

See discussions, stats, and author profiles for this publication at: <https://www.researchgate.net/publication/10606878>

# Aroma Composition of *Vitis vinifera* Cv. Tannat: the Typical Red Wine from Uruguay

ARTICLE in JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY · SEPTEMBER 2003

Impact Factor: 2.91 · DOI: 10.1021/jf030087i · Source: PubMed

CITATIONS

71

READS

32

7 AUTHORS, INCLUDING:



**Eduardo Boido**

University of the Republic, Uruguay

36 PUBLICATIONS 839 CITATIONS

SEE PROFILE



**Laura Fariña**

24 PUBLICATIONS 779 CITATIONS

SEE PROFILE



**Francisco Carrau**

Facultad de Química, University of the Repu...

52 PUBLICATIONS 1,083 CITATIONS

SEE PROFILE



**Eduardo Dellacassa**

University of the Republic, Uruguay

133 PUBLICATIONS 1,602 CITATIONS

SEE PROFILE

## Aroma Composition of *Vitis vinifera* Cv. Tannat: the Typical Red Wine from Uruguay

EDUARDO BOIDO,<sup>†</sup> ADRIANA LLORET,<sup>†,§</sup> KARINA MEDINA,<sup>†</sup> LAURA FARIÑA,<sup>†,§</sup>  
FRANCISCO CARRAU,<sup>†</sup> GIUSEPPE VERSINI,<sup>#</sup> AND EDUARDO DELLACASSA<sup>\*,§</sup>

Sección Enología and Cátedra de Farmacognosia y Productos Naturales, Facultad de Química,  
Universidad de la República, Avenida General Flores 2124, 11800 Montevideo, Uruguay, and  
Centro Sperimentale, Istituto Agrario di San Michele all'Adige, Via Edmondo Mach 1,  
38010 San Michele all'Adige, Trento, Italy

The free volatiles, as well as those released from the glycosidically bound forms by enzyme hydrolysis, have been analyzed to characterize young Tannat wines from two successive vintages. The Tannat wine showed some aroma profile peculiarities detected in the free forms but, above all, in the bound fraction for the level and profile of the norisoprenoidic fraction. Among the free volatile compounds, a rather low content of C<sub>6</sub> alcohols with a prevalence of *cis*-3-hexen-1-ol on the *trans* form and sometimes a remarkable level of *trans*-2-hexen-1-ol seem to be typical for the variety. C<sub>13</sub>-norisoprenoidic and monoterpenic volatiles made up ~42% of the total level of the volatiles observed in the glycosidase enzyme-released fraction. The other volatiles were C<sub>6</sub> alcohols (6%) and benzenoid compounds (51%). The dominating monoterpene alcohols were the *cis* and *trans* isomers of 3,7-dimethyl-1,6-octadiene-3,8-diol (8-hydroxylinalool). The C<sub>13</sub>-norisoprenoid pattern was composed by 3-hydroxy- $\beta$ -damascone, 3-oxo- $\alpha$ -ionol, vomifoliol, 4-oxo- $\beta$ -ionol, 3-oxo-7,8-dihydro- $\alpha$ -ionol, 4-oxo-7,8-dihydro- $\beta$ -ionol, grasshopper ketone, and 7,8-dihydrovomifoliol.

**KEYWORDS:** *Vitis vinifera* cv. Tannat wine aroma; free and bound forms; monoterpenes; norisoprenoids; shikimate derivatives

### INTRODUCTION

An overview of the amount of *Vitis vinifera* planted in Uruguay shows that vinifera grapes represent 60% of the vineyard (1). Moreover, since the 1970s, Uruguay has been producing fine wines with the *Vitis vinifera* cv. Tannat grape, which, although it originated in the south of France, is today almost unknown in Europe. The Tannat variety was introduced by Pascual Harriague, a Frenchman from the Basque region, who planted a vineyard of this variety in 1870, 400 km north of Montevideo, on the outskirts of the city of Salto (2). Nowadays, Tannat vineyards account for 27% of the area under vinifera grapes (3), Uruguay being the only country in the Americas where this grape is found. For this reason, the Uruguayan wine-making industry has established a strategy to produce Tannat wine using advanced viticultural and wine-making technologies (1).

Considerable research and development on the viticulture and enology of this variety are still necessary, in particular in relation to the chemical aroma composition of the wine. In the present

paper we contribute with the study of the aroma profile composition to a better understanding of the wines obtained exclusively from the Tannat grape and vinified in the traditional manner.

Wine aroma depends on numerous factors, with special importance being given to the variety of grape, vinification, maturation, and aging (4–6). It is well-known that the secondary metabolites of grapes are responsible for the principal aroma compounds in grape must and provide the basis of varietal character (4, 7). Fermentation increases the chemical and aroma complexity of wine by assisting in the extraction of compounds from solids present in the grape must, modifying some grape-derived compounds, and producing a substantial amount of yeast metabolites (8). The chemical compounds responsible for wine aroma are mainly alcohols, esters, acids, aldehydes, and ketones, and, although the same compounds are usually found in the majority of wines, the type of production carried out can cause important changes in their concentrations. These concentration differences may explain the variation between certain types of wines, which although they contain the same specific compounds, possess variations in their quantities.

In this research on the aroma components of wines of the Tannat variety, we focused the attention principally on compounds possibly linked to varietal peculiarities, extending in particular the investigation to isoprenoids and other compounds

\* Author to whom correspondence should be addressed (telephone 00598 2 9244068; fax 00598 2 9241906; e-mail edellac@fq.edu.uy).

<sup>†</sup> Sección Enología, Universidad de la República.

<sup>§</sup> Cátedra de Farmacognosia y Productos Naturales, Universidad de la República.

<sup>#</sup> Istituto Agrario di San Michele all'Adige.

such as monoterpenols and to the bound forms, with the aim to improve the characterization and classification of young Tannat wines.

## MATERIALS AND METHODS

**Sample Preparation.** Tannat wines of different vintages from experimental and estate vineyards, mostly located in the region of the southern part of Uruguay, have been investigated. Ten batches of 20 kg each of grapes of *Vitis vinifera* cv. Tannat from different clones were microvinified in the 1999 and 2000 vintages. SO<sub>2</sub> was added at 5 g/hL, and then each batch was inoculated with *Saccharomyces cerevisiae* strain CIVC 8130 (Gist Brocades, Santiago, Chile). Fermentation was carried out at 22–25 °C, and when the density had reached 1000 g/L, the wine was separated from the skins. The skins were then pressed and the two wine fractions combined. All of the samples were stabilized at 4 °C for 20 days and sterile-filtered (0.45 µm membrane), and then the free SO<sub>2</sub> content was adjusted to 40 mg/L. After bottling, the wines were conserved at 10 °C until analyzed.

**Isolation of Volatiles. Solid Phase Extraction Using ENV+ Cartridge.** Volatiles and precursors, the latter usually evaluated as aglycons among the bound forms, have been quantified after adsorption and separate elution from an Isolute (IST Ltd., Mid Glamorgan, U.K.) ENV+ cartridge packed with 1 g of highly cross-linked styrene–divinyl benzene (SDVB) polymer (40–140 µm, cod. no. 915–0100-C) as previously reported (9–11). The cartridges were sequentially conditioned with methanol (15 mL) and distilled water (20 mL). A sample of 50 mL of wine diluted with 50 mL of distilled water and containing 0.1 mL of internal standard (1-heptanol at 230 ppm in a 50% hydroalcoholic solution) was applied with an adequate syringe (4–5 mL/min), washing the residual with 15 mL of distilled water. The free aroma components were eluted with 30 mL of dichloromethane; the solution was dried with Na<sub>2</sub>SO<sub>4</sub> and concentrated to 1.5 mL on a Vigreux column, stored at –10 °C, and, immediately prior to GC analysis, further concentrated to 100 µL under a gentle nitrogen stream. The bound forms were eluted with 30 mL of methanol, and this solution was evaporated to dryness at the Rotavapor, then dissolved in 3 mL of citrate buffer at pH 5, added with Cytolase PCL5 (Gist-brocades, Lille Cédex, France), and reacted at 40 °C for 14 h. After the addition of the same internal standard (1-heptanol), the aglycons were extracted three times with 3 mL of pentane/dichloromethane 2:1, v/v, and the organic phase was dried with sodium sulfate and concentrated to 0.5 mL on a small Vigreux column and further reduced to 100 µL prior to GC analysis.

**HRGC Analysis.** For quantitative results, each sample was analyzed by HRGC, on a Shimadzu GC 14 B gas chromatograph equipped with a FID and Shimadzu data processor software EZ-Chrom, using two capillary columns. The first was a SE-52 (Mega, Legnano, Italy) cross-linked fused-silica capillary column (25 m × 0.32 mm i.d.), coated with 5% phenyl-polymethylsiloxane (0.40–0.45 µm phase thickness), column temperature, 60 °C for 8 min, rising to 180 °C at 3 °C/min, 180–250 °C at 20 °C/min, then 250 °C for 10 min; injector temperature, 250 °C; detector temperature, 280 °C; injection mode, split; split ratio, 1:30; volume injected, 1.0 µL; carrier gas, hydrogen, 55 kPa. The second was a Carbowax 20M (Ohio Valley, Marietta, Ohio) bonded fused-silica capillary column (25 m × 0.32 mm i.d.), coated with poly(ethylene glycol) (0.25 µm phase thickness), column temperature, 40 °C for 8 min, rising to 180 °C at 3 °C/min, then to 230 °C at 20 °C/min; injector temperature, 250 °C; detector temperature, 250 °C; injection mode, split; split ratio, 1:30; volume injected, 1.0 µL; carrier gas, hydrogen, 30 kPa.

**HRGC-MS Analysis.** GC-MS analyses were conducted using Shimadzu QP 5050 mass spectrometry equipped with reference libraries (12, 13) using a BP 20 (SGE, Ringwood, Australia) bonded fused-silica capillary column (25 m × 0.25 mm i.d.), coated with poly(ethylene glycol) (0.25 µm phase thickness), column temperature, 40 °C (6 min) to 180 °C at 3 °C/min, then to 220 °C at 10 °C/min, 220 °C (20 min); injector temperature, 250 °C; injection mode, split; split ratio, 1:40; volume injected, 1.0 µL; carrier gas, helium, 92.6 kPa (55.9 cm/s); interface temperature, 250 °C; energy, 70 eV; acquisition mass range, 40–400 amu.

**Identification and Quantification.** The components of the wine aroma were identified by comparison of their linear retention indices (LRI), determined in relation to a homologous series of *n*-alkanes, with those from pure standards or reported in the literature. Comparison of fragmentation patterns in the mass spectra with those stored on databases (12, 13) was also performed. In the cases that pure reference compounds were not used, the identification was indicated as tentative. Usually, HRGC-FID and HRGC-MS instrumental procedures using an internal standard (1-heptanol) were applied for quantitative purposes as previously described (14).

## RESULTS AND DISCUSSION

*V. vinifera* cv. Tannat acclimatized very well in Uruguay, where wines appreciated worldwide are produced. Early research has characterized the complex flavor described as “raspberry”, “plum”, “quince”, and “small-berry-like scents”, and in aged wines it changes to “smoked” and “licorice” character (15, 16), particularly through possible peculiarities in the free forms and possibly also in the heteroside fraction.

Tables 1 and 2 set out the data for the concentrations of free and bound components found in this study. The values represent the average of the two vintages 1999–2000.

**Analysis of Free Compounds.** About 51 volatile components, including alcohols, esters, carbonyl compounds, acids, terpenes, and norisoprenoids, were positively identified and quantitatively determined (Table 1).

The levels of the different groups of compounds released in these conditions from the Tannat wine samples are reported in Table 1. Among the compounds found, we give prominence to the ratio between *trans*- and *cis*-3-hexen-1-ol contents. As previously reported (17), the composition of the C<sub>6</sub> compounds is strongly dependent on four enzymes, which catalyze the biosynthesis of these compounds, and among these four, lipoyxygenase and hydroperoxide lyase are particularly important. Thus, the level and relationships between these compounds could be considered as characteristic of the *V. vinifera* variety (14). In particular for Tannat, *cis*-3-hexen-1-ol was higher than *trans* form in all of the samples analyzed from the two vintages considered.

In relation to fermentative compounds, several alcohols were identified, as shown in Table 1. Tannat wines contained remarkable amounts of 2-phenylethanol and low contents of higher alcohol acetates as well as fatty acid ethyl esters as in ageement with the profile reported for red wines and previous results for the sensory evaluation of this variety (16). These results are in close dependence with the yeast strain used in the vinification process (CIVC 8130, Gist-Brocades, Santiago, Chile). Although some of the most important compounds of the wine aroma are produced during the alcoholic fermentation as secondary products of yeast metabolism and the causal effects of *Saccharomyces cerevisiae* on wine flavor production are well documented, the effects of distinct strains differentially affecting wine flavor are less demonstrated (18). Although different strains have been found to differently affect both the volatile and macromolecule compositions in actual wine and model solutions (19), these differences have not been unambiguously evaluated, exceeding the scope of this work.

The levels of the monoterpene compounds, particularly mono-oxygenated monoterpenes, were all under their own thresholds, as is usually found for wines from neutral cultivars (20, 21); however, of relevance in some cases, rather high contents of limonene and geraniol were found.

With regard to the free norisoprenoids found in this wine, only small amounts of 3-oxo- $\alpha$ -ionol and vomifoliol were identified by searching for specific ions, *m/z* 108 and 124, respectively, on the full-scan mass spectra.

**Table 1.** Concentrations (Micrograms per Liter Equivalents of 1-Heptanol) of Free Volatile Compounds from Tannat Wines from 1999 and 2000 Vintages [Mean of Three Repetitions and Standard Deviation (SD) for 10 Samples Each Year]

compound	LRI <sup>a</sup>	identity assignment <sup>b</sup>	av content	SD
<b>esters</b>				
isobutyl acetate	1015	A	537	237
isoamyl acetate	1125	A	768	258
ethyl hexanoate	1237	A	354	120
ethyl pyruvate	1253	A	23	19
hexyl acetate	1275	A	27	16
ethyl lactate	1353	A	14465	10724
ethyl octanoate	1436	A	160	38
ethyl 3-hydroxybutyrate	1527	A	177	50
isoamyl lactate	1614	B (f)	114	43
ethyl decanoate	1684	A	70	24
diethyl succinate	1714	A	186	47
2-phenylethyl acetate	1815	A	135	54
ethyl 4-hydroxybutyrate	1822	B (f)	4018	1095
diethyl malate	2058	A	109	49
diethyl 2-hydroxyglutarate	2195	B (f)	194	112
ethyl phenyllactate	2249	B (f)	323	139
ethyl succinate	2370	B (f)	9219	3849
<b>Alcohols</b>				
2-methyl-1-propanol	1093	A	6935	2200
1-butanol	1155	A	561	447
2- and 3-methyl-1-butanol	1221	A	113545	54231
1-pentanol	1260	B (f)	78	30
4-methyl-1-pentanol	1328	B (f)	37	16
3-methyl-1-pentanol	1341	B (f)	131	67
3-ethoxy-1-propanol	1378	B (f)	285	86
3-(methylthio)-1-propanol	1725	B (f)	1242	396
benzyl alcohol	1882	A	58	45
2-phenylethanol	1918	A	32004	12011
<b>C6 compounds</b>				
1-hexanol	1368	A	904	254
<i>trans</i> -3-hexen-1-ol	1374	A	24	6
<i>cis</i> -3-hexen-1-ol	1382	A	49	15
<i>trans</i> -2-hexen-1-ol	1410	B (f)	76	47
<b>terpenes</b>				
limonene	1190	A	86	78
linalool	1558	A	tr	
$\alpha$ -terpineol	1718	A	tr	
nerol	1810	A	tr	
geraniol	1860	A	35	29
hodiol I ( <i>trans</i> -3,7-dimethyl-1,5-octadiene-3,7-diol)	1969	A	49	18
<b>acids</b>				
propanoic acid	1480	B (f)	63	18
2-methylpropanoic (isobutyric) acid	1608	B (f)	588	193
butyric acid	1670	B (f)	361	80
2- and 3-methylbutanoic (isovaleric) acid	1705	B (f)	765	223
hexanoic acid	1845	A	1245	256
octanoic acid	2072	A	1224	361
decanoic acid	2261	B (f)	51	41
<b>other compounds</b>				
acetoin	1266	B (f)	29	15
benzaldehyde	1535	B (f)	71	44
$\gamma$ -butyrolactone	1624	A	463	211
2-hydroxy-3,3-dimethyl- $\gamma$ -butyrolactone (pantolactone)	2028	B (f)	76	29
4-(carboethoxy)- $\gamma$ -butyrolactone	2168	B (f)	114	19
4-vinylguaiacol	2180	A	29	17
4-vinylphenol	2377	A	61	52

<sup>a</sup> Linear retention index based on a series of *n*-hydrocarbons reported according to their elution order on Carbowax 20M. <sup>b</sup> A, identities confirmed by comparing mass spectra and retention time with those of authentic standards supplied by Aldrich (Milwaukee, WI) and Fluka (Buchs, Switzerland); hodiol I was supplied by Versini (Istituto Agrario di San Michele all'Adige). B, identity tentatively assigned by comparing mass spectra with those obtained from the literature [(f) Adams, R. P. *Identification of Essential Oil Components by Gas Chromatography/Quadrupole Mass Spectroscopy*; Allured: Carol Stream, IL, 2001. McLafferty, F. W.; Stauffer, D. B. *The Wiley/NBS Registry of Mass Spectral Data*, 5th ed.; Wiley: New York, 1991]. C, tentatively identified.

An increase in the concentration of these compounds, and other norisoprenoids, is predictable during the conservation, which establishes a positive factor for the variety, although this must be demonstrated experimentally in conditions of aging of the wine.

Other remarkable components such as volatile phenols (4-vinylguaiacol and 4-vinylphenol), benzaldehyde,  $\gamma$ -butyrolactone, 2-hydroxy-3,3-dimethyl- $\gamma$ -butyrolactone (pantolactone), and 4-(carboethoxy)- $\gamma$ -butyrolactone have also been detected. The vinylphenols can be generated either from cinnamic acids as progenitors through yeast fermentation or as artifacts from the same acids in the GC injector (22).

**Analysis of Bound Compounds.** Glycoconjugates from Tannat wines were separated as indicated and the volatiles extracted with pentane/dichloromethane (2:1 v/v) from the hydrolysates and then analyzed using GC-MS. **Table 2** shows the concentration of components originating from the enzymatic hydrolysis of glycosidic precursors. C<sub>13</sub>-norisoprenoidic and monoterpenic volatiles released in these conditions from the Tannat wine samples were previously identified in other wine varieties; they made up ~42% of the total level of the volatiles released. The other volatiles were C<sub>6</sub> alcohols (6%) and benzenoid compounds (51%).

The dominating monoterpene alcohols were the monoterpenediols *cis* and *trans* isomers of 8-hydroxylinalool, with higher values for the *trans* isomer. It has been demonstrated that these monoterpenediols can play an important role as aroma precursors giving, by rearrangements under the acidic conditions of the wine, a number of volatile compounds, as in the case of 3,9-epoxy-*p*-menth-1-ene, the character impact compound of fresh dill herb (23). Furthermore, previous data reported in the literature also indicate that it is possible to assume a *cis/trans* relationship typical for this variety (24, 25). With regard to other monoterpenols, hotrienol,  $\alpha$ -terpineol, *trans*-linalool oxide (furanoid), *cis*-linalool oxide (furanoid), *trans*-linalool oxide (pyranoid), *cis*-linalool oxide (pyranoid), citronellol, nerol, geraniol, 7-hydroxygeraniol, *trans*-geranic acid, and linalool were identified, all of them in low concentrations as expected for a neutral variety.

The C<sub>13</sub>-norisoprenoid pattern was composed by 3-hydroxy- $\beta$ -damascone, 3-oxo- $\alpha$ -ionol, vomifoliol, and, in smaller concentrations, 4-oxo- $\beta$ -ionol, 3-oxo-7,8-dihydro- $\alpha$ -ionol, 4-oxo-7,8-dihydro- $\beta$ -ionol, grasshopper ketone, and 7,8-dihydro-vomifoliol. These compounds have been identified as heterosides also present in other varieties (26–28).

Like many fruits, mature grape berries contain numerous nonvolatile and nonodorant glycosidic compounds, the levels of which, particularly in grapes, are higher than those of volatile compounds (29). During wine-making, some of these compounds give rise to odorant compounds that play a role in certain of the aroma characteristics of wine. Of these compounds, norisoprenoid glycosides possessing 13 carbon atoms have been the most studied not only because of their structural diversity and aroma importance but also because their transformation into aroma compounds requires several steps.

The most abundant C<sub>13</sub>-norisoprenoids were vomifoliol, followed by 3-oxo- $\alpha$ -ionol, 3-hydroxy- $\beta$ -damascone, 3-oxo-7,8-dihydro- $\alpha$ -ionol, and 4-oxo-7,8-dihydro- $\beta$ -ionol. These norisoprenoid glycoconjugates are considered as degradation products of carotenoids in grape (30), and during wine aging they can give rise to potent odorants such as  $\beta$ -damascenone (31). Furthermore, the presence of 3-hydroxy- $\beta$ -damascone could be an index of a possible remarkable copresence of the acetylenic damascenone precursor (3-hydroxy-7,8-didehydro- $\beta$ -ionol), more



**Table 2.** Concentrations (Micrograms per Liter Equivalents of 1-Heptanol) of Glycosidically Bound Compounds Liberated by Enzymatic Hydrolysis from Tannat Wines from 1999 and 2000 Vintages [Mean of Three Repetitions and Standard Deviation (SD) for 10 Samples Each Year]

compound	LRI <sup>a</sup>	identity assignment <sup>b</sup>	av content	SD
<b>terpenes</b>				
<i>trans</i> -linalool oxide (furanoid)	1449	A	16.2	13.6
<i>cis</i> -linalool oxide (furanoid)	1474	A	36.5	17.6
linalool	1558	A	8.2	4.3
hottienol (3,7-dimethyl-1,5,7-octatrien-3-ol)	1620	C	4.5	2.2
$\alpha$ -terpineol	1718	A	9.7	6.4
<i>trans</i> -linalool oxide (pyranoid)	1739	B (7)	10.8	7.4
<i>cis</i> -linalool oxide (pyranoid)	1763	B (7)	8.9	12.0
nerol	1810	A	28.4	18.2
geraniol	1860	A	29.2	17.2
hodiol I ( <i>trans</i> -3,7-dimethyl-1,5-octadiene-3,7-diol)	1969	A	25.4	15.6
<i>trans</i> -8-hydroxylinalool ( <i>trans</i> -3,7-dimethyl-1,6-octadiene-3,8-diol)	2270	B (7)	146.0	124.2
<i>cis</i> -8-hydroxylinalool ( <i>cis</i> -3,7-dimethyl-1,6-octadiene-3,8-diol)	2310	B (7)	69.5	35.9
<i>trans</i> -geranic acid ( <i>trans</i> -3,7-dimethyl-2,6-octadien-1-oic acid)	2329	B (7)	68.0	42.0
<i>p</i> -menth-1-ene-7,8-diol	2528	B (2)	112.8	47.3
<b>norisoprenoids</b>				
3,4-dihydro-3-oxoactinidols (isomeric forms)	2456/66	B (3)	12.8	20.3
3-hydroxy- $\beta$ -damascone	2537	B (4)	374.9	239.1
3-oxo- $\alpha$ -ionol	2651	B (5)	496.4	275.3
4-oxo- $\beta$ -ionol	2658	B (5)	172.5	88.6
4-oxo-7,8-dihydro- $\beta$ -ionol	2694	B (4)	171.6	138.8
3-oxo-7,8-dihydro- $\alpha$ -ionol	2726	B (4)	250.3	204.8
grasshopper ketone <sup>c</sup>	3165	B (5)	5.8	2.9
vomifolol	3167	B (3)	1649.8	929.5
7,8-dihydrovomifolol	3262	B (5)	30.8	39.4
<b>shikimate derivatives</b>				
benzyl alcohol	1882	A	1130.9	498.0
2-phenylethanol	1918	A	975.1	544.8
4-vinylguaiacol	2180	A	65.1	95.0
4-vinylphenol	2377	A	39.7	33.1
methyl vanillate	2586	B (7)	6.4	2.2
zingerone [3-(4-hydroxy-3-methoxyphenyl)butan-2-one]	2779	B (7)	63.9	24.8
3-(4-hydroxy-3-methoxyphenyl)-1-propanol	2970	B (6)	613.0	151.9
methyl 2,5-dihydroxybenzoate	2993	B (7)	95.9	37.3
tyrosol [2-(4-hydroxy-phenyl)ethanol]	2999	B (6)	1290.6	593.7
3,4,5-trimethoxyphenol	3049	B (7)	87.2	33.9
<b>C6 compounds</b>				
1-hexanol	1368	A	224.6	97.0
<i>trans</i> -3-hexen-1-ol	1374	A	6.7	4.8
<i>cis</i> -3-hexen-1-ol	1382	A	45.6	24.1
<i>trans</i> -2-hexen-1-ol	1410	B (7)	28.3	9.8

<sup>a</sup> Linear retention index based on a series of *n*-hydrocarbons reported according to their elution order on Carbowax 20M. <sup>b</sup> A, identities confirmed by comparing mass spectra and retention time with those of authentic standards supplied by Aldrich (Milwaukee, WI) and Fluka (Buchs, Switzerland); hodiol I was supplied by Versini (Istituto Agrario di San Michele all'Adige). B, identities tentatively assigned by comparing mass spectra with those obtained from the literature [(7) Adams, R. P. *Identification of Essential Oil Components by Gas Chromatography/Quadrupole Mass Spectroscopy*; Allured: Carol Stream, IL, 2001. McLafferty, F. W.; Stauffer, D. B. *The Wiley/NBS Registry of Mass Spectral Data*, 5th ed.; Wiley: New York, 1991. (2) Versini, G.; Rapp, A.; Reniero, F.; Mandery, H. Structural identification and presence of some *p*-menth-1-ene diols in grape products. *Vitis* **1991**, 30, 143–149. (3) Strauss, C. R.; Wilson, B.; Williams, P. J. 3-Oxo- $\alpha$ -ionol, vomifolol, and roseoside in *Vitis vinifera* fruit. *Phytochemistry* **1987**, 26, 1995–1997. (4) Winterhalter, P. Bound terpenoids in the juice of the purple passion fruit (*Passiflora edulis*). *J. Agric. Food Chem.* **1990**, 38, 452–455. (5) Marais, J.; van Wyk, C. J.; Rapp, A. Effect of sunlight and shade on norisoprenoid levels in maturing Weisser Riesling and Chenin blanc grapes and Weisser Riesling wines. *S. Afr. J. Enol. Vitic.* **1992**, 13, 23–32. (6) Strauss, C. R.; Gooley, P. R.; Wilson, B.; Williams, P. J. Application of droplet countercurrent chromatography to the analysis of conjugated forms of terpenoids, phenols, and other constituents of grape juice. *J. Agric. Food Chem.* **1987**, 35, 519–524. C, tentatively identified. <sup>c</sup> Based on the ratio of mass fragments *m/z* 290 and 70 (1-heptanol).

efficient than the 3-hydroxy- $\beta$ -damascone to generate the potent rose-hay flavorant  $\beta$ -damascenone with a threshold level in water of  $\sim 0.009$  ppb (32, 33).

Multiple precursors to  $\beta$ -damascenone are present in wine, being 3-hydroxy-7,8-didehydro- $\beta$ -ionol (acetylenic diol), 3,5-dihydroxy-6,7-megastigmadien-9-ol (allenic triol), and their glycoconjugates reported as the main progenitors (34). Moreover, variations found in the concentration of  $\beta$ -damascenone for must and wine due to pH changes have been explained to be most likely caused by the presence of different precursors (free and bound forms), which are expected to show different reactivities (35, 36). For Tannat wine,  $\beta$ -damascenone was not detected in the experimental conditions above-

described, probably because this norisoprenoidic compound reaches its maximum level in wine within one year of aging (36).

The 4-oxo-ionol derivatives, even if found in other varieties (27), are peculiar, because theoretically they are not derivable from the xanthophylls, but probably from  $\beta$ -carotene through an allylic oxidation. In particular, 4-oxo-7,8-dihydro- $\beta$ -ionol could be the precursor of two isomeric megastigma-5,8-dien-4-ones, well-known flavor compounds of some plants such as *Osmantus* absolute, yellow passion fruit, and tobacco (37). Other interesting norisoprenoid aglycons are three isomers of 3,4-dihydro-3-oxoactinidol, possible precursors of actinidiolide forms.

Among the trioxxygenated C<sub>13</sub>-norisoprenoidic compounds, 7,8-dihydrovomifoliol and vomifoliol can be considered as precursors of theaspirane according to Heckman and Roberts (38). The same degradation pathway generates 3,4-dihydroxy-7,8-dihydro- $\beta$ -ionol precursor of vitispirane (30), this latter being reported as one of the key aromas of bottle-aged Riesling wines (23, 39).

Finally, among the shikimate-derived compounds we cite tyrosol [2-(4-hydroxyphenyl)-ethanol], benzyl alcohol, 2-phenylethanol, 3-(4-hydroxy-3-methoxyphenyl)-1-propanol, methyl 2,5-dihydroxybenzoate, and zingerone [3-(4-hydroxy-3-methoxyphenyl)butan-2-one]. Some of these compounds can be important aroma contributors such as zingerone, as mentioned by Williams et al. (40) in their research on aglycons with sensorial significance in grape. 4-Vinylphenol and 4-vinylguaiacol can be included in this group even with the quoted attention about their possible origin (21).

In conclusion, in the present work we characterized the aromatic profile of young Tannat wines from Uruguay having complex aromas that could be explained by some peculiarities detected in the free forms but, above all, in the bound fraction for the level and profile of the norisoprenoidic fraction. Although a complex variety of volatile components of wine, or hydrolysates of ENV+ extract of wines, have been observed in this study, very few of these compounds are likely to contribute to wine aroma. Nevertheless, sensory evaluation (16) indicates clearly that there are, within the overall pool of enzymatically released volatile compounds, some components which can make such a sensory contribution. To understand the impact of these individual compounds that can alter the aroma and flavor of Tannat wine, future investigations on this variety should involve sensory studies on individual constituents within the group of released flavorants. The application of GC-olfactometry technique could also be used for a more in-depth study of the impact of free aroma compounds in this variety.

#### ABBREVIATIONS USED

HRGC, high-resolution gas chromatography; FID, flame ionization detector; MS, mass spectrometry.

#### ACKNOWLEDGMENT

We are grateful for the assistance of Dr. M. Galli (Mega, Legnano, Italy), which made this work possible.

#### LITERATURE CITED

- Carrau, F. M. The emergence of a new Uruguayan wine industry. *J. Wine Res.* **1997**, *8*, 179–185.
- Galanti, A. N. *La Industria Vitivinícola Uruguaya*; Hemisferio Sur: Mendoza, Argentina; 1919.
- Instituto Nacional de Vitivinicultura. *Censo Vitícola del Uruguay*; INAVI: Las Piedras, Uruguay, 1995.
- Schreier, P. Flavour composition of wines: a review. *CRC Crit. Rev. Food Sci. Nutr.* **1979**, *12*, 59–111.
- Boulton, R. B.; Singleton, V. L.; Bisson, L. F.; Kunkee, R. E. *Principles and Practices of Winemaking*; Chapman and Hall: New York, 1995.
- Rapp, A. Volatile flavour of wine: correlation between instrumental analysis and sensory perception. *Nahrung* **1998**, *42*, 351–363.
- Rapp, A.; Versini, G. Influence of nitrogen compounds in grapes on aroma compounds of wine. In *Proceedings of the International Symposium on Nitrogen in Grapes and Wines*; Rantz, Ed.; American Society of Enology and Viticulture: Davis, CA, 1991; pp 156–164.
- Lambrecht, M. G.; Pretorius, I. S. Yeast and its importance to wine aroma—A review. *S. Afr. J. Enol. Vitic.* **2000**, *21*, 97–129.
- Carlin, S. Metodi di arricchimento di composti dell'aroma per una possibile tipicizzazione di vini Traminer. Thesis, Faculty of Agronomy, University of Udine, Italy, 1998.
- Boido, E.; Carrau, F.; Dellacassa, E.; Medina, K.; Lloret, A.; Versini, G. Modificación en la composición aromática producidas por la fermentación maloláctica en los vinos Tannat y su evolución durante la crianza. In *Proceedings of the VIII Congreso Latinoamericano de Viticultura y Enología*; Asoc. Enol. Uruguay: Montevideo, Uruguay, 2001.
- Boido, E.; Lloret, A.; Rodríguez, M.; Medina, K.; Carrau, F.; Dellacassa, E.; Disegna, E. Evolución del contenido polifenólico en la uva de *Vitis vinifera* L. cv. Tannat durante la madurez para dos manejos de viñedo, y el efecto en la composición polifenólica y de precursores aromáticos glicosidados de los vinos. In *Proceedings of the VIII Congreso Latinoamericano de Viticultura y Enología*; Asoc. Enol. Uruguay: Montevideo, Uruguay, 2001.
- Adams, R. P. *Identification of Essential Oil Components by Gas Chromatography/Quadrupole Mass Spectroscopy*; Allured: Carol Stream, IL, 2001.
- McLafferty, F. W.; Stauffer, D. B. *The Wiley/NBS Registry of Mass Spectral Data*, 5th ed.; Wiley: New York, 1991.
- Versini, G.; Orriols, I.; Dalla Serra, A. Aroma components of Galician Albariño, Loureira and Godello wines. *Vitis* **1994**, *33*, 165–170.
- Bayonove, C.; Baumes, R.; Gunata, Z.; Razungles, A.; Sapis, J. C. Il Potenziale aromatico dei vitigni rossi non aromatici. In *Atti X Incontro su: Contributi ed Influenza della Chimica nella Produzione, Conservazione e Commercializzazione del Vino*, Siena, 1990.
- Gámbaro, A.; Boido, E.; Zlotejablko, A.; Medina, K.; Lloret, A.; Dellacassa, E.; Carrau, F. Effect of malolactic fermentation on the aroma properties of Tannat wine. *Aust. J. Grape Wine Res.* **2001**, *7*, 27–32.
- Hatanaka, A. The biogenesis of green odour by green leaves. *Phytochemistry* **1993**, *34*, 1201–1218.
- Thorngate, J. H., III. Yeast strain and wine flavor: nature or nurture? In *Chemistry of Wine Flavor*; ACS Symposium Series 714; Waterhouse, L., Ebeler, S. E., Eds.; American Chemical Society: Washington, DC, 1998; pp 66–80.
- Lambrechts, M. G.; Pretorius, I. S. Yeast and its importance to wine aroma—a review. *S. Afr. J. Enol. Vitic.* **2000**, *21*, 97–129.
- Razungles, A.; Günata, Z.; Pinatel, S.; Baumes, R.; Bayonove, C. Etude quantitative de composés terpéniques, norisoprénoides et de leur précurseurs dans diverses variétés de raisins. *Sci. Aliments* **1993**, *13*, 59–72.
- 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.
- 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.
- Strauss, C. R.; Wilson, B.; Anderson, R.; Williams, P. Novel monoterpene diols and diol glycosides in *Vitis vinifera* grapes. *J. Agric. Food Chem.* **1988**, *36*, 569–573.
- Versini, G.; Carlin, S.; Nicolini, G.; Dellacassa, E.; Carrau, F. Updating of varietal aroma components in wines. In *Proceedings of the VII Congreso Latinoamericano de Viticultura y Enología. La Vitivinicultura del Hemisferio Sur*; Asoc. Enol. Argentina: Mendoza, Argentina, 1999; pp 323–349.
- Scienza, A.; Versini, G.; Romano, F. A. Considerations sur l'influence du genotype et du milieu sur la synthèse des arômes dans le raisin-cas particulier du Chardonnay. In *Atti Simp. Intern. sugli Aromi dell'Uva e del Vino, S. Michele all'Adige*; Scienza, A., Versini, G., Eds.; Manfrini: Trento, Italy, 1987; pp 9–53.

- (26) Bureau, S. M.; Baumes, R. L.; Razungles, A. J. Effects of vine or bunch shading on the glycosylated flavor precursors in grapes of *Vitis vinifera* L. Cv. Syrah. *J. Agric. Food Chem.* **2000**, *48*, 1290–1297.
- (27) Williams, P. J.; Sefton, M. A.; Leigh, F. Glycosidic precursors of varietal grape and wine flavor. In *Flavor Precursors: Thermal and Enzymatic Conversions*; ACS Symposium Series 490; Teranishi, R., Takeoka, G. R., Guntert, M., Eds.; American Chemical Society: Washington, DC, 1992; pp 74–86.
- (28) Wirth, J.; Guo, W.; Baumes, R.; Günata, Z. Volatile compounds released by enzymatic hydrolysis of glycoconjugates of leaves and grape berries from *Vitis vinifera* Muscat of Alexandria and Shiraz cultivars. *J. Agric. Food Chem.* **2001**, *49*, 2917–2923.
- (29) Baumes, R.; Wirth, J.; Bureau, S.; Gunata, Y.; Razungles, A. Biogenesis of C<sub>13</sub>-norisoprenoid compounds: experiments supportive for an apo-carotenoid pathway in grapevines. *Anal. Chim. Acta* **2002**, *458*, 3–14.
- (30) Winterhalter, P.; Skouroumounis, G. K. Glycoconjugated aroma compounds: occurrence, role and biotechnological transformation. In *Advances in Biochemical Engineering Biotechnology*; Scheper, T., Ed.; Springer: Berlin, Germany, 1997; pp 73–105.
- (31) Winterhalter, P. The generation of C<sub>13</sub> norisoprenoid volatils. In *Connaissance Aromatique des Cépages et Qualité des Vins*; Bayonove, C., Crouzet, J. Flanz, C., Martin, J. C., Sapis, J. C., Eds.; Rev. Fr. OEnol. Lettes: Paris, France, 1993; pp 65–73.
- (32) Baderschneider B.; Skouroumounis G.; Winterhalter P. Glucosidic precursors of  $\beta$ -damascenone in Riesling wine. In *Flavour Perception—Aroma Evaluation. Proceedings of the 5th Warburg Aroma Symposium*; Krause, H. P., Rothe, M. Eds.; Universität Potsdam: Potsdam, Germany, 1997; pp 473–476.
- (33) Sefton, M. A. Hydrolytically released volatile secondary metabolites from a juice sample of *Vitis vinifera* grape cvs. Merlot and Cabernet Sauvignon. *Aust. J. Grape Wine Res.* **1998**, *4*, 30–38.
- (34) Skouroumounis, G. K.; Sefton, M. A. The formation of  $\beta$ -damascenone in wine. In *Carotenoid-Derived Aroma Compounds*; Winterhalter, P., Rouseff, R. L., Eds.; American Chemical Society: Washington, DC, 2001; pp 285–299.
- (35) Kotseridis, Y.; Baumes, R. Identification of impact odorants in Bordeaux red grape juice, in the commercial yeast used for its fermentation, and in the produced wine. *J. Agric. Food Chem.* **2000**, *48*, 400–406.
- (36) Versini, G.; Carlin, S.; Dalla Serra, A.; Nicolini, G.; Rapp, A. Formation of 1,1,6-trimethyl-1,2-dihydronaphthalene and other norisoprenoid in wine: Considerations of the kinetics. In *Carotenoid-Derived Aroma Compounds*; Winterhalter, P., Rouseff, R. L., Eds.; American Chemical Society: Washington, DC, 2001; pp 285–299.
- (37) Winterhalter, P. Oxygenated C<sub>13</sub>-norisoprenoids: important flavour precursors. In *Flavor Precursors: Thermal and Enzymatic Conversions*; Teranishi, R., Takeoka, G. R., Guntert, M., Eds.; ACS Symposium Series 490; American Chemical Society: Washington, DC, 1992; pp 98–115.
- (38) Heckman, R. A.; Roberts, D. L. Synthesis of theaspirone, a component of tea essential oil. *Tetrahedron Lett.* **1969**, 2701–2704.
- (39) Winterhalter, P.; Sefton, M. A.; Williams, P. J. Two-dimensional GC-DCCC analysis of the glycoconjugates of monoterpenes, norisoprenoids, and shikimate-derived metabolites from Riesling wine. *J. Agric. Food Chem.* **1990**, *38*, 1041–1048.
- (40) Williams, P. J.; Sefton, M. A.; Wilson, B. Nonvolatile conjugates of secondary metabolites as precursors of varietal grape flavor components. In *Flavor Chemistry. Trends and Developments*; Teranishi, R., Buttery, R. G., Shahidi, F., Eds.; ACS Symposium Series 388; American Chemical Society: Washington, DC, 1988; pp 35–48.

---

Received for review February 4, 2003. Revised manuscript received June 8, 2003. Accepted June 9, 2003. This research was financially supported by the Instituto Nacional de Investigaciones Agropecuarias (INIA, Grant 125, INIA-LIA 055) and the Consejo Nacional de Investigación Científica y Tecnológica (CONICYT, Project 5058).

JF0300871