XV JORNADAS DE
BIOLOGIA DE LEVEDURAS
"PROFESSOR NICOLAU VAN UDEN"

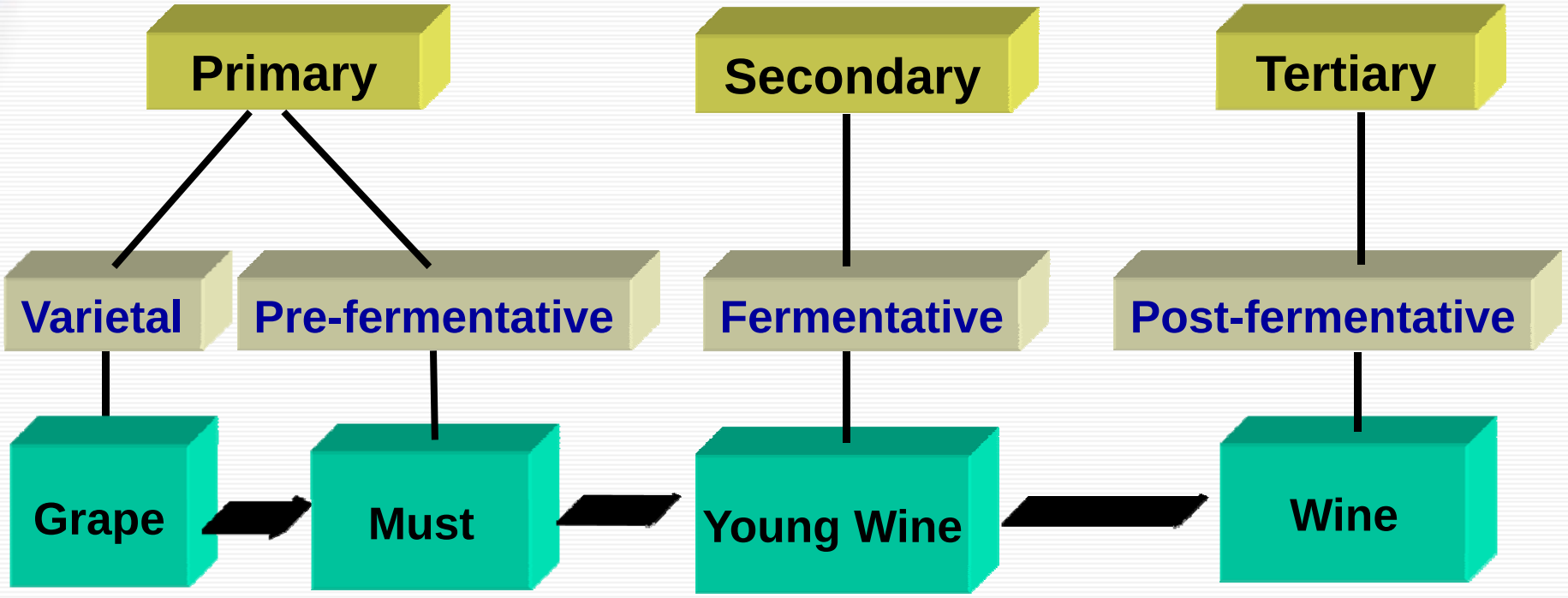
Porto 15-16.06.2007

The contribution of *Saccharomyces cerevisiae* strains to the aromatic profile of wines from the Vinhos Verdes Region

Claudia Lemos, Sofia Machado, Maria José Pereira, João Garrido, António Cerdeira, José Maria Oliveira, Dorit Schuller

Departamento/Centro de Biologia, Universidade do Minho
IBB - Institute for Biotechnology and Bioengineering, Centre for Biological Engineering, Universidade do Minho
Comissão de Viticultura da Região dos Vinhos Verdes (CVRVV)
Estação Vitivinícola Amândio Galhano (EVAG)

The origins of wine flavour



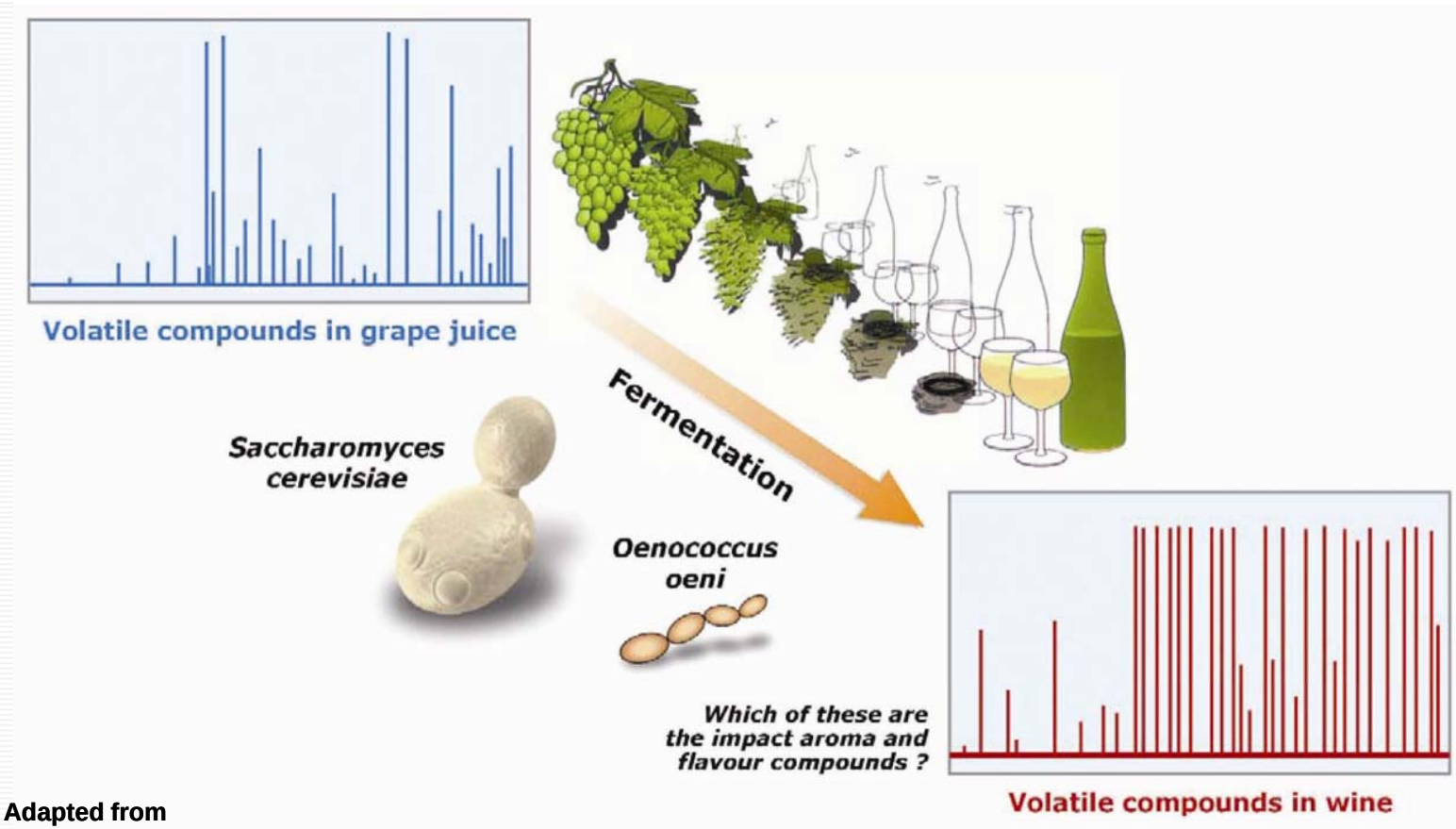
ripeness
phytotechny
sanitation
soil
climate

technology

alcoholic fermentation
malolactic fermentation

maturation
ageing
technology

The origins of wine flavour



Adapted from

Swiegers et al, Australian Journal of Grape and Wine Research, 2005

Objectives

To explore the diversity of natural *Saccharomyces cerevisiae* strains originating from grapes collected in the Vinho Verde Region in order to search for novel yeasts associated with aromatic diversification

- To investigate the predominance of inoculated strains during fermentation of musts obtained from "Trajadura" grapes;
- To determine secondary metabolites and volatile flavor compounds;
- To compare results from analytical determinations with preferences of a sensory panel.

Experimental procedure

Must from Trajadura grape variety, prepared according to Vinho Verde winemaking procedures

47 small-scale (500 ml) fermentations inoculated with 47 indigenous *S. cerevisiae* strains (DB-UM collection)

Fermentation (18 ºC) monitoring by daily must weight determination

Sensorial analysis and selection of the 10 most promising strains

10 medium-scale (50 l) fermentations

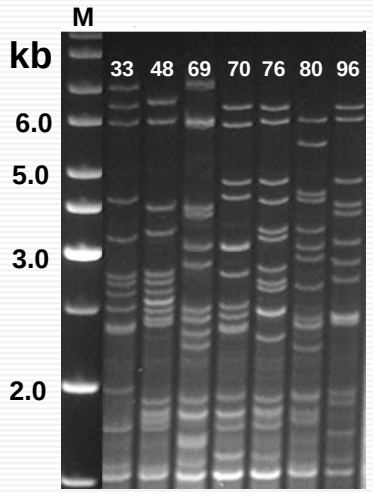
Evaluation of the inoculated strain's predominance during fermentation

Physicochemical analysis
(Secondary metabolites and aromatic profiling)

Sensory panel evaluation

Evaluation of inoculated strain's predominance

- Final fermentation stage (must weight reduction: 60 g/l)
- Plating of diluted must aliquots (YPD medium)
- Incubation (25ºC, 48 h)
- Random selection of 10 colonies from each fermentation
- Strain delimitation by mitochondrial DNA restriction fragment analysis (*Hin*f I)



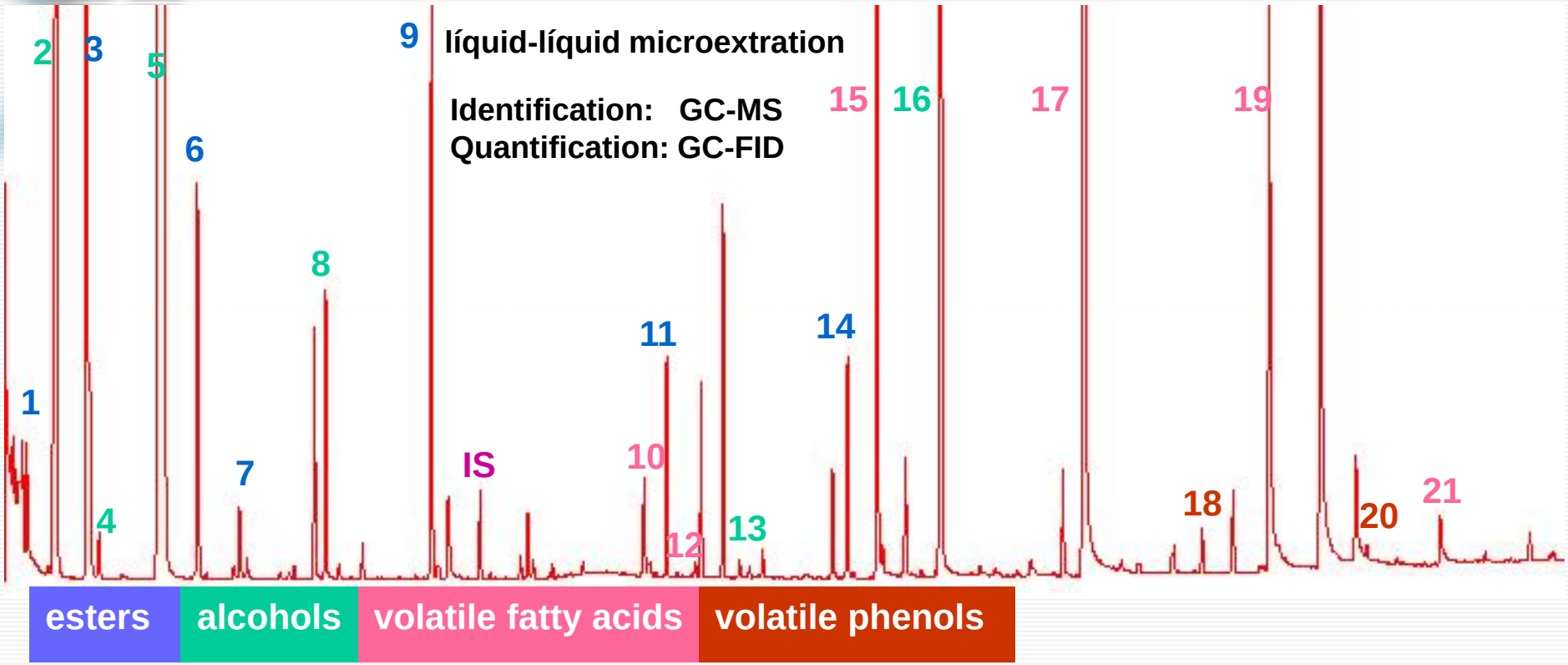
Examples of mtDNA RFLP patterns of inoculated strains

Physicochemical analysis

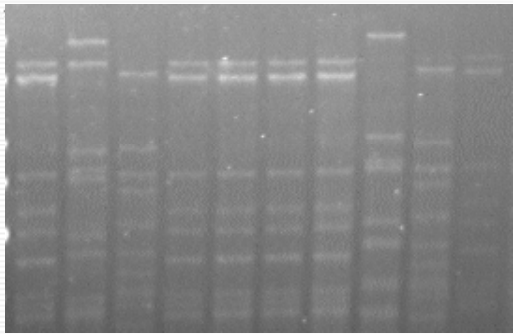
Parameter	*Method*
Ethanol	*Near-infrared spectroscopy; CVRVV - internal method*
Reducing sugars	*CVRVV - internal method 017 (segmented flux analysis)*
Total acidity	*Titulation (Regulation CEE2676/90; annex 13)*
Volatile acidity	*CVRVV - internal method 009 (segmented flux analysis)*
L-malic acid	*FTIR - CVRVV internal method 078*
Tartaric acid	*FTIR - CVRVV internal method 078*
pH	*Potentiometry (Regulation CEE2676/90; annex 24)*
Free SO_2	*Titulation - CVRVV internal method 104*
Total SO_2	*Titulation - CVRVV internal method 104*

CVRVV - Comissão de Viticultura da Região dos Vinhos Verdes

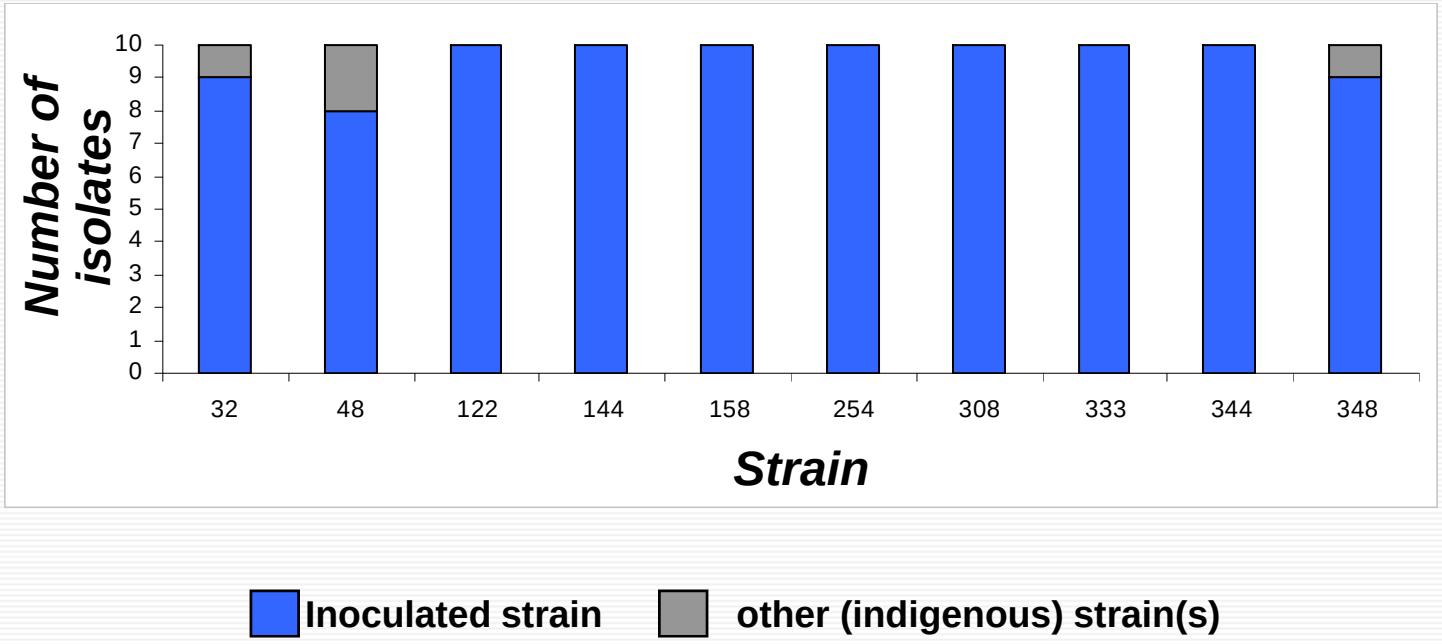
Chemical analysis - aromatic profiling



1 – Ethyl butyrate
2 – 2-Methyl-1-propanol
3 – 3-Methylbutyl acetate
4 – 1-Butanol
5 – 2-Methyl-1-butanol + 3-methyl-1- butanol
6 – Ethyl hexanoate
7 – Hexyl acetate
8 – 1-Hexanol
9 – Ethyl octanoate
10 – Butyric acid
11 – Ethyl decanoate
12 – 3-Methylbutyric acid + 2-Methylbutyric acid
13 – 3-(Methylthio)-1-propanol
14 – 2-Phenylethyl acetate
15 – Hexanoic acid
16 – 2-Phenylethanol
17 – Octanoic acid
18 – 4-Vinylguaiacol
19 – Decanoic acid
20 – 4-Vinylphenol
21 – Dodecanoic acid
IS – 4-Nonanol

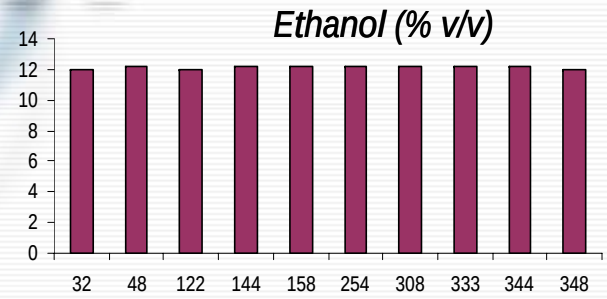


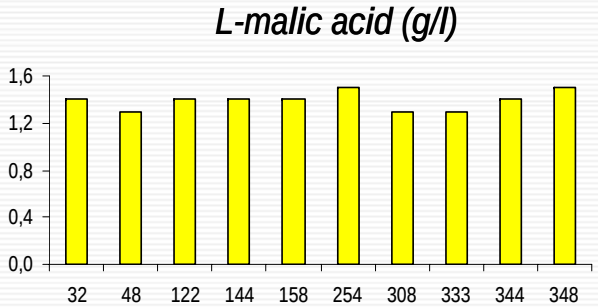
Evaluation of inoculated strain's predominance

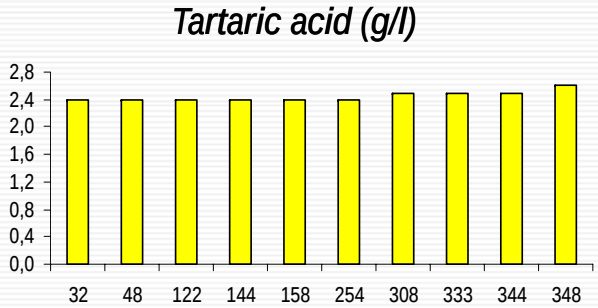


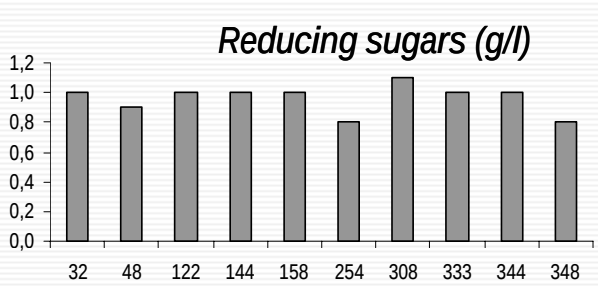
Results

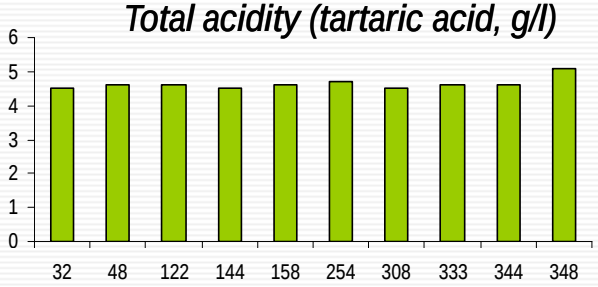
Physicochemical analysis

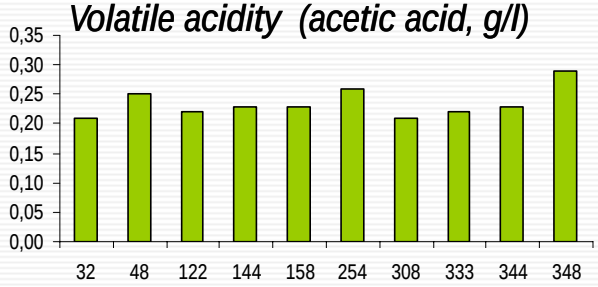


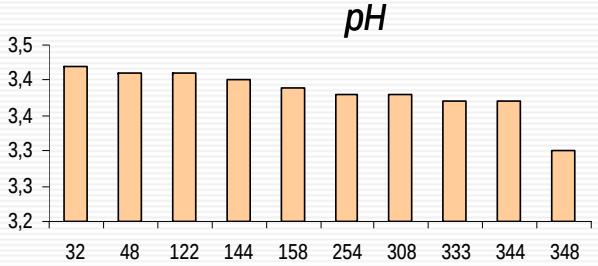


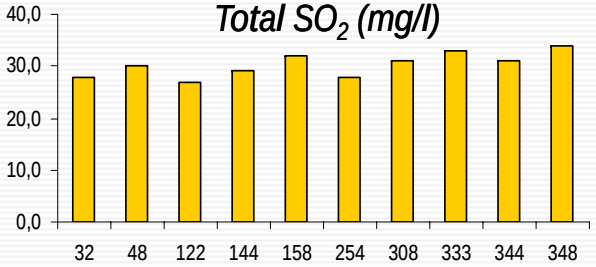


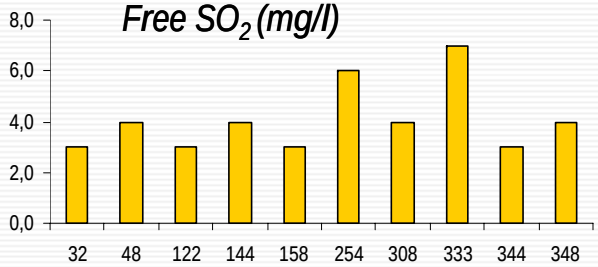






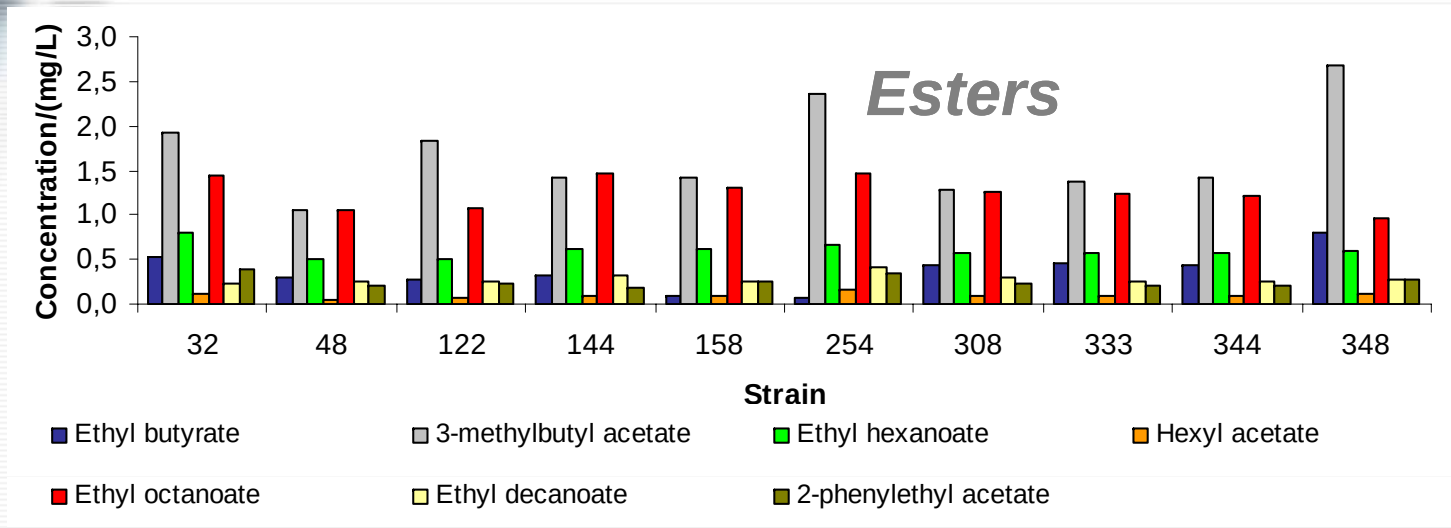


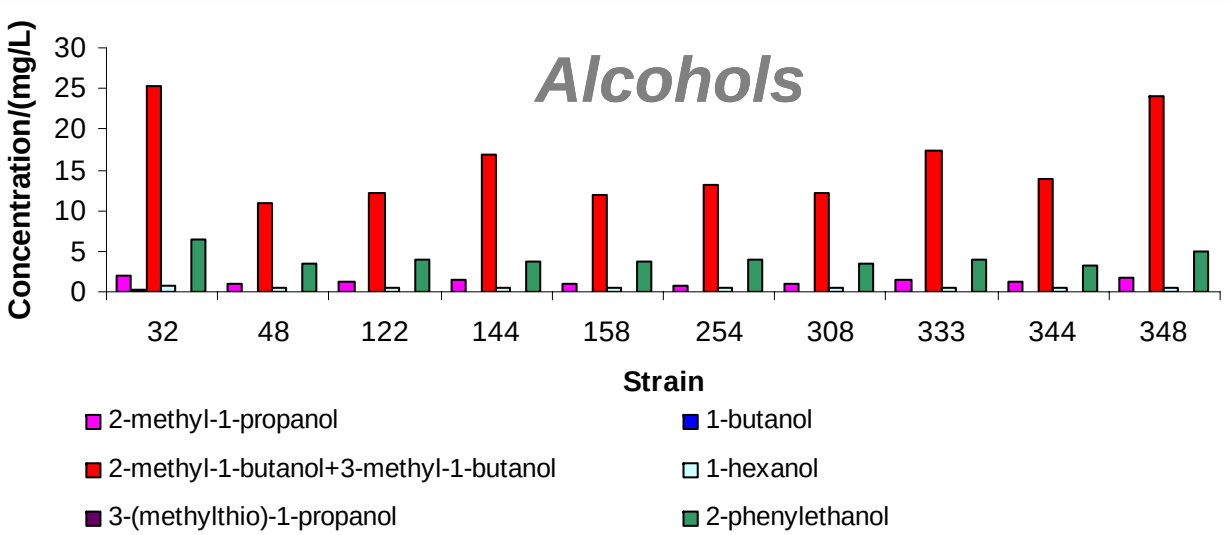




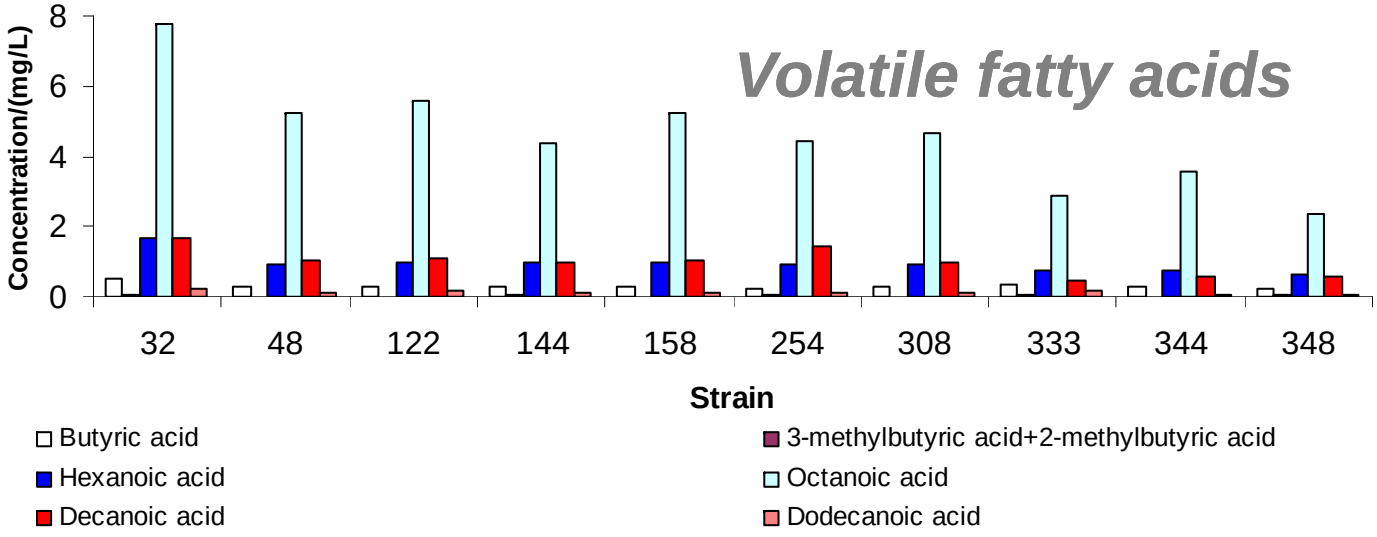
Strain

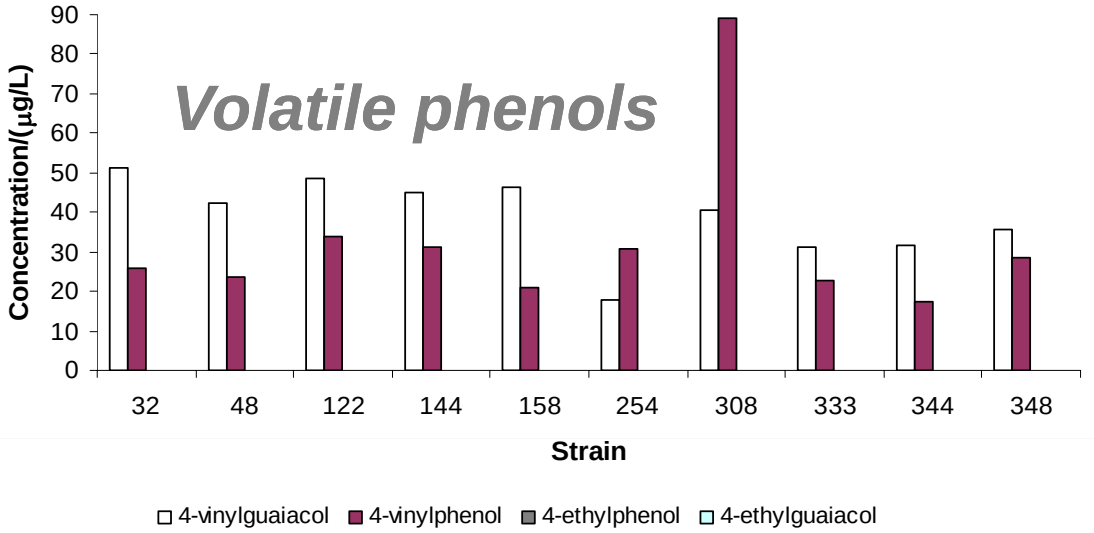
Results


Esters
Concentration/(mg/L)
3,0
2,5
2,0
1,5
1,0
0,5
0,0
32
48
122
144
158
254
308
333
344
348
Strain
Ethyl butyrate
3-methylbutyl acetate
Ethyl hexanoate
Hexyl acetate
Ethyl octanoate
Ethyl decanoate
2-phenylethyl acetate


Alcohols
Concentration/(mg/L)
30
25
20
15
10
5
0
32
48
122
144
158
254
308
333
344
348
Strain
2-methyl-1-propanol
1-butanol
2-methyl-1-butanol+3-methyl-1-butanol
1-hexanol
3-(methylthio)-1-propanol
2-phenylethanol

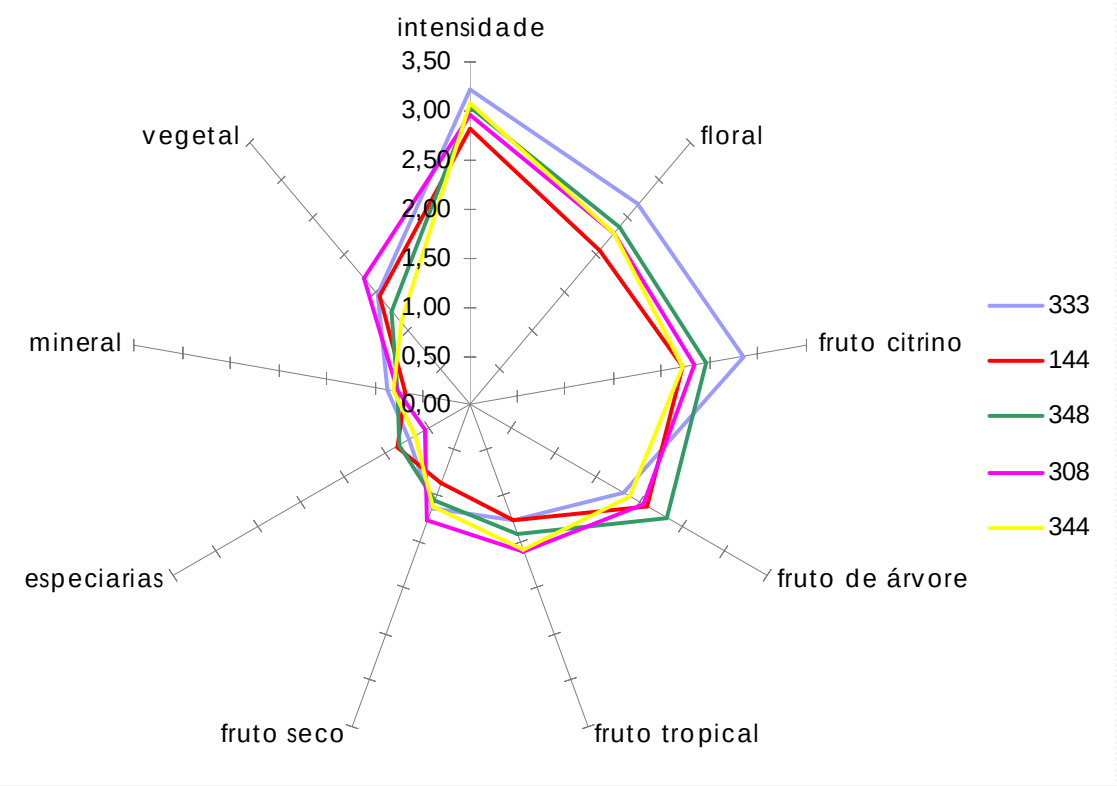
Results


Volatile fatty acids
Concentration/(mg/L)
0
2
4
6
8
32
48
122
144
158
254
308
333
344
348
Strain
Butyric acid
Hexanoic acid
Decanoic acid
3-methylbutyric acid+2-methylbutyric acid
Octanoic acid
Dodecanoic acid


Volatile phenols
Concentration/(µg/L)
0
10
20
30
40
50
60
70
80
90
32
48
122
144
158
254
308
333
344
348
Strain
4-vinylguaiacol
4-vinylphenol
4-ethylphenol
4-ethylguaiacol

Results

Evaluation by a sensory panel (12 members)



Global preference

144 > 333 = 348 > 308 = 344

Conclusions

Natural *Saccharomyces cerevisiae* strains originating from grapes collected in the Vinho Verde Region:

- Succeed to complete fermentation and clearly predominate in inoculated fermentations
- Produce wines with distinctive aromatic profiles;

Although differences in aromatic compounds were rather small, the sensory panel established a preference list, that was not associated with the predominance of specific aromatic compounds, but rather on the global wine appreciation

Acknowledgements

Departamento de Matemática - UM

Cecília Azevedo
Emília Athayde

Vinália
Spin-off da Universidade do Minho

IsabelAraújo

Financially supported by the POCI2010 program

Feder/FCT POCI /AGR/56102/2004