ATR-MIR AND MCR-ALS AS A TOOL FOR MONITORING WINE ALCOHOLIC FERMENTATION AND DETECTING BACTERIAL SPOILAGE

ricard.boque@urv.cat

www.isens.urv.cat/en

J. Cavaglia[1], S. Mas-Garcia[2,3], J.M. Roger[2,3], M. Mestres[1], R. Boqué[4]

[1]Universitat Rovira i Virgili. Instrumental Sensometry (iSens), Dept. of Analytical Chemistry and Organic Chemistry, Tarragona, 43007, Catalonia, Spain

[2]ITAP, Univ. Montpellier, INRAE, Institut Agro, 34196 Montpellier, France // [3]ChemHouse Research Group, 34196 Montpellier, France

[4]Universitat Rovira i Virgili. Chemometrics, Qualimetrics and Nanosensors Group, Dept. of Analytical Chemistry and Organic Chemistry, Tarragona, 43007, Catalonia, Spain

CAC 2022 — 29 AUGUST - 2 SEPTEMBER ROMA — XVIII CHEMOMETRICS IN ANALYTICAL CHEMISTRY

Introduction

Wine alcoholic fermentation is a complex biological process that involves the transformation of reducing sugars into ethanol and CO_2, among many other secondary products [1]. In this process, one of the most important deviations promoted by bacterial spoilage is malolactic fermentation, involving the transformation of malic acid into lactic acid by lactic acid bacteria (LAB). This deacidification of wine causes an undesirable loss of freshness notes in white wines [2].

Monitoring alcoholic fermentation using rapid analytical tools, such as Mid infrared (MIR) spectroscopy, has proven to be suitable for detecting deviations before the end of the process, allowing corrective actions to be taken in time [3].

Multivariate Curve Resolution Alternating Least-Squares (MCR-ALS) on spectroscopic data has been suggested as a monitoring tool during food processing. Using MCR-ALS, the extraction of relevant information from the MIR spectra during wine alcoholic fermentation allows determining if the process is developing correctly [4].

Aim of study

- To describe, using MCR-ALS models, the evolution of sugars and ethanol concentrations in wine fermentation and the appearance of a new compound (lactic acid) originating from lactic acid bacteria spoilage.
- To evaluate if the information obtained from the MCR-ALS models could be used as a process control tool to detect possible deviations during wine alcoholic fermentation.

Materials and Methods

Fermentation samples

Concentrated white must diluted with miliQ water to 200-228 g/L of sugars (glucose + fructose) + Fermentation activators and nutrient supply: Actimax Bio® Enovit®

+ *Saccharomyces Cerevisiae* yeast → NOC

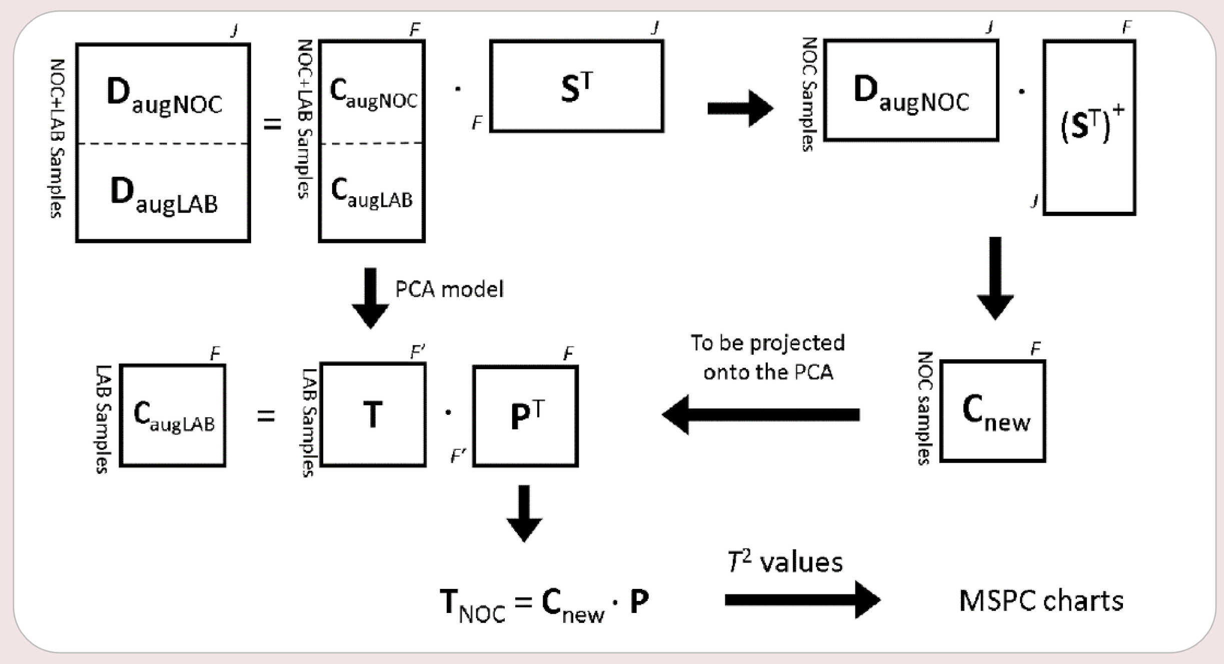
+ *Saccharomyces Cerevisiae* yeast + *Oenococcus oeni* & *Lactobacillus plantarum* lactic acid bacteria blend → LAB

Spectral data acquisition

Sampling at different time points → Centrifugation (1,5 mL) → A drop of sample → **Portable FTIR-ATR analysis:**
4000 – 649 cm^{-1}
3 replicates
32 scans
8 cm^{-1} resolution

Centrifugation → Determination of fermentation parameters (density, pH and malic acid)

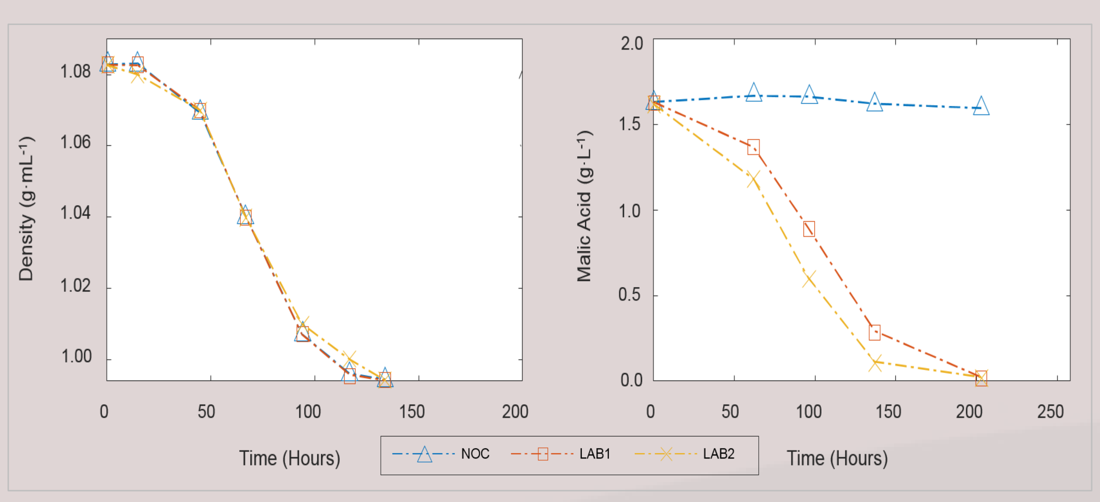
MCR-ALS models and MSPC charts



Results

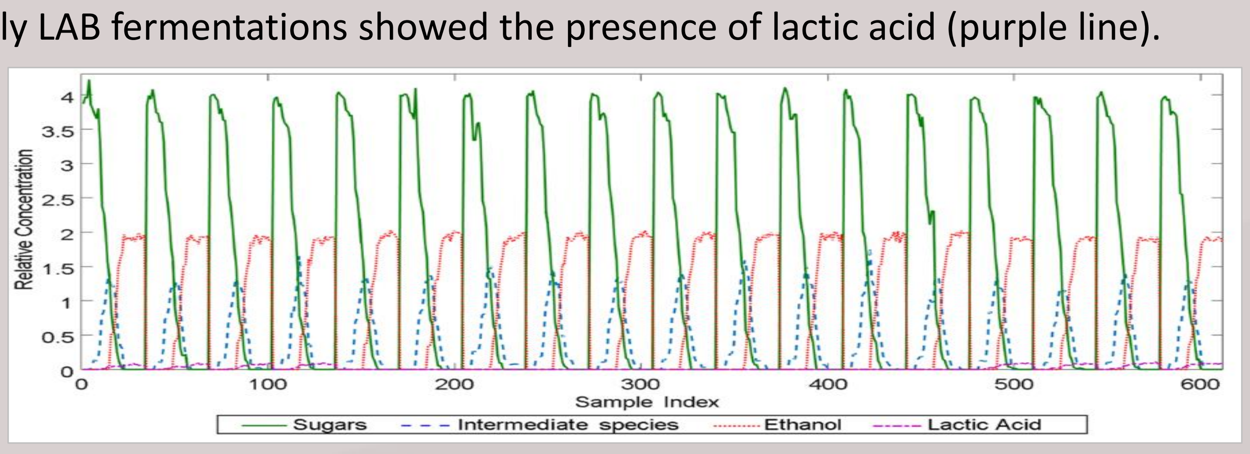
Evolution of the chemical parameters

- Density is an indirect measurement of the content of sugars in the must, so the density curves show the typical sigmoidal form of sugar consumption.
- Intentionally contaminated microvinifications with two different concentration of bacteria (LAB1 and LAB2) produced lactic acid confirming that those fermentations were deviated.



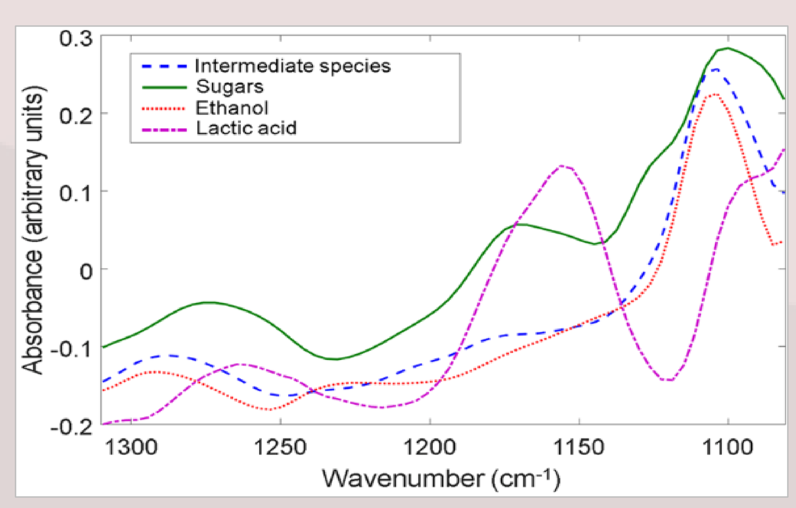
$C_{augNOCLAB}$ matrix for all fermentation batches

- All batches showed similar concentration trends for all components.
- Only LAB fermentations showed the presence of lactic acid (purple line).



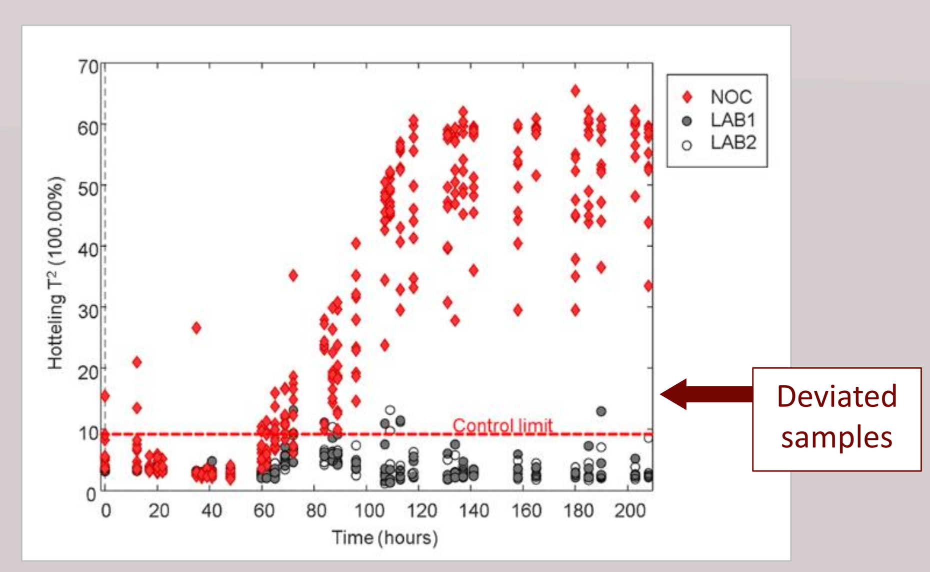
Pure spectral profiles in the resulting MCR-ALS model

- The pure spectral profiles (**S**^{T} matrix) obtained for the four components in the MCR-ALS model show absorbances in the expected regions for organic compounds.
- A peak for lactic acid is observed at 1150 cm^{-1}, which is related to C-O stretching from carboxylic acids.



'Inverse' MSPC chart

- An MSPC chart is proposed based on the contaminated samples rather than on control samples (NOC).
- Spectra from LAB samples were distinguishable from NOC samples before 100h, which is before the end of alcoholic fermentation.



Conclusions

- ATR-MIR data together with MCR-ALS models and MSPC charts could be used for the detection of lactic acid production during alcoholic fermentation.
- The use of MCR-ALS with ATR-MIR spectra enables to model the pure kinetic and spectra profiles of the main compounds involved in wine alcoholic and malolactic fermentations. This methodology becomes an improvement of the traditional MSPC charts for bacterial spoilage detection.
- If a fermentation batch is out of control in a traditional MSPC chart, but in control in this new 'inverse' MSPC chart, we could conclude not only that the sample is deviated, but also that the fermentation is deviated because of the production of lactic acid, as shown in the relative concentration profiles of MCR-ALS.

Bibliography

[1] P. Ribéreau-Gayon *et al.* Handbook of Enology, Vol.1: The Microbiology of Wine and Vinifications, 2nd ed, John Wiley & Sons (2006).

[2] L.F. Bisson & C.E. Butzke, Diagnosis and rectification of stuck and sluggish fermentations, *Am. J. Enol. Vitic.*, 51 (2000) 168-177.

[3] C.A.T. dos Santos et *al.* A review on the application of vibrational spectroscopy in the wine industry: From soil to bottle, *Trends Anal. Chem.*, 88 (2017) 100–118.

[4] J. Cavaglia et *al*. Detection of bacterial spoilage during wine alcoholic fermentation using ATR-MIR and MCR-ALS. *Food Control*, in press.

Acknowledgements

Grant PID2019-104269RR-C33 funded by MCIN/AEI/10.13039/501100011033

Authors would like to thank the scientific advice received from Anna de Juan (Universitat de Barcelona, Spain).