

Influence of must racking and fining procedures on the composition of white wine

by

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S u m m a r y : A study of the effects of two kinds of must clarification, settling or filtration by rotary vacuum filter and 5 juice fining treatments (bentonite, potassium caseinate, and combinations: bentonite + gelatin, and bentonite + caseinate + microcrystalline cellulose) on wine characteristics was carried out using must of the white cultivar Parellada. Although the general characteristics (ethanol, color, limpidity, residual sugars, pH and acidity) of Parellada wines were not modified, some wine components (e.g. nitrogenous substances, phenolic compounds and aroma) were lowered according to the type of must racking and juice fining agent added. The percentage of decrease of wine characteristics due to the pre-fermentative fining agent depends on the type of must racking. Highest losses were recorded when rotary vacuum filters were used for must racking, and when bentonite (0.3-0.5 g/l) was added to the juice as a fining agent before fermentation.

K e y w o r d s : white wine, Parellada variety, must clarification, racking, fining agents, bentonite, caseinate.

Introduction

In white wine-making, several techniques are used to remove dregs and sediments from juices and wines, since suspended solids may produce physical or chemical effects that influence certain organoleptic properties (WILLIAMS *et al.* 1978; FOSTER and DEAN COX 1984; DUBOURDIEU *et al.* 1986, TIENDA and HIDALGO 1990; SIMS *et al.* 1995).

The most common technique used to remove must solids, skin particles, wild yeast, oxidizing enzymes and colloids from juice is settling followed by racking (CASTINO *et al.* 1980; COLAGRANDE *et al.* 1986; HABA 1990; DÍAZ 1991; ALEIXANDRE and VELEZ 1992). This practice has several effects, which mainly depend on must composition and racking conditions, but it is accepted that juice clarification is an essential step in the production of quality white wines with the appropriate organoleptic characteristics (FOSTER and DEAN COX 1984; CASP and LÓPEZ 1986; CORDONNIER *et al.* 1988; ARTAJONA and BERTRAND 1990).

The most traditional and widely used kind of juice clarification is settling: for 12-24 h, with or without the addition of fining agents, at controlled temperature, and with the aid of sulphur dioxide. This is followed by racking to separate the juice from the solids.

Dynamic systems to clarify juice, such as centrifugation, flotation and filtration (FOSTER and DEAN COX 1984; DUBOURDIEU *et al.* 1986; CORDONNIER *et al.* 1988; TIENDA and HIDALGO 1990; PAETZOLD 1990; TROUSSEAU and CHAPRON 1991; BARDINI and MAGGI 1992) have become prevalent in the last few years since discontinuous racking, following settling, is a slow and inefficient system when applied in an industrial winery. Rotating vacuum filtration is the most common system of continuous clarification as it is rapid and efficient (GUY 1990). However, such filters are expen-

sive and some desirable compounds may be removed from the must (DE CASTRO 1986).

Fining is a technique that forms flocculated precipitates between some slightly soluble compounds (colloids) and the fining agents. The fining agents usually have an opposite charge to the colloids, so that they join the colloids and then sediment; as they fall, other particles are dragged down (GORINSTEIN *et al.* 1984; HSU and HEATHER-BELL 1987; BLADE and BOULTON 1988; CANTARELLI *et al.* 1989; ALEIXANDRE and VELEZ 1992). When fining agents take part in the fermentation process, they act as insoluble solids that promote yeast growth, and thus fermentation finishes faster and is more complete (GROAT and OUGH 1978; SIMS *et al.* 1995). However, some authors (VOILLEY *et al.* 1990; LUBBERS *et al.* 1993) describe interactions between aroma compounds and macromolecules, such as fining agents; these interactions may modify the volatility of aroma compounds by adsorption on the suspended solids, and, thus, change the organoleptic properties of the wine (MAIN and MORRIS 1994).

Although Parellada grapes have a neutral aroma (LÓPEZ-TAMAMES *et al.* unpubl.), their white wines from the Penedès AOC region have fruity characteristics and are used in the production of Spanish sparkling wine (DE LA PRESA-OWENS *et al.* 1995).

In our study, in which wines were produced for sparkling wine, the effect of two clarification methods combined with 4 different fining agents was studied. Thus, a 2x5 factorial statistical design with two replicates was performed. The 4 fining agents are those usually used in white wine-making of the Penedès AOC region: potassium caseinate, bentonite and two fining agent combinations: bentonite + gelatin and bentonite + potassium caseinate + microcrystalline cellulose (commercial preparation Microcel®).

Materials and methods

Fining agents: Potassium caseinate solution: 50 g/l of potassium caseinate (Alpha caseinate®, Chemiciperdomini SpA, Italy) in distilled water.

Bentonite suspension: 62.5 g/l of sodium bentonite ASB-60-S (ECC International, France) in distilled water.

Bentonite and gelatin suspension: 37.5 g/l of sodium bentonite ASB-60-S and 12.5 g/l of gelatin (Gelsol®, AEB Ibérica S.A., Spain) in distilled water.

Bentonite (45% w/w), potassium caseinate (50% w/w) and microcrystalline cellulose (5% w/w) (Microcel®, AEB Ibérica S.A., Spain) suspension: 50 g/l of Microcel® in distilled water.

Analytical methods: Conventional oenological parameters such as total and volatile acidities, °Brix, pH, total sulphur dioxide, ethanol % v/v and sugars were measured according to Office International de la Vigne et du Vin (OIV) methods (1990).

Color: absorbances at 420 and 520 nm were determined with a spectrophotometer HP-8452A.

Nitrogenous substances: Total soluble proteins were determined according to BRADFORD (1976), free amino acids and ethanolamine were determined by HPLC (PUIG-DEU and BUXADERAS 1994).

Polyphenol fraction: Total phenols were determined according to SINGLETON and ROSSI (1965), flavonoids and nonflavonoids according to KRAMLING and SINGLETON (1969), while catechin, epicatechin, procatechuic, caffeic, ferulic and coumaric acids were determined by HPLC (Merck-Hitachi HPLC system, L-5000 LC Controller, 655A-12 Liquid Chromatograph, L-4200 UV-VIS detector). A Nucleosil 120 C18 column, 250 x 4 mm, 5 mm particle size was used as stationary phase. Acetonitrile and water acidified (pH 2.5) with acetic acid were used as mobile phase; the flow rate was 1.5 ml/min. Detection was performed at 280 nm.

Volatile compounds: Ethyl propionate, ethyl butyrate, isoamyl acetate, hexyl acetate, isoamyl alcohols, ethyl hexanoate, hexyl alcohol, ethyl lactate, ethyl octanoate, linalool, ethyl decanoate, diethyl succinate, α -terpineol, nerol, 2-phenethyl acetate, ethyl dodecanoate, geraniol, benzyl alcohol, 2-phenethylethanol and ethyl tetradecanoate were isolated and quantified by discontinuous liquid-liquid extraction with dichloromethane. A Hewlett-Packard 5890 series II gas chromatograph equipped with a FID detector and a Supelcowax 10 (30 m x 0.25 mm i.d.) column were used.

Samples: Starting with the same juice from Parellada grapes, 20 winemaking processes were performed in parallel (2x5 factorial design with two replicates). The grape juice was obtained industrially with a Willmes® pneumatic press (10,000 kg capacity, pressure below 0.7 kg/cm²) and sulphur dioxide (ca. 70 mg/l) was added. One portion of the juice was racked by settling for 24 h, and the other part was passed through a rotary vacuum filter (filtrant coat formed with diatomaceous earth and perlites 50%). After settling or filtration, 20 winemaking processes were carried out in containers (25 l capacity).

Ten samples were taken from settled juice: two without fining agent (control) and 8 to which the 4 fining agents were added in duplicate (at 15 \pm 2 °C). Two controls and 8 treated juice samples were prepared in the same way from filtered juice. 200 ml of fining agent suspensions were added to the juice samples at the following concentrations (g/l): 0.4 of potassium caseinate (a), 0.5 of bentonite (b), 0.3:0.1 of bentonite:gelatin (c) and 0.4 of Microcel® (d). The fining agents were added together with *Saccharomyces cerevisiae* (strain P-154 selected by the winery Freixenet S.A.) (at 10⁶ cells/l). The 20 containers were left for 12 d in a cold-storage room at 14 °C. Density was measured as fermentation control; the wines were racked simultaneously when they had density of less than 1,000 (after 12 d of fermentation). Finally, the 20 wines were passed through a candle filter (diatomaceous earth and perlites 50%). Conventional wine composition parameters [mean and 95% confidence intervals for means (n=20)] were: ethanol [8.9% v/v (8.7–9.1)], sulphur dioxide [71.4 mg/l (65.1–77.7)], residual sugars [0.81 g/l (0.79–0.83)], volatile acidity [0.22 g acetic acid/l (0.20–0.25)], titratable acidity [5.47 g tartaric acid/l (5.40–5.55)], and pH [2.95 (2.93–2.97)].

Statistical procedures: The Statgraphics 4.2 program was used to perform a Multiple Analysis of Variance (MANOVA) to study the effects of racking and fining of juice on the white wine composition. The clarification (settling and filtration) and the type of fining (control, and a, b, c and d fining agent additions) were considered as qualitative variables. To compare the effects of the fining agents (a, b, c and d), ANOVA analysis was also used.

Results and discussion

The ratios of fermentation (density versus time) differ according to the racking type: filtered juices fitted a reciprocal model ($1/y = a+bx$), while settled juices fitted a multiplicative model ($y = ax^b$). Settled juices began fermentation later than filtered juices although oenological parameters were not different (16 °Brix; titratable acidity: 5.88 g tartaric acid/l and pH: 3.20).

MANOVA analysis (Tab. 1) shows that both variables, racking and fining, influenced many of the compounds studied. Moreover, the two variables frequently presented significant interactions. A probable explanation is that the fining agent effect depends on the extent of previous clarification of the juice. Tab. 2 demonstrates that the control wines derived from filtered musts are less rich in most compounds compared to the control wines from settled juice. Tab. 3 shows the decrease (%) from control wines to those from fined (a, b, c and d) juices. In the wines obtained from filtered juices, the decreases were usually lower than in those obtained from settled juices.

Must filtration produced wines with a lower content of phenols compared to those obtained from settled juices (Tab. 2). Flavonoids, which have a high capacity of oxidation and polymerization (SINGLETON 1987), were less in-

Table 1

Effects of the must racking type (settling and filtration), the must fining (control, potassium caseinate, bentonite, bentonite + gelatin, and Microcel®) and their interaction. MANOVA results expressed in terms of level of significance (p)

Parameter	Racking effect (a)	Fining agent effect (b)	Interaction (ab)
General parameters	N.S.*	N.S.	N.S.
Absorbance 420 nm	N.S.	N.S.	N.S.
Absorbance 520 nm	N.S.	N.S.	N.S.
Total polyphenols	0.001	N.S.	N.S.
Flavonoid phenols	0.007	0.001	0.001
Non flavonoid phenols	0.001	N.S.	N.S.
Protocatechuic acid	0.001	0.001	0.001
Caffeic acid	0.003	0.001	0.001
Ferulic acid	0.001	0.001	0.001
Coumaric acid	0.001	0.001	0.001
Catechin	N.S.	0.001	0.001
Epicatechin	0.001	0.001	0.001
Proteins	0.001	0.001	0.001
Ethanolamine	0.001	0.001	0.001
Proline	0.001	0.001	0.001
Phenylalantine	0.004	0.001	0.001
Glutamine	0.001	0.001	0.001
Histidine	0.001	0.01	N.S.
Arginine	0.02	0.001	N.S.
Leucine	0.001	0.001	0.001
Lysine	0.002	0.001	0.001
Asparagine	0.05	0.001	0.05
Alanine	0.001	0.02	0.001
Valine	0.04	0.001	0.001
Isoleucine	0.02	N.S.	0.05
Ethyl hexanoate	0.001	0.002	N.S.
Ethyl octanoate	0.001	0.01	N.S.
Ethyl decanoate	0.001	N.S.	N.S.
Isoamyl acetate	0.001	0.005	0.03
Hexyl acetate	0.001	0.02	0.03
2-phenethyl acetate	0.001	0.007	0.02
Isoamyl alcohols	N.S.	0.002	N.S.
Hexyl alcohol	N.S.	0.007	N.S.
2-phenethyl ethanol	0.001	0.001	0.001
Linalool	0.001	0.001	0.001
α-terpineol	0.001	0.001	N.S.
Nerol	N.S.	0.05	N.S.

* N.S.: not significant

fluenced by the racking type than other polyphenols. However, the absorbance at 420 nm, an indicator of color of white wines, was not significantly affected by the must racking type. Phenolic acids (protocatechuic, caffeic, ferulic and coumaric acids) and epicatechin were present at lower concentration in the white wines from filtered juice than those from settled juice (Tab. 2). Juice fining usually produced wines with lower polyphenolic content than controls. The greater effect was observed on the flavonoids

Table 2

Effects of settling and filtration on the composition of control wines (n=2)

Compound (mg/l)	Settling	Filtration
Total polyphenols	172.0	150.5
Non flavonoid phenols	127.8	110.7
Flavonoid phenols	44.2	39.8
Color (420 nm x 1000)	90	90
Phenolic acids	9.25	6.05
Epicatechin	3.29	1.63
Proteins	9.13	5.41
Amino acids (except Pro)	186.7	153.9
Proline	417.8	294.6
Ethanolamine	16.81	12.61
Ethyl esters	3.14	1.28
Acetates	0.98	0.69
Alcohols	28.60	27.91
2-phenethyl ethanol	6.00	4.24
Terpenes	0.78	0.29

(Tab. 1). In the wines obtained from settled must (which contains more flavonoid polyphenols, Tab. 2) the main difference was due to the use of potassium caseinate, alone or associated with other fining agents: wines from juices fined with a and d showed 20 and 23 % decrease, respectively, with respect to their control; whereas in the wines from juices fined with b and c (not clarified with potassium caseinate) the relative decreases were 6 and 4%, respectively. Protein fining agents (a, c or d) had an effect on epicatechin and phenolic acids (Tab. 3). The results for flavonoids coincide with those given by other authors (AMATI 1986; MANFREDINI 1989), indicating that the protein fining agents have a greater effect on flavonoids than on other polyphenols. However, when the juice phenolic content was low due to filtration (Tab. 2) this trend was not observed (Tab. 3).

Must racking and juice fining affected the concentration of nitrogenous substances (proteins, free amino acids and ethanolamine) (Tab. 1). Protein, free amino acids (Pro, Gln, His, Arg, Leu, Lys, Asn, Ala, Val, Phe and Ile) and ethanolamine contents were lower in wines obtained from filtered must (Tab. 2 and Figure) compared to those from settled must. The protein loss (Figure), due to fining before fermentation, was observed both in the wines obtained from settled (which contained more proteins) and in the wines from filtered juices. The bentonite concentration appeared to influence the protein loss: juices fined with 0.5 g/l of bentonite had approximately 45 % less protein than controls, wines with 0.3 g/l of bentonite between 21 and 32 %, and wines with 0.18 g bentonite/l showed no protein loss (Tab. 3, Figure). Free amino acid concentration (expressed as summation) decreased in wines derived from fined juice (Tab. 3). This depends on the must racking type: when the juice had higher amino acid concentrations (must racking by settling), losses were greater. Basic free amino acids of wines showed a greater decrease when

Table 3

Relative losses (%) of wine components due to fining agents added to the juice; a = potassium caseinate, b = bentonite, c = bentonite + gelatin, and d = Microcel[®]

Compound	Racking by settling				Racking by filtration			
	a	b	c	d	a	b	c	d
Total polyphenols	-9	-5	0	-3	0	-9	-4	0
Flavonoid phenols	-20	-6	-4	-23	-8	-14	-25	0
Non flavonoid phenols	-6	-4	0	0	0	-7	0	0
Epicatechin	-24	-3	-17	-21	0	0	0	0
Σ phenolic acids	-7	0	-8	-21	0	0	0	0
Proteins	-5	-47	-32	0	0	-44	-21	0
Σ free AA (without Pro)	-41	-41	-39	-34	-7	-16	-24	-16
Σ basic AA	-40	-51	-35	-35	-27	-38	-28	-28
Tyrosine	-41	0	0	-21	0	0	0	-5
Proline	-5	-55	-19	-38	0	0	-4	0
Ethanolamine	-15	-53	-20	-42	-6	0	0	0
Σ ethyl esters	0	-46	-45	-19	-13	-49	-47	-17
Σ acetates	-22	-47	-57	-21	0	-25	-38	-9
Σ alcohols	0	-47	-45	-18	0	-32	-28	-14
Σ terpenes	0	-42	-37	-20	0	-20	-18	0

bentonite was used (0.5 g/l). Bentonite has a negatively charged surface and adsorbs positively charged compounds such as the basic amino acids (MAIN and MORRIS 1994). In wines from settled must (Tab. 3), tyrosine, with a phenol group, was at lower concentration when potassium caseinate was added.

Wines obtained from settled musts had higher concentrations of volatile compounds than wines from filtered must (Tab. 2). Concentrations of ethyl esters (propionate, butanoate, hexanoate, octanoate, decanoate, dodecanoate and tetradecanoate), fermentative products typical for young and fruity wines (VERNIN 1986), and acetates (2-phenylethyl and isoamyl) were higher in wines whose must was settled. Concentrations of terpenes producing the varietal aroma and being responsible for floral notes (VERNIN 1986), and 2-phenethyl ethanol were also lower in wines whose juice was filtered (Tab. 2). The lower content of esters and terpenes in the wines from filtered must, compared to wines from settled must, could be due to losses

to retention in the filter or to oxidation of juice substances, e.g. terpene glycosides, amino acids, enzymes) that contribute to the formation of wine aroma. Wines from juices fined with bentonite (b and c) had lower concentrations of volatile compounds (Tab. 2). Wines with a volatile fraction similar to controls were obtained with the use of potassium caseinate (a). The fruity characteristics of the wines treated with caseinate were similar to those previously described by AMATI (1986) and GIACOMINI (1987).

Conclusions

Juice filtration was a more effective clarification system than settling. Wines from fined juices were poorer in most compounds than wines whose juice had not been treated. The losses due to the fining treatment observed in wines from settled musts were usually greater than the losses in wines from filtered musts. For wines from settled musts, the choice of the fining agent used before fermentation depended on the white wine type: potassium caseinate had less influence on aroma compounds and removed more phenolic compounds than other fining agents; bentonite removed more distinctly nitrogenous compounds and volatile substances (bentonite as juice fining agent at concentrations of 0.3-0.5 g/l is not recommended); protein fining agents or combinations such as Microcel[®], containing lower bentonite concentration are preferable.

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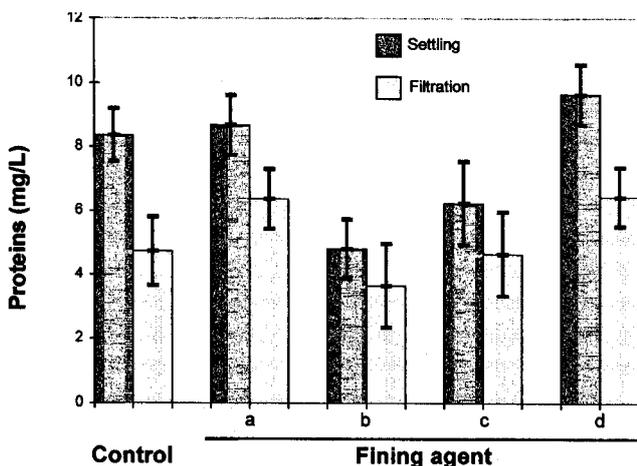


Figure: Effects of different fining agents on proteins (mg/l); a = potassium caseinate, b = bentonite, c = bentonite + gelatin, and d = Microcel[®].

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