*Scientific report*

# Observations concerning the increase of volatile acidity in red wines whilst ageing in barrels

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Summary : The increase in volatile acidity in red wines whilst being aged in barrels still poses problems and has led to numerous investigations. The authors focus on the two principle origins of acetic acid : firstly, the origin of the acidity may stem from oak wood which when burnt may increase acidity level, secondly due to the imputably essential microbiologic acetic acid bacteria. If the supply within the wood is limited (< 0.2 g/l $H_2SO_4$), the bacteria evoke a less noticeable increase (> 0.2-0.4 g/l $H_2SO_4$). Acetic acid bacteria only require a small amount of oxygen in order to develop and produce acetic acid. Meanwhile one can minimise microbiologically formed acetic acid through the rigorous control of the temperature of the storage area (12 to 16°C). In the case of used barrels having been left empty for some months, a two day cleansing with sulphur water will assure the elimination of the accumulated acetic acid in the surface layers of the wood.

*Key words : Ageing, red wines, barrels, volatile acidity, oxygen, lactic acid bacteria, acetic acid bacteria*

## INTRODUCTION

Ageing in barrels constitutes one of the major steps in the life cycle of red wines. Besides their classic properties of degasification, spontaneous clarification and the stabilisation of phenolic compounds and colour (PONTALLIER, 1981 ; VIVAS and *al.*, 1991), the workings of wine in barrels allow profound modifications of its chromatic, aromatic

and tasty characteristics (GLORIES, 1987 ; BOIDRON and *al.*, 1988 ; PEYRON and *al.*, 1994). These modifications contribute to the wines ability to age in bottles.

During ageing in barrels the wine receives small quantities of oxygen, increasing the potential of oxidoreduction ; this phenomenon is known as « low oxidation condition » (VIVAS and GLORIES, 1993). These particular oxidation conditions are at the origin of evolution of the coloured matter and the structure of the tannins in the wine. The penetration of oxygen in the wine is made possible by the porosity of the wood (FEUILLAT and *al.*, 1994) as well as the diffusion of air between the staves and the bunghole. It is enhanced by the creation of a depression of 100 to 120 mBar inside the barrel (MOUTOUNET and *al.*, 1994). However the ageing of wine in barrels is not without risk. Firstly the porous mass of the wood, particularly the surface 3 to 5 mm of the inner barrel, and which is impregnated with 4 to 6 l of wine (RIBÉREAU-GAYON and *al.*, 1976), is more or less loaded with contaminating yeasts and bacteria when coming in contact with the wine. There after the oxygen passing through the wood wall allows the environment to be microaerobic thus facilitating the oxidation phenomenon, reinforced by repetitive racking and topping and aerobic microorganisms (VIVAS and GLORIES, 1993). Amongst these phenomenons the most visible is the increase of volatile acidity. The measuring of the volatile acidity of wines is a classic measure, and is repeated regularly during the vinification and ageing of the wines (RIBÉREAU-GAYON and *al.*, 1976). The development of the volatile acidity is an important phenomenon even if some writers consider it minor (CHATONNET and *al.*, 1994). The greater majority of producers who age their wine in barrels, consider it a priority matter.

In this article we present the primary resulting risks associated with the increase of volatile acidity during the ageing of red wine in barrels. The work focuses on two fundamental principles : the supply of acetic acid by the oak wood and the production of volatile acidity by acetic acid bacteria, already mentioned by other authors (JOYEUX and *al.*, 1984, MARSAL, 1992).

## MATERIALS AND METHODS

### 1 — EXPERIMENTAL CONDITIONS

Under laboratory conditions, the studies were performed on bottles containing 100 ml of Merlot which was at the stage malolactic fermenta-



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tion, clarified by centrifugation (3000 tpm, 10 min) and sterilised by filtration (0.45 µm). The wine was either used directly, or after treatment with active charcoal, filtration through filter without ash and sterile filtration. The experiments in barrels were performed on homogeneous lots all being repeated five times. The experiments correlation was amplified by random samples being drawn from different cellars. Each experiment was reperformed over a period of three years.

## II — MICROBIOLOGIC ANALYSES

### 1) Acetic acid bacteria numeration

The numeration of acetic acid bacteria is realised using nutritious agar-agar. The nutritive medium being composed of half diluted raisin must, supplemented with 5 g/l of yeast extract and brought to a pH of 4.5. The nutritional must was added to an equal proportion of agar-agar DIFCO 50 g/l. The yeasts growth is inhibited by the addition of pimaricine (0.2 ml of solution of 5 mg/ml per dish of 10 ml) and the lactic acid bacteria by addition of penicillin (0.1 ml of solution to 2.5 mg/ml). The Pétri dishes were incubated for 5 to 7 days at 25°C under aerobic conditions. The nutritive mediums are sterilised for 10 min at 120°C.

### 2) Withdrawal of samples from barrels

The various samples were withdrawn through the bunghole once the barrels were open. The 5 ml samples of wine were drawn aseptically and collected in sterile test tubes. The given population values represent the average count results of 6 to7 agar-agar cultures.

## III — GENERAL ANALYSIS METHODS

1) The measure of alcohol, $SO_2$, volatile acidity and reducing sugars totals were in accordance with the protocol recommendations of the O.I.V.

### 2) Ethanal and acetic acid quantification

We use the enzymatic measuring kits (BOEHRINGER, Mannheim).

### 3) Measure of oxidoreduction potential and mix of the underlying oxygen

The oxidoreduction potential is measured using an electrode potentiometer (METTLER, Pessac), adapted for use on wine (VIVAS and *al.*,

1992). The measure of underlying oxygen was performed using a CLARK electrode (WTW, Oxy 96) under conditions prescribed by VIVAS and *al.* (1993).

### IV — ELECTRON MICROSCOPE SCANNING

Wood samples, drawn from different depths of the staves of used barrels (5 wines and more), were taken using a wood planer which had been washed with alcohol and then dissected with a scalpel under a flame to avoid contamination. The sliced sections were mounted and metal plated under vacuum using a mix of gold/palladium, before being covered with a fine coat of carbon.

The observations were performed using a scanning electron microscope SEM 515 (PHILIPS) equipped with a secondary electron detector with a electronic field of 15 kV.

## RESULTS

### I — INFLUENCE OF STORAGE METHODS ON THE VOLATILE ACIDITY CONTENT OF RED WINE

An homogeneous sample of red wine, which had reached the stage of malolactic fermentation, were placed in different recipients (stainless-steel vats of 10 hl, stainless-steel vats of 150 hl, stainless-steel vats with a floating top of 8 hl, concrete vats of 50 hl and lots of 5 new barrels, used barrels of 5 wines, used scraped and reburnt) were developed in a classic way for twelve months. The result is shown in figure 1.

In the low volume vats (stainless-steel 10 hl, AV : 0.39 g $H_2SO_4$/l) the volatile acidity level was greater than in the vats of larger capacity (figure 1, stainless-steel 150 hl, AV : 0.35 g $H_2SO_4$/l and concrete 50 hl, AV : 0.34 g/l $H_2SO_4$). This fact may be attributed to the insufficient quality of the sealing system. Similarly, for floating top vats the volatile acidity level is superior than for all the other conditions (AV : 0,34 g/l $H_2SO_4$). However this result should not be generalized, but rather used to emphasize the danger of bad control during the inflation on the airtight joint, and the subsequent irregular checking of its pressure.

Customarily we observe, whilst ageing in new barrels, a slight increase in volatile acidity compared to the same wine aged in a vat. The average increase varies from 0.10 to 0.15 g/l $H_2SO_4$ (120 to 180 mg acetic acid/l). In this experiment (figure1) the values are confirmed. In the new

barrels the volatile acidity is higher than in the vats (except vats with a floating top). In the used barrels it is at the same level as the vats (except vats with a floating top). Meanwhile, for the reconditioned used barrels, the burning of the shell after scraping allows for a greater quantity of acetic acid to form in the wood structure.

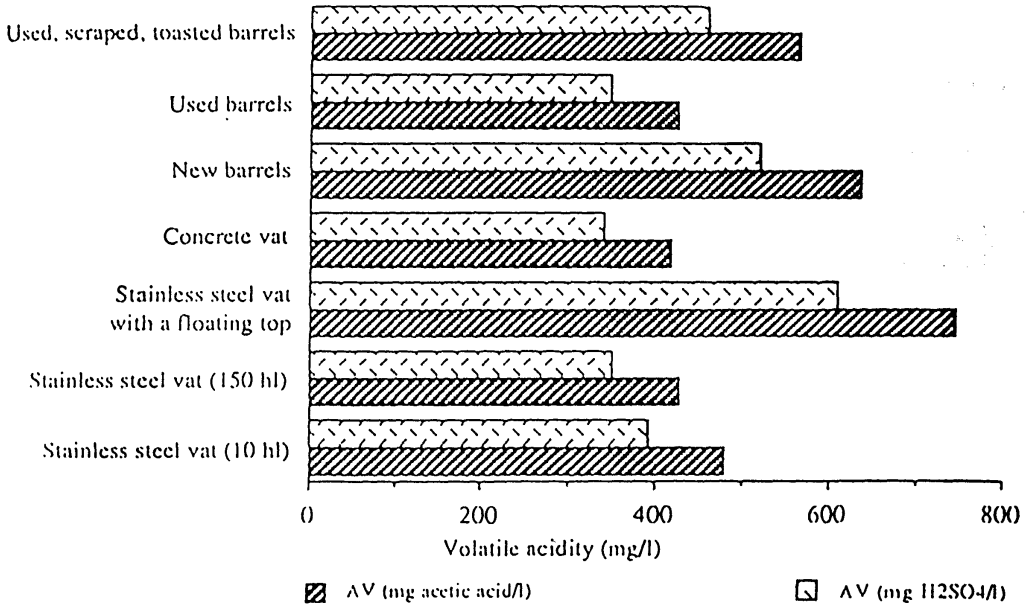


Figure 1 — Effect of the nature of the ageing recipient on the content of volatile acidity in red wines (example of a red wine ageing for 12 months).

## II — SUPPLY OF ACETIC ACID BY THE WOOD

### 1) Influence of toasting treatment

It has been known for a long time that the combustion of wood samples leads to the formation of acetic acid. Furthermore, when toasting the barrels during cooperage, one easily notices its characteristic smell. MARSAL (1992) accords great importance to this observation when determining the origin of accumulated volatile acidity ageing vinification in new barrels. We observe, in agreement with this author, that oak wood contains little free acetic acid (< 3 mg/g, figure 2), slightly more when lightly toasted (< 5 mg/g) and when moderately toasted on average (< 10 mg/g).

However when toasted strongly, the loss by volitization becomes superior on its formation. But besides this fact, the wood contains an significant fraction of acetic acid which can be released only after saponification (figure 2). This is due to acetate esterifying the polysaccharides of xylane type, generally present in the core of broad-leafed trees (JOSELEAU, 1980). This observation suggests the participation of esterified acetic acid in the volatile acidity of wine aged in new barrels.

In practice, moderate toasting creates between 0.1 and 0.15 g/l of acetic acid in the wine. However, slight and strong toastings limit the increase of volatile acidity to less than 0.1 g/l (table 1).

2) Influence of the origin of the wood

Oak from diverse French origins (Vosges, Limousin, Nevers, Allier, Bourgogne) and the United States (Oregon, Missouri, Ohio) do not show significant differences in their acetic acid content.

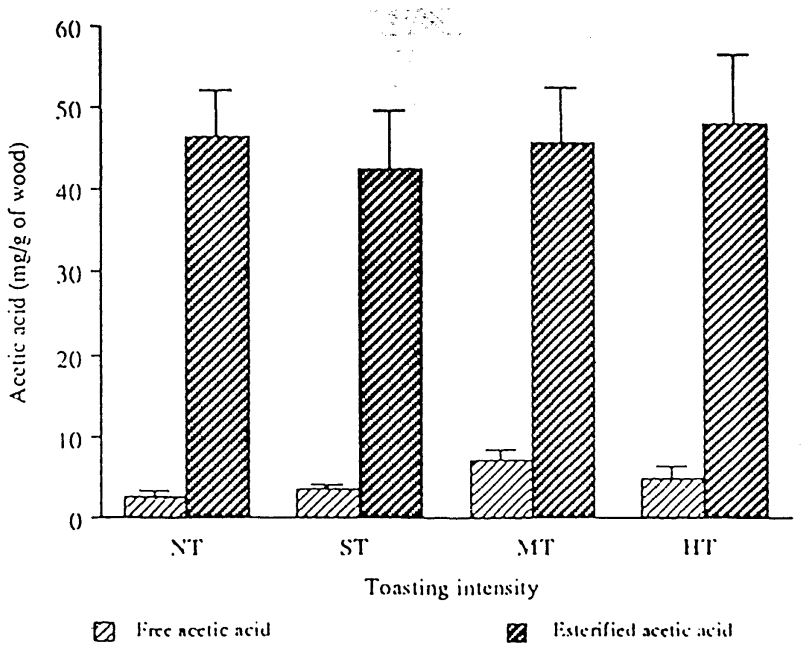


Figure 2 — Influence of toasting intensity of staves on the content of acetic acid (staves toasted at cooperage)

Toasting intensity : NT : not toasted - ST : slightly toasted
MT : medium toasted - HT : high toasted



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Table 1 - Effect of toasting intensity of the casks on the content of volatile acidity in wines after six months of storage

| Regions | Number of casks | Intensity of toasting | Volatile acidity (g $H_2SO_4$/l) | | Δ Volatile acidity |
|---|---|---|---|---|---|
| | | | t = 0 | t = 6 months | |
| Médoc | 10 | low | 0.36 | 0.44 | 0.08 |
| | 10 | moderate | 0.36 | 0.48 | 0.12 |
| | 10 | strong | 0.36 | 0.45 | 0.09 |
| Saint Emilion | 20 | moderate | 0.42 | 0.53 | 0.11 |
| Pomerol | 10 | moderate | 0.32 | 0.38 | 0.08 |

The origin of the wood and the type of oak does not appear to bear relation on the level of acetic acid. What is more the final amount of volatile acidity of wines aged in new barrels is not influenced by the origin of the wood (Allier, Tronçais, Vosges, Nevers, Limousin). Therefore we are not able to assign the cause of acetic acid in wine to the various type of wood used, be it free acetic acid or esterified.

3) Influence of barrel age

The age of barrels is of great importance on the increase of volatile acidity in wines. We have known for a long time that new barrels give wines a slightly superior level of volatile acidity as opposed to those aged in clean used barrels. In figure 3, one can verify this observation. In wine stored in a new barrel, the volatile acidity spreads noticeably more than in a barrel having contained between 2 and 5 wines. Meanwhile, in the used barrels constantly kept full, the volatile acidity increases only slightly, unless it is left empty for some months (table 2). This increase of microbiologic origin, associated with the accumulation in the bulk of the wood, with a population of acetic acid bacteria was observed with a scanning electron microscope (slide 1). During the repeated use of barrels, the proportion of esterified acid diminishes (figure 4). It is especially during the first year of utilisation that the esterified acetic acid diminishes mostly in the bulk of the wood (figure 4) : - 20 % the first year, - 5 % the second year, - 6 % the fifth year. In total after 5 wines, the wood looses approximately 30 % of its proportion of esterified acetic acid.

Tableau 2 — The effect of preparation method of used casks before barreling on the content of volatile acidity in red wine (analysed 6 month after being barreled ; results in g $H_2SO_4$/l)

| Level of withdrawal | Casks 4 years of age continiously used | Casks 4 years of age left empty for 4 months | | |
|---|---|---|---|---|
| | | Cleansing with sulphite water 48 h | Cleansing with water 48 h | Direct use |
| Surface wine | 0.54 | 0.56 | 0.6 | 0.65 |
| Middle of cask | 0.54 | 0.6 | 0.6 | 0.65 |
| Against the wood walls | 0.54 | 0.56 | 0.55 | 0.65 |
| Above the lees | 0.53 | 0.54 | 0.54 | 0.62 |
| Average | 0.54 | 0.56 | 0.57 | 0.64 |

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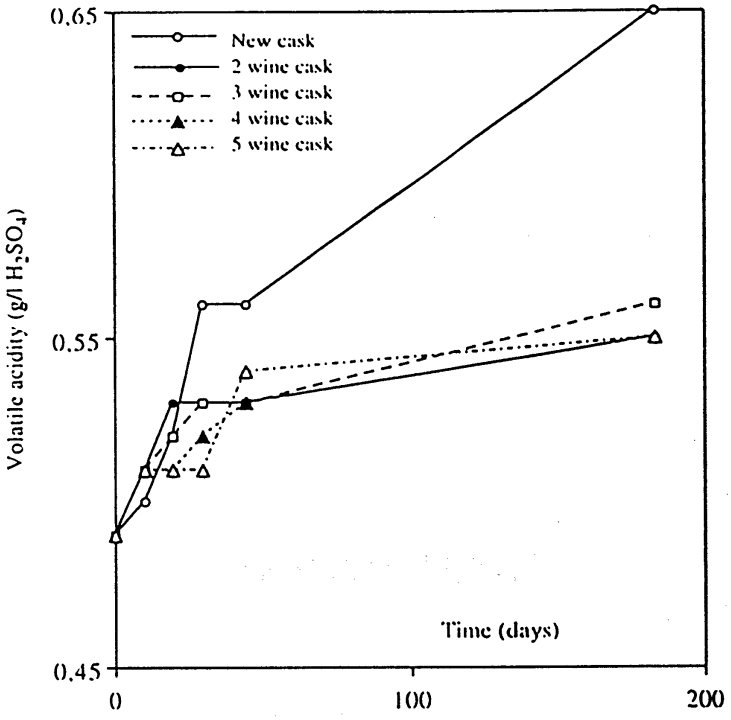


**Figure 3 — Effect of the age of the cask on the increase of volatile acidity in red wines (analysed after sixth month of ageing)**

## III — THE CONTRIBUTION OF ACETIC ACID BACTERIA TO THE PRODUCTION OF VOLATILE ACIDITY. THE EFFECTS OF OXIDOREDUCTION POTENTIAL

### 1) Studies in a controlled environment of the growth of acetic acid bacteria and the production of acetic acid

In a series of laboratory tests, the object was to study the behaviour of acetic acid bacteria over a period of time. Under different conditions, a red wine treated with charcoal and its control were saturated with oxygen (7 mg/l) and inoculated with a population of acetic acid bacteria to the order of $10^5$ cells/ml ; then after 9 days, a second weak supply of oxygen was effected (1.5 to 2 mg/l). During the course of the experiments the oxygen, ethanal and acetic acid was balanced and the populations of acetic acid bacteria numerated. The different results are shown in figure 5.

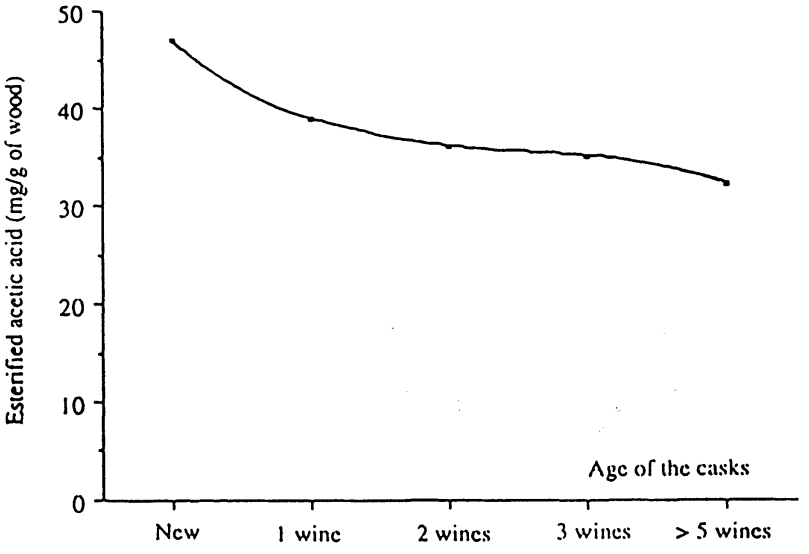


**Figure 4 — Incidence of the age of barrel on esterified acetic acid content**

The concentration of dissolved oxygen diminished rapidly in both the environments (treated and control wines). The bacteria population, very high at first, showed little variation in the two environments during the first few days. It attained, in both cases, its minimum at the same time as the oxygen. Despite the slight dissipation of the oxygen renewed on the $9^{th}$ day, a strong growth is triggered in both environments.

Between the beginning and the end of the experiment the concentration of acetic acid increased approximately 300 mg/l and 150 mg/l respectively in the purified and sample wines. Furthermore, the ethanal (the intermediary stage of the oxidation of the ethanol) had oxidised in the wine treated with charcoal.

On the whole, these results suggest that the biologic microoxidation of the ethanol to ethanal and thus to acetic acid is, for a given bacteria population, more active in a treated wine. One could argue that in such an environment, the dissolved oxygen is advantageously available to the bacteria due to oxidising substances (tannins, anthocyanins, phenolic acids) having been eliminated. These observations confirm the primary importance of dissolved oxygen in the increase of volatile acidity of microbial origin in wines.



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In a similar experiment, the environments were inoculated with initial populations of $10^3$, $10^4$, $10^5$ cells/ml, thereafter, we noted the lapse of the stationary stage and the maximum population attained in each environment. For populations of $10^3$, the lapse of the stationary stage was between 7 and 9 days, for $10^4$ cells/ml, it was limited to 3 days and, at $10^5$ cells/ml it was less than 3 days (table 3). The phenolic components did not seem to have a significant reaction on this development. In this experiment, the maximum population attained depended on the level of the initial population.

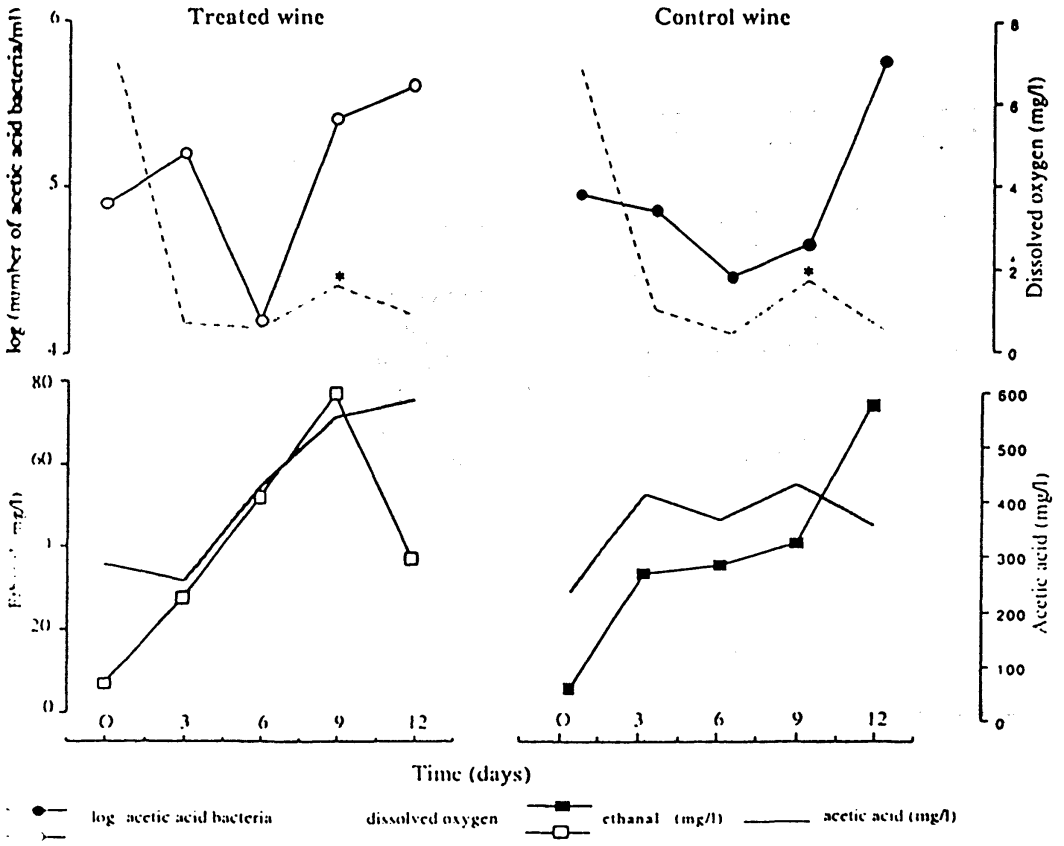


**Figure 5 — Laboratory study of the influence of oxygen consumed on the evolution of the acetic acid bacteria population and on the formation of acetic acid and ethanal in control wine and in wine treated with charcoal**

(initial oxygen content : 7mg/l ; initial bacteria population $10^5$ cells/ml ;
* indicates a second supply of oxygen)

Table 3 — The effect of the initial population of acetic acid bacteria, in control wine and in wine treated with charcoal, on the stationary stage and maximum population

| Samples | Initial population* | Stationary stage (days) | Maximum* population |
|---|---|---|---|
| Control wine | 3 | 9 (± 1) | 3.7 (± 1.2) |
| | 4 | 4 (± 1) | 4.7 (± 0.7) |
| | 5 | < 3 | 5.6 (± 0.2) |
| Treated wine | 3 | 7 (± 2) | 4.4 (± 0.4) |
| | 4 | 3 | 4.7 (± 0.6) |
| | 5 | < 3 | 5.6 (± 0.2) |

*log (acetic acid bacteria/ml)

2) Relation between the content of dissolved oxygen, the oxidoreduction potential and the viable acetic acid bacteria population

Meticulous measurements of the oxygen, oxidoreduction potential and acetic acid bacteria numerations were carried out on a large number of barrels (n = 74).We noticed that the level of acetic populations are very significantly associated with oxidoreduction potential (figure 6). The acetic acid bacteria have a great need for oxygen in order to develop, and it appears that the stationary phase depends on the state of the wines oxidation.

## IV — FACTORS INFLUENCING THE INCREASE OF VOLATILE ACIDITY

1) Influence of temperature, $SO_2$ and racking

In a series of preliminary observations (results not available) we demonstrated that the content of volatile acidity in barrels are spread in the bulk of the wine in a homogenous way. The proximity to the dregs (the source of microorganisms), the barrel walls and the surface of the wine (oxygen penetration zones) are not the preferential zones of acetic acid formation and acetic acid bacteria accumulation. The acetic acid bacteria are spread in the entire volume of the wine and form volatile acidity in equal quantity, no matter where they are localised. The samples were therefore drawn from the middle of the barrel.

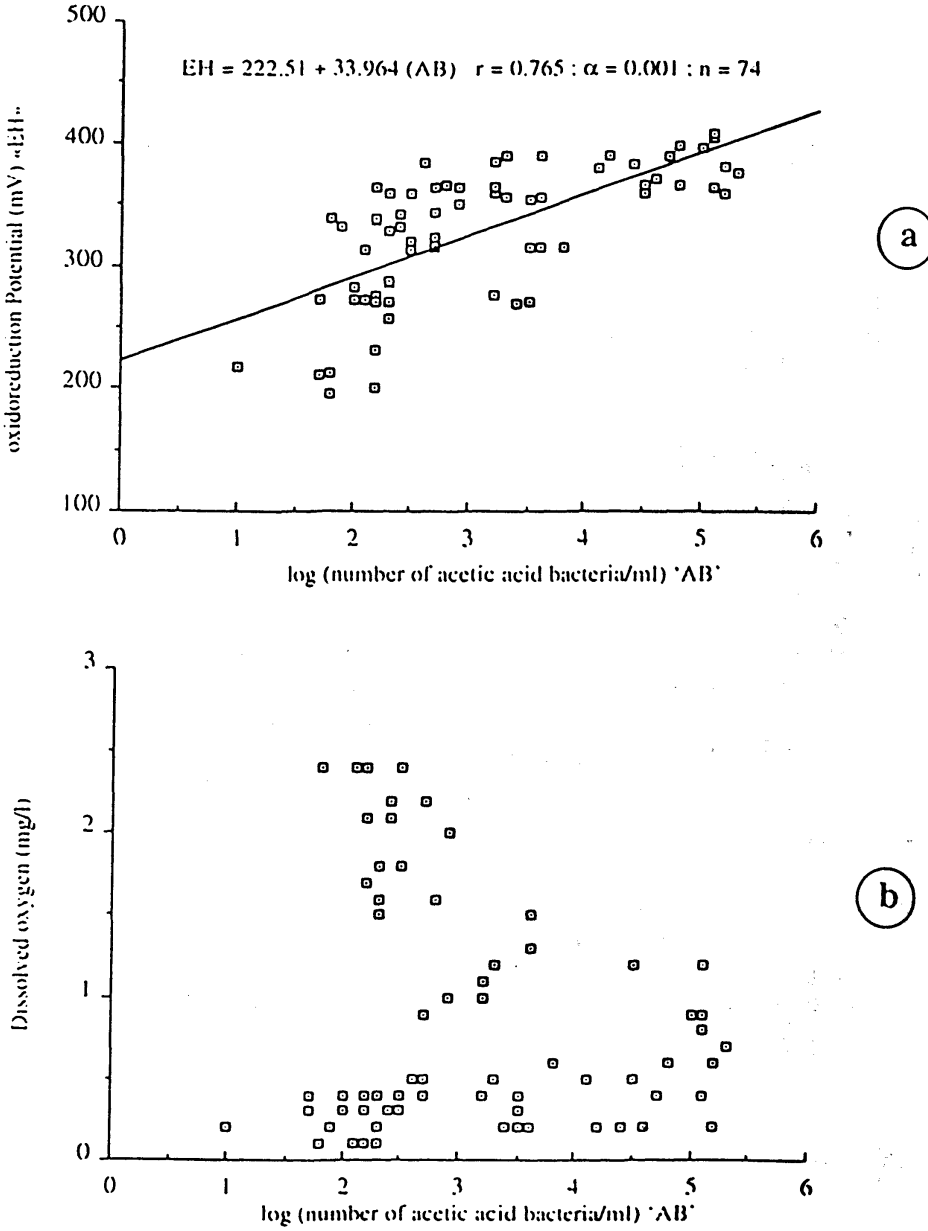


Figure 6 — Relation between oxidoreduction potential (a), dissolved oxygen (b) and the populations of acetic acid bacteria (measurements performed on 74 wines aged in new and used barrels)

Table 4 — The effect of the temperature of the cellar and the content of free $SO_2$ in the wine on the increase of volatile acidity (example of 3 cellars, wines analysed after ageing for 12 months in new barrels)

| Situation | Average temperature * of the cellar (°C) | free $SO_2$ (mg/l) | Volatile acidity (mg $H_2SO_4$/l) | | Increase of volatile acidity (mg $H_2SO_4$/l) |
|---|---|---|---|---|---|
| | | | Initial content | Content after 12 months | |
| Cellar A | 12 | 26 | 0.32 | 0.4 | 0.08 |
| | 16 | 25 | 0.34 | 0.5 | 0.16 |
| Cellar B | 13.5 | 17 | 0.28 | 0.39 | 0.11 |
| | 17.6 | 20 | 0.31 | 0.44 | 0.13 |
| Cellar C | 14 | 24 | 0.43 | 0.53 | 0.1 |
| | 16 | 25 | 0.36 | 0.46 | 0.1 |
| | 18.5 | 32 | 0.41 | 0.58 | 0.17 |

*average temperature whilst ageing

According to the example presented in table 4, it appears that temperature is the factor which determines the volatile acidity content once ageing is complete. $SO_2$ seems to have a secondary influence ; and in any case, the ageing of wine in temperatures in excess of 17-18°C provokes an abnormal increase of volatile acidity, thus even the content of free $SO_2$ is adjusted to more than 30-35 mg/l. In an experiment not presented, we have noticed that wines containing 36 and 41 mg/l of free $SO_2$ and stored for 3 months at 23-26°C formed 0.24 and 0.29 volatile acidity respectively (en g/l $H_2SO_4$) ; during the same period there was a very slight variation in the amount of free $SO_2$.

During the ageing of wine in barrels, racking constituted an important supply of oxygen. We have previously remarked that an aeration provokes the growth of acetic acid bacteria and leads to the formation of acetic acid (VIVAS and GLORIES, 1994). In order to confirm these results, we analysed, in a cellar, the influence of racking on the evolution of the dissolved oxygen, acetic acid, ethanal, $SO_2$ and acetic acid bacteria ; the results are represented in table 5. The acetic acid bacteria need, after racking, a time of latency before growth starts. It is interesting to emphasise that the free $SO_2$ is not an obstacle to the growth of acetic acid bacteria not even for their survival. 15 days after racking the volatile acidity and acetic acid have increased by 0.03 and 0.04 g/l respectively. The ethanal is subject to slight variations. After 60 days, the number of acetic acid bacteria stabilise ($10^3$ cells/ml), and the volatile acidity shows a weak variation.

2) Influence of barrel age and the conditions of preparation

In the case of new barrels, a special preparation is required before cooperage, only the cleansing with cold sulphite water is indicated. The content of acetic acid in the wood cannot be limited, not even that formed at the time of toasting. The imputable increase in volatile acidity in the barrel is low (< 0.2 g/l $H_2SO_4$). The microbiologic supply should be reduced by sufficient sulfiting (20-25 mg/l de $SO_2$) and a sufficiently low ambient temperature (12-15°C).

In the case of used barrels, especially those left empty for several months, the preparation influences the content of volatile acidity of the wine after having been aged (table 2). An empty barrel which has been left humid for a relatively long time, is the seat of development of bacteria. The accumulation of the bacteria is essentially localised in the 2 to 3 mm of the wine impregnated wood. They were localised using a scanning electron microscope to examine inner barrel surface samples taken from a depth of 1 mm to 10 mm (slide 1).


Slide 1 — Observation by scanning electron microscope of wood (depth 3 mm) of used barrels (5 wines)

1 : Detail of spring wood vessel (x 221) with little microorganisms. This is corresponding to used and sound barrels.

2 : Localisation of numerous microorganisms (bacteria and yeast) in spring wood vessel (x 925). This is corresponding to used barrels producing abnormal increases of volatile acidity.

Table 5 — The influence of racking on the content of $SO_2$, volatile acidity, acetic acid, ethanal, and the populations of acetic acid bacteria of red wines (after racking the free $SO_2$ is readjusted)

| Parameters | Before racking | After racking (4.5 mg/l of oxygène) | | |
|---|---|---|---|---|
| | | t = 3 days | t = 15 days | t = 60 days |
| Free $SO_2$ (mg/l ) | 20 | 26 | 22 | 20 |
| Total $SO_2$ (mg/l) | 54 | 79 | 68 | 64 |
| Volatile acidity (g $H_2SO_4$/l) | 0.47 | 0.47 | 0.5 | 0.5 |
| Acetic acid (mg/l )* | 540 | 560 | 580 | 580 |
| Ethanal (mg/l )* | 28 | 17 | 23 | 32 |
| Acetic acid bacteria (log (number of cells/ml)) | 3.2 | 3.4 | 4 | 3.8 |

*enzymatic quantification

When such barrels are filled directly with wine, they quickly release the acetic acid accumulated in the superficial layers of the wood into the wine (table 2). A 1 or 2 day cleansing of the barrels with sulphite water is sufficient to get rid of the acetic acid created by the bacteria (table 5). It is advisable, in addition, to sulphurise the barrel in order to sterilise the first layers of wood. The other proposed conditions for new barrels still apply in this case.

## DISCUSSION AND CONCLUSION

Whilst ageing wine in barrels, one regularly observes an increase of volatile acidity ; furthermore it is practically impossible to prevent this phenomenon. Customarily one tends to attribute the origin of the problem to microorganisms. Recently, MARSAL (1992) has emphasised the role of acetic acid found in oak wood, when it is added too that produced by lactic and acetic acid bacteria (RIBÉREAU-GAYON and *al.*, 1976). A porous recipient like a barrel which allows a slow and regular penetration of oxygen (VIVAS and GLORIES, 1993), is favourable for aerobic and microaerobic flora, which lead to the potential risk of an increase of volatile acidity in wine.

The wood naturally holds little free acetic acid (MARCO and *al.*, 1994) ; we found values of less than 3 mg/g of wood in our samples of unspecified origin. We also found the esterified acetic acid discharged after saponification. That this proportion of acetate results from strongly acetylised xylane type polysaccharides in the wood of broad leafed trees. (JOSELEAU, 1980). They are present in higher quantities (30 to 50 mg/g of wood) than the free acetate. The combined acetic acid can released by hydrolysis during storage of the wine in barrels, by following the same process prescribed for brandies (PUECH, 1987), prefered by wine in an environment which is more acid and rich in water. The burning of the inner shell of the barrel provokes a slight increase of free acetic acid resulting in the thermic degradation of the wood (PHILIPS and GOSS, 1932) and the hydrolysis of the acetyl and xylane groups (BIERMAN and *al.*, 1987). But these increases remain limited and represent 0.1 to 0.2 of the additional volatile acidity of wine aged in new barrels.

In wines, an aeration provokes the increase of the bacteria populations followed by the production and release of acetic acid. But over and above the oxygen necessary for the development of the acetic acid bacteria, it is essentially the value of oxidoreduction potential which will determine when the growth phase begins. Furthermore, the 2 mg/l supply of oxygen may be responsible for a multiplication of the populations of bacteria often more pronounced than when the wine is saturated (7 to 8 mg/l of oxygen). One proposed explanation is that a strong aeration provides the wine with the potential for maximised oxidoreduction which with the participation of all the oxidation-reducers compounds (tannins, anthocyanins, flavonoids, phenol acids...) lead to the consumption of the dissolved oxygen. The oxygen disappears rapidly and the bacteria are left with relatively little quantities for their multiplication. Of the small quantities of oxygen which limit the production of oxidoreduction potential, the oxidation-reducers compounds of wine are however required less and thus the bacteria use most of the oxygen for their development. The sulphur dioxide employed in normal doses is not enough to protect the wine against the development of acetic acid bacteria ; one must under all circumstances avoid extreme aerations and keep the wine at a sufficiently low temperature. These results are confirmed by practical work performed by us. We would like to mention that spring and summer are periods of high risk, particularly in premises which lack air conditioning and insulation.

When we use used barrels, a certain number of precautions are imposed. Generally speaking, the barrels should always be full with wine. Left empty, the wine impregnated superficial layers of wood are the origin of a substantial development of bacteria. The presence of residual humidity

and a confined atmosphere also favour the development of fungi. In this extreme situation, the interior of the casks acquires musty, acetic and stagnant odours and the barrels are irreparably lost.

A rinsing of the cask with water, followed by sulphurising, a draining for a few days and then left in a reasonably humid place (Hr = 80-85 %) after renewed sulphurising, assures reasonable conditions when storing casks for some months. Before reusing the barrels, it is necessary to cleanse them with sulphite water and sulphurise them again before being cooped with a wine. By respecting these few rules, one can avoid the insidious content of volatile acidity. The scraping and reburning of casks are prohibited because, on the one hand, the burning leads to the forming of acetic acid whilst degrading the wood, and on the other hand, because the impregnation of wine is much deeper than that simply indicated by the red coloured layer. The imperious rule when ageing in barrels remains the hygiene of material and the respect of elementary oenology.

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