Isolation and Characterization of Novel Benzoates, Cinnamates, Flavonoids, and Lignans from Riesling Wine and Screening for Antioxidant Activity

Beate Baderschneider and Peter Winterhalter*

Institut für Lebensmittelchemie der TU Braunschweig, Schleinitzstrasse 20, DE-38106 Braunschweig, Germany

A German Riesling wine has been fractionated with the aid of countercurrent chromatography. After purification by HPLC, the structures of 101 compounds were established by mass spectrometry and NMR spectroscopy. Seventy-three of the isolated compounds exhibited a phenolic or benzylic structure. Fifty-four compounds were reported for the first time as Riesling wine constituents. New compounds identified in this work included twelve benzoic and cinnamic acid derivatives. In addition to two isomeric (*E*)-caffeoyl ethyl tartrates, the glucose esters of (*E*)-cinnamic, (*E*)-*p*-coumaric, and (*E*)-ferulic acid, as well as the 4-*O*-glucosides of (*E*)- and (*Z*)-ferulic acid, have been identified for the first time in Riesling wine. The structures of two additional phenylpropanoids were elucidated as 3-hydroxy-1-(4-hydroxy-3-methoxyphenyl)-propan-1-one and 2,3-dihydroxy-1-(4-hydroxy-3-methoxyphenyl)-propan-1-one. Moreover, two ethyl esters, i.e., ethyl protocatechuate and ethyl gallate, as well as the glucose ester of vanillic acid, were newly detected in Riesling wine. Novel representatives in the flavonoid group were dihydrokaempferol, dihydroquercetin, and four dihydroflavonol glycoconjugates, i.e., the 3-*O*-glucosides of dihydrokaempferol and dihydroquercetin, as well as the 3-*O*-xyloside and the 3′-*O*-glucoside of dihydroquercetin. Additionally, six novel lignans, i.e., lariciresinol 4-*O*-glucoside, three isolariciresinol derivatives, and two secoisolariciresinols, as well as three neolignans were isolated. Structural elucidation of the newly isolated wine constituents is reported together with the determination of their antioxidant activity.

Keywords: *White wine; Riesling; benzoates; cinnamates; phenylpropanoids; flavonoids; dihydroflavonols; lignans; neolignans; antioxidant activity; TEAC values*

INTRODUCTION

Most studies on wine polyphenols carried out so far have focused on red wine. As a result, the crucial role of red wine constituents, e.g., flavonoids and stilbenes, in the prevention of degenerative diseases is well documented (*1*). In the case of white wine less information is available. In general it has been shown that white wine exhibits much less antioxidant activity than red wine. However, in a study by Vinson and Hontz (*2*) it was found that the phenolic compounds present in white wine are, on an equimolar basis, more effective than the red wine phenolics in inhibiting in vitro LDL oxidation. To obtain a better understanding of the antioxidant capacity of white wine, we decided to investigate the phenolic composition of a German Riesling wine. Determination of the structure and antioxidant activity of the phenolic Riesling constituents is a prerequisite to finally explain differences in bioavailability and bioactivity of the phenolic fractions of red and white wines. So far 101 pure compounds have been obtained, including seven novel stilbenes that were described in a preceding communication (*3*). Although stilbenes are often considered as the most important bioactive compounds in wine, these polyphenols are accompanied by many additional phenolic constituents, often with unknown physiological activity. The major group of phenolic constituents in white wine is the hydroxycinnamic acid derivatives (*4*). Despite the widespread occurrence of cinnamic acid derivatives, there are few reports about their contribution to the total antioxidant activity of white wine. Additional classes of wine constituents that are likely contributors to the overall antioxidant capacity of white wines are benzoates, flavonoids, and lignans. In the present study, twenty-seven novel compounds that belong to the above-mentioned classes of wine phenols are identified for the first time in Riesling wine. Furthermore, the antioxidant activities for a number of the isolated compounds are determined and compared with those of known antioxidants.

EXPERIMENTAL PROCEDURES

The Riesling wine (1992 vintage) was purchased in 1994. After workup the fractions were freeze-dried and kept at −18 °C. Details of preparation of isolates, fractionation, and purification of extracts, as well as testing of antioxidant activity, were given previously (*3*).

Isolation and Characterization of Novel Benzoic and Cinnamic Acid Derivatives. Besides known constituents (Figure 1, for complete spectral data cf. ref. *5*), i.e., (*E*)-*p*-coumaric acid **1a** (40.9 mg), (*E*)-*p*-coumaroyl tartaric acid **1b** (19.0 mg), 4-*O*-*β*-D-glucoside of (*E*)-*p*-coumaric acid **1c** (1.5 mg), (*Z*)-*p*-coumaric acid **2a** (2.0 mg), (*Z*)-*p*-coumaroyl tartaric acid **2b** (1.0 mg), 4-*O*-*β*-D-glucoside of (*Z*)-*p*-coumaric acid **2c** (4.7 mg), (*E*)-caffeic acid **3a** (44.2 mg), (*E*)-caffeoyl tartaric acid **3b** (41.5 mg), (*E*)-ethyl caffeate **3c** (5.8 mg), (*E*)-ferulic acid **4a**

* To whom correspondence should be addressed (email, P.Winterhalter@tu-bs.de; fax, ++49-531-3917230).

10.1021/jf010396d CCC: $20.00
Published on Web 05/22/2001

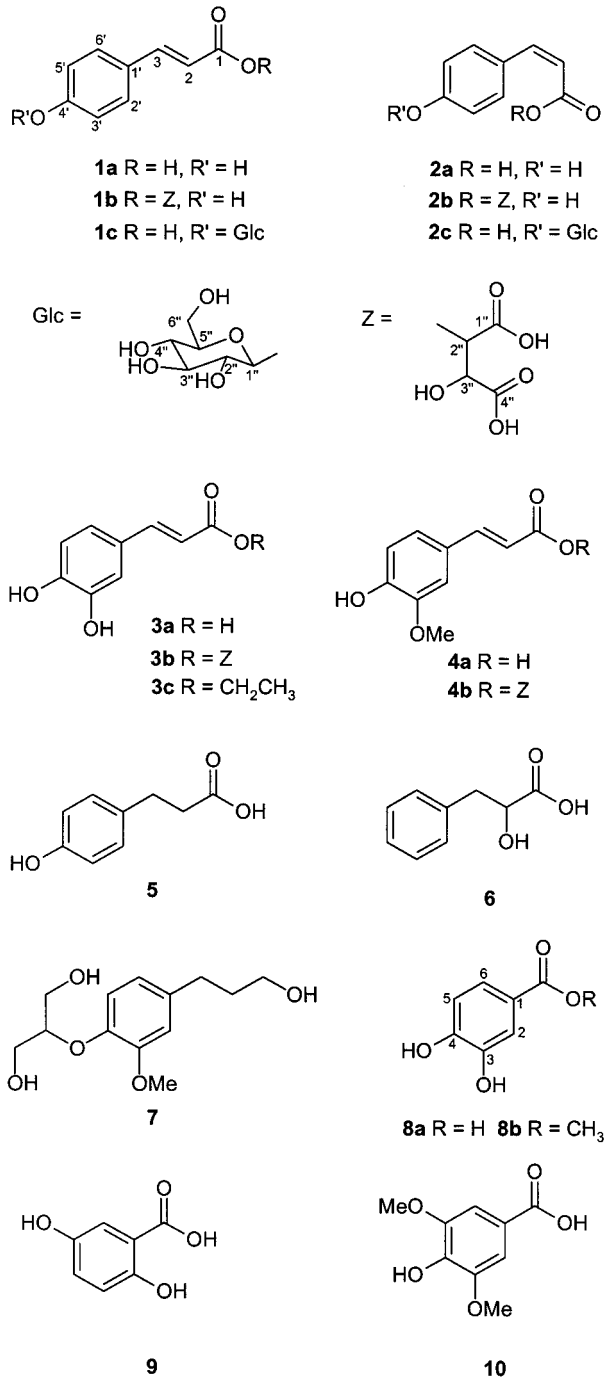

Figure 1. Structures of known phenylpropanoids **1**−**7** and benzoates **8**−**10** isolated from a German Riesling wine.

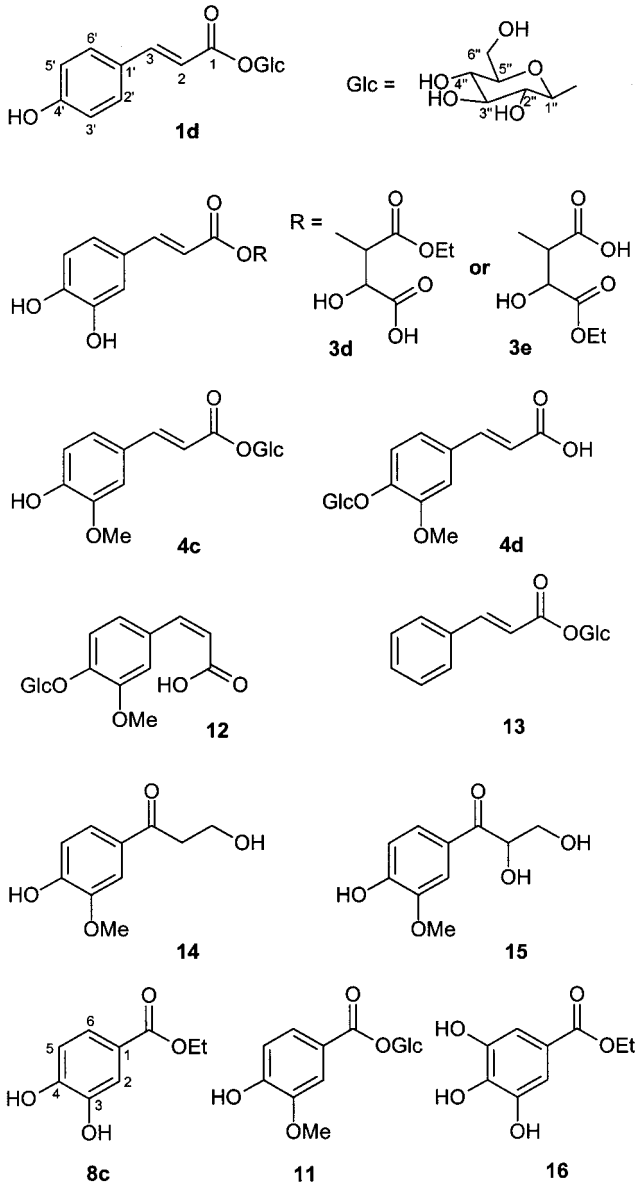

Figure 2. Structures of novel phenylpropanoids and benzoates isolated from a German Riesling wine: (*E*)-*p*-coumaroyl glucose ester **1d**; isomeric (*E*)-caffeoyl ethyl tartrates **3d/e**; (*E*)-feruloyl glucose ester **4c**; (*E*)-ferulic acid 4-*O*-*β*-D-glucoside **4d**; (*Z*)-ferulic acid 4-*O*-*β*-D-glucoside **12**; cinnamoyl glucose ester **13;** 3-hydroxy-1-(4-hydroxy-3-methoxyphenyl)-propan-1-one **14**; 2,3-dihydroxy-1-(4-hydroxy-3-methoxyphenyl)-propan-1-one **15;** ethyl protocatechuate **8c**; vanilloyl glucose ester **11**; and, ethyl gallate **16**.

(11.7 mg), (*E*)-feruloyl tartaric acid **4b** (65.3 mg), *p*-hydroxyphenylpropionic acid **5** (1.2 mg), 2-hydroxy-3-phenylpropionic acid **6** (12.9 mg), 2-[4-(3-hydroxypropyl)-2-methoxyphenoxy]-propan-1,3-diol **7** (2.4 mg), protocatechuic acid **8a** (4.4 mg), methyl protocatechuic acid **8b** (1.2 mg), gentisic acid **9** (2.1 mg), and syringic acid **10** (2.6 mg), the following cinnamic and benzoic acid derivatives were identified for the first time in Riesling wine (Figure 2).

Glucose Ester of (*E*)-*p*-Coumaric Acid (1d). 24.2 mg. UV (MeOH): λ_{max} 316, 225 nm. Thermospray−MS: pseudo molecular ion at *m*/*z* 344 $[M(326) + NH_4]^+$, 327 $[M + H]^+$. ^{1}H NMR (250 MHz, CD_3OD, ppm): δ 3.37−3.48 (4 H, m, H2″/H3″/H4″/H5″); 3.68 (1 H, dd, J = 12/4.5 Hz, H6″a); 3.85 (1 H, dd, J = 12/2 Hz, H6″b); 5.57 (1 H, d, J = 8 Hz, H1″); 6.37 (1 H, d, J = 15.5 Hz, H2); 6.81 (2 H, m, H3′/H5′); 7.48 (2 H, m, H2′/H6′); 7.73 (1 H, d, J = 15.5 Hz, H3). ^{13}C NMR (62.9 MHz, CD_3OD, ppm): δ 62.4 (C6″), 68.3 (C4″), 74.1 (C2″), 78.1 (C3″), 78.8 (C5″), 95.8 (C1″), 116.9 (C2/C3′/C5′), 127.0 (C1′), 131.4 (C2′/C6′), 147.9 (C3), 161.6 (C4′), 167.7 (C1).

Caffeoyl Ethyl Tartrate, Isomer I (3d). 1.1 mg. UV (MeOH): λ_{max} 330, 205 nm. ESI−MS (negative mode): pseudo molecular ion at *m*/*z* 339 $[M(340) - H^+]^-$, MS/MS of *m*/*z* 339 at *m*/*z* 293 $[M-H^+-C_2H_5OH]^-$ and *m*/*z* 177 [M − caffeoyl]$^-$. ^{1}H NMR (300 MHz, CD_3OD, ppm): δ 1.29 (3 H, t, J = 7 Hz, H1‴); 4.25 (2 H, q, J = 7 Hz, H2‴); 4.43 (1 H, d, J = 2 Hz, H3″); 5.49 (1 H, d, J = 2 Hz, H2″); 6.31 (1 H, d, J = 16 Hz, H2); 6.94 (1 H, d, J = 8 Hz, H5′); 6.97 (1 H, dd, J = 2/8 Hz, H6′); 7.05 (1 H, d, J = 2 Hz, H2′); 7.61 (1 H, d, J = 16 Hz, H3).

Caffeoyl Ethyl Tartrate, Isomer II (3e). 13.0 mg. UV (MeOH): λ_{max} 327, 301, 206 nm. ESI−MS (negative mode): pseudo molecular ion at *m*/*z* 339 $[M(340) - H^+]^-$, MS/MS of *m*/*z* 339 at *m*/*z* 293 $[M-H^+-C_2H_5OH]^-$ and *m*/*z* 177 [M − caffeoyl]$^-$. ^{1}H NMR (300 MHz, CD_3OD, ppm): δ 1.22 (3 H, t, J = 7 Hz, H1‴); 4.19 (2 H, m, H2‴); 5.45 (1 H, d, J = 2.5 Hz, H2″); 6.31 (1 H, d, J = 16 Hz, H2); 6.94 (1 H, d, J = 8 Hz,

H5′); 6.97 (1 H, dd, J = 2/8 Hz, H6′); 7.05 (1 H, d, J = 2 Hz, H2′); 7.61 (1 H, d, J = 16 Hz, H3); signal for H3″ obscured by solvent signal (approximately 4.8 ppm).

Glucose Ester of (*E*)-Ferulic Acid (4c). 23.9 mg. UV (MeOH): λ_{max} 327, 236, 218, 202 nm. Thermospray−MS: pseudo molecular ion at m/z 374 $[M(356) + NH_4]^+$, 357 $[M + H]^+$. ^{1}H NMR (360 MHz, CD_3OD, ppm): δ 3.37−3.48 (4 H, m, H2″/H3″/H4″/H5″); 3.69 (1 H, dd, J = 12/4.5 Hz, H6″a); 3.85 (1 H, dd, J = 12/2 Hz, H6″b); 3.90 (3 H, s, OCH_3); 5.58 (1 H, d, J = 7.5 Hz, H1″); 6.40 (1 H, d, J = 16 Hz, H2); 6.82 (1 H, d, J = 8 Hz, H5′); 7.10 (1 H, dd, J = 2/8 Hz, H6′); 7.20 (1 H, d, J = 2 Hz, H2′); 7.72 (1 H, d, J = 16 Hz, H3). ^{13}C NMR (90.6 MHz, CD_3OD, ppm): δ 56.5 (OCH_3), 62.4 (C6″), 71.2 (C4″), 74.1 (C2″), 78.1 (C3″), 78.8 (C5″), 95.8 (C1″), 111.9 (C2′), 114.8 (C5′), 116.6 (C2), 124.4 (C6′), 127.6 (C1′), 148.2 (C3), 149.4 (C4′), 151.0 (C3′), 167.7 (C1).

4-*O*-*β*-D-Glucoside of (*E*)-Ferulic Acid (4d). 1.0 mg. UV (MeOH): λ_{max} 284, 211 nm. Thermospray−MS: pseudo molecular ion at m/z 374 $[M(356) + NH_4]^+$, 357 $[M + H]^+$. ^{1}H NMR (360 MHz, CD_3OD, ppm): δ 3.39−3.51 (4 H, m, H2″/H3″/H4″/H5″); 3.69 (1 H, dd, J = 12/5 Hz, H6″a); 3.88 (1 H, dd, J = 12/2 Hz, H6″b); 3.90 (3 H, s, OCH_3); 4.96 (1 H, d, J = 7.5 Hz, H1″); 6.39 (1 H, d, J = 16 Hz, H2); 7.13 (1 H, dd, J = 2/8 Hz, H6′); 7.17 (1 H, d, J = 8 Hz, H5′); 7.23 (1 H, d, J = 2 Hz, H2′); 7.55 (1 H, d, J = 16 Hz, H3).

4-*O*-*β*-D-Glucoside of (*Z*)-Ferulic Acid (12). 3.8 mg. UV (MeOH): λ_{max} 267, 207 nm. ESI−MS: pseudo molecular ion at m/z 355 $[M(356) - H^+]^-$, MS/MS of m/z 355 at m/z 193 $[M - H^+ - \text{anhydroglucose}]^-$. ^{1}H NMR (360 MHz, CD_3OD, ppm): δ 3.39−3.49 (4 H, m, H2″/H3″/H4″/H5″); 3.68 (1 H, dd, J = 12/5 Hz, H6″a); 3.86 (1 H, dd, J = 12/2 Hz, H6″b); 3.88 (3 H, s, OCH_3); 4.90 (1 H, d, J = 7.5 Hz, H1″); 5.95 (1 H, d, J = 13 Hz, H2); 6.45 (1 H, d, J = 13 Hz, H3); 7.07 (1 H, dd, J = 2/8 Hz, H6′); 7.10 (1 H, d, J = 8 Hz, H5′); 7.55 (1 H, d, J = 2 Hz, H2′).

Glucose Ester of Cinnamic Acid (13). Isolated as its tetraacetate: 1.9 mg. UV (MeOH): λ_{max} 282, 218 nm. DCI−MS (reactant gas, NH_3): pseudo molecular ion at m/z 496 $[M(478) + NH_4]^+$. ^{1}H NMR (360 MHz, CD_3OD, ppm): δ 2.03−2.09 (4 × 3 H, 4 × s, 4 acetate); 3.90 (1 H, ddd, J = 10/4.5/2.5 Hz, H5″); 4.14 (1 H, dd, J = 12/2.5 Hz, H6″a); 4.32 (1 H, dd, J = 12/4.5 Hz, H6″b); 5.18 (1 H, dd, J = 10/9 Hz, H4″); 5.25 (1 H, dd, J = 9/8 Hz, H2″); 5.31 (1 H, t, J = 9 Hz, H3″); 5.86 (1 H, d, J = 8 Hz, H1″); 6.42 (1 H, d, J = 16 Hz, H3); 7.40−7.50 (5 H, m, H2′/H3′/H4′/H5′/H6′); 7.77 (1 H, d, J = 16 Hz, H2).

3-Hydroxy-1-(4-hydroxy-3-methoxyphenyl)-propan-1-one (14). 0.5 mg. UV (MeOH): λ_{max} 276, 228, 206 nm. ESI−MS (negative mode): pseudo molecular ion at m/z 195 $[M(196) - H^+]^-$, MS/MS of m/z 195 at m/z 165 $[M - H^+ - CH_2O]^-$. ^{1}H NMR (300 MHz, CD_3OD, ppm): δ 3.16 (2 H, t, J = 6.5 Hz, H2); 3.91 (3 H, s, OCH_3); 3.94 (2 H, t, J = 6.5 Hz, H3); 6.87 (1 H, d, J = 8 Hz, H5′); 7.55 (1 H, d, J = 2 Hz, H2′); 7.58 (1 H, dd, J = 2/8 Hz, H6′).

2,3-Dihydroxy-1-(4-hydroxy-3-methoxyphenyl)-propan-1-one (15). 2.9 mg. UV (MeOH): λ_{max} 306, 279, 231, 207 nm. ESIMS (negative mode): pseudo molecular ion at m/z 211 $[M(212) - H^+]^-$, MS/MS of m/z 211 at m/z 181 $[M - H^+ - CH_2O]^-$. ^{1}H NMR (300 MHz, CD_3OD, ppm): δ 3.73 (1 H, dd, J = 5.5/11.5 Hz, H3a); 3.89 (1 H, dd, J = 4/11.5 Hz, H3b); 3.92 (3 H, s, OCH_3); 5.10 (1 H, dd, J = 4/5.5 Hz, H2); 6.88 (1 H, d, J = 8.5 Hz, H5′); 7.53 (1 H, brs, H2′); 7.58 (1 H, dd, J = 2/8.5 Hz, H6′). ^{1}H NMR (300 MHz, CD_3COCD_3, ppm): δ 3.78 (1 H, dd, J = 4.5/11.5 Hz, H3a); 3.88 (1 H, dd, J = 3.5/11.5 Hz, H3b); 3.93 (3 H, s, OCH_3); 5.09 (1 H, dd, J = 4/5.5 Hz, H2); 6.94 (1 H, d, J = 8.5 Hz, H5′); 7.59 (1 H, d, J = 2 Hz, H2′); 7.63 (1 H, dd, J = 2/8.5 Hz, H6′). ^{13}C NMR (75.5 MHz, CD_3COCD_3, ppm): δ 57.1 (OCH_3), 67.1 (C3), 75.8 (C2), 113.1 (C2′), 116.2 (C5′), 125.4 (C1′/C6′), 149.2 (C3′), 153.8 (C4′), 199.7 (C1).

Ethyl Protocatechuate (8c). 2.0 mg. UV (MeOH): λ_{max} 299, 263, 219, 209 nm. ESIMS (negative mode): pseudo molecular ion at m/z 181 $[M(182) - H^+]^-$; MS/MS of m/z 181 at m/z 153 $[M - CH_2CH_3^+]^-$; MS/MS of m/z 153 at m/z 109 $[M - CH_2CH_3^+ - CO_2]^-$. ^{1}H NMR (300 MHz, CD_3OD, ppm): δ 1.35 (3 H, t, J = 7 Hz, H2′); 4.29 (2 H, q, J = 7 Hz, H1′); 6.79 (1 H, d, J = 8 Hz, H5); 7.41 (1 H, dd, J = 8/2 Hz, H6); 7.42 (1 H, d, J = 2 Hz, H2).

Glucose Ester of Vanillic Acid (11). 2.0 mg. UV (MeOH): λ_{max} 294, 265, 222, 205 nm. ESIMS (negative mode): pseudo molecular ion at m/z 329 $[M(330) - H^+]^-$; MS/MS of m/z 329 at m/z 167 $[M - H^+ - \text{anhydroglucose}]^-$. ^{1}H NMR (300 MHz, CD_3OD, ppm): δ 3.40−3.50 (4 H, m, H2″/H3″/H4″/H5″); 3.70 (1 H, dd, J = 12/5 Hz, H6″a), 3.86 (1 H, dd, J = 12/2 Hz, H6″b) 3.90 (3 H, s, OCH_3); 5.68 (1 H, d, J = 7.5 Hz, H1″); 6.86 (1 H, d, J = 8 Hz, H5); 7.62 (1 H, dd, J = 8/2 Hz, H6); 7.65 (1 H, d, J = 2 Hz, H2).

Ethyl Gallate (16). 5.8 mg. UV (MeOH): λ_{max} 281, 219 nm. ESIMS (negative mode): pseudo molecular ion at m/z 197 $[M(198) - H^+]^-$; MS/MS of m/z 197 at m/z 169 $[M - CH_2CH_3^+]^-$; MS/MS of m/z 169 at m/z 125 $[M - CH_2CH_3^+ - CO_2]^-$. ^{1}H NMR (300 MHz, CD_3OD, ppm): δ 1.35 (3 H, t, J = 7 Hz, H2′); 4.27 (2 H, q, J = 7 Hz, H1′); 7.04 (2 H, s, H2/H6).

An additional phenolic acid, the known wine constituent *p*-hydroxyphenylacetic acid **17** (1.0 mg, structure not shown) was identified (*6*).

Isolation and Characterization of Novel Dihydroflavonoids. Besides known constituents (Figure 3, for complete spectral data cf. ref. *5*), i.e., 2*R*,3*R*-dihydrokaempferol 3-*O*-α-L-rhamnoside **18a** (8.2 mg); 2*R*,3*R*-dihydroquercetin 3-*O*-α-L-rhamnoside **19a** (53.4 mg); catechin **20** (26.5 mg); epicatechin **21** (18.7); procyanidin B1 **22** (29.0 mg); procyanidin B3 **23** (4.0 mg); kaempferol 3-*O*-*β*-D-glucoside **24** (1.4 mg); and quercetin 3-*O*-*β*-D-glucuronide **25** (13.7 mg), the following dihydroflavonol derivatives were identified for the first time in Riesling wine (Figure 4).

2R,3R-Dihydrokaempferol (18). 1.0 mg. UV (MeOH): λ_{max} 208, 292 nm. ESIMS (negative mode): pseudo molecular ion at m/z 287 $[M(288) - H^+]^-$, MS/MS of m/z 287 at m/z 259 $[M - H^+ - CO]^-$. ^{1}H NMR (300 MHz, CD_3OD, ppm): δ 4.54 (1 H, d, J = 11 Hz, H3); 4.98 (1 H, d, J = 11 Hz, H2); 5.88 (1 H, d, J = 2 Hz, H8); 5.93 (1 H, d, J = 2 Hz, H6); 6.83 (2 H, m, H3′/H5′); 7.35 (2 H, m, H2′/H6′).

2R,3R-Dihydrokaempferol 3-*O*-*β*-D-glucoside (18b). 2.6 mg. UV (MeOH): λ_{max} 208, 291 nm; circular dichroism (CD) (c = 0.004% in MeOH): $[\theta]_{293} - 4.1 \times 10^4$; $[\theta]_{337} + 1.0 \times 10^4$. DCIMS (reactant gas, NH_3): pseudo molecular ion at m/z 468 $[M(450) + NH_4]^+$, m/z 451 $[M + H]^+$. ^{1}H NMR (360 MHz, CD_3OD, ppm): δ 2.98 (1 H, ddd, J = 9.5/5.5/2.5 Hz, H5″); 3.10 (1 H, t, J = 9 Hz, H3″); 3.21 (1 H, dd, J = 8/9 Hz, H2″); 3.25 (1 H, dd, J = 9/9.5 Hz, H4″); 3.59 (1 H, dd, J = 12.5/5.5 Hz, H6″a); 3.75 (1 H, dd, J = 12.5/2.5 Hz, H6″b); 3.82 (1 H, d, J = 8 Hz, H1″); 4.95 (1 H, d, J = 10 Hz, H3); 5.26 (1 H, d, J = 10 Hz, H2); 5.89 (1 H, d, J = 2 Hz, H8); 5.91 (1 H, d, J = 2 Hz, H6); 6.81 (2 H, m, H3′/H5′); 7.36 (2 H, m, H2′/H6′). ^{13}C NMR (90.6 MHz, CD_3OD, ppm): δ 62.6 (C6″), 71.2 (C4″), 74.6 (C2″), 77.2 (C3″), 77.6 (C5″), 78.2 (C3), 83.5 (C2), 96.9 (C6/C8), 102.2 (C10), 102.6 (C1″), 116.2 (C3′/C5′), 128.7 (C1′), 130.4 (C2′/C6′), 159.3 (C4′), 164.2 (C9), 165.6 (C5), 170.9 (C7), 195.5 (C4).

2R,3R-Dihydroquercetin (19). 5.6 mg. UV (MeOH): λ_{max} 207, 290 nm; CD (c = 0.01% in MeOH): $[\theta]_{333} + 1.3 \times 10^4$; $[\theta]_{296} - 5.2 \times 10^4$; $[\theta]_{253} + 0.8 \times 10^4$. ESIMS (negative mode): pseudo molecular ion at m/z 303 $[M(304) - H^+]^-$, MS/MS of m/z 303 at m/z 285 $[M - H^+ - H_2O]^-$. ^{1}H NMR (300 MHz, CD_3OD, ppm): δ 4.51 (1 H, d, J = 11.5 Hz, H3); 4.90 (1 H, d, J = 11.5 Hz, H2); 5.88 (1 H, d, J = 2 Hz, H8); 5.92 (1 H, d, J = 2 Hz, H6); 6.80 (1 H, d, J = 8 Hz, H5′); 6.84 (1 H, dd, J = 8/2 Hz, H6′); 6.94 (1 H, d, J = 2 Hz, H2′). ^{13}C NMR (75.5 MHz, CD_3OD, ppm): δ 73.7 (C3), 85.1 (C2), 96.3 (C6), 97.3 (C8), 102.0 (C10), 115.9 (C2′), 116.0 (C5′), 120.9 (C6′), 129.9 (C1′), 145.7 (C3′), 146.7 (C4′), 164.9 (C9), 165.7 (C5), 169.9 (C7), 198.1 (C4).

2R,3R-Dihydroquercetin 3-*O*-*β*-D-glucoside (19b). 37.6 mg. UV (MeOH): λ_{max} 205, 292 nm. CD (c = 0.0056% in MeOH): $[\theta]_{297} - 3.4 \times 10^4$; $[\theta]_{331} + 1.5 \times 10^4$. Thermospray−MS: pseudo molecular ion at m/z 484 $[M(466) + NH_4]^+$, m/z 467 $[M + H]^+$. ^{1}H NMR (360 MHz, CD_3OD, ppm): δ 3.01 (1 H, ddd, J = 9.5/5.5/2.5 Hz, H5″); 3.13 (1 H, t, J = 9 Hz, H3″); 3.22 (1 H, dd, J = 7.5/9 Hz, H2″); 3.24 (1 H, dd, J = 9/9.5 Hz, H4″); 3.61 (1 H, dd, J = 12.5/5.5 Hz, H6″a); 3.78 (1 H, dd, J = 12.5/2.5 Hz, H6″b); 3.89 (1 H, d, J = 7.5 Hz, H1″); 4.92 (1 H, d, J = 9.5 Hz, H3); 5.23 (1 H, d, J = 9.5 Hz, H2); 5.88 (1 H, d,

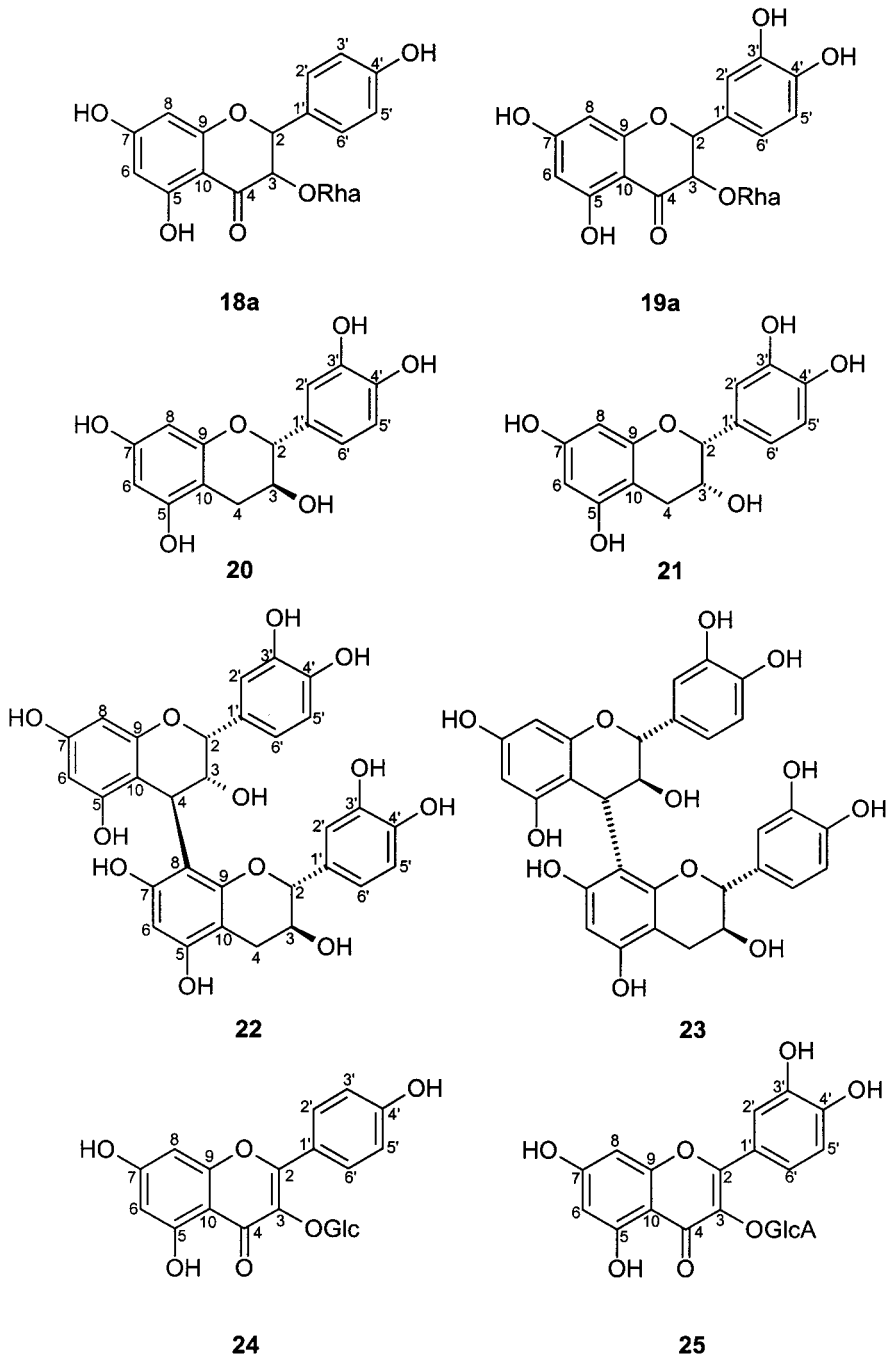

Figure 3. Known flavonoids isolated from a German Riesling wine (Rha, rhamnose; Glc, glucose; GlcA, glucuronic acid).

J = 2 Hz, H8); 5.90 (1 H, d, J = 2 Hz, H6); 6.78 (1 H, d, J = 8 Hz, H5′); 6.84 (1 H, dd, J = 8/2 Hz, H6′); 6.94 (1 H, d, J = 2 Hz, H2′). ^{13}C NMR (90.6 MHz, CD_3OD, ppm): δ 62.6 (C6″), 71.3 (C4″), 74.7 (C2″), 77.2 (C3), 77.7 (C5″), 78.2 (C3″), 83.6 (C2), 96.5 (C6), 97.3 (C8), 102.5 (C10), 102.6 (C1″), 115.9 (C2′), 116.2 (C5′), 121.1 (C6′), 129.1 (C1′), 146.4 (C3′), 147.3 (C4′), 164.1 (C9), 165.5 (C5), 169.8 (C7), 195.7 (C4).

Dihydroquercetin 3′-*O*-*β*-D-glucoside (19c). 1.0 mg. Thermospray−MS: pseudo molecular ion at m/z 484 [M(466) + NH_4]$^+$, 467 [M + H]$^+$. ^{1}H NMR (360 MHz, CD_3OD, ppm): δ 3.37 (1 H, t, J = 9.0 Hz, H4″); 3.44 (1 H, ddd, J = 9.5/5.5/2.5 Hz, H5″); 3.49 (1 H, t, J = 9 Hz, H3″); 3.52 (1 H, dd, J = 8.5/9 Hz, H2″); 3.67 (1 H, dd, J = 12.5/5.5 Hz, H6″a); 3.89 (1 H, dd, J = 12.5/2.5 Hz, H6″b); 4.57 (1 H, d, J = 11 Hz, H3); 4.83 (1 H, d, J = 8.5 Hz, H1″); 4.98 (1 H, d, J = 11 Hz, H2); 5.88 (1 H, d, J = 2 Hz, H8); 5.90 (1 H, d, J = 2 Hz, H6); 6.89 (1 H, d, J = 8 Hz, H5′); 7.10 (1 H, dd, J = 8/2 Hz, H6′); 7.38 (1 H, d, J = 2 Hz, H2′). ^{13}C NMR (90.6 MHz, CD_3OD, ppm): δ 62.6 (C6″), 71.5 (C4″), 73.4 (C3), 74.9 (C2″), 77.7 (C5″), 78.4 (C3″), 85.0 (C2), 96.3 (C8), 97.3 (C6), 102.5 (C10), 104.1 (C1″), 116.9 (C5′), 118.3 (C2′), 124.7 (C6′), 130.0 (C1′), 146.6 (C3′), 149.1 (C4′), 164.5 (C5), 165.3 (C9), 168.9 (C7), 198.5 (C4).

Dihydroquercetin 3-*O*-*β*-D-xyloside (19d). 0.9 mg. Thermospray−MS: pseudo molecular ion at m/z 454 [M(436) + NH_4]$^+$, m/z 437 [M + H]$^+$. ^{1}H NMR (360 MHz, CD_3OD, ppm): δ 3.06 (1 H, dd, J = 12/9 Hz, H5″a); 3.23 (2 H, m, H2″/H3″); 3.51 (1 H, dt, J = 4.5/9 Hz, H4″); 3.89 (1 H, d, J = 7 Hz, H1″); 3.94 (1 H, dd, J = 12/4.5 Hz, H5″b); 4.74 (1 H, d, J = 10 Hz, H3); 5.21 (1 H, d, J = 10 Hz, H2); 5.92 (1 H, d, J = 2 Hz, H8); 5.93 (1 H, d, J = 2 Hz, H6); 6.78 (1 H, d, J = 8 Hz, H5′); 6.82 (1 H, dd, J = 8/2 Hz, H6′); 6.94 (1 H, d, J = 2 Hz, H2′). ^{13}C NMR (90.6 MHz, CD_3OD, ppm): δ 65.9 (C5″), 70.8 (C4″), 73.5 (C2″), 75.7 (C3″), 77.5 (C3), 83.6 (C2), 96.3 (C8), 97.4 (C6), 101.9 (C10), 103.1 (C1″), 115.7 (C2′), 116.2 (C5′), 120.8 (C6′), 129.0 (C1′), 146.1 (C3′), 147.4 (C4′), 164.1 (C5), 165.4 (C9), 168.8 (C7), 195.6 (C4).

Isolation and Characterization of Novel Lignans and Neolignans. In addition to the known isolariciresinol 4′-*O*-

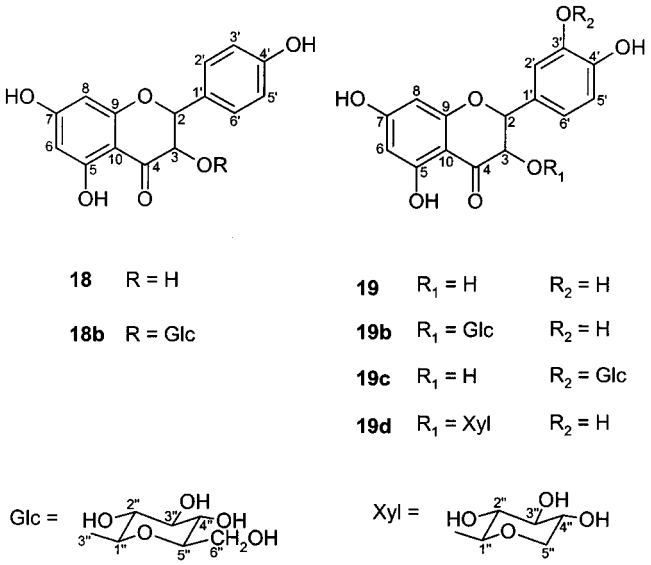

Figure 4. Structures of novel dihydroflavonol derivatives isolated from a German Riesling wine: 2*R*,3*R*-dihydrokaempferol **18**; 2*R*,3*R*-dihydrokaempferol 3-*O*-*β*-D-glucoside **18b**; 2*R*,3*R*-dihydroquercetin **19**; 2*R*,3*R*-dihydroquercetin 3-*O*-*β*-D-glucoside **19b**; dihydroquercetin 3′-*O*-*β*-D-glucoside **19c**; and dihydroquercetin-3-*O*-xyloside **19d**.

β-D-glucoside **30a** (21.2 mg), the following neolignans and lignan derivatives were isolated for the first time from Riesling wine (Figure 5).

2R,3R-2,3-Dihydro-2-(4′-hydroxy-3′-methoxyphenyl)-3-(glucosyloxymethyl)-7-methoxy-benzofuran-5-propanol (Dihydrodehydrodiconiferyl Alcohol *β*-D-Glucoside) (26). 5.2 mg. UV (MeOH): λ_{max} 282, 213 nm. CD (c = 0.05% in MeOH): $[\theta]_{294}$ +1.7 × 10^3; $[\theta]_{260}$ −0.05 × 10^3; $[\theta]_{243}$ +4.3 × 10^3. ORD (c = 0.05% in MeOH): $[\Phi]_{299}$ +1.6 × 10^3; $[\Phi]_{278}$ −0.6 × 10^3; $[\Phi]_{249}$ +1.6 × 10^3. ESIMS (positive mode): pseudo molecular ion at m/z 545 $[M(522) + Na]^+$, MS/MS of m/z 545 at m/z 383 $[M + Na - \text{anhydroglucose}]^+$, and m/z 365 $= [M + Na - \text{glucose}]^+$. ^{1}H NMR (300 MHz, CD_3OD, ppm): δ 1.82 (2 H, brqi, J = 8 Hz, H9); 2.62 (2 H, brt, J = 8 Hz, H8); 3.23 (1 H, dd, J = 8.5/9 Hz, H2″); 3.27−3.40 (3 H, m, H3″/H4″/H5″); 3.56 (2 H, t, J = 8 Hz, H10); 3.61−3.71 (2 H, m, H3/H6″a); 3.79−3.90 (2 H, m, H11a/H6″b); 3.83 (3 H, s, OCH_3−C3); 3.86 (3 H, s, OCH_3−C3′); 4.10 (1 H, dd, J = 9.5/8 Hz, H11b); 4.35 (1 H, d, J = 7.5 Hz, H1″); 5.58 (1 H, d, J = 6 Hz, H2); 6.72 (1 H, brs, H4 or H6); 6.76 (1 H, d, J = 8.5 Hz, H5′); 6.80 (1 H, brs, H6 or H4); 6.86 (1 H, dd, J = 8.5/2 Hz, H6′); 7.00 (1 H, d, J = 2 Hz, H2′). ^{13}C NMR (75.5 MHz, CD_3OD, ppm): δ 32.9 (C9), 35.8 (C8), 52.9 (C3), 56.5 and 56.8 (2 × OCH_3), 62.3 (C10), 62.8 (C6″), 71.7 (C4″), 72.4 (C11), 75.1 (C2″), 78.0 (C3″), 78.2 (C5″), 89.2 (C2), 104.3 (C1″), 111.0 (C2′), 114.4 (C4 or C6), 116.1 (C5′), 118.3 (C6 or C4), 119.8 (C6′), 129.8 (C5), 134.7 (C1′), 137.0 (C3a), 145.2 (C7), 147.5, 147.9 (C7a/C4′), 149.0 (C3′).

2R,2R-2,3-Dihydro-2-(4′-hydroxy-3′-methoxyphenyl)-3-(glucosyloxymethyl)-7-hydroxy-5-benzofuranpropanol (27). 1.6 mg. UV (MeOH): λ_{max} 282, 207 nm. CD (c = 0.05% in MeOH, d = 1 cm): $[\theta]_{294}$ +2.3 × 10^3; $[\theta]_{263}$ −0.9 × 10^3; $[\theta]_{240}$ +2.1 × 10^3; $[\theta]_{226}$ −1.7 × 10^3; $[\theta]_{210}$ +10.9 × 10^3. ORD (c = 0.05% in MeOH, d = 1 cm): $[\Phi]_{300}$ +2.1 × 10^3; $[\Phi]_{282}$ −0.8 × 10^3; $[\Phi]_{248}$ +1.2 × 10^3; $[\Phi]_{234}$ −1.7 × 10^3. Thermospray−MS: pseudo molecular ion at m/z 526 = $[M(508) + NH_4]^+$; m/z 329 $[M - \text{glucose} + H]^+$. ^{1}H NMR (360 MHz, CD_3OD, ppm): δ 1.78 (2 H, m, H9); 2.55 (2 H, m, H8); 3.23 (1 H, dd, J = 8/9.5 Hz, H2″); 3.28−3.40 (3 H, m, H3″/H4″/H5″); 3.55 (2 H, m, H10); 3.62 (1 H, m, H3); 3.69 (1 H, dd, J = 12/5.5 Hz, H6″a); 3.82 (3 H, s, H_3CO−C3′); 3.85 (1 H, dd, J = 12.5/2.5 Hz, H6″b); 3.86 (1 H, d, J = 9.5 Hz, H11a); 4.10 (1 H, dd, J = 9.5/8 Hz, H11b); 4.35 (1 H, d, J = 8 Hz, H1″); 5.57 (1 H, d, J = 6.5 Hz, H2); 6.56 (1 H, s, H6); 6.67 (1 H, s, H4); 6.75 (1 H, dd, J = 8/2 Hz, H6); 6.87 (1 H, d, J = 8 Hz, H5); 7.02 (1 H, d, J = 2 Hz, H2). ^{13}C NMR (90.6 MHz, CD_3OD, ppm): δ 32.7 (C9), 35.8 (C8), 53.2 (C3), 56.5 (OCH3), 62.3 (C10), 62.8 (C6″), 71.7 (C4″), 72.5 (C11), 75.2 (C2″), 78.1 (C3″), 78.2 (C5″), 88.9 (C2) 104.3 (C1″), 110.8 (C2′), 116.0 (C4), 117.0 (C5′), 119.8 (C6), 123.1 (C6′), 129.6 (C5), 135.0 (C1′), 136.8 (C3a), 141.9 (C7), 146.4 (C7a), 147.3 (C4′), 149.0 (C3′).

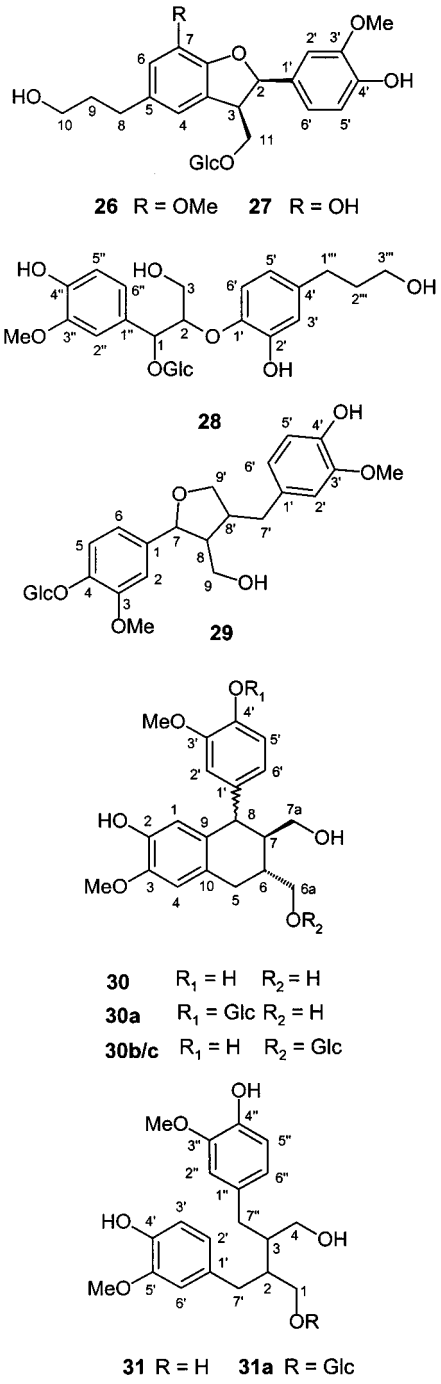

Figure 5. Structures of isolated neolignans **26**−**28** and lignans **29**−**31a**.

1-Glucosyloxy-2-[2-hydroxy-4-(3-hydroxy-propyl)-phenoxy]-1-(4-hydroxy-3-methoxy-phenyl)-propan-3-ol (28). 1.0 mg. UV (MeOH): λ_{max} 275, 208 nm. CD (0.01% in MeOH, d = 1 cm): $[\theta]_{312}$ −0.4 × 10^3; $[\theta]_{300}$ −0.0 × 10^3; $[\theta]_{271}$ +3.1 × 10^3; $[\theta]_{246}$ +0.4 × 10^3; $[\theta]_{235}$ +4.1 × 10^3; $[\theta]_{225}$ +0.7 × 10^3. ESIMS (negative mode): pseudo molecular ion at m/z 525 $[M(526) - H^+]^-$; MS/MS of m/z 525 at m/z 507 $[M - H^+ - H_2O]^-$ and m/z 345 $[M - H^+ - H_2O - \text{anhydroglucose}]^-$. ^{1}H NMR (300 MHz, CD_3OD, ppm): δ 1.77 (2 H, m, H2‴); 2.54 (2 H, t, J = 7 Hz, H1‴); 3.24 (1 H, dd, J = 8/9 Hz, H2‴′); 3.30−

3.45 (3 H, m, H3‴/H4‴/H5‴); 3.53 (2 H, t, J = 7 Hz, H3‴); 3.65 (1H, dd, J = 12.5/5 Hz, H6‴a); 3.84 (1 H, dd, J = 12.5/2 Hz, H6‴b); 3.85 (3 H, s, OCH_3); 3.8−3.9 (2 H, m, H3); 4.32 (1 H, d, J = 8 Hz, H1‴); 4.35 (1 H, m, H2); 4.99 (1 H, d, J = 5.5 Hz, H1); 6.55 (1 H, dd, J = 2/8 Hz, H5′); 6.67 (1 H, d, J = 2 Hz, H3′); 6.74 (1 H, d, J = 8 Hz, H5″); 6.88 (1 H, dd, J = 2/8 Hz, H6″); 6.89 (1 H, d, J = 8 Hz, H6′); 7.04 (1 H, d, J = 2 Hz, H2″).

1-{4-[4-(4-Hydroxy-3-methoxy-benzyl)-3-hydroxymethyl-tetrahydrofuran-2-yl]-2-methoxy-phenoxy}-*β*-D-glucopyranose (Lariciresinol 4-*O*-*β*-D-Glucoside) (29). 5.9 mg. UV (MeOH): λ_{max} 279, 225, 212 nm. CD (0.01% in MeOH, d = 1 cm): $[\theta]_{299}$ +0.4 × 10^3; $[\theta]_{293}$ 0; $[\theta]_{280}$ −1.8 × 10^3; $[\theta]_{259}$ −0.8 × 10^3; $[\theta]_{235}$ −9.5 × 10^3. ESIMS (negative mode): pseudo molecular ion at m/z 521 $[M(522) - H^+]^-$; MS/MS of m/z 521 at m/z 359 $[M - H^+ -$ anhydroglucose$]^-$. ^{1}H NMR (300 MHz, CD_3OD, ppm): δ 2.35 (1 H, qi, J = 6.5 Hz, H8); 2.55 (1 H, dd, J = 11/13 Hz, H7′a); 2.72 (1 H, m, H8′); 2.91 (1 H, dd, J = 5.5/13 Hz, H7′b); 3.66 (1 H, dd, J = 7/11 Hz, H9a); 3.68 (1 H, dd, J = 2.5/12 Hz, H6″a); 3.73 (1 H, dd, J = 6.5/8 Hz, H9′a); 3.83 and 3.86 (2 × 3 H, 2 × s, 2 × OCH_3); 3.86 (2 H, m, H9b/H6″b); 4.0 (1 H, dd, J = 6.5/8 Hz, H9′b); 4.82 (1 H, d, J = 6.5 Hz, H7); 4.87 (1 H, d, J = 7.5 Hz, H1″); 6.66 (1 H, dd, J = 2/8 Hz, H6′); 6.72 (1 H, d, J = 8 Hz, H5′); 6.79 (1 H, d, J = 2 Hz, H2′); 6.88 (1 H, dd, J = 2/8 Hz, H6); 6.99 (1 H, d, J = 2 Hz, H2); 7.14 (1 H, d, J = 8 Hz, H5).

6R,7S,8S-8-(4-Hydroxy-3-methoxyphenyl)-6,7-bis-hydroxymethyl-3-methoxy-5,6,7,8-tetrahydro-naphthalen-2-ol ((+)-Isolariciresinol) (30). 4.2 mg. UV (MeOH): λ_{max} 284, 212 nm. CD (0.01% in MeOH, d = 1 cm): $[\theta]_{293}$ −8.3 × 10^3; $[\theta]_{277}$ +5.3 × 10^3; $[\theta]_{253}$ +0.3 × 10^3; $[\theta]_{240}$ +10.2 × 10^3. ORD (0.01% in MeOH, d = 1 cm): $[\Phi]_{320}$ 0; $[\Phi]_{299}$ -3.2 × 10^3; $[\Phi]_{295}$ 0; $[\Phi]_{284}$ +9.4 × 10^3; $[\Phi]_{267}$ + 0.3 × 10^3; $[\Phi]_{244}$ + 6.2 × 10^3; $[\Phi]_{239}$ 0. ESIMS (negative mode): pseudo molecular ion at m/z 359 $[M(360) - H^+]^-$; MS/MS of m/z 359 at m/z 344 $[M - H^+ - CH_3]^-$. ^{1}H NMR (300 MHz, CD_3OD, ppm): δ 1.78 (1 H, m, H7); 2.0 (1 H, m, H6); 2.78 (2 H, brd, J = 8 Hz, H5); 3.40 (1 H, dd, J = 4.5/11 Hz, H7aa); 3.62−3.71 (3 H, m, H7ab/H6a); 3.78 (3 H, s, OCH_3−C3′); 3.79 (1 H, d, J = 10 Hz, H8); 3.81 (3 H, s, OCH_3−C6); 6.18 (1 H, brs, H1); 6.61 (1 H, dd, J = 2/8 Hz, H6′); 6.67 (1 H, brs, H4); 6.68 (1 H, d, J = 2 Hz, H2′); 6.74 (1 H, d, J = 8 Hz, H5′).

6R,7S,8S-6-Glucosyloxymethyl-8-(4-hydroxy-3-methoxyphenyl)-7-hydroxymethyl-3-meth-oxy-5,6,7,8-tetrahydronaphthalen-2-ol ((+)-Isolariciresinol 6a-*O*-*β*-D-Glucoside) (30b). 1.3 mg. UV (MeOH): λ_{max} 283, 206 nm. CD (c = 0.01% in MeOH, d = 1 cm): $[\theta]_{293}$ +3.3 × 10^3; $[\theta]_{277}$ +11.7 × 10^3; $[\theta]_{256}$ −2.4 × 10^3; $[\theta]_{240}$ −29.9 × 10^3; $[\theta]_{224}$ −4.3 × 10^3; $[\theta]_{218}$ −7.4 × 10^3. ESIMS (negative mode): pseudo molecular ion at m/z 521[M(522) − H^+]$^-$; MS/MS of m/z 521 at m/z 359 $[M - H^+ -$ anhydroglucose$]^-$. ^{1}H NMR (400 MHz, CD_3OD, ppm): δ 1.78 (1 H, m, H7); 2.17 (1 H, m, H6); 2.84 (1 H, brd, H5); 3.20 (1 H, dd, J = 8/9 Hz, H2″); 3.27−3.40 [4 H, m, obscured at 3.35 (1H, m, H7aa); (3H, m, H3″/H4″/H5″)]; 3.64 (1H, m, H6aa); 3.68 (1H, m, H7ab); 3.72 (1 H, dd, J = 4/12 Hz, H6″a); 3.78 (3 H, s, OCH_3); 3.80 (3 H, s, OCH_3); 3.84 (1 H, brd, J = 10 Hz, H1); 3.87 (1 H, dd, J = 2/12 Hz, H6″b); 4.02 (1 H, dd, J = 6/10 Hz, H6ab); 4.29 (1H, d, J = 8 Hz, H1″); 6.18 (1 H, s, H8); 6.61 (1 H, dd, J = 2/8 Hz, H6′); 6.64 (1 H, s, H5); 6.68 (1 H, d, J = 2 Hz, H2′); 6.73 (1 H, d, J = 8 Hz, H5′). ^{13}C NMR (90.6 MHz, CD_3OD, ppm): δ 33.1 (C5), 37.4 (C6), 45.3 (C7), 47.9 (C8), 56.4 (2 × OCH_3), 61.6 (C7a), 62.8 (C6″), 65.2 (C6a), 71.7 (C4″), 73.9 (C6a), 75.2 (C2″), 78.0 (C3″), 78.2 (C5″), 104.6 (C1″), 112.4 (C4), 113.9 (C2′), 116.0 (C5′), 117.4 (C1), 123.3 (C6′), 129.1 (C9), 134.2 (C1′), 138.7 (C10), 145.2 (C4′), 145.9 (C2), 147.2 (C3), 149.0 (C3′).

6-Glucosyloxymethyl-8-(4-hydroxy-3-methoxyphenyl)-7-hydroxymethyl-3-methoxy-5,6,7,8-tetrahydronaphthalen-2-ol (Isolariciresinol 6a-*O*-*β*-D-Glucoside) (30c). 0.9 mg. UV (MeOH): λ_{max} 283, 206 nm. CD (c = 0.1% in MeOH, d = 1 cm): $[\theta]_{293}$ +3.3 × 10^3; $[\theta]_{277}$ +11.7 × 10^3; $[\theta]_{256}$ −2.4 × 10^3; $[\theta]_{240}$ −29.9 × 10^3; $[\theta]_{224}$ −4.3 × 10^3; $[\theta]_{218}$ −7.4 × 10^3. ESIMS (negative mode): pseudo molecular ion at m/z 521 $[M(522) - H^+]^-$; MS/MS of m/z 521 at m/z 359 $[M - H^+ -$ anhydroglucose$]^-$. ^{1}H NMR (300 MHz, CD_3OD, ppm): δ 2.11 (1 H, m, H7); 2.21 (1 H, m, H6); 2.72 (1 H, dd, J = 9.5/17 Hz, H5a); 2.99 (1 H, dd, J = 6/17 Hz, H5b); 3.16 (1 H, dd, J = 8/9 Hz, H2″); 3.23−3.35 (4 H, m, H7aa/H3″/H4″/H5″); 3.56 (1 H, dd, J = 5.5/9.5 Hz, H6aa); 3.65 (1 H, dd, J = 6.5/11 Hz, H7ab); 3.65 (1 H, dd, J = 5.5/12 Hz, H6″a); 3.75 (3 H, s, OCH_3); 3.83 (3 H, s, OCH_3); 3.84 (1 H, dd, J = 2.5/12 Hz, H6″b); 3.91 (1 H, dd, J = 6/9.5 Hz, H6aa); 4.21 (1 H, d, J = 4.5 Hz, H8); 4.23 (1 H, d, J = 8 Hz, H1″); 6.35 (1 H, brs, H1); 6.45 (1 H, dd, J = 2/8 Hz, H6′); 6.64 (1 H, d, J = 8 Hz, H5′); 6.69 (1 H, brs, H4); 6.70 (1 H, d, J = 2 Hz, H2′). ^{13}C NMR (90.6 MHz, CD_3OD, ppm): δ 33.0/33.2(C6/C5), 44.7/46.2 (C7/C8), 56.3/56.4 (2 × OCH3), 62.8/62.9 (C7a/C6″), 71.6 (C4″), 73.4 (C6a), 75.1 (C2″), 77.1 (C3″), 78.2 (C5″), 104.7 (C1″), 112.4 (C4), 115.3/115.5 (C2′/C5′), 117.1 (C1), 124.1 (C6′), 128.6 (C9), 133.0 (C1′), 135.9 (C10), 145.5 (C4′), 145.8 (C2), 147.8 (C3), 148.3 (C3′).

2R,3R-2,3-Bis-(4-hydroxy-3-methoxy-benzyl)-butan-1,4-diol ((−)-Secoisolariciresinol) (31). 1.0 mg. UV (MeOH): λ_{max} 282, 209 nm. ESIMS (negative mode): pseudo molecular ion at m/z 361 $[M(362) - H^+]^-$; MS/MS of m/z 361 at m/z 346 $[M - H^+ - CH_3]^-$. ^{1}H NMR (300 MHz, CD_3OD, ppm): δ 1.91 (2 H, m, H3/H4); 2.56 (2 H, dd, J = 7.5/14 Hz, H7a/H7′a); 2.67 (2 H, dd, J = 7/14 Hz, H7b/H7′b); 3.59 (4 H, m, H1/H4); 3.74 (6 H, s, 2 × OCH_3); 6.55 (2 H, dd, J = 2/8 Hz, H6′/H6″); 6.59 (2 H, d, J = 2 Hz, H2′/H2″); 6.66 (2 H, d, J = 8 Hz, H5′/H5″).

1-Glucosyloxy-2,3-bis-(4-hydroxy-3-methoxy-benzyl)-butan-4-ol (Secoisolariciresinol *β*-D-Glucoside) (31a). 1.2 mg. UV (MeOH): λ_{max} 282, 210 nm. ESIMS (negative mode): pseudo molecular ion at m/z 523 $[M(524) - H^+]^-$; MS/MS of m/z 523 at m/z 361 $[M - H^+ -$ anhydroglucose$]^-$. ^{1}H NMR (300 MHz, CD_3OD, ppm): δ 2.01 (1 H, m, H3); 2.08 (1 H, m, H2); 2.57−2.75 (4 H, m, H7′/H7″); 3.21 (1 H, dd, J = 8/9 Hz, H2‴); 3.30−3.40 (3 H, m, H3‴/H4‴/H5‴); 3,56 (1 H, dd, J = 5.5/10.5 Hz, H1a); 3.57 (1 H, dd, J = 6/11 Hz, H4a); 3.65 (1 H, dd, J = 5.5/11 Hz, H4b); 3.68 (1 H, dd, J = 5.5/12 Hz, H6‴a); 3.76 (6 H, s, 2 × OCH_3); 3.87 (1 H, dd, J = 2/12 Hz, H6‴b); 3.90 (1 H, dd, J = 5.5/10.5 Hz, H1a); 4.19 (1 H, d, J = 8 Hz, H1‴); 6.57 and 6.58 (2H, 2 × dd, J = 2/8 Hz, H6′/H6″); 6.63 and 6.65 (2H, 2 × d, J = 2 Hz, H2′/H2″); 6.66 and 6.67 (2H, 2 × d, J = 8 Hz, H5′/H5″).

Nuclear Magnetic Resonance (NMR) Spectroscopy. ^{1}H and ^{13}C NMR spectral data were recorded on Fourier transform Bruker AM 360, AC 250, and AMX 300 spectrometers with TMS as internal reference standard.

Mass Spectrometry (MS). Electronic impact ionization MS (EIMS) and desorption chemical ionization MS (DCIMS) were carried out with a Finnigan TSQ 70 mass spectrometer at 70 eV using ammonia as reactant gas for chemical ionization. Thermospray−MS data were recorded on a Finnigan SSQ 710 using ammonium acetate (0.1 mM in 10% methanol) as buffer (vaporizer, 85 °C; aerosol, 260 °C; SEV, 1200 V). Electrospray ionization ion trap multiple mass spectrometry (ESIMS) data were obtained using a Bruker Esquire LC MS/MS system with electrospray ionization.

Circular Dichroism (CD) and Optical Rotatory Dispersion (ORD). Spectra were recorded with a JASCO J-710 polarimeter at 20 °C (d = 1 cm).

Capillary Electrophoresis (CE). Sugar analysis was performed by capillary electrophoresis using a Beckman P/ACE DNA system with a diode array detector according to the method of Hirsch and Maier (*7*). Prior to analysis, sugars were liberated from their glycoconjugates by acidic hydrolysis and derivatized with ethyl *p*-aminobenzoate to give UV-active compounds.

RESULTS AND DISCUSSION

In the course of an ongoing study on antioxidants in white wine, a German Riesling wine has been fractionated by chromatographic techniques and 101 polar wine constituents have been obtained (*3*). Among the isolated compounds seven new stilbene derivatives have been identified, which have been discussed in detail in a recent publication (*3*). Here we report the isolation and structural elucidation of additional novel wine constitu-

ents which belong to the classes of cinnamates, benzoates, flavonoids, and lignans.

Whereas stilbenes have been recognized as potent bioactive compounds in wine, the class of simple phenolic acids, such as cinnamates and benzoates, has attracted minor attention so far. Cinnamic acid derivatives are present in the vacuolar fluid of grapes in substantial amounts. As free acids they are less abundant but as esters with tartaric acid they are quite common and constitute a major source of phenolic substances in white wine. Caffeoyl tartaric acid **3b** (Figure 1) is known to be the major hydroxycinnamate ester in grape juices and wines, accounting for more than 50% of the total hydroxycinnamates (*4*). Somers et al. (*8*) identified 10 hydroxycinnamic acid derivatives in a young Riesling wine. Six of them were also isolated during the present study. In addition to (*E*)-caffeoyl tartaric acid **3b**, (*E*)-*p*-coumaric acid **1a**, (*E*)-*p*-coumaroyl tartaric acid **1b**, (*E*)-caffeic acid **3a**, (*E*)-ethyl caffeate **3c**, and (*E*)-feruloyl tartaric acid **4b** were found. The presence of the *cis*-isomers of *p*-coumaric acid **2a** and the tartaric acid ester **2b** in Riesling wine has previously been reported by Baranowski and Nagel (*9*). (*E*)-Ferulic acid **4a** has been detected in wine by Drawert et al. (*10*). The 4-*O*-glucosides of (*E*)- and (*Z*)-*p*-coumaric acid **1c** and **2c** have been identified in white wine by Biau et al. (*51*). Of the additional compounds with phenylpropanoid structure, i.e., compounds **5**−**7**, the first two of these have previously been reported by Drawert et al. (*6*), whereas the glycerol adduct **7** was most recently identified in a French Gewürztraminer wine (*11*).

In comparison with cinnamic acid derivatives, benzoates are present at much lower levels in wine. For gallic acid, for example, concentrations were determined to be in a range of 1−3 mg/L (*12*). The composition of benzoic acid derivatives has mainly been studied by GC after preparation of trimethylsilyl derivatives or after methylation (*6*, *10*). Four benzoates which have been reported as wine constituents by Drawert et al. (*6*, *10*) and Güntert et al. (*13*) were also isolated in the present study, i.e., protocatechuic acid **8a** and its methyl ester **8b**, gentisic acid **9**, and syringic acid **10** (Figure 1).

Isolation and Characterization of Novel Phenylpropanoids and Benzoates. Twelve novel members in this category have been identified during the present study, the structures of which (as shown in Figure 2) have been elucidated on the basis of MS and NMR spectroscopic data.

Glucose Esters and Glucosides of Cinnamic Acids. Besides the esters of cinnamic acid derivatives with tartaric acid, esters of (*E*)-*p*-coumaric acid and (*E*)-ferulic acid with β-glucose (compounds **1d** and **4c**) were obtained. The occurrence of *p*-coumaroyl and feruloyl glucose esters in grapes has been reported by Reschke and Herrmann (*14*). Ong and Nagel (*15*) tentatively identified two compounds in White Riesling juice which consisted of either caffeic acid or *p*-coumaric acid, glucose, and tartaric acid. However, up to now there have been no reports about the presence of glucose esters **1d** and **4c** in wine. Additionally, the glucose ester of cinnamic acid **13** as well as the 4-*O*-glucoside of (*E*)-ferulic acid **4d** have been identified for the first time in Riesling wine. The majority of the glucoconjugates have been isolated in their free form; only the purification of cinnamoyl glucose ester **13** required a derivatization step. Glucoconjugate **13** was acetylated and characterized as its peracetate. The position of the linkage of the glucose moiety in the above-mentioned compounds was deduced from the chemical shift of the anomeric sugar proton. In glycosides the resonances for the β-anomeric protons usually appear as doublet ($J = 8$ Hz) between 4.4 and 4.8 ppm. In glucose esters this resonance is shifted downfield by about 1 ppm. Additionally, a characteristic feature of glucose esters is the appearance of the signal of the anomeric carbon of the sugar residue at a remarkably upfield position (93−97 ppm) in the carbon spectrum (*16*). For the carboxylic resonances an upfield shift by 2−5 ppm along with a downfield shift (0.5−2.0 ppm) of the resonances of the adjacent carbon atoms caused by glycosylation can be observed.

(*E*)-Caffeoyl Ethyl Tartrates and Additional Phenylpropanoids. Whereas Somers et al. (*8*) identified the diethyl ester of (*E*)-caffeoyl tartaric acid as a minor constituent in a young Riesling wine, we were able to isolate two compounds whose structures were elucidated as isomeric (*E*)-caffeoyl tartrates esterified with a single molecule of ethanol. Both compounds gave an identical MS/MS fragmentation pattern and almost equivalent proton NMR spectra. Differences were observed for the resonance signals of the methylene group in the ethyl moiety. Whereas for compound **3d** a quartet was obtained, the proton NMR spectra of compound **3e** revealed a multiplet signal indicating a hindered rotation for the ethyl ester group. This observation led us to conclude that compounds **3d** and **3e** are two isomeric (*E*)-caffeoyl ethyl tartates differing in the positions of their ethyl ester linkages.

Moreover, two structurally related phenylpropanoids were identified which are new to Riesling wine, i.e., 3-hydroxy-1-(4-hydroxy-3-methoxyphenyl)-propan-1-one (**14**) and the dihydroxy-derivative **15** (Figure 2). Spectral data obtained for compounds **14** and **15** were in good agreement with previously published data (*17*−*19*).

Ethyl Esters of Protocatechuic and Gallic Acid and Glucose Ester of Vanillic Acid. Compounds **8c** and **16** were identified as ethyl esters of protocatechuic and gallic acid, respectively. The structure of compound **11** was elucidated as glucose ester of vanillic acid. The occurrence of the ethyl esters of (*E*)-*p*-coumaric acid and (*E*)-caffeic acid in Riesling wine has been reported by Somers et al. (*8*). Güntert et al. (*13*) identified ethyl vanillate, ethyl 4-hydroxybenzoate, and ethyl 4-hydroxyphenylacetate, as well as the methyl esters of protocatechuic and vanillic acid, in Riesling wine. However, to the best of our knowledge the presence in Riesling wine of ethyl protocatechuate **8c** and ethyl gallate **16**, as well as the occurrence of the glucose ester of vanillic acid **11**, has not been reported up to now.

Occurrence of Flavonoids in Wine. The increasing interest in flavonoids in recent years is mainly linked to the recognition of their physiological properties. Besides their antibacterial, antiviral, antiinflammatory, antiallergenic, and vasodilatory activities (*20*), antioxidant and anticarcinogenic properties of flavonoids have also been reported (*21*−*23*). The potential role of flavonoids in coronary heart disease prevention was underpinned by epidemiological studies which showed that the consumption of a flavonoid-rich diet is inversely associated with mortality from coronary heart disease (*24*). The potential health effects of moderate wine consumption have also been linked to the flavonoid content of red wine (*25*). Whereas nonflavonoid phenolics are almost equally distributed in white and red wine, the flavonoid content differs significantly. In red

wines, flavonoids commonly constitute more than 85% of the phenolic substances, whereas in white wine they attain only approximately 20% of the total polyphenol content. Because flavonoids originate from the skins, seeds, and stems of grapes, the extraction yield during vinification is mainly influenced by factors such as temperature and length of skin contact. Therefore, the flavonoid content in red wine is about 20−30-fold (> 1000 mg/L) the content in white wine (*20*). Lea et al. (*26*) showed the presence of (+)-catechin (**20**) and (−)-epicatechin (**21**) in almost equal amounts in white wine, as well as the occurrence of four procyanidins. Also, the existence of minor dimeric, trimeric, oligomeric, and polymeric procyanidins was reported. The gallic acid ester of catechin and two catechin-catechin-gallate isomers were tentatively identified in white grapes by Lee and Jaworski (*27*). Singleton and Trousdale (*28*) isolated two dihydroflavonol derivatives, i.e., the 3-*O*-rhamnosides of dihydrokaempferol and dihydroquercetin, engeletin **18a** and astilbin **19a**, repectively, for the first time from skins of white grapes. Their presence in white wine was proven by HPLC analysis. Concentrations were in a range of < 0.02 to 2 mg/L (*28*). Flavonol derivatives are found in white wines at very low levels and very few papers deal with the flavonol composition of white grapes or wines. White grape varieties have been shown by Cheynier and Rigaud (*29*) to contain quercetin 3-glucoside and kaempferol 3-glucoside along with the corresponding 3-glucuronides and trace amounts of diglycosylated flavonols. Strauss et al. (*30*) reported the presence of kaempferol-3-*O*-glucoside (**24**) in Muscat of Alexandria grapes. Quercetin 3-*O*-glucuronide (**25**) was the only flavonol derivative which was detected in Spanish white wines by Betés-Saura et al. (*31*).

Of the known wine constituents, engeletin (**18a**), astilbin (**19a**), catechin (**20**), epicatechin (**21**), procyanidins B1 and B3 (**22** and **23**), kaempferol 3-*O*-glucoside (**24**), and quercetin 3-*O*-glucuronide (**25**) (Figure 3) could be isolated and unambiguously identified by comparison with published spectral data.

Isolation and Characterization of Novel Dihydroflavonol Derivatives from Riesling Wine. Besides the already known dihydroflavonol 3-*O*-rhamnosides, astilbin **18a** and engeletin **19a**, six additional dihydroflavonol derivatives were isolated for the first time from Riesling wine (see Figure 4). The newly identified dihydroflavonols were dihydrokaempferol **18** and dihydroquercetin **19**, as well as four glycosylated derivatives. The structures of compounds **18b** and **19b** were elucidated as the 3-*O*-glucosides of dihydrokaempferol and dihydroquercetin, respectively. The spectral data for **19b** were in good agreement with literature data (*32*). Proton-NMR signals for compound **18b** were almost identical with the signals obtained for compound **19b** except that the signals for the trisubstituted aromatic ring were replaced by signals for a *p*-hydroxyphenyl unit. The proton NMR spectra revealed a downfield shift for H2 and H3 caused by glycosylation at C3 for compounds **18b** and **19b**, **19d**. For **19c**, a significant downfield shift of H2′ by 0.42 ppm indicated the linkage of the glucose unit via carbon atom C3′. The stereochemistry of glycoconjugates **18b** and **19b**−**d** was deduced from the coupling constants between H2 and H3 ($J \approx 11$ Hz) indicating a *trans*-configuration. The absolute stereochemistry was clarified with the help of the CD data. A positive maximum at about 330 nm and a negative maximum at about 295 nm are typical for 2*R*,3*R*-configuration (*33*). The sugar units of dihydroquercetin derivatives **19c** and **19d** were identified by capillary electrophoresis after acid hydrolysis of the glycoconjugates and derivatization of the liberated sugars (*7*). These compounds could be identified as dihydroquercetin 3′-*O*-glucoside (**19c**) and dihydroquercetin 3-*O*-xyloside (**19d**).

Isolation of Novel Lignan and Neolignan Derivatives. Up to the present, knowledge about the occurrence of lignans and neolignans in white wine has been very limited. In 1992, Marinos and co-workers (*34*) were able to identify isolariciresinol 4′-*O*-*β*-glucopyranoside **30a** and cedrusin 4′-*O*-*β*-D-glucoside in an Australian Riesling wine. Moreover, on the basis of FABMS/MS data, Marinos (*35*) tentatively identified a further lignan glucoconjugate, i.e., *seco*-isolariciresinol *β*-D-glucoside **31a**. In addition to glucoconjugates **30a** and **31a**, eight novel lignan and neolignan derivatives were identified in the course of our investigation.

Neolignans 26−28. The structures of 2*R*,3*R*-2,3-dihydro-2-(4′-hydroxy-3′-methoxyphenyl)-3-(glucosyloxymethyl)-7-methoxy-5-benzofuranpropanol (**26**), 2*R*,3*R*-2,3-dihydro-2-(4′-hydroxy-3′-methoxyphenyl)-3-(glucosyloxymethyl)-7-hydroxy-5-benzofuranpropanol (**27**), and 1-glucosyloxy-2-[2-hydroxy-4-(3-hydroxypropyl)-phenoxy]-1-(4-hydroxy-3-methoxyphenyl)-propan-3-ol (**28**) were deduced from mass spectrometric, and one and two-dimensional NMR, as well as chiroptical data. ^{1}H- and ^{13}C NMR data for the aglycone moiety of glucoside **26** were in good agreement with data published for the neolignan dihydrodehydrodiconiferyl alcohol (*36*, *37*). A downfield shift of the resonance for C-11 of approximately 7 ppm, compared to that of the nonglycosylated neolignan alcohol (*37*), indicated glycosylation via the hydroxymethylene group C-11. On the basis of the chiroptical data (CD- and ORD spectra) the 2*R*,3*R*-configuration could be assigned (36). The NMR data for neolignan glucoside **27** were almost identical with data obtained for compound **26**. The missing signal for the methoxy group at C-7 and a difference of 14 amu in the molecular mass indicated the presence of a hydroxyl group instead of the methoxy function. With regard to the absolute stereochemistry, ORD data clearly allowed assignment of the 2*R*,3*R*-configuration (*36*). The structure of the third neolignan was established as compound **28**, a glucoconjugate previously identified by Popoff and Theander in *Pinus sylvestris* (*38*). A positive Cotton effect in the CD spectra of **28** at $\lambda = 235$ nm revealed an *S*-configuration at C-2 (*39*, *40*). The low amounts of **28** available excluded determination of the stereochemistry at C-1.

Lariciresinol 4-O-*β*-D-Glucoside 29. In the carbon spectrum of **29** two aromatic systems and eight additional nonolefinic signals, together with signals for a glucose moiety, were apparent. The molecular mass determined by ESIMS was 522 amu. Comparison with published data allowed the identification of lariciresinol 4-*O*-*β*-D-glucoside **29** (*17*, *41*). Because **29** was present in the Riesling wine in only trace amounts, it was not possible to isolate enough material for clarification of the absolute stereochemistry.

Isolariciresinol 30 and Isolariciresinol Glucoconjugates 30a−c. NMR data for lignan **30** were in agreement with data published for isolariciresinol (*38*, *42*). However, the CD spectrum was inverse to that published by Lundgren et al. for (−)-isolariciresinol (*36*). Hence, lignan **30** is the (+)-isomer with 6*R*,7*S*,8*S*-

stereochemistry. In addition to the known Riesling wine constituent **30a** (*34*), two additional glucoconjugates of isolariciresinol **30** could be isolated. On the basis of NMR and CD data, the aglycon moiety of **30b** was established to be the (+)-isomer of isolariciresinol **30**. With regard to the glycosidic linkage, a downfield shift of 8 ppm for C6a in the carbon spectrum of glucoconjugate **30b,** compared to that of the nonglycosylated **30**, indicated glycosylation via the hydroxymethylene group in position C6a. Also in the case of **30c**, NMR data revealed the structure of an isolariciresinol *β*-D-glucopyranoside. The downfield shift for C6a revealed glycosylation via this position. Because of slight downfield shifts in the resonances for the protons H1, H6, H7, and H8, and a highfield shift for H6′ compared to that reported in published data (*43*), the presence of (+)-isolariciresinol or its enantiomer is unlikely. Determination of the stereochemistry of glucoconjugate **30c** failed because of the low amount of isolated material and the lack of reference data for this specific stereoisomer.

(−)-Secoisolariciresinol 31 and Secoisolariciresinol *β*-D-Glucoside 31a. The structures of the two remaining lignans were elaborated as 2*R*,3*R*-2,3-bis-(4-hydroxy-3-methoxy-benzyl)-butan-1,4-diol (secoisolariciresinol, **31**) and its 1-*O*-*β*-D-glucopyranoside **31a**. Spectral data were in good agreement with those published by Achenbach et al. *(41)* for lignan **31** previously identified in *Carissa edulis*. The negative sign of optical rotation indicated the presence of the 2*R*,3*R*-isomer. Glucoconjugate **31a** was first detected by Popoff and Theander (*38*) in conifer needles.

Antioxidant Activity of Isolated Cinnamates and Benzoates. Initially antioxidant activity was principally associated with flavonoids and stilbenes. Evidence is now increasing that hydroxycinnamates and their conjugates are similarly active. As cinnamates are significant components of the human diet they may provide beneficial health effects (*44*−*46*). Potential health effects of ferulic and caffeic acids have been demonstrated in many animal models and in vitro assays (*45*). Natella et al. (*47*) investigated the relationship between the structures of benzoic and cinnamic acid derivatives and their antioxidant activity. The authors compared the antioxidant capacity of four derivatives of benzoic acid and their homologous derivatives of cinnamic acid in their quenching activity toward peroxyl radical and in modulating the in vitro resistance of human low-density lipoprotein (LDL) to oxidative modification. It was found that the antioxidant efficiency of monophenols is strongly enhanced by the introduction of a second hydroxy group and is increased by one or two methoxy substitutions in the ortho position. It was also demonstrated that substitution of the carboxylic group of the benzoic acid derivative by the propenoic side chain leading to the homologous cinnamic acid derivative enhances the antioxidant capacity of the aromatic ring considerably. Our results for the radical scavenging capacity of cinnamic and benzoic acid derivatives obtained in the Trolox equivalent antioxidant capacity test (TEAC test; for details cf. *3, 50*) were comparable to the findings of Natella et al. (*47*). As shown in Table 1 the radical scavenging capacity of hydroxycinnamic acid derivatives does not differ significantly from that of other phenolic antioxidants. The TEAC values for hydroxycinnamic acid derivatives were found to be in a range of 0.5 to 1.7 Trolox equivalents. The respective values for the flavonoids quercetin and catechin are 3.8 and 2.7. Surprisingly, in the TEAC test the cinnamic acid derivative with two hydroxy groups, caffeic acid (**3a**), was less active than the monosubstituted *p*-coumaric acid (**1a**). In contrast, the antioxidant capacity of the esters of caffeic acid exceeded the activity of *p*-coumaroyl esters. Benzoic acids were somewhat less active in comparison with the homologous derivatives of cinnamic acid, but they provide antioxidant activity as well. Esterification of the carboxylic group of hydroxycinnamic acids slightly decreased the antioxidant activity. Chen and Ho (*44*) obtained similar results when they studied the antioxidant activities of caffeic acid and related hydroxycinnamic acid derivatives in different testing systems.

Table 1. Antioxidant Activity (expressed as Trolox Equivalents) of Isolated Cinnamic and Benzoic Acid Derivatives

compound	Trolox equivalents (mmol Trolox/mmol)
(*E*)-*p*-coumaric acid **1a**	1.4
(*E*)-*p*-coumaroyl tartaric acid **1b**	0.5
(*E*)-*p*-coumaroyl glucose ester **1c**	0.7
(*Z*)-*p*-coumaroyl tartaric acid **2b**	0.8
(*E*)-caffeic acid **3a**	1.1
(*E*)-caffeoyl tartaric acid **3b**	1.1
(*E*)-ethyl caffeate **3c**	1.5
(*E*)-ferulic acid **4a**	1.7
(*E*)-feruloyl tartaric acid **4b**	1.2
(*E*)-feruloyl glucose ester **4c**	1.3
p-hydroxyphenyl propionic acid **5**	0.7
protocatechuic acid **8a**	1.0
protocatechuic acid methyl ester **8b**	0.8
gentisic acid **9**	1.0
syringic acid **10**	1.2
p-hydroxyphenyl acetic acid **17**	0.2

Antioxidant Activity of Isolated Flavonoids and Lignans. Flavonoids have been identified as antioxidant principles of plant-derived foodstuffs in a very large number of investigations. Frankel et al. (*49*) reported that the inhibition of low-density lipoprotein oxidation by various Californian wines correlated with their contents of gallic acid, catechin, myricetin, quercetin, caffeic acid, rutin, epicatechin, cyanidin, and malvidin-3-*O*-glucoside. The activity of flavonoids in inhibiting lipid peroxidation is due to several kinds of action. Flavonoids are able to scavenge superoxide anions and hydroxyl radicals. They may donate hydrogen atoms to peroxy radicals, forming a flavonoid radical, and this flavonoid radical in turn is able to react with free radicals thereby terminating the radical chain reaction (*48*). The relationship between the structure of flavonoids and their antioxidant potential has been intensively studied by Rice-Evans and co-workers (*50*). These authors reported the chemistry of flavonoids to be predictive for their free radical scavenging activity. Three criteria were found to enhance radical scavenging effectiveness: (i) an *ortho*-dihydroxy structure in the B ring; (ii) a 2,3-double bond in conjugation with a 4-oxo function in the C ring, and (iii) hydroxy groups in positions 3 and 5 in the A ring (*50*). Our results concerning the antioxidant capacity of white wine flavonoids agree with these findings. A summary of the antioxidant testing is given in Table 2. Quercetin, with the 2,3-double bond in conjugation to the 4-oxo group, exhibited highest TEAC values. Blockage of the 3-OH group by glycosylation decreased the antioxidant activity as shown for, e.g., quercetin-3-*O*-glucuronide **25**. Hydrogenation of the 2,3-double bond leading to dihy-

Table 2. Antioxidant Activity (expressed as Trolox Equivalents) of Isolated Flavonoids and Reference Substances (Quercetin, Rutin, Quercitrin, and Kaempferol Rutinoside)

compound	Trolox equivalents (mmol Trolox/mmol)
dihydrokaempferol 3-*O*-rhamnoside **18a**	1.0
dihydrokaempferol 3-*O*-glucoside **18b**	1.0
dihydroquercetin **19**	1.7
dihydroquercetin 3-*O*-rhamnoside **19a**	1.8
dihydroquercetin 3-*O*-glucoside **19b**	1.3
catechin **20**	2.7
epicatechin **21**	3.0
procyanidin B1 **22**	4.3
procyanidin B3 **23**	4.8
quercetin 3-*O*-glucuronide **25**	2.6
quercetin	3.8
quercetin 3-*O*-rutinoside (rutin)	2.9
quercetin 3-*O*-rhamnoside (quercitrin)	2.5
kaempferol 3-*O*-rutinoside	1.3

droquercetin derivatives also reduced the antioxidant effectiveness. Kaempferol derivatives, with only one hydroxy group in ring B, were less active in scavenging the ABTS radical compared the corresponding quercetin derivatives. On a molar basis, the dimeric procyanidins **22** and **23** were the most active antioxidants among the flavonoids tested, revealing about twice the activity of monomeric catechin or epicatechin, respectively. This is easily explained by twice the number of phenolic hydroxy groups in the case of procyanidins in comparison with the monomers. TEAC values determined for catechin **20** and epicatechin **21** are in the same range as that obtained for glycosylated quercetin.

Because of the low amount of material available, only three representatives of the isolated neolignans and lignans were tested. TEAC values (mmol TEAC/mmol) for neolignan **26**, isolariciresinol **30**, and isolariciresinol 4′-β-D-glucoside **30a** were determined to be 2.5, 2.5, and 1.9, respectively.

Evidence is increasing that the antioxidant capacity of white wine is much more associated with the presence of hydroxy cinnamates than with any other class of wine constituents. Cinnamic acid derivatives represent the major group of white wine phenols and possess remarkable antioxidant activity. Final conclusions about the contribution of the different classes of white wine constituents to the overall antioxidant activity must await the outcome of an ongoing monitoring of German Riesling wines.

LITERATURE CITED

(1) Watkins, T. R., Ed. *Wine − Nutritional and Therapeutic Benefits*. ACS Symposium Series 661; American Chemical Society: Washington, DC, 1997.

(2) Vinson, J. A.; Hontz, B. A. Phenol antioxidant index: comparative antioxidant effectiveness of red and white wines. *J. Agric. Food Chem*. **1995**, *43*, 401−403.

(3) Baderschneider, B.; Winterhalter, P. Isolation and characterization of novel stilbene derivatives from Riesling wine. *J. Agric. Food Chem*. **2000**, *48*, 2681−2686.

(4) Macheix, J.-J.; Sapid, J.-C.; Fleuriet, A. Phenolic compounds and polyphenoloxidase in relation to browning in grapes and wines. *Crit. Rev. Food Sci. Nutr*. **1991**, *30*, 441−486.

(5) Baderschneider, B. Isolierung und Strukturaufklärung antioxidativ wirksamer Verbindungen aus Weisswein. Ph.D. Thesis, Universität Braunschweig, Germany, 2000.

(6) Drawert, F.; Lessing, V.; Leupold, G. About the separation and gas-chromatographic determination of low molecular weight phenolic compounds. *Chem. Mikrobiol. Technol. Lebensm*. **1977**, *5*, 65−70.

(7) Hirsch, D.; Maier, H. G. Capillary electrophoretic determination of sugars as indicator for adulteration in coffee extracts. *Lebensmittelchemie* **1999**, *6*, 149−150.

(8) Somers, T. C.; Vérette, E.; Pocock, K. F. Hydroxycinnamate esters of *Vitis vinifera*: changes during white vinification, and effects of exogenous enzymic hydrolysis. *J. Sci. Food Agric*. **1987**, *40*, 67−78.

(9) Baranowski, J. D.; Nagel, C. W. Isolation and identification of 10 hydroxycinnamic acid derivatives in white Riesling wine. *Am. J. Enol. Vitic*. **1981**, *32*, 5−13.

(10) Drawert, F.; Schreier, P.; Scherer, W. Gas-chromatographic-mass-spectrometric investigation of volatile wine constituents. III. Acids of the wine flavor. *Z. Lebensm. Unters. Forsch*. **1974**, *155*, 342−347.

(11) Baltenweck-Guyot, R.; Trendel, J.-M., Albrecht, P.; Schaeffer, A. Glycosides and phenylpropanoids in *Vitis vinifera* cv. Gewurztraminer wine. *J. Agric. Food Chem*. **2000**, *48*, 6178−6182.

(12) Singleton, V. L.; Trousdale, E. White wine phenolics: Varietal and processing differences as shown by HPLC. *Am. J. Enol. Vitic*. **1983**, *34*, 27−34.

(13) Güntert, M.; Rapp, A.; Takeoka, G. R.; Jennings, W. HRGC and HRGC-MS applied to wine constituents of lower volatility. *Z. Lebensm. Unters. Forsch*. **1986**, *182*, 200−204.

(14) Reschke, A.; Herrmann, K. Occurrence of 1-*O*-hydroxycinnamoyl-β-D-glucoses in fruits. 15. Phenolic constituents of fruits. *Z. Lebensm. Unters. Forsch*. **1981**, *173*, 458−463.

(15) Ong, B. Y.; Nagel, C. W. High-pressure liquid chromatographic analysis of hydroxycinnamic acid-tartaric acid esters and their glucose esters in *Vitis vinifera*. *J. Chromatogr*. **1978**, *157*, 345−355.

(16) Agrawal, P. K. NMR Spectroscopy in the structural elucidation of oligosaccharides and glycosides. *Phytochemistry* **1992**, *31*, 3307−3330.

(17) Jerz, G. Phytochemical analysis of *Ongokea gore* (Olacaceae). Ph.D. Thesis, University of Erlangen, Germany, 1999.

(18) Crestini, C.; D'Auria, M. Singlet oxygen in the photodegradation of lignin models. *Tetrahedron* **1997**, *53*, 7877−7888.

(19) Goméz-Cordovés, C.; Bartolomé, B.; Jimeno, M. L. Identification of 2,3-dihydroxy-1-guaiacylpropan-1-one in brandies. *J. Agric. Food Chem*. **1997**, *45*, 873−876.

(20) Soleas, G. J.; Diamandis, E. P.; Goldberg, D. M. Wine as a biological fluid: History, production, and role in disease prevention. *J. Clin. Lab. Anal*. **1997**, *11*, 287−313.

(21) Rice-Evans, C. A.; Miller, N. J.; Paganga, G. Structure − antioxidant activity relationships of flavonoids and phenolic acids. *Free Radical Biol. Med*. **1996**, *20*, 933−956.

(22) Verma, A. K.; Johnson, J. A.; Gould, M. N.; Tanner, M. A. Inhibition of 7,12-dimethylbenz[*a*]anthracene and *N*-nitrosomethylurea induced rat mammary cancer by dietary flavonol quercetin. *Cancer Res*. **1988**, *48*, 5754−5788.

(23) Wei, H.; Tye, L.; Bresnick, E.; Brit, D. F. Inhibitory effect of apigenin, a plant flavonoid, on epidermal ornithine decarboxylase and skin tumor promotion in mice. *Cancer Res*. **1990**, *50*, 499−502.

(24) Hertog, M. G. L.; Feskens, E. J. M.; Hollman, P. C. H.; Katan, M. B.; Kromhout, D. Dietary antioxidant flavonoids and risk of coronary heart disease: the Zutphen elderly study. *Lancet* **1993**, *342*, 1007−1011.

(25) Waterhouse, A. L. Wine and heart disease. *Chem. Ind. (London)* **1995**, 338−341.

(26) Lea, A. G. H.; Bridle, P.; Timberlake, C. F.; Singleton, V. L. The procyanidins of white grapes and wines. *Am. J. Enol. Vitic*. **1979**, *30*, 289−300.

(27) Lee, C. Y.; Jaworski, A. Phenolic compounds in white grapes grown in New York. *Am. J. Enol. Vitic.* **1987**, *38*, 277−281.
(28) Singleton, V. L.; Trousdale, E. K. White wine phenolics: Varietal and processing differences as shown by HPLC. *Am. J. Enol. Vitic.* **1983**, *34*, 27−34.
(29) Cheynier, V.; Rigaud, J. Identification et dosage de flavonols du raisin. *Proceedings of the 9th International Conference of Group Polyphenols*, Montpellier, France, November 1986; Royal Society of Chemistry: London, 1986.
(30) 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.
(31) Betés-Saura, C.; Andrés-Lacueva, C.; Lamuela-Raventós, R. M. Phenolics in white free run juices and wines from Penedès by high-performance liquid chromatography: Changes during vinification. *J. Agric. Food Chem.* **1996**, *44*, 3040−3046.
(32) Dübeler, A.; Voltmer, G.; Gora, V.; Lunderstädt, J.; Zeeck, A. Phenolics from *Fagus sylvatica* and their role in defence against *Cryptococcus fagisuga*. *Phytochemistry* **1997**, *45*, 51−57.
(33) Gaffield, W.; Waiss, A. C., Jr. Structural relationships and interconversions of isomeric astilbins. *J. Org. Chem.* **1995**, *40*, 1057−1061.
(34) Marinos, V. A.; Tate, M. E.; Williams, P. J. Lignan and phenylpropanoid glycerol glucosides in wine. *Phytochemistry* **1992**, *31*, 4307−4312.
(35) Marinos, V. A. Application of fast atom bombardment mass spectrometry to the examination of glycoconjugates in grape juice and wine. Thesis, University of Adelaide, Australia, 1992.
(36) Lundgren, L. N.; Popoff, T.; Theander, O. Dilignol glycosides from needles of *Picea abies*. *Phytochemistry* **1981**, *20*, 1967−1970.
(37) Fang, J.-M.; Lee, C.-K.; Cheng, Y.-S. Lignans from leaves *of Juniperus chinensis*. *Phytochemistry* **1992**, *31*, 3659−3661.
(38) Popoff, T.; Theander, O. The constituents of conifer needles. VI. Phenolic glycosides from *Pinus sylvestris*. *Acta Chem. Scand. Ser. B* **1977**, *31*, 329−337.
(39) Matsuda, N.; Kikuchi, M. Studies on the constituents of *Lonicera* species. X. Neolignan glycosides from the leaves of *Lonicera gracilipes* var. *glandulosa* Maxim. *Chem. Pharm. Bull.* **1996**, *44*, 1676−1679.
(40) Della Greca, M.; Molinaro, A.; Monaco, P.; Previtera, L. Neolignans from *Arum italicum*. *Phytochemistry* **1994**, *35*, 777−779.
(41) Achenbach, H.; Waibel, R.; Addae-Mensah, I. Lignans and other constituents from *Carissa edulis*. *Phytochemistry* **1983**, *22*, 749−753.
(42) Okuyama, E.; Suzumura, K.; Yamazaki, M. Pharmacologically active components of Todopon Puok (*Fagraea racemosa*), a medicinal plant from Borneo. *Chem. Pharm. Bull.* **1995**, *43*, 2200−2204.
(43) Achenbach, H.; Löwel, M.; Waibel, R.; Gupta, M.; Solis, P. New lignans from *Stemmadenia minima*. *Planta Med.* **1992**, *58*, 270−272.
(44) Chen, J. H.; Ho, C.-T. Antioxidant activities of caffeic acid and its related hydroxycinnamic acid compounds. *J. Agric. Food Chem.* **1997**, *45*, 2374−2378.
(45) Clifford, M. N. Chlorogenic acids and other cinnamates − nature, occurrence and dietary burden. *J. Sci. Food Agric.* **1999**, *79*, 362−372.
(46) Kroon, P. A.; Williamson, G. Hydroxycinnamates in plants and wood: Current and future perspectives. *J. Sci. Food Agric.* **1999**, *79*, 355−361.
(47) Natella, F.; Nardini, M.; Di Felice, M.; Scaccini, C. Benzoic and cinnamic acid derivatives as antioxidants: Structure−activity relation. *J. Agric. Food Chem.* **1999**, *47*, 1453−1459.
(48) Cook, N. C.; Samman, S. Flavonoids − chemistry, metabolism, cardioprotective effects, and dietary sources. *Nutr. Biochem.* **1996**, *7*, 66−76.
(49) Frankel, E. N.; Waterhouse, A. L.; Teissedre, P. L. Principal phenolic phytochemicals in selected Californian wines and their antioxidant activity in inhibiting oxidation of human low-density lipoproteins. *J. Agric. Food Chem.* **1995**, *43*, 890−894.
(50) Rice-Evans, C. A.; Miller, N. J.; Paganga, G. Structure − antioxidant activity relationships of flavonoids and phenolic acids. *Free Radical Biol. Med.* **1996**, *20*, 933−956.
(51) Biau, S.; Dumon, M. C.; Vercauteren, J.; Glories, Y. Étude des constituants des vins blancs de Bordeaux. In *Polyphénols Communications 96; XVIIIth International Conference on Polyphenols, Bordeaux (France), Groupe Polyphenols*; Vercauteren, J., Cheze, C., Dumon, M. C., Weber. J. F., Eds.; 1996; pp 37−38.

Received for review March 22, 2001. Revised manuscript received April 11, 2001. Accepted April 12, 2001. DFG (Bonn) is thanked for funding the research (Wi 901/5-1 and 5-2).

JF010396D