Formation of Furfurylthiol Exhibiting a Strong Coffee Aroma during Oak Barrel Fermentation from Furfural Released by Toasted Staves

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Furfurylthiol (FFT) is formed in white wines during alcoholic fermentation in the barrel from the furfural released by toasted staves. The quantity of furfural released into the must has a decisive effect on the quantity of FFT in the finished wine. Wines fermented in new barrels thus contain larger quantities of FFT than those fermented in used barrels. Fermentation conditions favorable to an excess production of H_2S (hydrogen sulfide) by the yeast promote the formation of this volatile thiol. The presence of this volatile thiol in white wines is, therefore, closely related to the yeast's sulfur metabolism.

INTRODUCTION

Furfurylthiol (FFT) is an extremely odorous compound (*1*), first identified in roasted coffee (*2*). It has also been identified in other foods such as roasted meat, wheat bread, and popcorn (*3*). In a previous paper, we reported the identification of FFT in white Petit Manseng wines, red Bordeaux wines, and toasted barrel staves. However, the quantities of FFT formed in barrel staves during toasting were too small to account for the FFT content in the wines (*4*).

This paper reports on the presence of FFT in barrel-fermented white wines and conditions that promote its formation.

MATERIALS AND METHODS

Wines, Must, and Fermentation Medium Used. The white wines analyzed were all from the 1999 vintage. The Petit Manseng wines were from Jurançon (Domaine Cauhapé). The Sauvignon Blanc and Semillon wines were from Graves (Bordeaux, Clos Floridène). The white Chardonnay wine was from Puligny Montrachet (Domaine Leflaive). FFT was assayed in these wines that had been fermented and aged, either in stainless steel tank or in new or used barrels for 1 month after the end of alcoholic fermentation. In the case of Sauvignon Blanc must fermented in a new barrel, the FFT content was monitored during alcoholic fermentation and aging on lees.

A medium-toasted French oak barrel (*5*) was filled with a hydroalcoholic model solution (12% ethanol, 5 g/L tartaric acid, pH 3.5). The furfural content of the model solution was analyzed regularly from the time it was first put into the barrel during 15 days.

A Sauvignon Blanc must was fermented with *Saccharomyces cerevisiae* (strain Zymaflore VL3) in four barrels of different origins (age and cooperage). Wine samples were taken from each barrel at the end of alcoholic fermentation. FFT, furfural,

and furfuryl alcohol in the wines were assayed. The wines' FFT concentrations were plotted on a graph with those of the furfural extracted from the barrels. The amounts of furfural released by the barrels [∑ furfural] were equal to the sum of the furfural and furfuryl alcohol concentrations in the wines (*6*).

The must used for the microfermentation experiments was made from Sauvignon Blanc grapes from the Bordeaux area, 1999 vintage. They were pressed and sterile-filtered on a membrane filter (0.40 μm) and then sulfured (30 mg/L free SO_2) and maintained at 4 °C in 50 L stainless steel tanks befor use. The model fermentation medium consisted of the following solution: 85 g of glucose, 85 g of fructose, 3 g of tartaric acid, 0.3 g of citric acid, 2 g of potassium phosphate, 0.3 g of mesoinositol, 100 mg of alanine, 300 mg of arginine, 100 mg of glutamine, 50 mg of serine, 50 mg of threonine, and 110 mg of ammonium sulfate, all in 1 L of distilled water adjusted to pH 3.5 with solid potassium hydroxide, and 10 mL/L of vitamin solution (4 mg/L *d*-biotin, 100 mg/L thiamin hydrochloride, 100 mg/L pyridoxine hydrochloride, 100 mg/L nicitinic acid, 100 mg/L *d*-panthothenic acid hemicalcium salt, and 100 mg/L *p*-aminobenzoic acid) was added. Sauvignon Blanc must and model medium were fermented with *S. cerevisiae* (strain Zymaflore VL3) in the laboratory, either supplemented with furfural (15 mg/L) or not. The alcoholic fermentation took place in 750 mL bottles, filled three-fourths full and closed with bubbling stoppers, in an air-conditioned room at 20 °C. An unfermented control was obtained by adding 50 mg/L pimaricine to each type of must.

A fermentation medium with the composition specified above, containing furfural (15 mg/L) but no ammonium sulfate (i.e., no sulfur compounds), was fermented with increasing quantities of cysteine (0–20 mg/L).

A fermentation medium, containing 70 mg/L assimilable nitrogen with 15 mg/L added furfural, was supplemented with increasing quantities of nitrogen, by adding either asparagines (0.9, 1.4, and 1.9 g/L) or ammonium sulfate (0.6, 0.9, and 1.4 g/L).

Assaying Furfural, Furfuryl Alcohol, and FFT in Various Wines and Medium. FFT is assayed in wines and model medium using the method described by Tominaga (*4*), and the coefficient of variation is <5%. To assay the furfural and furfuryl alcohol, the method described by Boidron (*7*) was used. This method has a coefficient of variation of <5%.

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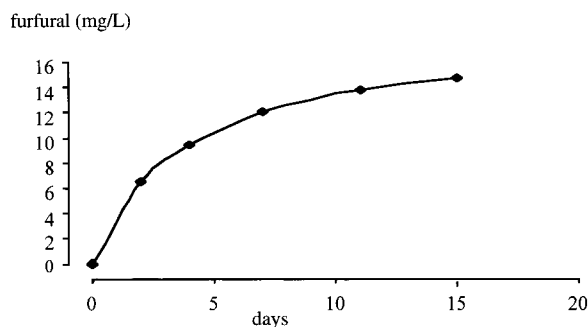[†] In charge of research for Seguin Moreau (Merpins, F-16103 Cognac, France) at Faculté d'Oenologie.

Figure 1. Kinetics for the release of furfural into a model medium in contact with a barrel.
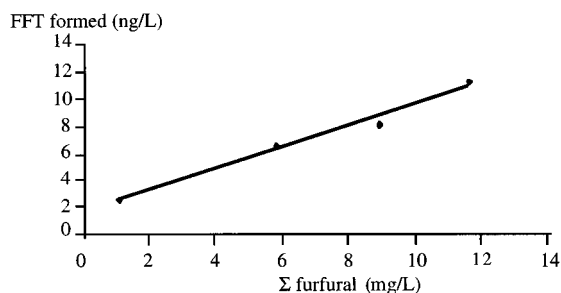

Figure 2. Relationship between furfural released by the barrels and FFT content of white wines after fermentation.

Table 1. Contribution of Oak Barrels to FFT Formation (Nanograms per Liter)

	Petit Manseng	Sauvignon blanc	Chardonnay	Semillon
new barrel	23	8	20	47
used barrel	2	3	6	4
vat	0	0	na[a]	0

[a] na, sample not analyzed.

RESULTS AND DISCUSSION

Impact of New Barrels on FFT Formation. Assays of the volatile thiols in white wines made from various grape varieties confirmed the presence of FFT in the wines, irrespective of the grape variety (Table 1). However, the FFT content in the wines was considerably affected by the container used. The wines fermented in new barrels had a higher FFT content than those fermented in used barrels. No FFT was found in the wines fermented in stainless steel vats.

Relationship between Furfural Released by the Barrels and FFT Formed in the Wines. Fermentation in new barrels apparently promotes the formation of FFT in wines. These results suggest the presence of a precursor of FFT in the toasted wood of new barrels. The important role played by furfural in the formation of FFT in roasting coffee beans is now well documented (*8*, *9*). As the furfural was rapidly diffused in the medium in the barrel (Figure 1), it was, therefore, present in the must at the start of alcoholic fermentation. Assaying the furfural and FFT contents of wines made from the same Sauvignon Blanc must fermented in barrels of different origins and ages highlighted a significant correlation ($r^2 = 0.98$) between the concentrations of these two compounds in the wines (Figure 2). It would appear, therefore, that FFT is formed in white wines from furfural released by the barrels during alcoholic fermentation.

Role of Yeast Fermentation Activity in FFT Formation. FFT was formed only in media fermented
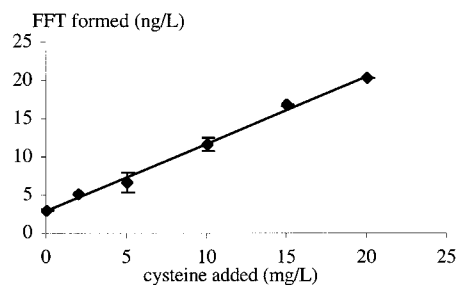

Figure 3. Relationship between FFT content formed during alcoholic fermentation and cysteine concentration of the fermentation medium.

Table 2. Role of Furfural in FFT Formation (Nanograms per Liter) during Alcoholic Fermentation

	Sauvignon	fermentation medium
fermented, without furfural	0	0
fermented, with furfural	7	6
not fermented, with furfural	0	0

Table 3. FFT Content during Alcoholic Fermentation and in the First Month of Aging

days	ng/L FFT	days	ng/L FFT	days	ng/L FFT
0	0	4	14	9	17
2	12	7	16	35	18

in the presence of furfural (Table 2), thus confirming the role of furfural in FFT formation. This reaction apparently requires the fermentation action of the yeast to take place. FFT formation was observed from the beginning of alcoholic fermentation (Table 3). Concentrations continued to increase until alcoholic fermentation was completed and then remained stable for the first few months of aging on the lees. FFT formation in white wines is, therefore, closely linked to alcoholic fermentation.

Impact of Adding Cysteine to Fermentation Medium on FFT Production. The quantities of FFT produced during alcoholic fermentation were proportional ($r^2 = 0.92$) to the amounts of cysteine added to the medium (Figure 3). Conversion of furfural into FFT requires the replacement of a carbonyl group with a thiol group. The sulfide synthesis of yeast during alcoholic fermentation may be involved in the FFT formation mechanism. Increased FFT production in media with a high cysteine content is thought to be due to an excessive production of sulfide, leading to the release of large amounts of H_2S (*10*), due to the yeast's cysteine desulfydrase activity (*11*). FFT production is, therefore, apparently related to the yeast's sulfur metabolism, particularly the production of HS^-.

Impact of Assimilable Nitrogen in Fermentation Medium on FFT Formation. Enhancing a fermentation medium's nitrogen content by adding asparagine (a must is considered to have a high assimilable nitrogen content at values >200 mg/L) clearly led to a decrease in FFT production (Figure 4). However, increasing the nitrogen content by adding ammonium sulfate had little impact on FFT production (Figure 4). In this case, the FFT content remained stable, irrespective of the quantity of nitrogen added. This tendency was confirmed by duplicate experiments.

The nitrogen content of a must has a decisive impact on H_2S production (*12*–*14*). It is difficult to ferment nitrogen-deficient must, and excessive amounts of H_2S are sometimes produced. In a nitrogen-deficient must, amino acid synthesis is slowed and the sulfides produced

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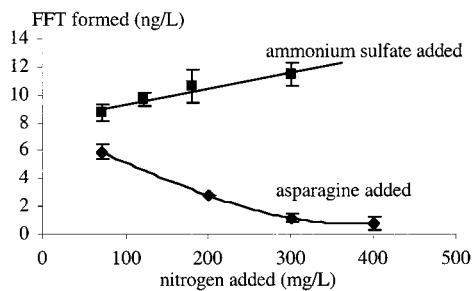*J. Agric. Food Chem.*, Vol. 49, No. 10, 2001 **4835**

Figure 4. Changes in FFT content according to the total nitrogen content of a fermentation medium, adjusted by adding asparagine and ammonium sulfate.

by the yeast are released into the medium as H_2S. Production of the HS^- anion in excess of the amounts required for the yeast's protein synthesis promotes the formation of FFT, as shown in Figure 4. In practice, ammonium sulfate is often used to correct excessive H_2S production. As this adds both sulfur and nitrogen to the must (*15*), it is logical that FFT production should be less impacted, even if the nitrogen level is high.

Conclusion. FFT was present only in barrel-fermented wines. Furfural is a precursor of FFT, released by toasted wood when it comes into contact with wine. The wines fermented in new barrels logically had a higher FFT content.

The formation of FFT by yeast in the presence of furfural increased when the conditions were favorable for the production of HS^- anions in excess of the amounts required for protein synthesis: high sulfur content (cysteine, sulfates) and relatively low nitrogen levels. It is, therefore, closely related to the yeast's sulfide synthesis mechanism. Future publications will investigate whether the reaction substituting an SH group for the furfural's carbonyl group takes place inside or outside the yeast cells.

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