JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

Resveratrol Content of *Palomino fino* Grapes: Influence of Vintage and Fungal Infection

ANA ROLDÁN,* VICTOR PALACIOS, ILDEFONSO CARO, AND LUIS PÉREZ

Departament of Chemical Engineering, Food Technology and Environmental Technology, Faculty of Sciences, University of Cádiz, Apartado 40, Polígono Río San Pedro, 11510 Puerto Real, Cádiz, Spain

The objective of this study was to determine the influence of certain factors on the resveratrol content of *Palomino fino* grapes, cultivated in the Jerez-Xérèz-Sherry area, at the moment of harvest. The results show that the resveratrol content is highly influenced by the climatic conditions prior to the period of maturation of the fruit. On the other hand, the gray mold pressure in the vineyard, a fungal infection caused by *Botrytis cinerea*, increased the resveratrol contents at the early stages of fungal development. When *Botrytis* development was extensive, the resveratrol content tended to decrease in the juice but tended to increase in the skin. Physiological stress of the plant leads to increases in the resveratrol content, caused as much by the climatic conditions of the vintage as by biotic factors. In this case resveratrol is present mainly in the glycosylated form.

KEYWORDS: Resveratrol; grape; juice; *Palomino fino*; *Botrytis cinerea*

INTRODUCTION

The great interest shown in recent years in resveratrol and its derivatives has led to numerous studies aimed at the determination of these compounds in all types of commercial wines (*1*). These studies established that resveratrol levels were higher in red wines than in white wines, due fundamentally to the vinification techniques. Prefermentative practices such as the addition of SO_2 and ascorbic acid before grape crushing (*2*) or operations such as maceration (*3*−*5*), alcoholic fermentation (*4*), inoculation with different yeast strains (*6*−*8*), malolactic fermentation (*4*, *8*), precipitation phenomena, oxidation, or adsorption (*9*, *10*), together with the β-glucosidase activity (*8*) and clarification with some fining agents (used in wine clarification and filtration operations) (*2*, *5*, *8*, *9*), can influence the levels of resveratrol and its derivatives during the wine-making process.

On the other hand, many of these factors are closely related with other parameters that have a direct influence on the grape, such as the variety, climate, soil, and sanitary stage, to name but a few. These factors can modify the concentrations of resveratrol and its derivatives in the grape at the moment of harvest and, therefore, its content in the final wine.

According to Jeandet et al. (*3*) the resveratrol content of wines made with red grapes (var. Pinot noir) is ∼3 times higher than in those made with white grapes (var. Chardonnay blanc) when vinted under the same conditions. Some authors have shown that the resveratrol content in wines differs according to the variety of grape; however, within the same variety, significant differences can be found in the resveratrol content from one vintage to another (*5*, *11*). Much of this variance can be explained in terms of the intrinsic properties of the cultivars such as the age of the vines (*5*, *11*, *12*), the grape maturation (*5*), and the degree of infection by *Botrytis cinerea* (*3*, *13*−*15*). In this way, the radiation, temperature, humidity, precipitation, and evaporation phenomena—characteristics of the production area—all have an effect on the fruit composition and, as a consequence, the resveratrol content at the moment of harvest (*16*, *17*) and, finally, in the wine (*5*, *9*, *12*, *18*).

These factors would also influence the presence of infections in the plant and, as a consequence, the production of resveratrol. In this sense, it is thought that the resveratrol synthesis is related to the activation of the plant defense mechanism before damage or attack caused by some pathogen. Nevertheless, in the years when there was a high or moderate *B. cinerea* pressure in vineyards, a decrease was observed in the resveratrol content of wines (*15*). In these cases, the resveratrol concentration in juices and wines is determined by a balance between resveratrol production by the plant and resveratrol degradation by the pathogen (*3*). According to Schouten and colleagues (*19*), who caracterized two different laccase genes from *B. cinerea* (Bclcc1 and Bclcc2), only laccase gene Bclcc2 in liquid cultures in the presence of resveratrol was strongly expressed. Thus, Bclcc2 does not detoxify resveratrol but, rather, converts it into compounds that are more toxic for the fungus itself.

Given the great importance of these compounds, as well as the existence of a few studies that provide evidence for the direct influence of certain factors on the resveratrol content in the grape, the work described here was designed to determine the effect of climatic conditions of the vintage on the glycosylated and isomeric free resveratrol contents in both the juice and skin of the *Palomino fino* grape. In addition, the special interest in the defense mechanism of this variety in the *B. cinerea*/grapevine interaction led us to determine the production levels

* Author to whom correspondence should be addressed (fax 00 34-956-016411; e-mail ana.roldan@uca.es).

10.1021/jf020774u CCC: $25.00
Published on Web 01/24/2003

of the isomeric forms with respect to different degrees of infection by gray mold.

MATERIALS AND METHODS

Sampling. Samples of *Palomino fino* grapes were taken during the course of two vintages (1996−1997) in 24 plots located in the Jerez-Xérèz-Sherry D.O. wine production area (latitude 36° 41′ N) included in the maturation monitoring program conducted by the Laboratorio Agroalimentario y Estación Enológica of Jerez de la Frontera (Cádiz, Spain).

In Jerez the classic pruning system called "stick and thumb" is used, which consists of cutting the two branches that make up each vine to leave in alternate years a single "stick" with eight buds (from which the bunches of grapes develop) and a "thumb" with just one bud. The bud on the thumb sprouts into the following year's stick, whereas this year's stick will then be cut back to just a thumb. The stick and its bunches are held up by two wires, 20 and 40 in., respectively, above the ground, which make up the espalier (*20*, *21*). The work of manuring is based on the use of organic fertilizer, animal manure fundamentally, which is applied at 40000−50000 kg/ha to organic manure each of 3 or 4 years (*20*).

In each of the plots, samples of ∼2 kg per plot were taken in clusters of 10−15 berries. The grape samples were collected at the stage of finishing on the day of commercial harvest (maturity). Vines were selected from different parts of the plots in order to ensure that the size and general characteristics of the plots were as representative as possible.

To study the incidence of gray mold (*B. cinerea*) on *Palomino fino* grapes, four different lots were taken from healthy grapes and from grapes affected at 50, 75, and 100% by *Botrytis*. The lots were obtained from a plot located in the Jerez production area. The vintage used in this study was 1997.

Characterization of Juices. The samples collected were crushed mechanically to extract the juice. The resulting juice was then filtered (pore size ≤ 0.1 mm), and the analytical determinations required to monitor commercial grape maturity were conducted together with the determination of resveratrol and its derivatives. In parallel, a sample of the residual skin was taken and immediately frozen until analysis could be performed.

The mean fresh berry weight was determined by gravimetry. The Baume grade was determined by using the OIV procedure (*22*) with a Dujardin-Salleron density meter. The titratable acidity was determined by titration according to the method of the American Society of Enologists (*9*). The pH was measured with a digital pH meter (micro-pH 2001, Crison Instruments, S.A., Barcelona, Spain) equipped with a combined electrode. The phenols index (PI) of the juice was determined by measuring the sample absorbance at 280 nm (*9*) with a UV−visible spectrophotometer (Perkin-Elmer 200, Perkin-Elmer Corp., Norwalk, CT). The gluconic acid and glycerol contents of juices were determined by enzymatic test (*23*).

Extraction of Resveratrol and Its Glycosylated Derivatives. Given the different characteristics of the two types of sample (the juice and the skin), each was subjected to a different extraction process prior to analysis by high-performance liquid chromatography (HPLC).

For the juice samples, a solid phase extraction (SPE) was performed using C_{18}/600 mg LiChrospher cartridges (Supelco Inc., Bellefonte, PA) and following the methodology proposed by Mattivi (*11*) (but using 10 mL of diluted sample that had been neutralized). Prior to the extraction stage the sample was centrifuged and filtered using 0.45 μm Teflon syringe filters (Millex-LCR, Millipore, Bedford, MA). A multiple collector—Visiprep-DL solid phase extraction vacuum manifold (Supelco)—was used for the extraction of solid samples, and this allowed 12 samples to be extracted simultaneously.

For analysis of the skins, samples of skin (25 g) were macerated with 50 mL of a mixture MeOH/HCl at 0.1% for 30 min with ultrasound. After the extraction, the sample was centrifuged and filtered prior to analysis by direct injection according to the methodology described by Mattivi (*11*) with some modifications: a LiChrospher 100 RP reversed-phase column (250 × 4 mm; 5 μm, Merck) was used at a temperature of 40 °C. The operating conditions were as follows: eluent A, H_3PO_4 10^{-3} M; eluent B, CH_3CN; flow, 1 mL/min; elution gradient, at 0 min, 100% A, at 25 min, 50% B.

The HPLC equipment (Waters, Milford, MA) consisted of a model 2690 separation module, a model 996 aligned photodiode UV−visible light detector (190−800 nm), and a PC running Millennium 2010 software for the control and processing of the chromatographic data.

trans- and *cis*-resveratrol were identified by comparison of their retention times and their UV spectra with those of the commercial *trans*-resveratrol standard and with those of *cis*-resveratrol obtained by photoisomerization of the trans compound. The glycosylated forms of the two resveratrol isomers were identified on the basis of the work carried out by Roggero (*24*) on the retention times corresponding to the various commercially available compounds.

Resveratrol and its isomers were quantified by means of a multiple-level calibration curve (external standard) using pure *trans*-resveratrol (Sigma). Because glucoside standards are not commercially available, quantification was based on the assumption that they have the same molar extinction coefficients as *trans*- and *cis*-resveratrol.

Meteorological Data. Mean monthly temperature, relative humidity, radiation, and precipitation data for the 1996−1998 period were compiled from the Experimental Vine-culture Station of CIFA Rancho de la Merced, located in the municipal distric of Jerez de la Frontera (Cádiz, Spain). The results corresponding to the vintages studied are shown in **Table 1**.

Table 1. Average Climatic Data Corresponding to the Growth (February–June) and Maturation (July–August) Periods for the 1997 and 1998 Vintages

	growth period		maturation	
	1997	1998	1997	1998
mean temp (°C)	16.3	16.0	23.4	25.5
mean radiation (MJ/m^2)	603.9	554.7	160.8	141.3
rel humidity (%)	69.4	67.2	59.9	54.7
precipitation (mm)	211.1	711.4	1.2	0.1

RESULTS AND DISCUSSION

Influence of Climatic Conditions of the Vintage on the Resveratrol Content. It can be observed from the results in **Table 2** that the mean total resveratrol contents in the berry, at the moment of harvest, were different for each vintage. In the first vintage, the total resveratrol content in the berry was higher (84.7 nmol) by almost a factor of 2 than in the second vintage (47.9 nmol). The isomeric distribution of the different compounds in the berry also showed significant differences between the vintages. In the first vintage (1997) there was a greater content of glucosides in the skin than in the second one. However, in the 1998 vintage the free and glycosylated isomers were present in greater quantities in the juice, although the total content of resveratrol was lower. This indicates that the grape composition and, to a greater extent, the resveratrol content and its distribution in the berry are clearly influenced by the climatic conditions experienced by the vintage.

In the 1997 vintage, during the stage of vine growth, a low level of precipitation (211.1 mm) (**Table 1**) occurred with regard to the suitable precipitation level for the maturation of the *Palomino fino* grape variety, which is typically ∼600 mm (*23*). As a consequence, prior to reaching the maturation period, the plant was experiencing hydric stress. During the maturation, low temperatures, as expected given the mean temperatures of the area (24−25 °C) (*25*), and a high radiation level were registered. Under these conditions, a decrease in photosynthesis and the general metabolism of the plant took place in addition to a delay in the migration caused by bases coming from the soil (*25*). All of these factors led to high total acidity and polyphenol values in the juice (**Table 2**). Due to the occurrence

Table 2. *Palomino fino* Grape Composition for the 1997 and 1998 Vintages (Mean ± SD)[a]

								RJ		RS	
vintage	°Be	pH	MFB	TA	TPI	G	GA	GL	FI	GL	FI
1997	10.6 ± 0.6	3.6 ± 0.1	2.65 ± 0.31	3.46 ± 0.42	37.6 ± 16.7	0.09 ± 0.04	0.11 ± 0.09	4.8 ± 3.3	nd	65.5 ± 21.0	14.4 ± 9.5
1998	10.7 ± 0.9	3.8 ± 0.1	2.07 ± 0.39	2.67 ± 0.37	19.5 ± 8.43	0.13 ± 0.06	0.19 ± 0.10	6.0 ± 2.5	2.9 ± 0.7	25.5 ± 13.2	13.5 ± 10.0

[a] °Be, density unit; MFB, mean fresh berry weight (g); TA, total acidity (g of TH_2/L); TPI, total polyphenols index; G, glycerol (g/L); GA, gluconic acid (g/L); RJ, resveratrol in juice (nmol/berry); RS, resveratrol in skin (nmol/berry); GL, glucosides; FI, free isomers; nd, not detectable.

Table 3. Glycerol, Gluconic Acid, and Resveratrol Contents for the Different Degrees of Infection (Mean ± SD)[a]

	RJ					RS	
VA (%)	G	GA	G/GA	GL	FI	GL	FI
0	1.47 ± 0.87	0.28 ± 0.06	5.25	2.13 ± 1.8	0.27 ± 0.15	14.20 ± 8.10	1.45 ± 0.92
50	3.53 ± 0.95	0.66 ± 0.11	5.35	6.93 ± 2.3	0.95 ± 0.34	32.71 ± 12.20	0.64 ± 0.30
75	4.71 ± 0.51	1.26 ± 0.03	3.74	5.09 ± 2.6	0.39 ± 0.10	51.56 ± 22.13	1.70 ± 0.61
100	6.03 ± 0.92	5.93 ± 0.09	1.02	1.45 ± 0.77	0.09 ± 0.03	63.60 ± 20.04	2.06 ± 1.04

[a] VA, visual analysis (infection degree %); G, glycerol (g/L); GA, gluconic acid (g/L); RJ, resveratrol in juice (mg/L); RS, resveratrol in skin (mg/kg); GL, glucosides; FI, free isomers.

of physiologic stress, the plant activates its defense mechanisms and, as a consequence, the total resveratrol content in this vintage was higher.

In contrast, the climatic conditions experienced by the 1998 vintage prior to maturation were more favorable. This situation is reflected both in the resveratrol content and also in the physicochemical composition of the grape. In this case, the phenomena of perspiration and migration were favored, and the fruit therefore reached a good stage of maturity, which is reflected in the better quality of the juices (lower acidity and a lower polyphenol concentration) (**Table 2**).

Thus, it can be concluded that a situation of physiologic stress, such as that caused by the limited precipitation experienced by the first vintage, causes resveratrol accumulation to take place in the berry, and this is present mainly in the glycosylated form. These glucosides accumulate in the skin and are stored for use as part of the defense mechanism of the plant, whereas the free isomers in the juice spread and disappear.

Influence of Biotic Factors (Gray Mold). The nature of the *Palomino fino* variety (which has a thin skin) and the special climatic conditions of the Jerez area, as well as certain elicitation factors—such as the pricking of insects (*Lobesia botrana*) or the precipitation level during the grape harvest—make this variety particularly susceptible to attack by *B. cinerea* and, as a consequence, to the appearance of gray mold. The determination of the glycosylated and free isomers in the resveratrol levels, for different degrees of infection, is of special interest in order to assess the influence of the fungus on the variety under investigation as well as to gain information about its defense mechanism.

As indicated in the bibliography (*26*−*29*), when grapes are affected by *Botrytis*, an increase in the levels of glycerol and gluconic acid in the juice is observed. On the basis of this information, we can take the levels of glycerol and gluconic acid, which are shown in **Table 3**, as being indicative of the degree of infection in the berry. Consequently, a relationship between the glycerol/gluconic acid ratio and the infection degree determined by visual analysis was established for each of the selected samples. Comparing the data in **Table 3** with the average levels of glycerol and gluconic acid at the moment of the grape harvest (**Table 2**), it can be seen that the glycerol/gluconic acid ratio (G/GA) in both vintages was lower (0.82 and 0.68, respectively). According to Pérez et al. (*27*), the glycerol and gluconic acid levels in the juice of healthy grapes in the Jerez area are in the range of 130−190 mg/L for glycerol and 80−310 mg/L for gluconic acid, and these levels vary according to the maturity of the fruit. The results shown in **Table 3** indicate that, even though some berries are apparently healthy within an infected cluster, the presence of *B. cinerea* can cause initial physiologic effects on the fruit, which are reflected in the presence of compounds associated with the defense mechanism of the plant, among which are resveratrol and its derivatives.

As indicated by Ribereau-Gayon et al. (*30*), during the internal and initial development of the fungus upon grape skin, a significant accumulation of glycerol takes place due to the degradation of sugars by the causal agent of gray mold. When the fungal agent emerges on the exterior of the fruit and reaches to its stationary phase in terms of growth, a significant accumulation of gluconic acid, which could be the result of oxidative stress induced by *B. cinerea*, and an increase in the laccase enzymatic activity occurs. Therefore, the glycerol/gluconic acid ratio can be taken as indicative of the development stage of *B. cinerea* and its internal or external phase of performance.

As can be observed from the results in **Table 3**, the glycosylated forms of resveratrol prevail over the free ones both in the juice and in the skin of the *Palomino fino* grape regardless of the degree of infection. However, the evolution of the resveratrol content in the juice and the skin as related with the glycerol/gluconic acid ratio is very different. In the juice, during the internal phase of the fungal agent development, a progressive accumulation of the total content of resveratrol is observed as the glycerol/gluconic acid ratio increases. At more advanced infection stages, when *Botrytis* had reached its external development, a widespread decrease of the total resveratrol content is observed until levels of the same order as the initial ones are attained. These results confirm the hypothesis put forward by Jeandet et al. (*15*), who indicated that wines obtained from grapes that have suffered a high *Botrytis* infestation (80%) have lower resveratrol contents due to degradation by the laccase enzymes such as stilbene oxidase (*3*, *12*, *15*). Thus, the resveratrol content in the juice, and therefore in the wine, seems to be the result of a balance between resveratrol synthesis by the plant and the degradation of resveratrol by the pathogen.

In the skin (**Table 3**), where according to a number of authors (*1*, *31*−*33*) the resveratrol synthesis occurs, one would expect an accumulation of resveratrol in response to infection by *B. cinerea*. However, the results show that even before attack by *B. cinerea* (when the berries did not exhibit disease symptoms), that is, in the first stage of the internal development of the pathogen, a sharp decrease in the resveratrol content occurs. This phenomenon could be due to the fact that, before the fungal presence and before resveratrol synthesis is stimulated, the resveratrol already present in the skin is used by the grape as part of its defense mechanism. This compound would become a precursor of stilbenes such as ϵ-viniferins (*14*), which are more toxic to the pathogen. For this reason, the free and glycosylated forms of resveratrol decrease. In the second stage of pathogen development, an accumulation of resveratrol takes place until certain levels are reached, and these are maintained throughout its external development. These results are according to Romero et al. (*34*), who indicated that in grape berry skins infected by powdery mildew the resveratrol and piceid isomers were considerably increased and the degree of infection was positively related to their stilbene content. In principle it would be hoped that the resveratrol levels would diminish the performance of the laccase, although these results seem to indicate that, at skin level, the degree of resveratrol synthesis is greater than the degree of degradation by the enzyme. Consequently, the laccase activity of *B. cinerea* appears to be more important within the pulp than in the skin. In liquid cultures the oxidative inactivation of resveratrol is facilitated by rapid distribution of the laccases (*32*).

Situations of physiologic stress, caused as much by unfavorable climatic conditions as by biotic factors (infection by *B. cinerea*), cause the resveratrol content to increase, and this is present mainly in its glycosylated forms.

On the other hand, during attack by this fungus, the rot factor and the resveratrol levels in the juice and skin are parameters that can be used to indicate the evolution of the pathogen in the berry.

ACKNOWLEDGMENT

We express our gratitude to the Consejería de Agricultura y Pesca de la Junta de Andalucía for the technical and professional assistance provided by the staff of the Laboratorio Agroalimentario y Estación Enológica de Jerez de la Frontera, without whose help it would not have been possible to carry out this work.

LITERATURE CITED

(1) Goldberg, D. M.; Ng, E.; Karumanchiri, A.; Yan, J.; Diamandis, E. P.; Soleas, G. J. Assay of resveratrol glucosides and isomers in wine by direct-injection high-performance liquid chromatography. *J. Chromatogr. A* **1995**, *708*, 89−98.

(2) Castellari, M.; Spinabelli, U.; Riponi, C.; Amati, A. Influences of some technological practices on the quantity of resveratrol in wine. *Z. Lebensm. Unters. Forsch. A* **1998**, *206*, 151−155.

(3) Jeandet, P.; Bessis, R.; Sbaghi, M.; Meunier, P.; Trollat, P. Resveratrol content of wines of different ages: Relationship with fungal disease pressure in the vineyard. *Am. J. Enol. Vitic*. **1995**, *46*, 1−4.

(4) Pezet, R.; Cuenat, P. Resveratrol in wine: Extraction from skin during fermentation and post-fermentation standing of must from Gamay grapes. *Am. J. Enol. Vitic.* **1996**, *47*, 287−290.

(5) Threlfall, R. T.; Morris, J. R.; Mauromoustakos, A. Effect of variety, ultraviolet light exposure, and enological methods on the *trans*-resveratrol level of wine. *Am. J. Enol. Vitic*. **1999**, *50*, 57−64.

(6) González-Candelas, L.; Gil, J. V.; Lamuela-Raventós, R. M.; Ramón, D. The uses of transgenic yeasts expressing to gene encoding to glucosyl-hydrolase as a tool to increase resveratrol content in wine. *Int. J. Food Microbiol*. **2000**, *59*, 179−183.

(7) Vacca, V.; Leccis, L.; Fenu, P.; Pretti, L.; Farris, G. A. Wine yeasts and resveratrol content. *Biotechnol. Lett*. **1997**, *19*, 497−498.

(8) Vrhovsek, U.; Wendelin, S.; Eder, R. Effects of various vinification techniques on the concentration of cis- and trans-resveratrol and resveratrol glucoside isomers in wine. *Am. J. Enol. Vitic*. **1997**, *48*, 214−219.

(9) Argilés, J. P.; López-Soriano, J. Resveratrol, a magic molecule? The grape, the wine and their beneficent effects about the health. *Sci. World* **1998**, 192.

(10) Zapata, J. M.; Calderón, A.; Ros Barceló, A. *p*-Hydroxybenzoic acids as inhibitors of the grapevine peroxidase oxidation of 4-hydroxystilbene. *Vitic. Enol. Sci*. **1994**, *49*, 114−116.

(11) Mattivi, F. Solid phase extraction of *trans*-resveratrol from wines for HPLC analysis. *Z. Lebensm. Unters. Forsch.* **1993**, *196*, 522−525.

(12) Martínez, M. V.; García, M. C.; Troncoso, A. M. Factores que afectan a los niveles de resveratrol en el vino. *Aliment., Equip. Tecnol.* **2000**, *2*, 113−118.

(13) Barlass, M.; Miller, R. M.; Douglas, T. J. Development of methods for screening grapevines for resistance to infection by downy mildew. II. Resveratrol production. *Am. J. Enol. Vitic*. **1987**, *38*, 65−68.

(14) Bavaresco, L.; Petegolli, D.; Cantú, E.; Fregoni, M.; Chiusa, G.; Trevisan, M. Elicitation and accumulation of stilbene phytoalexins in grapevine berries infected by *Botrytis cinerea*. *Vitis* **1997**, *36*, 77−83.

(15) Jeandet, P.; Bessis, R.; Sbaghi, M.; Maume, B. F.; Meunier, P.; Peyron, D.; Trollat, P. Effect of enological practices on the resveratrol isomer content of wine. *J. Agric. Food Chem*. **1995**, *43*, 316−319.

(16) Celotti, E.; Ferrarini, R.; Zironi, R.; Be, L. S. Resveratrol content of some wines obtained from dried Valpolicella grapes: Recioto and Amarone. *J. Chromatogr. A* **1996**, *730*, 47−52.

(17) Lamuela Raventós, R. M.; Rosemary, A. I.; Waterhouse, A. L.; De la Torre, C. Direct HPLC analysis of *cis*- and *trans*-resveratrol and piceid isomers in Spanish red *Vitis vinifera* wines. *J. Agric. Food Chem*. **1995**, *43*, 281−283.

(18) Bessis, R.; Blache, D.; Adrián, M.; Breuil, B. C., Boudon, E.; Jeandet, P. Typicité du Pinot Noir. Le resvératrol dans les phénomènes de défense naturelle de la vigne et de santé de consommateurs. Relations avec le terroir de Bourgogne. *Rev. Fr. Oenol.* **1998**, 170.

(19) Schouten, A.; Wagemakers, L.; Stefanato, F.; van der Kaaij, R. M.; van Kan, J. A. L. Resveratrol acts as a natural profungicide and induces selfintoxication by a specific laccase. *Mol. Microbiol.* **2002**, *43*, 883−894.

(20) García Luján, A. *La Viticultura del Jerez*; Mundi-Prensa: Madrid, Spain 1997.

(21) Piérni, R. Sherry, the heart of Spanish soul—Part I. *Mattoni Grand Drink* **2002**, *11*, 1−3.

(22) Office International of the Vigne et du Vin. *Recueil des Methodes Internationales d'Analysis des Vin*; OIV: Paris, France, 1978.

(23) Möllering, H.; Bergmeyer, H. U. *Methods of Enzymatic Analysis*, 3rd ed.; 1984; Vol. VI, pp 220−227.

(24) Roggero, J. P. Wine phenolics analysis via direct HPLC injection. *Am. Lab. News* **1997**, Jan.

(25) Palacios, V. M.; Nebot, E.; Pérez, L. Uses of factor analysis for the characterization and modelling of maturation of *Palomino fino* grapes in the Jerez region. *Am. J. Enol. Vitic*. **1997**, *48*, 317−322.

(26) Fregoni, M.; Iacono, F.; Zamboni, M. Influence du *Botrytis cinerea* sur les caracteristiques physico-chimiques du raisin. *Bull. OIV* **1986**, *667*−*668*, 996−1013.

(27) Pérez, L.; Valcarcel, M. J.; González, P.; Domecq, B. Influence of *Botrytis* infection of the grapes on the biological aging process of fine sherry. *Am. J. Enol. Vitic*. **1991**, *42*, 58−62.

(28) Pineau, J. Modification de la composition du raisin parn attaque du *Botrytis cinerea*. *Rev. Fr. Oenol*. **1987**, *108*, 17−20.
(29) Ravji, R. G. Glycerol production by four common grape molds. *Am. J. Enol. Vitic*. **1988**, *39*, 77−82.
(30) Ribereau-Gayon, P.; Dubordieu, D.; Donèche, B.; Lonvaud, A. *Traité d'oenologie. 1. Microbiologie duvin Vinifications*; Dunod: Paris, France, 2000.
(31) Hoos, G.; Blaich, R. Metabolism of stilbene phytoalexins in grapevines: Oxidation of resveratrol in sail-cell cultures. *Vitis* **1988**, *27*, 1−12.
(32) Hoos, G.; Blaich, R. Influence of resveratrol on germination of conidia and micelyal growth of *Botrytis cinerea* and *Phomopsis viticola*. *J. Phytopathol*. **1990**, *129*, 102−110.
(33) Sotheeswaran, S.; Pasupathy, V. Distribution of resveratrol oligomers in plants. *Phytochemistry* **1993**, *32*, 1083−1092.
(34) Romero, A. I.; Lamuela-Raventós, R. M.; Adrés-Lacueva, C.; de la Torre-Boronat, M. Method for the quantitative extraction of resveratrol and piceid isomers in grape berry skins. Effect of powdery mildew on the stilbene content. *J. Agric. Food Chem*. **2001**, *49*, 210−215.

Received for review July 15, 2002. Revised manuscript received November 11, 2002. Accepted November 11, 2002.

JF020774U