Aroma Potential of Two Bairrada White Grape Varieties: Maria Gomes and Bical

S. Rocha,*,† P. Coutinho,† A. Barros,† M. A. Coimbra,† I. Delgadillo,† and A. Dias Cardoso‡

Departamento de Química, Universidade de Aveiro, 3810-193 Aveiro, Portugal, and Estação Vitivinícola da Bairrada, Apartado 7, 3781 Anadia Codex, Portugal

Maria Gomes and Bical are the main white grape varieties in Portuguese Bairrada Appellation, which represent 80% and 15%, respectively, of white vineyard. To estimate their aroma potentialities, free and potential volatile components from the musts were examined. The free volatile components were extracted using a liquid−liquid continuous method and were analyzed by gas chromatography−mass spectrometry. The potential volatile compounds were determined after elimination of the free components by heat and enzymatic treatments. Principal component analysis was used to establish relations between the compounds and the varieties and also the form (free or in potential). Maria Gomes has 11.5 mg/L volatile compounds, of which 33% are in free form; Bical has 4.4 mg/L volatile compounds, of which 46% are in free form. A total of 59 compounds was identified and quantified. In Maria Gomes, the sum of the terpenoids is within the perception limits for hotrienol (0.21 mg/L) and linalool (0.20 mg/L). In Bical, benzyl alcohol and phenylethylethanol represent 20% of the volatile compounds. Considering that the volatile composition pattern of Maria Gomes and Bical varieties are different, wine-making technologies should be developed specifically for each variety.

Keywords: *Maria Gomes variety; Bical variety; must; free volatile compounds; potential volatile compounds; PCA*

INTRODUCTION

The aroma is one of the most important factors in determining wine character and quality. Several studies recognized a relation between the wine character and the grape and musts volatile compounds, namely terpenoids (Cordonnier and Bayonove, 1974; Gunata et al., 1985; Strauss et al., 1986; Wilson et al., 1986). Volatile compounds appear in the free or glycosidically linked (or both) forms. The study of the volatile components originating from the nonvolatile precursors has been the object of several investigations (Cordonnier and Bayonove, 1974; Williams et al., 1982a,b; Gunata et al., 1985; Voirin et al., 1992a,b). These precursors have been reported as glycosides having the aroma compounds as their aglycons. They may be released by acid or enzymatic treatments (Gunata et al., 1990; Razungles et al., 1993).

Bound aroma, potentially developed during wine making, is unknown for the majority of grape cultivar appellations. French varieties such as Chardonnay (Sefton et al., 1993; Arrhenius et al., 1996), Cabernet Sauvignon (Gómez et al., 1995), and Muscat (Carro-Mariño et al., 1995), German varieties such as Riesling (Skouroumounis and Winterhalter, 1994), and several Spanish varieties (Versini et al., 1995; López-Tamames et al., 1997) have been studied to establish databases of flavor compounds. Bairrada is one of the ancient wine-making regions in Portugal, and its characteristics were recently (1979) recognized in the definition of the Bairrada Appellation. Maria Gomes and Bical are the main white grape varieties in Bairrada, which represent 80 and 15%, respectively, of white vineyard; nevertheless the aroma composition of these varieties is not yet fully characterized. Maria Gomes is a variety that exists spread throughout the Portuguese Appellations, where it is also known by the names “Fernão Pires” and “Gaeiro”. To estimate the aroma potentialities of these varieties, the free and potential volatile components (PVC) from the musts were examined using liquid−liquid continuos extraction and gas chromatography coupled with mass spectrometry (GC-MS).

MATERIALS AND METHODS

Materials. *Vitis vinifera* var. healthy-state Maria Gomes and Bical grapes from the 1998 harvest were collected in the Bairrada Appellation, and the musts production (microvinification) was carried out in the Estação Vitivinícola da Bairrada, Anadia. The musts were treated with SO_2 (60 mg/L), followed by the skin contact process for 6 h at room temperature, and were clarified by centrifugation. The samples were stored at −20 °C until use.

Heat Treatment and Enzyme Hydrolysis. To inactivate the endogenous enzymes and to eliminate the free volatile components, the musts of each variety were heated under stirring for 15 min at 100 °C (Cordonnier and Bayonove, 1974). A commercial enzymatic preparation usually used in white wine making, Lallzyme de Lalvin (14.1 mg), obtained from Lallemand/Proenol (Vila Nova de Gaia, Portugal), was added to 250 mL of must, and the mixture was incubated at 35 °C for 24 h. This enzymatic preparation is reported by the producer to have activity of β-glucosidase, pectinase, arabinosidase, and rhamnosidase. After incubation, the released aglycons were extracted as described below.

Extraction Method. The extraction procedure was a modification of the method described by Eteviant (1987). The musts without and with heat and enzymatic treatments were

* Author to whom correspondence should be addressed (telephone + 351 234370718; fax + 351 234370084; e-mail scarrico@dq.va.pt).

† Universidade de Aveiro.

‡ Estação Vitivinícola da Bairrada.

10.1021/jf000175s CCC: $19.00
Published on Web 08/15/2000

submitted to a process of liquid−liquid continuos extraction with dichloromethane. Six independent extractions were done for each one of the four must samples, in a total of 24 extractions. The must (250 mL), supplemented with internal standard (130 and 65 ng of 3-octanol to Maria Gomes and Bical varieties, respectively), and 75 mL of dichloromethane were placed in the extractor. Extractions were carried out for 24 h at ca. 50 °C. The dichloromethane extracts were cooled to −20 °C to separate the frozen water from the organic phase by decantation and then dried over anhydrous sodium sulfate. The excess of low-boiling solvent was removed by distillation at low pressure using a trap with liquid nitrogen. The concentrate (about 1 mL) was stored in a glass screw-top vial at −20 °C.

GC-MS. The must extracts were analyzed by GC-MS on a Hewlett-Packard 5890 series II gas chromatograph, equipped with a 30 m × 0.32 mm (i.d.) DB-FFAP fused silica capillary column, connected to a Hewlett-Packard mass selective detector, according to the method described by Rocha et al. (1996). Splitless injections were used. The oven temperature was programmed from 35 to 220 °C at 2 °C/min, the injector temperature was 255 °C, and the transfer line was heated at 250 °C. Helium carrier gas had a column head pressure of 12 psi. The mass spectrometer was operated in the electron impact mode at 70 eV, scanning the range *m*/*z* 30−300 in a 1-s cycle. Identification of volatile compounds was achieved by comparison of the GC retention times and mass spectra with those, when available, of the pure standard compounds. All mass spectra were also compared with those of the data system library (Wiley 275) and other published spectra (*Eight Peak Index of Mass Spectra*, 1974). Estimated concentrations for all compounds were made by peak area comparisons with the area of a known amount of internal standard (3-octanol). The reproducibility of the extracts was expressed as coefficient of variation (CV) in Table 1.

Principal Component Analysis (PCA). A PCA was applied to the normalized areas of the 59 compounds identified by GC-MS (Maria Gomes and Bical varieties, both in free form and as PVC, in a total of four samples, each with six extraction replicates). PCA, as an exploratory technique, allows one to study the main sources of variability present in the data sets, to detect clustering formation, and to establish relations between samples (objects) and compounds (variables) (Jolliffe, 1986).

RESULTS AND DISCUSSION

Table 1 shows the volatile composition of the Maria Gomes and Bical musts, and its different distribution in the free form and in potential. The PVC were determined after elimination of the free components by heat treatment followed by enzymatic treatment. The PVC fraction contains the glycosidically linked components released by the enzymes plus the compounds produced by the heat treatment (70 °C, 15 min) at the must pH (3.2) and also those compounds that arise from the thermal degradation of sugars. Some of these compounds are known to be developed during the wine making and aging, contributing to the final wine aroma (Cordonnier and Bayonove, 1978; Williams et al., 1980; Strauss et al., 1987). This fact is particularly interesting to the neutral varieties such as these under study.

The free volatile compounds isolated from Maria Gomes accounted for 3.84 mg/L, a value higher than that obtained for Bical (2.04 mg/L). These values do not include the furan-derived compounds because the majority of furans are products of sugar degradation (Tu et al., 1992) due to the thermal treatment. These compounds are higher in Maria Gomes than in Bical, which is consistent with the higher amount of sugars present in former musts. For these reasons these compounds were not considered. Both varieties contain aliphatic and aromatic alcohols, ketones, terpenoid compounds, aliphatic acids, and C_{13} norisoprenoids. Because of the considerable significance of volatile monoterpenes to flavor and varietal character of *V. vinifera* varieties (Strauss et al., 1986), particular attention was devoted to these compounds. Alcohols were also the object of particular considerations because, quantitatively, the alcohol fraction was the main chemical group present in the musts.

Free Alcohol and Terpenoid Volatile Compounds. The composition of terpenoid fraction was different in the two varieties studied: the must of Maria Gomes contains 11 terpenoids that represent 1.10 mg/L, and the must of Bical exhibits 8 terpenoids, corresponding to 0.23 mg/L. Quantitatively, the terpenols (3,7-dimethylocta-1,5-dien-3,7-diol, 2,6-dimethylocta-2,7-dien-1,6-diol, 3,7-dimethylocta-1-en-3,7-diol, linalool, hotrienol, and geraniol) are the main terpenoids present in Maria Gomes musts; the terpendiols (3,7-dimethylocta-1,5-dien-3,7-diol and 2,6-dimethylocta-2,7-dien-1,6-diol) are the main terpenoids present in Bical. Many of these compounds, such as geraniol and linalool, are fragrant and are doubtless important to the general enhancement of the floral and fruity aromas (Marais, 1983). The hotrienol and linalool have been reported as having a determinant role in the wine aroma profile due to their aroma properties and low sensorial perception level (Simpson, 1979; Marais, 1983). The 3,7-dimethylocta-1,5-dien-3,7-diol was the dominant monoterpene of Maria Gomes and Bical musts; although odorless, it can represent a major potential source of grape flavor as precursors of flavorants such as hotrienol (Marais, 1983; Wilson et al., 1984). For both varieties, all free terpenoid compounds determined are under the limits of perception, reported by Marais (1983).

The alcohol fraction is the major one, although its composition is different in the two varieties. This fraction is composed mainly by n-alcohols of C_6 chain length and aromatic compounds such as benzyl alcohol and phenylethylethanol. The aliphatic alcohols were more abundant in the must from Maria Gomes (84% of the alcohols extracted), whereas the aromatic alcohols were present in higher amount in the must from Bical (52% of the alcohols extracted). The presence of benzyl alcohol and phenylethylethanol may cause the sweet and flowery notes (Belitz and Grosch, 1999), what could be considered as a positive characteristic for the Bical variety. C_6 alcohols have herbaceous and greasy odors, which seem related to deleterious effect in the wine (Baumes et al., 1986; Cordonier, 1989), although, in white wines, a small herbaceous perception is appreciated by some consumers (Dubois, 1994). Their origin was reported as being related mainly to the lipooxygenase activity of the grape (Cordonier, 1989) or must aeration (Cordonnier and Bayonove, 1978). C_6 alcohols accounts for 18% of total volatile compounds of Maria Gomes and Bical, which represents 725 μg/L in Maria Gomes musts and 383 μg/L in Bical musts. These values indicate, mainly for the Maria Gomes variety, that attention should be paid to avoid the deleterious effect associated with the presence of these components. The more abundant thiol in the musts is 4-methyl-5-thiazoethanol. It accounts for 15% of total free aroma compounds in Maria Gomes and 3% in Bical.

Alcohol and Terpenoid Volatile Compounds in Potential Form. The PVC are 67% of the total volatile compounds in Maria Gomes (7.66 mg/L) and 54% in

Table 1. Free and Potential Volatile Components Identified in Dichloromethane Extracts of Maria Gomes and Bical Musts, Grouped by Chemical Classes

peak	compound	ident.[a]	concentration[b] (μg/L)			
			MG_F	MG_{PVC}	BIC_F	BIC_{PVC}
		Terpenoids				
6	*trans*-linalool oxide	A, B, C	- - -	25.7 (7)	- - -	- - -
8	*cis*-linalool oxide	A, B, C	- - -	36.2 (4)	16.2 (17)	- - -
15	linalool	A, B, C	64.2 (3)	133.0 (9)	6.7 (19)	17.3 (27)
20	hotrienol	B, C	56.6 (12)	152.9 (8)	9.9 (10)	20.9 (8)
23	α-terpineol	A, B, C	22.4 (5)	148.6 (3)	3.2 (6)	5.5 (4)
25	linalool *E*-pyranic oxide	B, C	23.9 (5)	23.6 (6)	3.8 (9)	1.6 (8)
27	linalool *Z*-pyranic oxide	B, C	10.1 (3)	13.3 (8)	- - -	- - -
31	nerol	A, B, C	- - -	18.1 (2)	- - -	- - -
33	geraniol	A, B, C	53.6 (3)	66.8 (4)	- - -	- - -
38	3,7-dimethylocta-1,5-dien-3,7-diol	B, C	621.8 (3)	249.3 (6)	123.4 (4)	41.8 (3)
39	3,7-dimethylocta-1-en-3,7-diol	B, C	94.4 (3)	234.3 (5)	- - -	- - -
43	3,7-dimethylocta-1,7-dien-3,6-diol	B, C	23.5 (5)	53.9 (7)	- - -	- - -
50	2,6-dimethylocta-2,7-dien-1,6-diol	B, C	95.2 (8)	93.5 (6)	48.8 (8)	55.0 (8)
52	farnesol	A, B, C	35.5 (7)	85.2 (9)	17.0 (6)	tr.
	subtotal (μg/L)		1101.2	1334.4	229.0	142.1
	subtotal (%)[c]		28.7	17.4	11.3	6.1
		Alcohols				
2	1-hexanol	A, B, C	447.7 (1)	190.5 (5)	203.9 (1)	11.4 (5)
3	*trans*−3-hexen-1-ol	A, B, C	15.5 (4)	9.5 (4)	10.7 (3)	tr.
4	*cis*−3-hexen-1-ol	A, B, C	16.7 (2)	12.5 (6)	13.6 (1)	2.4 (7)
5	*trans*−2-hexen-1-ol	A, B, C	245.6 (2)	160.2 (3)	154.8 (1)	28.6 (2)
12	2-(methylthio)ethanol	B	- - -	- - -	1.2 (9)	1.3 (9)
14	(D,L)-2,3-butanediol	A, B, C	29.9 (9)	414.2 (6)	15.8 (14)	38.3 (13)
18	(*R*,*S*)-2,3-butanediol	A, B, C	73.6 (6)	477.0 (2)	52.9 (9)	102.7 (4)
24	methionol	A, B, C	13.3 (8)	21.4 (4)	26.7 (6)	30.6 (5)
29	2-(2-butoxyethoxy)ethanol	B	27.0 (5)	34.7 (10)	5.3 (3)	4.0 (7)
34	benzyl alcohol	A, B, C	78.0 (3)	142.2 (8)	153.4 (4)	149.7 (6)
35	phenylethylethanol	A, B, C	191.2 (3)	267.8 (5)	298.2 (5)	255.3 (7)
45	4-vinyl-2-methoxyphenol	A, B, C	- - -	- - -	128.6 (8)	392.6 (5)
48	4-methyl-5-thiazoethanol	B	577.6 (9)	586.0 (6)	51.7 (5)	122.1 (11)
	subtotal (μg/L)		1716.1	2316.0	1112.8	1135.0
	subtotal (%)[c]		44.7	30.2	54.6	48.3
		Acids				
7	acetic acid	A, B, C	99.3 (8)	467.3 (6)	114.6 (8)	201.8 (5)
13	propanoic acid	A, B, C	- - -	- - -	2.2 (7)	3.6 (5)
16	isobutyric acid	A, B, C	22.0 (6)	39.4 (5)	16.2 (9)	20.3 (9)
21	butyric acid	A, B, C	- - -	- - -	3.2 (9)	7.8 (8)
32	hexanoic acid	A, B, C	115.0 (3)	157.2 (4)	50.4 (5)	43.1 (9)
37	*trans*-2-hexanoic acid	A, B, C	- - -	128.8 (9)	- - -	48.8 (4)
42	octanoic acid	A, B, C	40.9 (4)	43.4 (4)	31.5 (5)	22.9 (7)
44	nonanoic acid	A, B, C	- - -	- - -	18.4 (6)	17.1 (6)
47	decanoic acid	A, B, C	- - -	- - -	51.1 (8)	18.9 (3)
55	dodecanoic acid	A, B, C	49.7 (7)	134.6 (9)	32.7 (9)	20.5 (9)
	subtotal (μg/L)		326.9	970.7	320.3	404.8
	subtotal (%)[c]		8.5	12.7	15.7	17.2
		Ketones				
1	3-hydroxy-2-butanone	A, B, C	155.7 (2)	389.1 (7)	148.9 (2)	152.4 (6)
19	γ-butyrolactone	A, B, C	100.1 (4)	365.1 (3)	71.8 (5)	136.6 (5)
28	tetrahydro-2H-pyran-2-one	B	7.4 (3)	19.9 (4)	7.4 (8)	11.4 (6)
40	3-hydroxy-2-methyl-4H-pyran-4-one	B, C	tr.	25.4 (7)	- - -	- - -
41	2-pyrrolidinone	B	32.8 (6)	60.6 (6)	16.6 (8)	29.1 (9)
46	2,3-dihydro-3,5-dihydroxy-6-methyl-4H-pyran-4-one	B, C	31.4 (8)	1252.2 (2)	- - -	208.3 (9)
49	4-(1-hydroxyethyl)-γ-butanolactone	B, C	152.9 (2)	345.2 (4)	122.6 (3)	113.7 (11)
57	tetrahydro-4-hydroxy-4-methyl-2H-pyran-2-ona	B, C	77.0 (6)	242.4 (7)	- - -	- - -
	subtotal (μg/L)		557.3	2699.9	367.3	651.5
	subtotal (%)[c]		14.5	35.2	18.0	27.7
		C_{13} Norisoprenoids				
10	vitispyrane	A, B, C	- - -	12.1 (5)	- - -	5.4 (11)
30	β-damascenone	A, B, C	94.6 (2)	17.1 (8)	6.7 (8)	5.9 (7)
59	dihydro-β-ionone	B, C	23.9 (7)	93.4 (4)	- - -	- - -
	subtotal (μg/L)		118.5	122.6	6.7	11.3
	subtotal (%)[c]		3.1	1.6	0.3	0.5
		Furans				
9	furfural	A, B, C	tr.	159.9 (2)	- - -	40.1 (3)
17	5-methylfurfural	A, B, C	tr.	62.2 (5)	- - -	- - -
22	furfuryl alcohol	A, B, C	13.7 (5)	53.8 (9)	15.0 (8)	23.5 (7)
26	2-(5H)-furanone	B, C	11.7 (5)	40.1 (4)	10.1 (6)	17.6 (7)
51	1 (3H)-isobenzofuranone	B	69.5 (6)	213.6 (6)	- - -	- - -
53	2,3-dihydrobenzofuran	B, C	66.0 (9)	596.9 (8)	38.0 (4)	383.2 (9)
54	2-furancarboxylic acid	B, C	tr.	85.3 (8)	- - -	- - -
56	5-hydroxymethylfurfural	A, B, C	tr.	12350.5 (3)	38.7 (9)	3579.5 (9)
	subtotal (μg/L)		160.9	13562.3	101.8	4043.9

Table 1 (Continued)

peak	compound	ident.[a]	concentration[b] (μg/L) MG_F	MG_{PVC}	BIC_F	BIC_{PVC}
		Others				
11	benzaldehyde	A, B, C	10.6 (2)	19.4 (8)	2.2 (8)	3.9 (7)
36	benzothiazole	A, B, C	8.7 (6)	tr.	tr.	tr.
58	vanillin	A, B, C	tr.	197.5 (8)	tr.	tr.
	subtotal (μg/L)		19.3	229.0	2.2	9.3
	subtotal (%)[c]		0.5	2.9	0.1	0.2
	TOTAL (μg/L)		4000.2	21222.8	2140.1	6392.5
	TOTAL without furans (μg/L)		3839.3	7660.5	2038.3	2348.6

MG_F free volatile components of Maria Gomes must; MG_{PVC} potential volatile components of Maria Gomes must; BIC_F free volatile components of Bical must; BIC_{PVC} potential volatile components of Bical must. [a] The reliability of the identification or structural proposal is indicated by the following: A mass spectrum and retention time consistent with those of an authentic standard; B structural proposals are given on the basis of mass spectral data (Wiley 275); C mass spectrum consistent with spectra found in the literature. [b] Mean of six replicates; numbers in parentheses correspond to the CV (%). [c] Furans were not considered.

Bical (2.35 mg/L). These values do not include the furan-derived compounds. The major classes of PVC in Maria Gomes are ketones (35.2%), alcohols (30.2%), terpenoids (17.4%), and acids (12.7%); in Bical are alcohols (48.3%), ketones (27.7%), acids (17.2%), and terpenoids (6.1%). For the two varieties, the amount of PVC is higher than the corresponding free forms.

In Maria Gomes musts, the amount of PVC terpenoids is 21% higher than the amount of terpenoids in free form. Conversely, the level of PVC terpenoids of Bical is 61% lower. As observed in the free form, the PVC terpendiols are the major terpenoid component of the musts (26% of total terpenoids in Maria Gomes and 68% in Bical). The terpendiol 3,7-dimethylocta-1-en-3,7-diol is significant in Maria Gomes variety, although it is absent in Bical. The PVC levels of linalool, hotrienol, and α-terpeniol are higher than in the free forms. Maria Gomes contains twice more linalool, three times more hotrienol, and seven times more α-terpeniol. The *trans*-linalool oxide and *cis*-linalool oxide are present mainly in the PVC fractions. The terpenoid oxide exhibits a higher sensorial perception level compared with terpenols. The presence of a higher amount of terpenoids in the PVC fraction compared with the free one shows the potential properties of Maria Gomes variety due to the significant importance of terpenoids in the aroma as well as their role as precursors of other aroma compounds (Williams et al., 1980; Marais, 1983; Strauss et al., 1988). In Maria Gomes variety, the sum of the free and PVC forms of hotrienol (0.21 mg/L) and linalool (0.20 mg/L) are over the limits of perception for these compounds (0.11 and 0.10 mg/L, respectively—Marais, 1983), which may allow one to infer its contribution to the improvement of the wine aroma quality.

The amount of PVC alcohols are 35% higher than the free alcohols in Maria Gomes; on the other hand, in Bical, the level of free and PVC alcohols is similar. However, it is observed that the level of C_6 PVC alcohols is lower than their free forms, 29 and 89% respectively for Maria Gomes and Bical. The 2,3-butanediol cis and trans isomers are abundant in PVC form in Maria Gomes musts, as they account for 38% of the total alcohols; in Bical, these compounds account only for 12%. However, they seem not to have influence in the sensorial wine properties (Webb et al., 1967; Radler and Zorg, 1986). The level of PVC aromatic alcohols (benzyl alcohol and phenylethylethanol) is 52% higher in Maria Gomes, but in Bical, these compounds are 10% lower. Voirin (1992b) indicates that the presence of aromatic alcohols are associated with the neutral cultivars. The 4-methyl-5-thiazoethanol accounts for 8% of total PVC in Maria Gomes and 5% in Bical. This compound exists in similar amounts in free and PVC forms in Maria Gomes, and in Bical, the amount of PVC doubles the amount of free form. Thiazoles may occur naturally in foods such as tomato and wine or as a result of heat treatment (Belitz and Grosch, 1999). In the case of the musts of the varieties under study, especially Maria Gomes, 4-methyl-5-thiazoethanol seems to be related to its varietal character, as it occurs in significant amounts in the free form and as PVC.

The limit of perception for 1-hexanol, *trans*-2-hexen-1-ol, and *cis*-3-hexen-1-ol is estimated as 4, 15, and 13 mg/L in beer and 4.8, 6.7, and 0.07 mg/L in water, respectively (Dubois, 1994). In Maria Gomes and Bical musts the values are, respectively, 0.64 and 0.22 mg/L for 1-hexanol, 0.41 and 0.18 mg/L for *trans*-2-hexen-1-ol, and 0.03 and 0.02 mg/L for *cis*-3-hexen-1-ol. These concentrations are within the limits expected to occur in wines and are not expected to have any negative sensorial contribution to the wine aroma; nevertheless the levels of these compounds need to be kept under control.

Other Compounds. The odor threshold value for ketones is substantially higher than for other compounds studied; thus this chemical group was not the object of an exhaustive study. Considering the two varieties, the amount of free ketones was lower than their PVC amount, due mainly to 2,3-dihydro-3,5-dihydroxy-6-methyl-4H-pyran-4-one, a product usually associated with thermal degradation (Belitz and Grosch, 1999). The major norisoprenoid present in the musts is β-damascenone (111.7 μg/L in Maria Gomes and 12.6 μg/L in Bical). Because of its very low sensorial threshold of 0.002 μg/L in water (Betitz and Grosch, 1999), this compound seems to be important for these must aroma characteristics. In Maria Gomes, β-damascenone is 85% in free form; however, in Bical, the amount of free form is comparable with the amount of the PVC. Vitispyrane is only found as a PVC, both in Maria Gomes (12.1 μg/L) and Bical (5.4 μg/L). These values are lower than the sensorial odor threshold in water, estimated as 800 μg/L (Rapp and Pretorius 1990; Belitz and Grosch, 1999). Dihydro-β-ionone was only detected in Maria Gomes, mainly as PVC (80%). C_{13} norisoprenoids are grape-derived compounds but are usually not present in the free form and arise in juice and wines by hydrolytic degradation of precursor substances (Williams et al., 1982b; Simpson and Miller, 1983; Strauss et al., 1987). This is the reason for the higher amount of vitispyrane and dihidro-β-ionone as PVC.

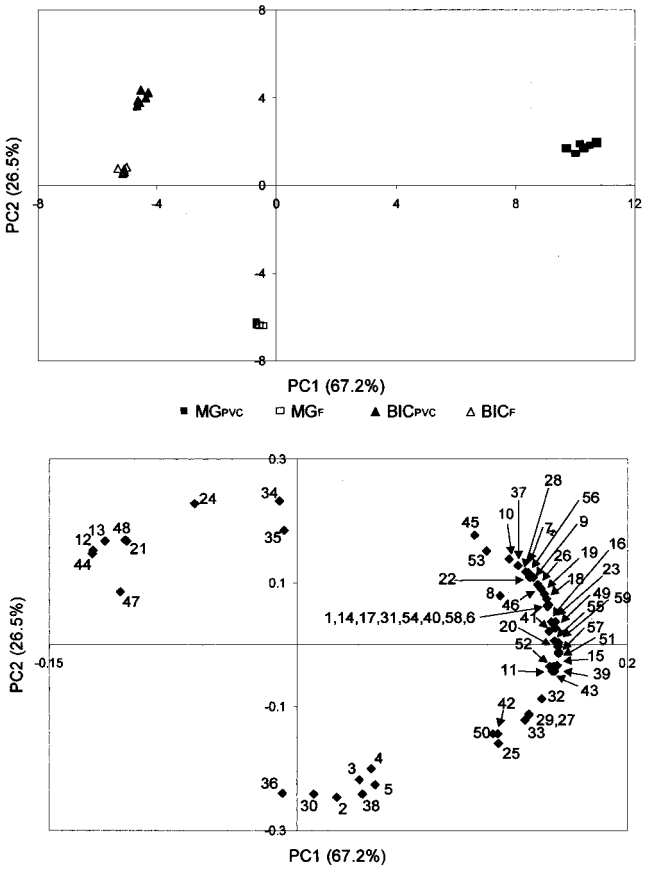

Figure 1. PC1 × PC2 Scatter plot of the main sources of variability between the Maria Gomes and Bical musts. (a) Distinction between the samples MG_F, MG_{PVC}, BIC_F, and BIC_{PVC} (scores); (b) relation between the 59 volatile components (loadings).

PCA. The PCA was used to study the main sources of variability between the different must varieties (in the free and PVC forms), and to establish relations between the varieties (in both forms) and volatile components. Figure 1a shows the scores scatter plot of the two first principal components (which contains 93.7% of the total variability) that represents the distinction among the 24 samples. Figure 1b represents the corresponding loadings plot that establishes the relative importance of each volatile component, and it is therefore useful for the study of relations among the volatile compounds and relations between volatile compounds and samples. The first quadrant contains the PVC of Maria Gomes (MG_{PVC}). These samples are characterized by the furans and pyranones, products of thermal degradation (Table 1, peaks 9, 10, 17, 22, 26, 28, 40, 41, 46, 51, 53, 54, 56−58), terpenoid compounds (6, 8, 15, 20, 23, 25, 27, 31, 33, 39, 43, 50, 52), acids (7, 16, 32, 37, 42, 55), alcohols (14, 18, 29, 45), ketones (1, 19, 49), dihydro-β-ionone (59), and benzaldehyde (11). The free volatile components of Maria Gomes (MG_F) are related to the negative PC2 side. These samples are characterized by C_6 alcohols (2−5), β-damascenone (30), 3,7-dimethylocta-1,5-dien-3,7-diol (38), and benzothiazole (36). Bical samples, both free form and PVC, are represented in the second quadrant. These samples are characterized by the propanoic (13), butyric (21), nonanoic (44), and decanoic (47) acids, the thioalcohols 2-(methylthio)ethanol (12), methionol (24), and 4-methyl-5-thiazoethanol (48), and the aromatic alcohols (34, 35). Furthermore, the analysis of the PC1 × PC3 scores and loadings plots (data not shown) allows one to relate nonanoic and decanoic acids, 2-(methylthio)ethanol (12), and phenylethylethanol (35) with the free volatile components of Bical (BIC_F) and 4-methyl-5-thiazoethanol and propanoic and butyric acids with the PVC of Bical (BIC_{PVC}).

Conclusions. The results of this work show that Maria Gomes and Bical varieties have different aroma profiles. Maria Gomes has a higher amount of volatile compounds and in higher concentration than Bical. The sum of the terpenoids in free form and as PVC is beyond the perception limit for hotrienol and linalool. Furthermore, the occurrence of odorless terpendiols allows us to infer that this variety might be an important one if new strategies of wine-making technology are used, such as enzymatic treatments to release the aroma components. The presence of aromatic alcohols, both in free form and as PVC, may be an interesting characteristic of the musts from Bical. These properties may profit the development of an adequate wine-making technology to release these flowery and sweet compounds and increase the aroma quality. Furthermore, as a consequence of the fact that the Maria Gomes and Bical varieties exhibit different volatile composition patterns, wine-making technologies should be developed specifically for each variety.

LITERATURE CITED

Arrhenius, S. P.; McCloskey, L. P.; Sylvan, M. Chemical markers for aroma of *Vitis vinifera* var. Chardonnay regional wines. *J. Agric. Food Chem*. **1996**, *44*, 1085−1090.

Baumes, R.; Cordonnier, R.; Nitz, S.; Drawert, F. Identification and determination of volatile constituents in wines from different wine cultivars. *J. Agric. Food Chem*. **1986**, *37*, 927−943.

Belitz, H.-D.; Grosch, W. *Food Chemistry*; Springer-Verlag: Berlin, 1999.

Carro-Mariño, N.; López-Tamames, E.; García-Jarés, M. C. Contribution to the study of the aromatic potential of three Muscat *Vitis vinifera* varieties: Identification of new compounds. *Food Sci. Technol. Int*. **1995**, *1*, 105−116.

Cordonnier, R. Mécanismes et facteurs de formation des composés à flaveurs herbacées (Mechanisms and factors of formation of herbaceous flavor compounds). *Rev. Oenol*. **1989**, *53S*, 25−27.

Cordonnier, R.; Bayonove, C. Mise en évidence dans le baie de raisin, variété muscat d'Alexandrie, de monoterpènes liés révélables par une ou plusieurs enzymes du fruit (Highlight, in muscat d'Alexandrie grapes, of bound monoterpens released by one or more fruit enzymes). *C. R. Acad. Sci*. **1974**, *278*, 3387−3390.

Cordonnier, R.; Bayonove, C. Les composants variétales et préfermantaires de l'arôme des vins (Varietal and prefermentative components of the wine aroma). *Parfum. Cosmet. Arômes* **1978**, *24*, 67−77.

Dubois, P. Les arômes des vins et leurs défauts (Wines aromas and their defects). *Rev. Fr. Oenol*. **1994**, *145*, 27−40.

Eight Peak Index of Mass Spectra, 2nd ed.; The Mass Spectra Data Centre: Nottingham, U.K., 1974.

Eteviant, P. X. Mise au point sur les techniques d'extraction et séparation des constituants volatiles du vin (Improvement of extraction and separation techniques of wine volatile components). *Connais. Vigne Vin* **1987**, *21*, 247−265.

Gómez, E.; Martínez, A.; Laencina, J. Changes in volatile compounds during maturation of some grape varieties. *J. Sci. Food Agric*. **1995**, *67*, 229−233.

Gunata, Y. Z.; Bayonove, C. L.; Baumes, R. L.; Cordonier, R. E. The aroma of grapes. Localization and evolution of free and bound fractions of some grape aroma components cv. Muscat during first development and maturation. *J. Sci. Food Agric*. **1985**, *36*, 857−862.

Gunata, Y. Z.; Bayonove, C. L.; Tapiero, C.; Cordonier, R. E. Hydrolysis of grape monoterpenyl β-D-glucosides by various β-glucosidases. *J. Agric. Food Chem*. **1990**, *38*, 1232−1236.

Jolliffe, I. T. *Principal Component Analysis*; Springer-Verlag: New York, 1986.

López-Tamames, E.; Carro-Mariño, N.; Gunata, Y. Z.; Sapis, C.; Baumes, R.; Bayonove, C. Potential aroma in several varieties of Spanish grapes. *J. Agric. Food Chem*. **1997**, *45*, 1729−1735.

Marais, I. Terpenes in the aroma of grapes and wines: a review. *S. Afr. J. Enol. Vitic*. **1983**, *4*, 49−60.

Radler, F.; Zorg, J. Characterization of the enzyme involved in formation of *z*-butanol from meso-2,3-butanodiol by lactic bacteria. *Am. J. Enol. Vitic*. **1986**, *37*, 206−210.

Rapp, A.; Pretorius, P. I. Foreign and undesirable flavours in wine. In *Flavours and Off-flavours*; Charalambous, G., Ed.; Elsevier: Amsterdam, 1990.

Razungles, A.; Gunata, Z.; Pinatel, S.; Baumes, R.; Bayonove, C. Etude quantitative de composés terpéniques, norisoprénoïdes et de leurs précurseurs dans diverses variétés de raisins (Quantitative studies on terpenes, norisoprenoids and their precursors in several varieties of grapes). *Sci. Aliments* **1993**, *13*, 59−72.

Rocha S.; Delgadillo, I.; Ferrer-Correia, A. J. GC-MS study of volatiles of normal and microbiologically attacked cork from *Quercus suber* L. *J. Agric. Food Chem*. **1996**, *44*, 865−871.

Skouroumounis, G. K.; Winterhalter, P. Glycosidically bound norisoprenoids from *Vitis vinifera cv*. Riesling. *J. Agric. Food Chem*. **1994**, *42*, 1068−1072.

Sefton, M. A.; Francis, I. L.; Williams, P. J. The volatile composition of Chardonnay juices: A study by flavor precursor analysis. *Am. J. Enol. Vitic*. **1993**, *44*, 359−370.

Simpson, R. F. Some important aroma components of white wine. *Food Technol. Aust*. **1979**, *31*, 516−522.

Simpson, R. F.; Miller, G. C. Aroma composition of aged Riesling wine. *Vitis* **1983**, *22*, 51−63.

Strauss, C. R.; Wilson, B.; Gooley, P. R.; Williams, P. J. *Role of Monoterpenes in Grape and Wine Flavor*; Parliment, T. H., Croteua, R., Eds.; ACS Symposium Series 317; American Chemical Society: Washington, DC, 1986; pp 222−242.

Strauss, C. R.; Wilson, B.; Anderson, R.; Williams, P. J. Development of precursors of C_{13} nor-isoprenoid flavorants in Riesling grapes. *Am. J. Enol. Vitic*. **1987**, *38*, 23−27.

Strauss, C. R.; Wilson, B.; Williams, P. J. Novel monoterpene diols and diol glycosides in *Vitis vinifera* grapes. *J. Agric. Food Chem*. **1988**, *36*, 569−573.

Tu, D.; Xue, S.; Meng, C.; Mansilla, A. E.; Peña, A. M.; Lopez, F. S. Simultaneous determination of 2-furfuraldehyde and 5-(hydroxymethyl)-2-furfuraldehyde by derivative spectrophotometry. *J. Agric. Food Chem*. **1992**, *40*, 1022−1025.

Versini, G.; Orriols, I.; Serra-Adalla, D. Aroma components of Galician Albariño, Loureira and Godello wines. *Vitis* **1995**, *33*, 165−170.

Voirin, S. V.; Baumes, R.; Gunata, Y.; Bitteur, S.; Bayonove, C. Analytical methods for monoterpene glycosides in grape and wine. I. XAD-2 extraction and gas chromatographic−mass spectrometric determination of synthetic glycosides. *J. Chromatogr*. **1992a**, *590*, 313−328.

Voirin, S. V.; Baumes, R. L.; Sapis, J. C.; Bayonove, C. Analytical methods for monoterpene glycosides in grape and wine. II. Qualitative and quantitative determination of monoterpene glycosides in grapes. *J. Chromatogr*. **1992b**, *595*, 269−281.

Webb, A. B.; Kepner, R. E.; Maggiora, L. Sherry aroma. VI. Some volatile components of flor sherry of spanish origin. *Am. J. Enol. Vitic*. **1967**, *18*, 190−199.

Williams, P. J.; Strauss, C. R.; Wilson, B. Hydroxylated linalool derivatives as precursors of volatile monoterpenes of muscat grapes. *J. Agric. Food Chem*. **1980**, *28*, 766−771.

Williams, P. J.; Strauss, C. R.; Wilson, B.; Massy-Westropp, R. A. Novel monoterpene disaccharides glycosides of *Vitis vinifera* grapes and wines. *Phytochemistry* **1982a**, *21*, 2013−2020.

Williams, P. J.; Strauss, C. R.; Wilson, B. Use of C18 reversed-phase liquid chromatography for the isolation of monoterpene glycosides and norisoprenoid precursors from grape juice and wine. *J. Chromatogr*. **1982b**, *235*, 471−480.

Wilson, B.; Strauss, C. R.; Williams, P. Changes in free and glycosidically bound monoterpenes in developing muscat grapes. *J. Agric. Food Chem*. **1984**, *32*, 919−924.

Wilson, B.; Strauss, C. R.; Williams, P. C. The distribution of free and glycosidically bound monoterpenes among skin, juice, and pulp fractions of some white grape varieties. *Am. J. Enol. Vitic*. **1986**, *37*, 107−111.

Received for review February 9, 2000. Revised manuscript received June 21, 2000. Accepted June 26, 2000. We appreciate the financial support of PAMAF (Project 6039), and the Research Unit 62/94, QOPNA. S.R. was supported by a postdoctoral grant from FCT (PRAXIS XXI/BPD/16331/98).

JF000175S