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Effect of juice turbidity on fermentative volatile compounds in white wines

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Summary

'Chardonnay' (n = 4), 'Pinot gris' (n = 3) and 'Müller-Thurgau' juices (n = 3), each at 6 turbidity levels (15, 45, 86, 141, 215 and 350 NTU) obtained by adding increasing amounts of their own fine juice lees, were fermented using 'Montrachet Red Star' yeast. The main volatile compounds in free form which may have a sensory role were measured using GC-FID, with a DB-WAX column, after fixing onto Isolute ENV+ resin. Changes for around 40 volatile compounds and fermentation parameters are shown. Juice turbidity levels just below 100 NTU are the best compromise for obtaining adequate fruity notes and minimising languishing fermentation and off-flavours in white wine, if correct microbiology management at the winery is guaranteed, whereas slightly higher NTU levels could contribute to a slightly more complex aroma. However, variability due to juice turbidity in the range investigated is lower than variability due to yeast strain observed in a previous experiment. Thus the choice of yeast strain to direct white wine aroma must be overriding as compared to NTU levels

Key words: juice, turbidity, wine, aroma compounds, nephelometric units.

Introduction

Since the late Seventies it has been well-known (Ribereau-Gayon *et al.* 1975, Groat and Ough 1978, Bertrand *et al.* 1978, Bertrand 1978, Houtman *et al.* 1980 and extensive literature cited) that adequate transparency in white juice - almost independently of the technological approach used to achieve it and variety - is a fundamental prerequisite for obtaining - freely citing Houtman and du Plessis (1986a) - a fine "basic fermentation bouquet, ... being a major wine quality determining factor in the cellar". According to these last authors, juice clarification must be "effective but not too drastic", in order to avoid detrimental effects both on the course of fermentation, largely related to sterols and fatty acids (Lafon-Lafourcade *et al.* 1977, Cocito and Delfini 1997), and on aroma quality, mainly caused by higher production of volatile acidity when clarity is excessive (Delfini and Cervetti 1991, Delfini and Costa 1993) or volatile sulphur compounds when there is a lack of clarity (Karagiannis and Lanaridis 2002).

In principle, the effects of juice turbidity on white wine quality are known, but the level of aforementioned severity needs to be better defined, as few papers specify nephelometric turbidity unit (NTU) levels, work within turbidity ranges which are genuinely consistent with proper white wine processing or take into account numerically relevant case studies and varieties (Gerbaux and Meurgues 1996, Karagiannis and Lanaridis 2002, Bosso and Guaita 2008). Hence this work aims to provide more detail about the effects of juice turbidity on wine composition, focusing on turbidity levels below 400 NTU and volatile compounds potentially affecting specific aroma characteristics. Moreover, variability caused by juice turbidity is compared with variability due to the yeast strain.

Material and Methods

Four different 'Chardonnay', three 'Pinot gris' and three 'Müller-Thurgau' sulphited juices (80 mg·L^{-1}), each at 6 turbidity levels (T0 = 15 NTU, T1 = 45 NTU, T2 = 86, T3 = 141, T4 = 215, T5 = 350) were fermented in 2-L bottles at 19-21 °C using 'Montrachet Red Star' yeast (500 mg·L^{-1}). Turbidity levels were obtained by adding increasing amounts of their own fine juice lees into statically settled brilliant juice (T0). As regards basic analysis of juices, total soluble solids ranged between 18.5 and 21.9 °Brix, pH 3.06-3.29 and assimilable nitrogen 60-224 mg·L^{-1}. After the completion of fermentation, the wines were decanted, sulphited (70 mg·L^{-1}), analysed for basic composition within a week and stored at 4 °C until analysis of volatile compounds, 3 months later.

Assimilable nitrogen was measured according to Nicolini *et al.* (2004 a, b). Methanol, higher alcohols, acetaldehyde and ethyl acetate were measured using GC-FID with a Carbopack C packed column according to the classic method proposed by Gabri and Salvagiotto (1980) and Usseglio-Tomasset and Matta (1983); vinylphenols were quantified using a HPLC-ECD approach with RP-18e Purospher column (Larcher *et al.* 2007); higher alcohol acetates, fatty acid ethyl esters, fatty acids, methionol, 2-phenylethanol and other aroma compounds in free form were measured using GC-FID, with a DB-WAX column, after fixing onto Isolute ENV+ resin, expressing data as n-eptanol, R.F. = 1 (Boido *et al.* 2003).

ANOVA and Fischer's LSD test (main effects: juice and turbidity level) were applied for data processing, using STATISTICA v. 8.0 (StatSoft Inc., Tusla, OK, USA).

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Results and Discussion

The Table shows the mean values and relevant statistical significance (Fischer's LSD test, $p < 0.05$) of basic compositional parameters and volatile compounds of wines given *per* juice turbidity level. Differences between juices are not shown even if statistically significant, being obvious given the experimental design.

Our attention will focus on analytical parameters with a potential role in terms of sensory characteristics and technology. All turbidity levels were statistically well differentiated. Greater turbidity gave shorter fermentations and lower residual sugars, as expected according to the aforementioned literature and Houtman and du Plessis (1986 b), and this cannot be traced back to assimilable nitrogen, since no statistical differences or trends were found for this parameter between turbidity levels (data not shown). As regards the basic analytical parameters, higher turbidity levels also resulted in lower volatile acidity (with a maximum difference of 0.09 $g \cdot L^{-1}$ between the extreme means) and acetaldehyde (15 $mg \cdot L^{-1}$), and higher glycerol (0.9 $g \cdot L^{-1}$). The differences observed for ethyl acetate (3.2 $mg \cdot L^{-1}$) are technologically negligible.

As regards C_6 chain length alcohols, typical pre-fermentative compounds, we found a statistically significant increase in hexanol and an increasing trend, though not significant, of *trans* 3-hexen-1-ol and the sum of C_6 alcohols with respect to juice turbidity. This figure agrees with the literature and our previous results, being consistent with the contribution of juice lees to content in linoleic and linolenic acids and related enzymes, and with SO_2 treatments (Cordonnier and Bayonove 1977, Di Stefano and Ciolfi 1982, Bayonove *et al.* 1987, Herraiz *et al.* 1990, Gomez *et al.* 1993, Nicolini *et al.* 1996). The statistically significant decrease in *cis* 3-hexen-1-ol could be related to higher isomerases favouring the more stable *trans* form, but this is worth further investigation. Since C_6 aldehydes, characterised by much lower sensory thresholds than the corresponding alcohols (Meilgaard 1975, Hatanaka 1993), and C-2 unsatured alcohols were not measured, it is difficult to forecast the effect of turbidity in terms of intensity of perceptible green, vegetal-like notes.

As regards fermentative compounds, 2-methyl-1-propanol and amyl alcohols increased with turbidity, while propanol showed only marginal variations. The maximum difference for the sum of these higher alcohols was about 60 $mg \cdot L^{-1}$, with a small direct sensory relevance.

The sum of acetates of higher alcohols (acetates), responsible for fruity notes (Romano *et al.* 1987), only differed statistically between the excessively clarified level T0, with the lowest content, and T1, with the highest, whereas a decreasing trend appeared at increasing turbidity levels. This behaviour substantially agrees with that observed by Houtmann *et al.* (1980), though here with a less marked decrease. In any case, variations between T1 and T5 were limited, around 250 $\mu g \cdot L^{-1}$. T1, T2 and T0 in decreasing order of content differed significantly from T3 and T5 for fatty acid ethyl esters (esters). Thus fruity aroma could reach its maximum in correspondence with T1, due to acetates and esters, while the contribution of the former, expressed as the acetates/esters ratio, rises slightly with turbidity from about 1.3 up to 1.5.

Greater turbidity resulted in a clear decrease in hexanoic, octanoic, decanoic and to some extent also butanoic acid, at least from T1 to T5, confirming a previous work of Edwards *et al.* (1990) where NTU levels were not specified. The difference in the sum of quoted fatty acids was about 3 $mg \cdot L^{-1}$, probably not significant in direct sensory terms. On the other hand, isovaleric acid increased significantly with turbidity, with a difference of about 200 $\mu g \cdot L^{-1}$ in the turbidity range tested, which can probably penalise wine quality by masking more pleasant aromas.

Focusing on other major compounds in terms of sensory relevance, vinylphenols, 2-phenylethanol and methionol increased with turbidity. As regards vinylphenols and 2-phenylethanol, their increase with turbidity - about 90 $\mu g \cdot L^{-1}$ and 9 $mg \cdot L^{-1}$, respectively - could contribute to enhancing spicy-floral notes, while a direct methionol-related detrimental effect on wine quality (*e.g.* cabbage notes) seems unlikely given the quantity involved, about 300 $\mu g \cdot L^{-1}$.

Ethyl lactate, as well as γ-butyrolactone, increased significantly with turbidity while ethyl succinate, diethyl succinate and diethyl malate showed the same behaviour with maximum in correspondence with the T3 level, but effects in sensory terms are unimportant.

Spread between turbidity levels (expressed as the maximum average / minimum average ratio) for each analytical parameter was below 1.5, with the exclusion of residual sugars (3.08), methionol (1.85), volatile acidity (1.82) and *cis* 3-hexenol (1.75). The variability of the content of each compound due to juice turbidity is definitely lower than variability due to the yeast strain, as proved by comparing the spread values gathered from a previous experiment (Nicolini *et al.* 2009) in which 10 yeast strains were used for the fermentation of 6 varietal juices. Thus the choice of yeast strain to direct white wine aroma must be overriding as compared to NTU levels.

In conclusion - in the light of the results of this experiment, which took into account different grape varieties and assimilable nitrogen levels - juice turbidity levels just below 100 NTU are probably the best compromise for obtaining adequate fruity, fermentation notes for white wine, when the risk of languishing fermentation is carefully avoided by correct microbiological management at the winery, but slightly higher NTU levels would not really seem to penalise aroma quality, sometimes contributing to a slightly more complex aroma.

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T a b l e

Volatile compound content in wine with respect to the turbidity of the relevant juice.
Spread values (maximum average / minimum average) are also given.
(§ = spread due to yeast strain, re-calculated from Nicolini *et al.* 2009)

Parameter		Turbidity level						Spread	Yeast spread (§)
		mean value (n=10); Sign. Fisher's LSD test ($p<0.05$)							
Juice turbidity level		T0	T1	T2	T3	T4	T5		
	NTU	15 a	45 b	86 c	141 d	215 e	350 f		
Fermentation duration		T5	T4	T3	T2	T1	T0		
	day	15.6 a	16.7 ab	17.6 bc	18.0 bcd	19.3 cd	19.6 d	1.26	
Alcohol		T0	T1	T2	T4	T3	T5		
	% vol	11.78 a	11.98 b	12.05 b	12.05 b	12.06 b	12.07 b	1.02	
Total acidity		T0	T1	T2	T3	T4	T5		
	g l^{-1}	6.34 a	6.36 a	6.45 ab	6.51 b	6.53 b	6.55 b	1.03	
Volatile acidity		T5	T3	T2	T4	T1	T0		
	g l^{-1}	0.11 a	0.12 ab	0.12 ab	0.13 ab	0.15 b	0.20 c	1.82	
Glycerol		T0	T1	T2	T3	T4	T5		
	g l^{-1}	5.73 a	6.13 b	6.35 bc	6.55 c	6.60 c	6.64 c	1.16	
Sugar		T5	T3	T4	T2	T1	T0		
	g l^{-1}	2.4 a	2.7 ab	3.1 ab	3.2 ab	4.7 b	7.4 c	3.08	
Acetaldheyde		T5	T3	T2	T4	T1	T0		
	mg l^{-1}	49.1 a	51.3 ab	52.1 ab	52.4 ab	56.3 b	64.4 c	1.31	2.38
Ethyl acetate		T5	T3	T4	T2	T0	T1		
	mg l^{-1}	26.8 a	26.9 a	27.2 a	28.9 ab	29.1 ab	30.0 b	1.12	2.04
Hexanol		T0	T1	T3	T2	T4	T5		
	µg l^{-1}	846 a	861 ab	889 abc	896 abc	926 bc	936 c	1.11	1.89
trans 3-hexenol		T0	T3	T1	T2	T4	T5		
	µg l^{-1}	89.7 a	94.4 a	94.7 a	99.4 a	99.9 a	100.5 a	1.12	1.14
cis 3-hexenol		T5	T4	T3	T2	T1	T0		
	µg l^{-1}	38.1 a	43.4 ab	49.6 bc	58.4 cd	65.3 d	66.8 d	1.75	1.41
Sum of C6 alcohols		T0	T1	T3	T2	T4	T5		
	µg l^{-1}	1002 a	1021 a	1033 a	1053 a	1069 a	1074 a	1.07	1.78
Propanol		T3	T1	T4	T5	T2	T0		
	mg l^{-1}	17.9 a	18.1 a	18.1 a	18.1 a	18.2 a	20.3 b	1.13	9.36
2-methyl-1-propanol		T0	T1	T2	T4	T3	T5		
	mg l^{-1}	28.7 a	30.6 ab	31.2 b	32.4 bc	33.2 c	35.2 d	1.23	2.04
2-methyl-1-butanol		T0	T1	T2	T3	T4	T5		
	mg l^{-1}	38.1 a	40.5 ab	41.2 bc	43.9 cd	45.8 de	48.4 e	1.27	1.47
3-methyl-1-butanol		T0	T1	T2	T4	T3	T5		
	mg l^{-1}	154 a	164 b	171 b	181 c	183 c	199 d	1.29	1.32
Sum of higher alcohols		T0	T1	T2	T4	T3	T5		
	mg l^{-1}	241 a	253 ab	262 b	277 c	278 c	300 d	1.24	1.94
Isobuthyl acetate		T1	T0	T3	T4	T5	T2		
	µg l^{-1}	14.2 a	15.0 a	15.5 a	15.7 a	15.9 a	16.2 a	1.14	5.82
Isoamyl acetate		T0	T5	T4	T3	T2	T1		
	µg l^{-1}	2219 a	2570 ab	2614 ab	2638 ab	2651 ab	2724 b	1.23	5.46
Hexyl acetate		T5	T4	T3	T2	T0	T1		
	µg l^{-1}	147 a	155 ab	178 bc	194 cd	209 d	209 d	1.42	1.75
2-phenylethyl acetate		T0	T5	T2	T1	T3	T4		
	µg l^{-1}	458 a	518 ab	531 ab	547 b	551 b	555 b	1.21	6.79
Sum of acetates		T0	T5	T4	T3	T2	T1		
	µg l^{-1}	2900 a	3250 ab	3339 ab	3382 ab	3392 ab	3494 b	1.20	5.04
Ethyl butanoate		T0	T5	T4	T3	T2	T1		
	µg l^{-1}	123 a	125 ab	129 ab	134 ab	143 b	144 b	1.17	1.62
Ethyl hexanoate		T3	T0	T4	T5	T2	T1		
	µg l^{-1}	691 a	718 ab	747 ab	751 ab	804 b	826 b	1.20	1.66
Ethyl octanoate		T5	T4	T3	T2	T0	T1		
	µg l^{-1}	921 a	989 ab	1007 ab	1070 bc	1118 c	1139 c	1.24	1.93
Ethyl decanoate		T5	T4	T3	T2	T1	T0		
	µg l^{-1}	253 a	263 a	281 ab	318 bc	355 cd	374 d	1.48	1.79
Sum of ethyl esters		T5	T3	T4	T0	T2	T1		
	µg l^{-1}	2051 a	2113 a	2128 ab	2333 bc	2336 bc	2463 c	1.20	1.80
Isobutanoic acid		T0	T1	T5	T2	T4	T3		
	µg l^{-1}	815 a	881 ab	903 b	930 b	931 b	931 b	1.14	2.96
Butanoic acid		T5	T0	T3	T4	T2	T1		
	µg l^{-1}	651 a	676 ab	688 ab	701 ab	722 b	734 b	1.13	1.42

Table, continued

Parameter		Turbidity level						Spread	Yeast spread (§)
		mean value (n=10); Sign. Fisher‘s LSD test ($p<0.05$)							
Isovaleric acid		T0	T1	T2	T3	T4	T5		
	µg l^{-1}	771 a	832 ab	845 bc	907 cd	933 d	945 d	1.23	2.39
Hexanoic acid		T5	T4	T3	T2	T1	T0		
	µg l^{-1}	3328 a	3593 ab	3613 ab	3798 bc	4117 c	4125 c	1.24	1.61
Octanoic acid		T5	T4	T3	T2	T1	T0		
	µg l^{-1}	5306 a	5836 ab	6123 bc	6326 bc	6691 c	6815 c	1.28	1.97
Decanoic acid		T5	T4	T3	T2	T1	T0		
	µg l^{-1}	1453 a	1524 a	1604 a	1703 ab	1888 bc	1986 c	1.37	1.72
4-vinylphenol		T0	T3	T1	T2	T5	T4		
	µg l^{-1}	195 a	201 a	221 ab	226 ab	250 b	253 b	1.30	50.8
4-vinylguaiacol		T0	T1	T2	T3	T4	T5		
	µg l^{-1}	68 a	85 b	88 b	90 b	93 b	100 b	1.47	23.1
Sum of vinylphenols		T0	T3	T1	T2	T4	T5		
	µg l^{-1}	263 a	291 ab	306 ab	315 ab	347 b	351 b	1.33	33.7
Methionol		T0	T1	T2	T3	T4	T5		
	µg l^{-1}	349 a	425 b	489 b	576 c	580 cd	647 d	1.85	9.56
2-phenylethanol		T0	T1	T2	T5	T4	T3		
	mg l^{-1}	31.89 a	35.37 b	37.12 b	39.80 c	40.03 c	40.32 c	1.26	2.14
Ethyl lactate		T0	T1	T2	T3	T4	T5		
	µg l^{-1}	489 a	507 a	559 b	592 bc	602 bc	625 c	1.28	1.84
Ethyl succinate		T0	T1	T2	T5	T4	T3		
	µg l^{-1}	5646 a	6134 ab	6340 bc	6652 cd	6699 cd	6936 d	1.23	2.81
Diethyl malate		T0	T2	T1	T5	T4	T3		
	µg l^{-1}	38.2 a	46.5 ab	47.4 b	47.7 b	47.7 b	49.1 b	1.29	1.46
Diethyl succinate		T0	T1	T2	T5	T4	T3		
	µg l^{-1}	84.5 a	95.4 b	95.9 b	96.2 b	97.2 b	99.8 b	1.18	2.57
γ-butyrolactone		T0	T1	T2	T3	T4	T5		
	µg l^{-1}	204 a	215 ab	245 bc	263 cd	280 d	287 d	1.41	4.08

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