

Effect of the harvest time on the population and diversity of wine yeasts

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INTRODUCTION & AIM

The microbial composition of the grape berry influences the performance of alcoholic fermentation, thus impacting on the characteristics of the final wine [1].

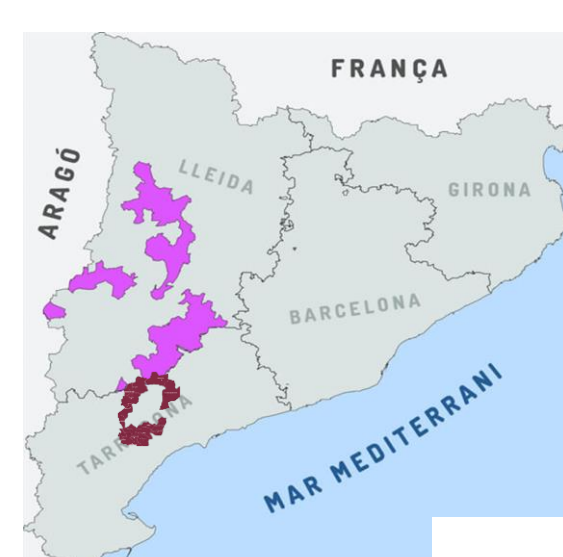
The optimal timing for grape harvest is critical for wine elaboration; it is decided based on climate conditions and physico-chemical characteristics [2].

There is little information on the dynamics of grape microbiota during its maturation.

The **aim** of the present study is to elucidate if the grape surface fungal microbiome evolves during the ripening process, and characterize its evolution.

METHOD

Harvest grapes at two maturation stages



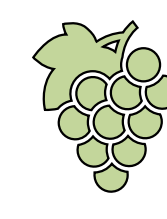
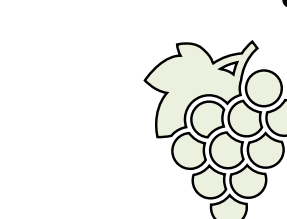
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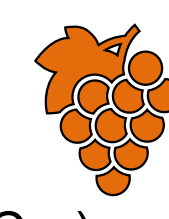
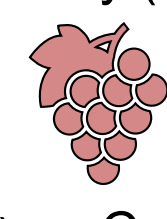
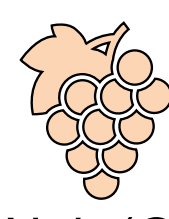


Green grapes

Mature grapes



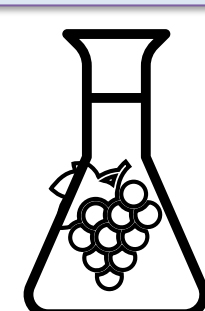
Chardonnay (Cha)



Grenache Noir (Gre)

Carignan (Car)

Monitor the spontaneous fermentation for each must typology

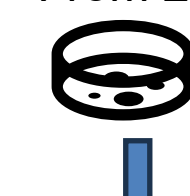


Physico-chemical analysis of musts and final wines
Density measurement
Plate count

Monitor fungal diversity of the stages: ➔

- Must (I)
- Mid-fermentation (density 1040–1060; M)
- End of fermentation (density 1010; F)

From 24 isolated colonies



PCR primers ITS1/ITS4 [3]
Yeast species identification

PCR primers Delta12/Delta21 [4]

S. cerevisiae genotyping

RESULTS & DISCUSSION

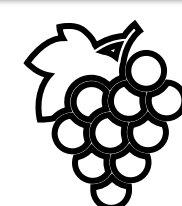


Table 1. Chemical characterization of musts. PA stands for Potential Alcohol and TA for Titratable Acidity

	Green grapes			Mature grapes		
	Chardonnay	Grenache Noir	Carignan	Chardonnay	Grenache Noir	Carignan
°Brix	18.0	23.6	19.6	22.0	27.2	22.7
PA (% v/v)	10.0	13.7	11	12.8	16.1	13.0
Density (g/cm ³)	1075	1100	1082	1093	1118	1096
TA (g/L)	8.1	8.2	5.5	5.2	4.8	5.7
pH	3.30	3.22	3.20	3.59	3.42	3.53

Table 2. Chemical analysis of the obtained wines. AC stands for Alcoholic Content.

	Cha	Gre	Car
Green grapes			
AC (% v/v)	12.32	-	11.68
TA (g/L)	7.97	-	8.20
pH	3.25	-	3.04
L-lactic acid (g/L)	0.67	-	< 0.1
L-malic acid (g/L)	2.86	-	1.30
Glycerol (g/L)	5.90	-	5.50

Mature grapes			
AC (% v/v)	13.25	16.78	14.33
TA (g/L)	5.61	5.10	5.80
pH	3.53	3.36	3.27
L-lactic acid (g/L)	<0.1	< 0.1	< 0.1
L-malic acid (g/L)	1.810	0.20	1.20
Glycerol (g/L)	7.230	9.90	7.10

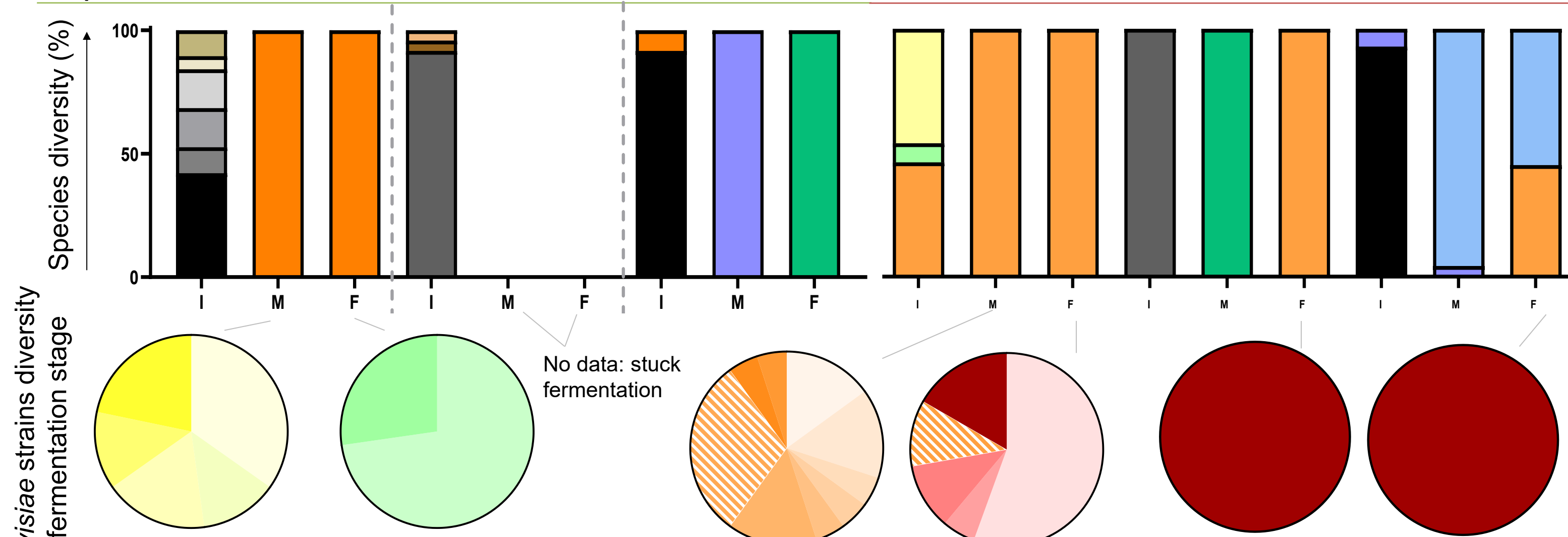


Figure 1. Diversity of species isolated and identified in the spontaneous fermentations. At the points where *S. cerevisiae* is present, the abundance of *S. cerevisiae* strains is represented in percentage. Identical strains are indicated by the same pattern; coincidence of colors does not indicate that are the same strain.

CONCLUSION

The community of the grape surface evolves during the ripening process.

- In the first stages predominate non-yeast fungi.
- In the mature grape generally predominates yeast, and the variability of *S. cerevisiae* increases.

FUTURE WORK / REFERENCES

Collect more data to have a better understanding of the microbiota evolution, as well as to correlate this evolution with varietal, physico-chemical, climate or geological factors.

- Loureiro, V.; Ferreira, M. M.; Monteiro, S.; Ferreira, R. B. The Microbial Community of Grape Berry. In *The Biochemistry of the Grape Berry*; Hernâni, G.; Chaves, M.M.; Delrot, S., Eds. 2012; pp 241–268.
- Fugelsang, K. C.; Edwards, C. G. *Wine Microbiology: Practical Applications and Procedures*, 2nd ed.; Springer: New York, 2007.
- Esteve-Zarzoso, B.; Belloch, C.; Uruburu, F.; Querol, A. Identification of Yeasts by RFLP Analysis of the 5.8S rRNA Gene and the Two Ribosomal Internal Transcribed Spacers. *Int. J. Syst. Bacteriol.* 1999, 49, 329–337.
- Legras, J.-L.; Karst, F. Optimisation of Interdelta Analysis for *Saccharomyces cerevisiae* Strain Characterisation. *FEMS Microbiol. Lett.* 2003, 221, 249–255.