D2 showed a very similar profile, regardless of the *SEGs/MOX* combination used, with slight differences between them in red fruits or vanillas notes. As for tannins, tasters described them as bitter, but also silkier at bottling time. In terms of volatile compounds, the highest concentration of esters, aldehydes or norisoprenoids, among others, was observed mainly in those wines elaborated with the highest doses of *SEGs* and after bottle time. As for phenolic compounds, a general decrease in their content was observed.

PHOTO OXIDATION OF LUGANA WINES: INFLUENCE OF YEASTS AND RESIDUAL NITROGEN ON VSCS PROFILE

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Keywords: Light-induced oxidation, Lugana wine, VSCs profile, Nitrogen

Lugana wines are made from Turbiana grapes. In recent times, many white and rosé wines are bottled and stored in flint glass bottles because of commercial appeal. However, this practice could worsen the aroma profile of the wine, especially as regards the development of volatile sulfur compounds (VSCs). This study aims to investigate the consequences of exposure to light in flint bottles on VSCs profile of Lugana wines fermented with two different yeasts and with different post-fermentation residual nitrogen.

Wines were produced with a standard protocol with Turbiana grapes with two different yeasts. During the alcoholic fermentation of the must additions of inorganic or organic nitrogen supply were made. Wines were bottled in inert conditions in flint bottles and exposed for 30 days to light at controlled temperature of 20°C. Subsequently the VSCs profile of the wines was analyzed using GC-MS techniques. Wines were then subjected to the sorting task sensory analysis.

The VSCs profile analyzed showed significant differences for carbon disulfide, methanethiol, dimethyl sulfide and dimethyl trisulfide. The variability given by the yeasts leads to statistically significant differences only for diethyl sulfide and dimethyl disulfide. Regarding the differences given by the residual nitrogen, the samples in the transparent bottles with higher residual nitrogen showed a greater increase of sulfur compounds. Wines with a higher organic residual nitrogen showed significant differences for carbon disulfide, methanethiol, dimethyl sulfide, diethyl sulfide and dimethyl disulfide. Linear correlations were found between residual nitrogen in wines and carbon disulfide, methanethiol and dimethyl sulfide. The sensory analysis sorting task highlighted the formation of two main classifications made up of wines with a low residual nitrogen and wines with a high residual organic nitrogen.

This study showed the great impact of light exposure in transparent bottles has on the aromatic and sensory quality and how the post-fermentation residual nitrogen, especially for organic nitrogen, in the wines can worsen this qualitative deterioration. This underlines the impact of the presence of residual nitrogen on the stability of the wine during maturation, placing the attention on the dose of nitrogenous nutrition introduced during alcoholic fermentation. The choice of yeast strain seems to have an influence, albeit minor, on the development of VSCs compounds in wines exposed to light.

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IV.P.41 POTENTIAL OF PEPTIDASES FOR AVOIDING PROTEIN HAZES IN MUST AND WINE

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Keywords: protein haze, peptidases, wine protein, wine stabilization

Haze formation in wine during transportation and storage is an important issue for winemakers, since turbid wines are unacceptable for sale. Such haze often results from aggregation of unstable grape proteinaceous colloids. To date, foreseeably unstable wines need to be treated with bentonite to remove these, while excessive quantities, which are often required, affect the wine volume and quality (Cosme et al. 2020). One solution to avoid these drawbacks might be the use of peptidases. Marangon et *al.* (2012) reported that Aspergillopepsins I and II were able to hydrolyse the respective haze-relevant proteins in combination with a flash pasteurisation. In 2021, the OIV approved this enzymatic treatment for wine stabilisation (OIV-OENO 541A and 541B).

Herein, we aimed to gain an improved understanding of the influence of this peptidase treatment on the colloids and the quality of must and wine. For this purpose, naturally present colloids were removed from a must and wine by ultrafiltration and replaced by protein-rich, well-characterized must and wine colloids, respectively. Subsequent enzymatic treatments were performed in duplicate on technical scale (ca. 60 L for must, 16 L for wine) by adding two aspergillopepsins separately to the musts and wine followed by a brief heating to 80 °C. Control batches were treated identically, except for enzyme addition. Aliquots (each 30 L) of the treated musts were fermented. The composition and concentration of the colloids in the treated musts and wines were determined chromatographically. Haze forming potential was assessed by the heat test. The influence of the peptidases on the quality of the resulting musts and wines was investigated by sensory trials (triangle tests).

Size exclusion chromatography showed a reduction of the proteins in the musts by the enzyme treatment of about 80% as compared to a 15% reduction for the heat treatment without enzyme. Fermentation of the enzyme-treated musts resulted in stable wines, while the wines from the must without enzyme addition were unstable. The treatment of the wine showed only minor reductions of proteins (19%) in all wines. An effect of the enzyme treatment on the carbohydrates or sensory differences were not observed in comparison to the treatment without enzyme.

In brief, we provide new insights into the influence of the OIV-approved peptidase treatment with aspergillopepsin on colloids and wine quality, which will help achieve greater acceptance from wine makers.

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REMEDIATION OF SMOKE TAINTED WINE USING MOLECULARLY IMPRINTED POLYMERS

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Keywords: smoke taint, volatile phenols, volatile phenol glycoconjugates, wine sensory

In recent years, vineyards in Australia, the US, Canada, Chile, South Africa and Europe have been exposed to smoke from wildfires. Wines made from smoke-affected grapes often exhibit unpleasant smoky, ashy characters, attributed to the presence of smoke-derived volatile compounds, including volatile phenols (which occur in free and glycosylated forms). Various strategies for remediation of smoke tainted wine have been evaluated. The most effective strategies involve the removal of smoke taint compounds via the addition of adsorbent materials such as activated carbon, which can either be added directly or used in combination with nanofiltration. However, these treatments often simultaneously remove wine constituents responsible for desirable aroma, flavour and colour attributes.

This study sought to evaluate molecularly imprinted polymers (MIPs) as a novel adsorbent developed specifically to target the removal of smoke-derived volatile phenols from wine. MIPs were initially added to a smoke tainted Pinot Noir wine, and their capacity to remove volatile phenols (by 40 to 50%) demonstrated by gas chromatography-mass spectrometry analysis of wine before and after treatment. A semi-commercial scale trial was subsequently undertaken and involved passing smoke tainted Chardonnay, rosé and Cabernet Sauvignon wines through a column packed with MIPs. The impact of treatment was evaluated by comparing the colour, volatile phenol composition and sensory profiles of wines, before and after treatment. Findings from this study afford winemakers valuable insight into the potential for MIPs to be used as a novel additive for the remediation of smoke tainted wine. Results from a kinetic study comparing the binding efficacy of different volatile phenols to MIPs will also be presented

TARTARIC STABILIZATION MAY AFFECT THE COLOR AND POLYPHENOLIC COM-POSITION OF TANNAT RED WINES FROM URUGUAY

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Keywords: anthocyanin, color intensity, tannins, Tannat

Tartrate precipitation affects the properties of wines, due to the formation of crystals that cause turbidity, even after being bottled. The forced tartaric stabilization is carried out frequently for young wines, through various physicochemical procedures. The traditional treatment for tartaric stabilization is refrigeration, but it can have a negative effect on wine's sensory properties, and particularly on the color of red wines. The aim of this study was to evaluate the effect of different tartaric stabilization options on the color and phenolic composition of Tannat red wines from Uruguay. Cold treatment (C: 14 days at 4 ° C), mannoproteins (M: 100 mL / HL), arabic gum (AG: 200 g / HL), carboxymethylcellulose (CMC: 10 g / HL) and carboxymethylcellulose + arabic gum (AG CMC + 10 g / HL + 200 g / HL) were tested. The chromatic properties, basic composition, polyphenolic indexes, and anthocyanins and derived-anthocyanin pigments contents were analyzed. The initial impact of the cold treatment was significant, but differences with the other wines were attenuated over time. At 15 days of the start of the essay, C wine had significantly lower color intensity and was much brighter and less red than control wine. C had too the lowest anthocyanin and proanthocyanidin contents. Anthocyanin profile of this wine shows an increase in the proportion of non-acylated glucosides and malvidin, and a decrease in the percentages of delphinidin, cyanidin and coumaryl-glucosides. Anyway, the typical anthocyanin profile of the variety was slightly modified. All wines showed low differences in color and polyphenolic composition at five months from stabilization. However, the contents of free anthocyanins were decreased by all stabilization treatments in relation to the control wines. CMC+GA wines had the highest color intensity and proanthocyanidin levels while M and CMC wines had the highest catechin contents. At this time, it was verified that the contents of free anthocyanins were diminished by all treatments in relation to the control wine. At 14 months, CMC+GA wines had the highest colour intensity, and the lowest luminosity (L*). Colour intensities of C and CMC wines had not differences respect to those of control wines. However, all wines showed precipitation of tartaric salts at 5 and at 14 months from stabilization. Therefore, the tested options (products and doses) do not stabilize red wines adequately.

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THE EFFECT OF BENTONITE FINING ON THE VOLATILE AND NON-VOLATILE PROFILE OF ITALIAN WHITE WINES

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Keywords: volatiles, macromolecules, fining, quality

Marselan wines have an unusual high proportion of seed derived tannins from grapes having high proportions of skins, which are rich in tannins. But the causes behind this characteristic have not yet been identified. In vintage 2023 wines were made at experimental scale (9 kg by experimental unit) from Arinarnoa, Marselan and Tannat Vitis vinifera grape cultivars by traditional maceration, and by techniques aimed to increase the wine content in skin derived tannin: addition of extraction enzymes, addition at vatting of grape-skin enological tannins, or by extended maceration, known to increase the seed derived tannin contents of wines. Macerations were of 7 days, except in the extended macerations that were of 15 days. Additionally, samples of seeds and skins from each cultivar were separately macerated in a wine-like solution for 15 days. All treatments were made by triplicate. The contents of anthocyanins and tannins were analysed along macerations spectrophotometrically (tannins reactive to methyl cellulose, total anthocyanin) and using a HPLC-DAD system (pigments, flavan-3-ols). During the first 3 days of winemaking, Arinarnoa and Tannat musts had similar tannin contents that were much higher than those in Marselan musts. But at day 5, Arinarnoa had reached its maximum tannin content while in Tannat and Marselan it continued to increase until day 7. At this point, Marselan had as much tannin contents as Arinarnoa while Tannat had much higher concentrations. Along the post-fermetative macerations, Tannat tannin contents decreased while they continued to increase in Marselan. Thus, from day 13 to 15 of maceration Marselan and Tannat had similar tannin contents that were at devatting significantly higher than in Arinarnoa. The addition of skin tannins did not significantly increase the tannin concentrations of wines. Noteworthy, just in Marselan, the maceration enzymes significantly increased the anthocyanin and particularly the tannins concentrations of musts relative to the other treatments in a magnitude that increased with the maceration time. The macerations in wine-like solutions showed that the extraction of anthocyanins and particularly of skin tannins was very low in Marselan related to the observed in Arinarnoa and Tannat, while the seed tannins were extracted at similar rate in the three cultivars. This research proved that the high proportion of seed tannins in Marselan wines is due to a limited extraction of these compounds from the skins.

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THE EFFECT OF DIFFERENT TERROIRS ON AROMA COMPOUNDS OF 'KALECIK KARASI' WINES

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Keywords: Aromatic compounds, Kalecik Karası, Terroir, SPME GC-MS

Kalecik Karası is a domestic grape variety of Turkey, originating from Kalecik district, 80 km from Ankara. Although there is no definite evidence, it is known that it was used in wine production by many civilizations that lived in the Anatolian region, especially the Hittites. Compared to other black wine grapes, it stands out with its low tannin content, rich fruity aroma and complex structure. In good vintages, red fruits such as strawberries, cherries and raspberries stand out in the aroma profile. Although its structure is elegant, it has the potential to age and develop similar to the 'Pinot Noir' wine of the Burgundy region. This offers a complex aroma structure including red flowers, earth and ripe fruits.

The concept of terroir, which explains that the characteristic features of the wine are due to the limited geographical region where the vine is grown and the wine production techniques, plays an important role on the aroma of the wine. Terruar becomes a complex concept with the effect of agricultural practices and production techniques as well as the effects of grape variety, climate and soil. Therefore, there is not much information and clarity about the aromatic effect of terroir in Kalecik Karası wines. This study aimed to investigate the effect of terroir on aroma differentiation in Kalecik Karası wines.

The study was carried out on the wines of the Kalecik Karası grapes grown in three different regions of the 2021 vintage, each of which has different growing conditions, different climate and soil structure. The aroma compounds of the samples taken from the wines produced in Ankara, Cappadocia, Denizli-Çal regions with different climate and soil structures were analyzed as duplicate (n=2) in SPME (Solid Phase Micro Extraction) technique in GC-MS. In addition, the wines were evaluated by sensory analysis.

It was determined that the total amount of aroma compounds of Kalecik Karası wine samples taken from Denizli region was higher than the samples taken from Ankara and Nevşehir regions. In addition, in the sensory analysis, it was revealed that the Kalecik Karası wines grown in the Ankara region have red fruit, flower and cotton candy odors, while the ripe fruit and spice notes are dominant in the wines from the Denizli region. As a result, it has been determined that terroir plays a key role on the aromas of 'Kalecik Karası' wines.

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TOWARDS THE SHELF-LIFE PREDICTION OF OLD CHAMPAGNE VINTAGES DEPENDING ON THE BOTTLE CAPACITY

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Keywords: Carbone dioxide, Champagne, Aging on lees, Effervescence

Today, nearly one billion bottles of different sizes and capacities are aging in Champagne cellars while waiting to be put on the market. Among them, several tens of thousands of prestigious cuvees elaborated prior the 2000s are potentially concerned by prolonged aging on lees. However, when it comes to champagne tasting, dissolved CO_2 is a key compound responsible for the very much sought-after effervescence in glasses [1]. Yet, the slow decrease of dissolved CO_2 during prolonged aging of the most prestigious cuvees raises the issue of how long a champagne can age before it becomes unable to form CO_2 bubbles during tasting [2].

Measurements of dissolved CO_2 concentrations were done on a collection of 13 successive champagne vintages, stored in standard 75 cL bottles and 150 cL magnums, showing prolonged aging on lees ranging from 25 to 47 years. The vintages elaborated in magnums were found to retain their dissolved CO_2 much more efficiently during prolonged aging than the same vintages elaborated in standard bottles. A multi-variable exponential decay-type model was proposed for the theoretical time-dependent concentration of dissolved CO_2 and the subsequent CO_2 pressure in the sealed bottles during champagne aging. The CO_2 mass transfer coefficients through the crown caps used to seal champagne bottle prior the 2000s was thus approached in situ, with a global average value m3 s-1 [3]. Moreover, the shelf-life of a champagne bottle was examined in view of its ability to still produce CO_2 bubbles in a tasting glass. A formula was proposed to estimate the shelf-life of a bottle having experienced prolonged aging on lees, which combines the various relevant parameters at play, including the geometric parameters of the bottle [3]. Increasing the bottle size is found to tremendously increase its capacity to preserve dissolved CO_2 and therefore the bubbling capacity of champagne during tasting.

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OPTIMISATION OF THE AROMATIC PROFILE OF UGNI BLANC WINE DISTILLATE THROUGH THE CONTROL OF ALCOHOLIC FERMENTATION

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Keywords: Online monitoring of aromas, Anisothermal temperature, Lees, Distillation

The online monitoring of fermentative aromas provides a better understanding of the effect of temperature on the synthesis and the loss of these molecules. During fermentation, gas and liquid phase concentrations as well as losses and total productions of volatile compounds can be followed with an unprecedented acquisition frequency of about one measurement per hour. Access to instantaneous production rates and total production balances for the various volatile compounds makes it possible to distinguish the impact of temperature on yeast production (biological effect) from the loss of aromatic molecules due to a physical effect³. Up to now, this innovative approach has been carried out successfully on isothermal fermentations¹². Elucidating the role of anisothermal fermentation temperature profiles is a crucial issue that may lead to a deeper understanding of the influence of temperature on yeast metabolism in relation to the synthesis of aromatic molecules. In this study, results from different temperature control strategies of fermentation with increasing and decreasing profiles are explored. These fermentations were carried out on a laboratory scale with the online monitoring tool for alcoholic fermentation leading to a powerful dataset concerning higher alcohols, acetate and ethyl esters. The anisothermal control of the fermentation temperature shows that the production of higher alcohols is slowed down with the lowering of the temperature profiles and inversely for the ascending profiles. For isoamyl acetate and ethyl hexanoate, with ascending temperature profiles, larger losses are entailed with increasing temperature during fermentation and therefore the concentration in the liquid decreases. Obviously, the phenomenon is reversed for the descending profiles which allow to combine a better production of esters with an optimized conservation in liquid phase until the end of the alcoholic fermentation while minimizing the synthesis of higher alcohols. In strong concentrations, these alcohols may represent an organoleptic defect, especially for the distillation wines in Charente⁴. After the fermentation step, the wines were microdistilled with their lees in order to reproduce the conditions of distillation in Charente. Thanks to this step, it was possible to note the aromatic richness of the lees concerning the heavy ethyl esters⁵. Moreover, the impact of the anisothermal temperature profiles quoted above is also confirmed on the aromas released from the lees by the heating process of the distillation..

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USE OF COLD LIQUID STABULATION AS AN OENOLOGICAL TECHNIQUE IN WHITE WINEMAKING: EFFECTS ON PHENOLIC, AROMATIC AND SENSORIAL COMPOSITION

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Keywords: pre-fermentative technique, polyphenolic compounds, volatiles compounds, antioxidant power

The application of different winemaking techniques helps to modify the basic parameters, phenolic profile, and aroma components influencing the final wine quality. In particular, pre-fermentative processes aim to increase the extraction and preservation of grape native compounds. Among them, cold liquid stabulation (macération sur bourbes) consists in maintaining the grape juice on its lees, in suspended condition at low temperature (0-8 °C) for a variable time (generally from 7 to 21 days). The aim of this work is to apply the cold liquid stabulation on two Italian white grape varieties, Arneis and Cortese, to evaluate the impact on basic parameters, color, polyphenolic compounds (TPI), antioxidant power (DPPH), total polysaccharides, and free and glycosylated volatile compounds (GC-MS analysis) during and after the process. Cortese and Arneis grape juices were kept at 4 °C on their lees (manually suspended twice a day) during three different periods (7, 14, 21 days) and then compared to a control without stabulation. After the stabulation period, the lees were discarded and the juices fermented, cold stabilized, and bottled. The analyses were carried out at the end of stabulation, of the alcoholic fermentation and after one month from bottling. The chemical data obtained were supported with sensory analysis done by a trained panel on the wines after fermentation and bottling. The results showed that the cold liquid stabulation has an impact on the acidic composition of the produced wines for both varieties. The low temperature affected tartaric acid content, being it found lower already after 7 days of stabulation. Nevertheless, pH decreased in the samples stabulated for the longest time (21 days). Differences have been found on TPI of wines, even if in a different extent depending on the grape variety. In fact, on Arneis samples an increasing trend of TPI alongside antioxidant capacity was found, meanwhile in Cortese the stabulation led to a decrease in TPI, without differences in the antioxidant capacity among stabulated samples. This behaviour may be connected to the grape phenolic composition. After bottling, the produced wines were not sensory perceived different in terms of bitterness, astringency, and body. Nevertheless, Cortese stabulated wines at 14 and 21 days were preferred in terms of overall judgement with respect to control, in agreement with the higher content of volatile compounds. An increasing liking trend was found also for Arneis, whereas the highest content of volatile compounds corresponded to 7 days stabulation

VALORIZATION OF GRAPE WINE POMACE USING PULSED ELECTRIC FIELDS (PEF) AND SUPERCRITICAL CO₂ (SC CO₂) EXTRACTION

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Keywords: Grape pomace, Grape seed oil, Pulsed electric fields (PEF), Supercritical CO₂ (SC CO₂)

Wine grape pomace quantitatively and qualitatively represents the most important fraction of wine waste. Namely, this by-product makes ~ 20% of the total mass of vinified grapes, and it is characterized with high concentrations of polyphenolic antioxidants, as well as grape seed oil. Hence, valorization of wine pomace, as an alternative to traditionally employed disposal, has drown considerable interest in recent years. Earlier studies were mostly focused on the extraction of phenolics, while mechanisms enhancing the extraction of lipid fraction from grape pomace, as well as their impact on the grape seed oil quality are far less investigated. In addition, opposed to conventional solvent extraction methods, new trends lead to the employment of eco-friendly extraction technologies as supercritical CO_2 (SC CO_2) extraction. The aim of this research was to study impact of low and high intensity pulsed electric fields (PEF) pretreatments prior to SC CO₂ extraction of grape seed oil, from Graševina grape pomace, on the oil yield and chemical composition. Results showed that PEF assisted SC CO₂ extracted more than 95% of pomace lipids and contributed to significantly higher concentrations of both lipophilic (sterols and tocochromanols) and hydrophilic antioxidants (polyphenolic compounds) in grape seed oil. These concentrations were up to 10% higher for total sterols, but even more than 50% higher for total tocochromanols and total individual polyphenols, respectively. PEF pretreated samples showed significantly higher concentrations of stigmasterol, β -sitosterol, Δ 5-avenasterol, Δ 5,24-stigmastadienol and Δ 7-avenasterol. Moreover, significantly higher concentrations of all analyzed tocochromanols were also found in these samples, primarily of β -tocopherol, plastochromanol-8 and α -tocotrienol that showed more than two times higher values. In addition, PEF pretreatments significantly contributed to the extraction of all individual polyphenolic compounds, while more than two times higher concentrations were found for gallic, p-coumaric and ferulic acids. Moreover, PEF assisted SC CO₂ extraction showed favorable effect on the extraction of the most abundant fatty acid, linoleic acid. Finally, the highest concentrations of both lipophilic and hydrophilic compounds were extracted by PEF pretreatment of higher intensity.

VOLTAMETRIC PROFILING OF RED WINE COMPOSITION DURING MACERATION: A STUDY ON FOUR GRAPE VARIETIES

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Keywords: Maceration, Voltammetry, Polyphenols, Red wine

During red wine vinification, maceration allows the must, and consequently the wine, to be enriched with several compounds that contribute to the creation of the typical organoleptic characteristics of red wines. Among these, extraction of polyphenols (PPs) during maceration is a major process of enological interest.

The purpose of this study was the evaluate the suitability of a rapid analytical approach based in linear sweep voltammetry to monitor PPs extraction during vinification. Four red grape varieties were investigated, and fermentations were carried out with three different yeasts in triplicate. The density was evaluated daily and every 48h samples were taken to monitor changes in voltametric profiles and in the anthocyanins, polyphenols and precipitable tannins content.

The voltametric profiles of the wines were monitored using disposable screen-printed carbon electrodes with the working and counter electrode in carbon paste and an Ag/AgCl reference electrode. A drop of sample was loaded onto the sensor, and linear sweep voltammograms were acquired between 0-1200mV at a scan rate of 100mV/s. Analyzing the voltammograms, it was possible to observe differences between varieties and, within each variety, the temporal evolution of maceration.

In the second-derivative voltammograms, a positive peak was observed at low potentials, it increases initially showing a maximum after 7 days of fermentation. This peak is associated with the more easily oxidized compounds in wine. Another region of the voltammogram that shows a trend associated with the progress of fermentation is that around 440mV; in this region the negative peak reaches a maximum after 24 h after the start of maceration and then slowly decreases. This region has been associated with the concentration of monomeric anthocyanins and flavanols. The negative peak around 780mV results initially influenced by the presence of free SO₂, when SO₂ is bound the peak decreases in intensity and then increases again during maceration. By constructing PLS-R models for the concentration of anthocyanins, polyphenols and precipitable tannins the best pre-processing method resulted the second derivative and good regression models were obtained (R2 from 0.75 to 0.95).

In conclusion, this study provides a first proof of concept of the suitability of a simple analytical approach based on linear sweep voltammetry to monitor the evolution of phenolic composition during red wine maceration.

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WINE RACKING IN THE WINERY AND THE USE OF INERT GASES: CONTROL AND OPTIMIZATION OF THE PROCESS

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Keywords: Inert gases, racking wine, purging, blanketing, oxygen

Atmospheric oxygen (O_2) generates oxidation in wines that affect their physicochemical and sensory evolution. The O_2 uptake in the different winemaking processes is generally considered to be negative for the sensory characteristics of white and rosé wines. Wine racking is a critical point of O_2 uptake, as the large surface area of the wine exposed during this operation and the inability to maintain an effective inert gas blanket over it.

The aim was to study the uptake of O_2 during the racking of a model wine as a reference and to compare with purging the destination tank with different inert gases. In addition, inert gases were also used to protect the wine in the racking tank by blanketing the wine. Finally, a full-scale inerting study was carried out in a commercial winery during the racking of a white wine to evaluate the effectiveness of the use of different inert gases. Dissolved oxygen (DO) and Head Space Oxygen (HSO) was monitored in different points during the wine racking.

Purging an empty tank with different inert gases was effective being the CO_2 :Ar (20:80) mixture clearly the most effective, requiring less gas volume to displace O_2 . The opposite result was found with N_2 because it worked in dilution mode. Although from an economic viewpoint, the most recommendable gas was CO_2 .

The level of protection of the racked wine and the headspace in the empty destination tank differed depending on the gas used and the thickness (% of the tank volume) of the blanket formed with each gas. Based on the results obtained, purging with 25% of the empty tank volume of each inert gas is recommended to protect racked wine in a good cost-benefit way. To keep the headspace of the racking tank inert, blanketing with 50% of tank volume of Ar, CO_2 or the mixture of both were sufficient. Applying different volumes of gas had little effect on the DO of the wine at the tank outlet.

The study of a white wine racking in a commercial winery demonstrated the greater efficacy of Ar versus N_2 in the purging of the destination tank, while for the hoses inerting, the differences between both gases were minor. In addition, Ar was able to maintain the wine at lower DO levels as well as to provide a higher level of HSO protection in the destination tank during the racking process.

The results obtained allow us to recommend the appropriate type and volume of inert gas to minimize O_2 uptake during wine racking.

YEAST LEES OBTAINED AFTER *STARMERELLA BACILLARIS* FERMENTATION AS A SOURCE OF POTENTIAL COMPOUNDS TO IMPROVE SUSTAINABILITY IN WINE-MAKING

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Keywords: non-saccharomyces yeast, Yeast cell walls, Yeast protein extracts, Yeast polysaccharides

The yeast residue left over after wine-making, known as wine yeast lees, is a source of various compounds that are of interest for wine and food industry. In winemaking, yeast-derived glycocompounds and proteins represent an example of circular economy approach since they have been proven to reduce the need for bentonite and animal-based fining agents. This leads to a reduced environmental impact in the stabilization and fining processes in winemaking. (de Iseppi et al., 2020, 2021). The recent discovery of the wine-making potential of the non-Saccharomyces yeast Starmerella bacillaris has sparked new interest in the use of this species for lees valorization, due to its potential difference in cellular composition from the conventional wine yeast Saccharomyces cerevisiae (Lemos et al., 2016; Moreira et al., 2022). To investigate the cell compositions of yeasts present in the lees, 5 strains of Starmerella bacillaris and Saccharomyces cerevisiae were grown in winemaking conditions. After cells harvesting, different cell components (from cell wall and cytoplasm) were separated by means of cell breakage with glass beads and further enzymatic or chemical treatments. The fractions were characterized in respect of sugar and protein content, by means of HPLC and SDS-PAGE separation, evidencing differences between the species in terms of mannose, glucose and N-acetylglucosamine profile, protein content and protein molecular size. To investigate the practical implications on winemaking, the fractions were tested on wine as agents of protein stabilization and fining. This allowed to make some preliminary evaluation about the potential applications of Starmerella bacillaris as yeast derivatives, obtained from yeast lees.

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LARGE SURVEY OF THE CHEMICAL COMPOSITION OF WINES RESULTING OF THE PRESSING OF RED WINE MARC. FIRST RESULTS

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Keywords: Press-wine, Phenolic composition, Aromatic composition, Sensorial categorization

In the Bordeaux vineyards, press red wine represents about 15% of the volume of wines. Valuing this large volume of press wine is necessary from an economic point of view, of course, but also because of their organoleptic contribution to the blend. Nevertheless, there is a lack of recent knowledge on the composition of press wines. This work aims to establish an initial assessment of their composition (aromatic and polyphenolic) and to set up hypothesis on to the links with their sensorial identity.

Measurements and dosages were done in 50 press wines and their associated free-run wines. Wines are monovarietal batch from: cabernet-sauvignon and merlot from Saint-Estèphe, Médoc, France. The vintage is 2021. The production of wines was done in the estate to the classical process. The grapes, picked up and harvested by hand, were destemmed, sorted using an optical sorter and crushed. During vinification, extractions were adapted to each batch by daily tastings. Maceration did not exceed 21 days. After pressing, the wines were kept in oak-barrels. Three months after pressing, all the wines were tasted and were categorized according to their aptitude to be incorporated in the blend of the premium wine. Samples were kept at 12°C in bottles.

For the study of the aromatic composition: dimethyl sulphide (DMS) and its precursors (HS-SPME-GC-MS); higher alcohols (GC-FID) and 33 esters (HS-SPME-GC-MS) were measured. Concerning the analysis of phenolic compounds: anthocyanin monomers (HPLC-UV) and flavonols (HPLC-fluo.) were determined. Indices such as IPT, CieLAB, pH, AT have also been measured.

As expected, the results showed a significant difference between the two grape varieties. For each grape variety, PCA suggest differences between the press wines and the drop wines for all compounds. For the aromatic compounds, total tannins and flavonols: the press wines are more concentrated than the free-run wines. There are no significant differences between the press and free-run wines concerning anthocyanins. More statistical analysis permits to highlight unexpected separation of compounds according to the pressing step. The data also highlight links between the composition and the sensorial categorization.

In conclusion, the study permits to propose a first molecular database and to explore the origins of the sensorial categorization of that wines.

The experiment is renewed during the 2022 harvest and new compounds are added to the database.

IV.P.54 ADDITION OF OAK WOOD ALTERNATIVE PRODUCTS: QUALITATIVE AND SENSORIAL EFFECTS FOR A WHITE WINE OF ALIGOTE.

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Keywords: Aligoté white wine, oak chips, phenolic compounds, aroma markers

Wines matured in contact with wood are extremely popular with consumers all over the world. Oak wood allows the organoleptic characteristics of wine to be modified. Wines are enriched with volatile and non-volatile compounds extracted from the wood. The aromas extracted from oak wood contribute to the construction of the wine's aromatic profile and the main polyphenols extracted can modify taste perceptions such as astringency and bitterness. All the compounds extracted from the wood thus contribute to the balance and quality of the wines. The maturation of wine in vats with the addition of alternative oak products has become increasingly popular in all wine producing countries of the world. The main reasons for the development of such products are the optimisation of their production, the reduction of the cost of wine ageing as well as the increase of the level of hygiene in the production. This study is part of this context and focuses on oak chips: an alternative wood product to barrels. It aims to evaluate the optimum dose and the best level of toasting of the oak wood for the addition of these chips during alcoholic fermentation in a white Aligoté wine. During our experiment, the white Aligoté must before alcoholic fermentation was added with different doses of chips (1-2-3-4-5 g/L) at different toasting levels (5 levels: fresh, light toasting, medium toasting, medium + toasting, strong toasting). A control wine could was also made without the addition of chips for comparison. In order to determine the optimal dose and toasting of the oak chips used, the classic oenological parameters (Foss: pH, Alcoholic Strength, Total Acidity, Volatile Acidity, Sugars), colour (A420nm and CIELAB parameters), total phenolic compounds (TPI, total tannins and Folin index), monomeric and dimeric proanthocyanidin, phenolic acid and ellagitannin composition (HPLC-UV/MS), as well as fruity and woody aroma markers (GC/MS) were analysed. Sensory analyses were also carried out for each wine.

VOLATILE COMPOUNDS AND SENSORY PROFILE OF NEBBIOLO RED WINES TREATED WITH WOOD FORMATS ALTERNATIVE TO BARRELS

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Keywords: oak aromas, whisky lactone, alternative oak formats, wine aging

In winemaking, the use of wood products alternative to barrels, has become a useful tool for the achievement of numerous oenological objectives, including the fast release of desirable volatile and polyphenolic compounds, colour stabilization, and important economic advantages if compared to the traditional barrel production. Among a huge array of variables, the wood format, the vinification protocol, especially the moment of the infusion of the woods and the exposed surface area of the alternative woods are of relevant significance, since they may influence the speed and intensity of the aroma transfer from the wood to the wine defining different sensory profiles.

The oak wood formats used during this research study were sticks, cubes, and chips, added in commercial doses to red Nebbiolo wines, in two different stages of the vinification process: during and after the alcoholic fermentation. Samples were analyzed for their aromatic composition at the racking and after seven months of aging. The sample preparation was carried out by Solid Phase Extraction (SPE) using polymeric cartridges with high load capacity and multiple retention mechanisms. Extracts were analyzed by gas chromatography-mass spectrometry (GC-MS) and gas chromatography-flame ionization detector (GC FID). The sensory profiles of the wines were assessed after seven months by a trained panel.

The quantitative and qualitative profile of oak aromas resulted strongly dependent on both the moment of application and the type of wood. Statistically higher values of vanillin and furan compounds were noted both in the wines where wood was added after the fermentation and in the wines treated with sticks. Wines treated with cubes showed elevated values of benzoic aldehydes and wines with chips statistically higher levels of whisky lactone, a key molecule for the expression of the boisée note in wines.

Sensory analysis results confirmed a significative greater intensity of descriptors such as, vanilla, or oak wood related to boisée notes, in the wines treated with cubes and chips, well recognizable if compared to those with sticks which had significantly higher violet highlights. Furthermore, the use of cubes accentuated the perception of some spicy scents such as cloves and pepper. In summary, considering the overall results, oak cubes appear to impart intermediate characteristics if compared to sticks and chips, conferring in wines a more balanced aroma and sensory profile.

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IV.P.56 TANNINS AND ANTHOCYANINS KINETICS OF EXTRACTION FROM ARINARNOA, MARSELAN AND TANNAT UNDER DIFFERENT WINEMAKING TECHNIQUES

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Keywords: Marselan, Tannat, Arinarnoa, Tannins

Marselan wines have an unusual high proportion of seed derived tannins from grapes having high proportions of skins, which are rich in tannins. But the causes behind this characteristic have not yet been identified. In vintage 2023 wines were made at experimental scale (9 kg by experimental unit) from Arinarnoa, Marselan and Tannat Vitis vinifera grape cultivars by traditional maceration, and by techniques aimed to increase the wine content in skin derived tannin: addition of extraction enzymes, addition at vatting of grape-skin enological tannins, or by extended maceration, known to increase the seed derived tannin contents of wines. Macerations were of 7 days, except in the extended macerations that were of 15 days. Additionally, samples of seeds and skins from each cultivar were separately macerated in a wine-like solution for 15 days. All treatments were made by triplicate. The contents of anthocyanins and tannins were analysed along macerations spectrophotometrically (tannins reactive to methyl cellulose, total anthocyanin) and using a HPLC-DAD system (pigments, flavan-3-ols). During the first 3 days of winemaking, Arinarnoa and Tannat musts had similar tannin contents that were much higher than those in Marselan musts. But at day 5, Arinarnoa had reached its maximum tannin content while in Tannat and Marselan it continued to increase until day 7. At this point, Marselan had as much tannin contents as Arinarnoa while Tannat had much higher concentrations. Along the post-fermetative macerations, Tannat tannin contents decreased while they continued to increase in Marselan. Thus, from day 13 to 15 of maceration Marselan and Tannat had similar tannin contents that were at devatting significantly higher than in Arinarnoa. The addition of skin tannins did not significantly increase the tannin concentrations of wines. Noteworthy, just in Marselan, the maceration enzymes significantly increased the anthocyanin and particularly the tannins concentrations of musts relative to the other treatments in a magnitude that increased with the maceration time. The macerations in wine-like solutions showed that the extraction of anthocyanins and particularly of skin tannins was very low in Marselan related to the observed in Arinarnoa and Tannat, while the seed tannins were extracted at similar rate in the three cultivars. This research proved that the high proportion of seed tannins in Marselan wines is due to a limited extraction of these compounds from the skins.

Sensory properties : psychophysics-cognitive psychology, experimental economy, connexions with neurosciences

V.P.1 HOW DO ROOTSTOCKS AFFECT CABERNET SAUVIGNON AROMATIC EXPRESSION?

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Keywords: rootstock, Cabernet Sauvignon, sensory analysis, aromatic expression

Grape quality potential for wine production is strongly influenced by environmental parameters such as climate and agronomic factors such as rootstock. Several studies underline the effect of rootstock on vegetative growth of the scions [1] and on berry composition [2, 3] with an impact on wine quality. Rootstocks are promising agronomic tools for climate change adaptation and in most grape-growing regions the potential diversity of rootstocks is not fully used and only a few genotypes are planted. Little is known about the effect of rootstock genetic variability on the aromatic composition in wines; thus further investigations are needed.

The purpose of this communication is to highlight how rootstock influences Cabernet Sauvignon red wine aromatic expression.

This study was conducted in 2021 in the GreffAdapt plot (55 rootstocks × 5 scions × 3 blocks) focusing on *Vitis vinifera* cv. Cabernet Sauvignon and on 20 rootstocks [4]. Grape samples were collected and fermented in triplicate at laboratory scale under standardized conditions; wines were stabilized and stored at the end of alcoholic fermentation.

Sensory analyses were performed to evaluate rootstock impact on aromatic expression. Conventional sensory profiles were carried out following the methodology used by Pelonnier-Magimel et al. (2020) [5], divided into three main steps: descriptor generation, specific training on the generated vocabulary and final evaluation. A panel with similar wine knowledge and previous sensory training was selected for this purpose.

During the first step of sensory evaluation, the tasters generated a defined number of descriptors on a wine selection and following this session 11 terms were chosen based on with panel agreement.

A specific session was carried out before the start of the training in order to validate the general consensus for the proposed references (or descriptors). A control sensory profile was organized after several weeks of training to verify the consensus of the panel.

Sensory analysis data did not allow to highlight a difference in Cabernet Sauvignon red wine aromatic expression for this specific vintage, characterized by excessive rainfall and mean temperatures below the seasonal average. In conclusion, the exploration of other sensory approaches would be interesting to complete this work, as well as a complementary study of other vintages characterized by contrasting climatic conditions compared to 2021.

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V.P.2 SENSORY EVALUATION OF WINE AROMA: SHOULD COLOR-DRIVEN DESCRIPTORS BE USED?

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Keywords: Aroma, Cognition, Color, Expertise

The vocabulary used to describe wine aroma is commonly organized according to color, raising the question of whether they reflect the reality of olfactory perception. Previous studies have assumed this convention of color-aroma matching, and have investigated color's influence on the perception of aroma only in dyed white wine or in red wine from particular places of origin. Here 48 white and red varietal wines from around the world were evaluated in black glasses then in clear glasses by a panel of wine experts, who gave intensity ratings for aroma attributes commonly used by wine professionals. In black glasses, aromas conventionally associated with white wine were perceived in the red wines, and vice versa. When wine color was made visible, ratings for green fruit, citrus fruit, and stone fruit generally decreased among the red wines and increased among the white wines, while the opposite occurred for red fruit, dark fruit, and oak. This dependence of aroma perception on visual input suggests the usage of certain descriptors by experts is more cognitive than purely sensorial. The influence of color was indirectly evident even in black glasses: three oaked Chardonnays were rated highly in red fruit and dark fruit, relative to the unoaked white wines, suggesting the judges here associated oak with red wine and consequently used oakiness to deduce wine color before rating the aromas believed to be appropriate. Findings suggest color-driven descriptors, used when wine color can be seen or surmised, do not foster objective assessments of wine aroma.

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V.P.3 USING CHECK-ALL-THAT-APPLY (CATA) TO CATEGORIZE WINES: A DECISION-MAKING TOOL FOR WINE SELECTION

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Keywords: Check-All-That-Apply, Hierarchical Cluster Analysis (HCA), multiple factor analysis (MFA), sensory characterization

Bordeaux is the largest appellation vineyard in France. This contrasting vineyard with varied terroirs offers all styles of wine, resulting from the blending of several grape varieties. If these different profiles make the renown of Bordeaux wines, it can appear as a constraint when the aim is to study Bordeaux wines in their diversity. The selection of a representative sample can be performed by a sensory analysis carried out by trained panelists or by wine professionals, which can take several forms: consensus among experts, conventional descriptive analysis, typicality or quality evaluation. However, because of time, economic, and logistical constraints, these methods have limited applications. As an alternative to classical descriptive analysis, more intuitive methods that do not require training have been proposed recently to describe wines using an expert panel such as Napping, Free Choice or Flash Profiling, CATA or RATA. However, in order to categorize a large number of wines, the CATA method seems to be the most appropriate, especially when working with wine professionals. CATA was used in order to define the distinct profile of 143 red Bordeaux wines sold at less than 8€ and to select the wines that best represent each profile. The wines were evaluated by 62 descriptors divided into 12 groups comprising 6 visual, 33 aroma, 5 flavors, 3 taste, and 15 mouthfeel attributes, as well as overall quality perception by 48 wine experts. The results were analyzed by Correspondence Analysis (CA) followed by Hierarchical Cluster Analysis (HCA) leading to the categorization of the wines into twelve groups. One to three representative wines of each group were selected to reach 20 wines in total. In order to validate the approach, trained panelists then analyzed the selected wines with a conventional descriptive analysis and these results were compared to those obtained with CATA questions by Multiple Factor Analysis (MFA). Both methods highlighted the same main sensory characteristics as well as a similar overall quality score. Color, woody character, vegetal notes, sweetness and pleasant mouthfeel were evaluated similarly for both panels. In contrast, fruity note evaluation seems to be more complicated and highlighted limitations for the two sensory analysis approaches. Nevertheless, CATA appears as a fast and reproducible technique for categorizing a large number of wines in order to select a representative sample of the products to be studied.

V.P.4 CONSENSUS AND SENSORY DOMINANCE ARE DEPENDENT ON QUALITY CONCEPT DEFINITIONS

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Keywords: quality, multisensory integration, wine experts, holistic approaches

The definition of the term "quality" in sensory evaluation of food products does not seem to be consensual. Descriptive or liking methods are generally used to differentiate between wines (Lawless et al., 1997). Nevertheless, quality evaluation of a product such as wine can also relate to emotional aspects. As exposed by Costell (2002), product quality is defined as an integrated impression, like acceptability, pleasure, or emotional experiences during tasting. According to the 'modality appropriateness' hypothesis which predicts that wine tasters weigh the most suitable sensory inputs for a specific assessment (Freides, 1974; Welch & Warren, 1980), the nature of the quality definitions may modulate sensory influences.

The aim of the present study was to evaluate whether the consensus, the discrimination between wines and the multisensory integration process during wine assessment depend on the types of questions or on the definition of the quality concept. The study was carried out among wine-tasting professionals who are able not only of technically judging the wines, but also of giving an appreciation of their emotional states (arousal and imagination elicited by wine) during the tasting. This served as a benchmark for comparing holistic technical, liking, emotional and aesthetic responses to wines. Expert wine tasters assessed holistic questions on twenty wines repeatedly, under different tasting conditions: global (all senses), unimodal (visual, smell and taste), and combined senses (visual/smell, visual/in-mouth sensations and olfaction/in-mouth sensations). After classical analysis (correlation between these different holistic questions, consensus and discrimination between wines according to the question), regression models suggested a dominance of smell in arousal, image and hedonism decisions, and of visual dominance for technical quality decisions. Visual cues dominated in the more technical quality questions, whereas smell cues prevailed in the emotional (representational) questions. A modulating view of multisensory integration was thus reflected which depends on the quality concept definitions being assessed.

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V.P.5 CONSUMER PERCEPTION OF INTERSPECIFIC HYBRID RED WINE COLOR IN RE-LATION TO ANTHOCYANIN PROFILE AND CHEMICAL COLOR PARAMETERS

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Keywords: interspecific hybrid wines, color, consumer perception, anthocyanin

Interspecific hybrid winegrapes are of growing interest in the context of climate change based on their disease resistance and cold hardiness. In addition to a need for increased understanding of their chemical composition, there is little empirical evidence on the consumer perception of non-*vinifera* wine. Phenolic compounds, and particularly color, play an important organoleptic and quality determination role in wine, but can vary significantly in interspecific hybrid wines compared to wines produced from *Vitis vinifera* cultivars [1, 2, 3]. Anecdotally, the variation in anthocyanin species, interactions, and concentrations in interspecific hybrids could result in a variance from *"vinifera*-like" wine color.

Eight commercial wines were sourced from regions across the United States, including five non-*vinifera* monovarietal wines (Corot noir, Maréchal Foch, Marquette, Norton, and St. Croix) and three Pinot noirs from France, Oregon, and New York. The selection of Pinot noir from three representative regions was to create a context of a *Vitis vinifera* cultivar with a unique anthocyanin profile. The eight wines were used in a consumer perception sensory analysis, where red wine consumers reported hedonic liking for a selection of color parameters as well as 'expected liking' prior to tasting the wine and 'actual liking' following in-mouth evaluation. Chemical analyses of these eight wines and eleven other non-vinife-ra wines included UV-Vis spectrophotometry, CIEL*a*b* colorimetry, and anthocyanin analysis via high-pressure liquid chromatography (HPLC) and mass spectrometry (MS).

For all color parameters, the wines ranked highest for liking included Maréchal Foch, St. Croix, and the Pinot noirs from France and Oregon. Wines with higher L* values (lighter color) and higher b* values (more brown hues), including Corot noir, Norton, and Pinot noir from New York, were less liked than darker wines with less brown hues. Notably, panelists reported that quality expectations formed from visual inspection did not match their actual liking of the wine.

Interestingly, this work suggests that color is only a weak predictor for actual liking of a wine. Furthermore, it is possible that the potential diversity of color from interspecific hybrid wines falls within the range of colors of the different cultivars and styles of wine produced around the world, allowing further research to move away from the goal of "vinifera-like" color in optimizing interspecific red wine production.

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V.P.6 HOW TO EVALUATE THE QUALITY OF NATURAL WINES?

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Keywords: Natural wines, Tasting sheet

The movement of Natural wines has clearly increased in the last few years, to reach a high demand from consumers nowadays. Switzerland has not been left out of this movement and has created a dedicated association in 2021. This association has the ambition to develop a specific tasting sheet for natural wines. The study of the tasting notes shows that the olfactory description of wines is recent but predominant today. But wine is a product makes to be drunk and not (just) to smell it. Based on these findings, a new 100-point tasting sheet has been developed. The main characteristics are 1) an evaluation in the mouth before the description of the olfaction, 2) to give 50% of the points on the judgment for the mouth characteristics, 3) to pejorate the visual aspects only if the wine is judged as "not drinkable" and 4) to express personal emotions. This sheet was tested during a national wine competition. In order to validate the capability of it to discriminate good and bad wines, several wines were selected; gold or silver medal wines and other wines with different scores. Chemical and microbiological analyses were performed on these wines. They were also described by an sensory expert panel according to the Check-all-that-apply method. This study highlights the sensory and chemical characteristics of "good" and "bad" natural wines. Moreover, we illustrate that it is possible to make natural wines without oenological deviations or that sometimes, these deviations can also bring a little complexity, as long as the intensity remains low. Analytic should be always completed by tasting !

V.P.7 PERCEPTUAL INTERACTIONS PHENOMENA INVOLVING VARIOUS VOLATILE COMPOUND FAMILIES LINKED TO SOME FRUITY NOTES IN BORDEAUX RED WINES

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Keywords: Red wine, fruity aroma, Perceptual interactions

Fruity notes play a key role in the consumer's appreciation of Bordeaux red wines. If literature provides a lot of knowledge about the nature of volatile compounds involved in this fruity expression, the sensory phenomena involving these compounds in mixture still need to be explored. Considering previous sensory works about the impact of esters and some overripening compounds, the goal of this work was to study the implication of perceptual interactions involving red wine odorant compounds of diverse origins and described as potentially affecting fruity aromatic expression

Six wines with particular profiles have been selected for this study. All the considered volatile compounds were assayed and sensory analysis consisting of triangular tests were conducted on these six wines in which supplementations by aromatic families have been done to reach the minimal, mean, and maximal concentrations found in Bordeaux typical wines. These concentrations were determined thanks to quantification in 20 Bordeaux red wines selected for their typicity.

For impacting additions, sorting tasks have been performed to evaluate the level of similarity between samples and sensory profiles were established in order to explicit which character was modified with additions.

Our results showed that the addition as well as the level of supplementation of some compounds have a significant impact on wine aroma. Volatile compounds from different origins, depending on their concentration, influence the perception of wine being involved in perceptual interaction. Further studies must now be conducted to explain more specifically how they affects red wine fruity notes.

V.P.8 GRAPE SPIRITS FOR PORT WINE PRODUCTION: SCREENING THEIR AROMA PROFILE

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Keywords: Grape spirits, Port wine, odorant compounds, sensory analysis

Port is a fortified wine, produced from grapes grown in the demarcated Douro region. The fortification process consists in the addition of a grape spirit (77% v/v) to the fermenting juice for fermentation interruption, resulting in remaining residual sugars in the wine and increased alcohol content (19–22%). The approval of grape spirits follows the Appellation (D.O. Port wine) rules1 and it is currently carried out based on analytical control and on sensory evaluation done by the public Institute that upholds the control of the quality of Douro Appellation wines. However, the producers of Port wines would like to have more information about quality markers of grape spirits. Thus, this work intends to characterize the aroma profile of several samples (23) of grape spirits for Port wine production. That characterization was done by using aroma descriptive analysis with a sensory panel and by using olfactometry (GC-O) in order to screen, with a sniffers panel, the most potent odorant compounds across the several volatile compounds of the samples. It was also determined the sensory thresholds of some of the identified compounds in order to determine the odorant activity value of each compound.

The aroma profile results revealed different grape spirits aroma profiles. The PCA applied to the average results (from a sensory panel) of aroma attributes intensities allows the separation of the samples across the two principal components, which explain more than 50% of the variability. The overall quality appears to be linked to the positive side of component 1 more associated with the fruity, floral and sweet odor notes. The samples with low quality are placed in the opposite side of this component, and linked to higher intensity of odour notes such as tails, herbaceous and oily.

The chromatographic analysis (GC–O and GC–MS) of several grape spirit samples pointed out as potent odorants several compounds that belong to different chemical families, namely esters, alcohols, terpenic compounds, acids and ketones. Based on the sensory thresholds, determined by the sensory panel in hydroalcoholic solutions (20% v/v), the odorant activity values were calculated for the different odorant compounds. The obtained results showed, that the compounds, which presented the higher odorant activity values were esters and terpenic group compounds.

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