

Yeast diversity during spontaneous fermentation of Tostado wine

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INTRODUCTION & AIM

Tostado wine is a prestigious sweet wine produced in Galicia (NW Spain) from air-dried grapes of traditional grape varieties grown in this region. The peculiar characteristics of the must, mainly its high sugar concentration, make its fermentation particularly difficult.

The **aim** of this study was to characterize the dynamics of the yeast populations involved in Tostado fermentation.

Key words: Tostado wine, spontaneous fermentation, yeast diversity, non-*Saccharomyces*, *Saccharomyces cerevisiae* strains



METHODS

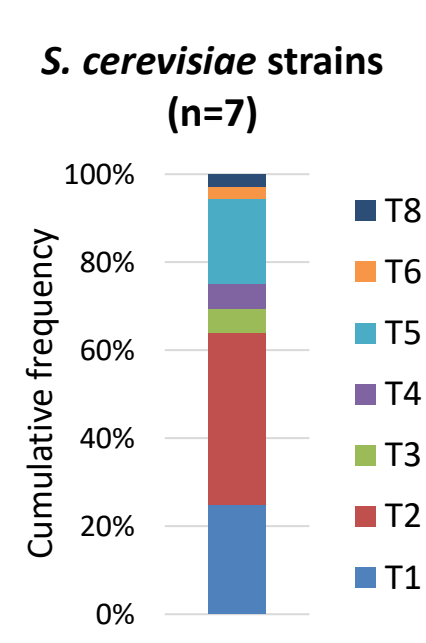
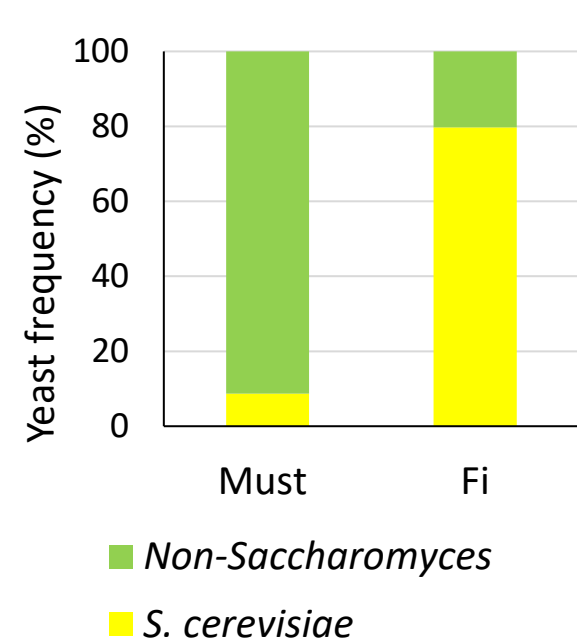
- White and red Tostado musts, supplied by local winegrowers
- **Spontaneous fermentation**
- **Yeasts isolation** at different stages of the process using conventional methodology.
- **Yeasts identification:**
 - At the species level - PCR amplification of the 5.8S rRNA gene and the two internal spacers ITS1 and ITS2.
 - At the strain level of *Saccharomyces cerevisiae* isolates- by mtDNA-RFLP.

RESULTS & DISCUSSION

Diversity of yeast population in musts from air-dried grapes and during their fermentation for Tostado wine elaboration

Winery 1: Grapes from ALBARIÑO

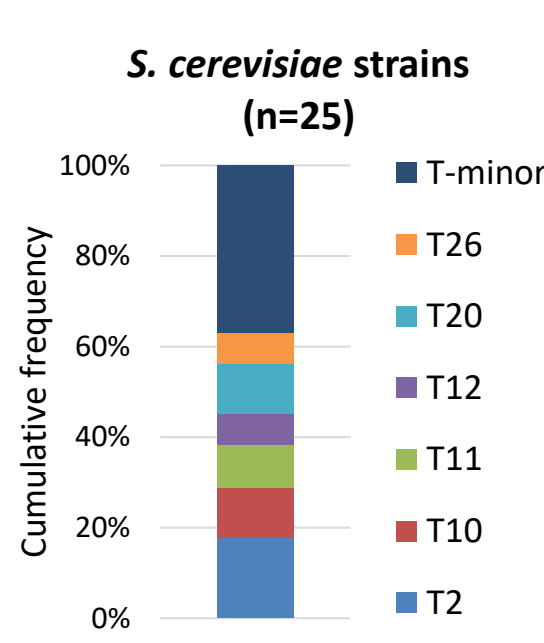
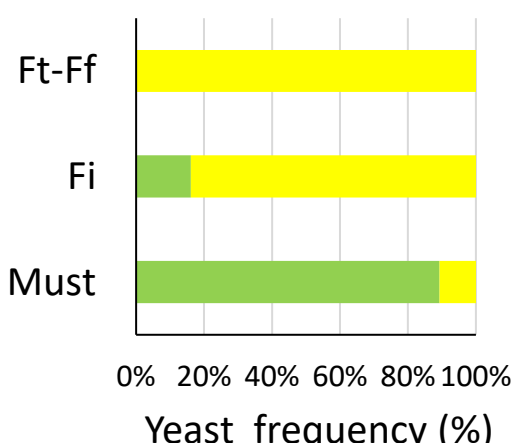
Yeast species (%)	Must	Fermentation
<i>Hanseniaspora uvarum</i>	30,43	20,26
<i>Metschnikowia</i> spp.	6,52	
<i>Starmerella bacillaris</i>	33,70	
<i>Candida californica</i>	10,87	
<i>Saccharomyces cerevisiae</i>	8,70	79,74
<i>Issatchenkia terricola</i>	1,09	
<i>Pichia anomala</i>	1,09	
<i>Zygosascus meyeriae</i>	2,17	
<i>Candida apicola</i>	5,43	



Winery 2: Grapes from TREIXADURA

Yeast species (%)	Must	Fi*	Ft-Ff*
<i>Hanseniaspora uvarum</i>	18		
<i>Metschnikowia</i> spp.	40	0,30	
<i>Starmerella bacillaris</i>		1,52	
<i>Saccharomyces cerevisiae</i>	11	83,89	100
<i>Pichia anomala</i>	1		
Other species	30	5,47	
<i>Lachancea thermotolerans</i>	1	8,81	

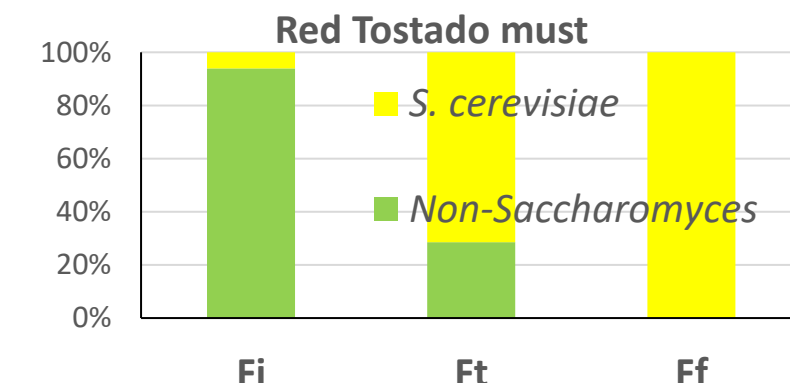
*Fi, Ft and Ff are initial, middle and final stage of fermentation; T-minor include those strains at frequency <5%



Winery 3: Mix of white grape varieties and mix of red grape varieties

Winery 3	White tostado must			Red tostado must		
Yeast Species (%)	Must	Fi*	Ft*	Fi*	Ft*	Ff*
<i>Hanseniaspora uvarum</i>	2,1	35,36		68,98	21,51	
<i>Metschnikowia</i> spp.	10,53	3,04		6,02		
<i>Starmerella bacillaris</i>	7,37	8,2	31,89			
<i>Candida californica</i>	14,21		4,98			
<i>Saccharomyces cerevisiae</i>				6,02	71,43	100
<i>Lachancea thermotolerans</i>	51,58	44,26	61,13	17,13		
<i>Debaryomyces hansenii</i>	14,21					
<i>Zygosaccharomyces</i> spp.		9,13				
Other species		1,99		0,92	7,06	

- The **must of white grapes** mix fermented with non-*Saccharomyces* species, mainly *Lachancea thermotolerans* and *Starmerella bacillaris*.



- Non-*Saccharomyces* yeasts, including *Hanseniaspora uvarum*, *Metschnikowia* spp., *Starmerella bacillaris*, *Lachancea thermotolerans* and *Candida* spp., among other yeasts predominated in the early stages of fermentation.
- As fermentation progressed, non-*Saccharomyces* yeasts were substituted by *Saccharomyces cerevisiae*.
- A codominance between different *S. cerevisiae* strains was observed in all fermentations.

CONCLUSION

The fermentation of Tostado musts shows a high diversity of yeast species and, at the strain level of *Saccharomyces cerevisiae*. These yeasts are assumed to be well adapted to the high sugar concentration of Tostado wines.

FUTURE WORK

The oenological properties of the isolated non-*Saccharomyces* species and *S. cerevisiae* strains will be evaluated to select suitable starter cultures for Tostado production.

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