Identification of a Precursor of β-Methyl-γ-octalactone in the Wood of Sessile Oak (*Quercus petraea* (Matt.) Liebl.)

Eric Masson, Raymond Baumes, Christine Le Guernevé, and Jean-Louis Puech*

Unité de Recherches Biopolymères et Arômes, Institut des Produits de la Vigne, Institut National de la Recherche Agronomique, 2 place Viala, 34060 Montpellier Cedex, France

The 6′-O-gallate derivative of (3*S*,4*S*)-4-β-D-glucopyranosyloxy-3-methyloctanoic acid was isolated for the first time from the wood of Sessile oak (*Quercus petraea* (Matt.) Liebl.) and shown to be a precursor of *cis*-β-methyl-γ-octalactone. The structure of this precursor was determined by HRFAB-MS, NMR, LCMS, and chiral analysis of the liberated (3*S*,4*S*)-β-methyl-γ-octalactone on a chiral fused silica capillary column. Optical rotation was shown to be identical to that of the same compound previously isolated from the wood of *Platycarya strobilacea* Sieb. et Zucc. by Tanaka and Kouno in 1996. Moreover, the 6′-O-gallate derivative of a *threo*-4-β-D-glucopyranosyloxy-3-methyloctanoic acid was tentatively identified as a minor precursor of *trans*-β-methyl-γ-octalactone in the same wood of Sessile oak.

Keywords: *Sessile oak; Quercus petraea; β-methyl-γ-octalactone; whiskey lactone; precursor*

INTRODUCTION

First identified in whiskey by Suomalainen and Nykänen (1970) and subsequently in oak wood by Masuda and Nishimura (1971), β-methyl-γ-octalactone (also known as whiskey or oak lactone) is one of the major volatile compounds of oak wood extracted by oak-aged wines and brandies (Guymon and Crowell, 1972; Kepner et al., 1972; Otsuka et al., 1974). Of the four stereoisomers of β-methyl-γ-octalactone, only the 3*S*,4*S* (cis) and 3*S*,4*R* (trans) forms are present in oak wood (Masuda and Nishimura, 1981; Günther and Mosandl, 1987; Guichard et al., 1995; Masson et al., 1995) (Figure 1). These two stereoisomers, characterized by a distinctive odor of coconut, celery, and fresh wood, have different flavor perception thresholds. Detection thresholds determined by GC-sniffing of a racemic mixture were 1 μg/L for the cis form and 20 μg/L for the trans form (Abbott et al., 1995).

The increase in the level of β-methyl-γ-octalactone in oak wood extracts heated in a strong acidic medium indicates the existence of a precursor in oak wood (Otsuka et al., 1974), the structure of which was reported to be 3-methyl-4-(3′,4′-dihydroxy-5′-methoxybenzoyloxy)octanoic acid (Otsuka et al., 1980 a,b) (Figure 2). More recently, two other precursors, (3*S*,4*S*)-4-β-D-glucopyranosyloxy-3-methyloctanoic acid and its derivative 6′-*O*-gallate (Figure 3), have been isolated and identified by Tanaka and Kouno (1996) from the wood of *Platycarya strobilacea* Sieb. et Zucc. (Juglandaceae).

The scientific literature has systematically cited the compound isolated by Otsuka et al. (1980a,b) as the precursor of β-methyl-γ-octalactone in oak wood. However, neither the structure nor the presence of this compound in oak wood have been confirmed.

In this study we show that the 6′-*O*-gallate derivative of (3*S*,4*S*)-4-β-D-glucopyranosyloxy-3-methyloctanoic acid, isolated by Tanaka and Kouno (1996) from the wood of *Platycarya strobilacea* Sieb. et Zucc., is present in the wood of Sessile oak (*Quercus petraea* (Matt.) Liebl., Fagaceae), a European white oak species used for cooperage, and that it is indeed a precursor of β-methyl-γ-octalactone.

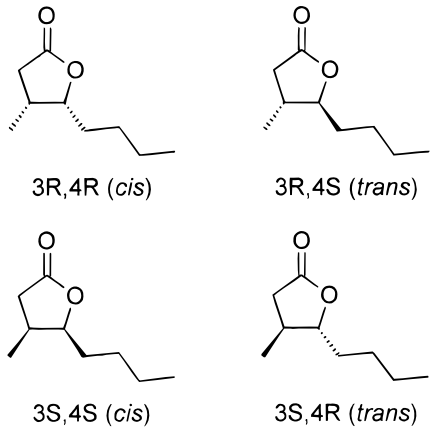

Figure 1. Structure of the cis and trans isomers of β-methyl-γ-octalactone.

MATERIALS AND METHODS

Plant Material. The wood used for the extraction of the precursor was from a 196 year old Sessile oak (*Quercus petraea* (Matt.) Liebl., Fagaceae) from the forest of Bercé (Sarthe, France). The species was determined from the morphological characteristics of twigs, leaves, and fruits collected from the tree (Dupouey and Badeau, 1993).

Extraction of the Precursor. A sample of fresh wood was ground into a powder of particles less than 0.5 mm. In a stoppered Erlenmeyer flask, 500 mL of methanol was added to 50 g of wood powder. Extraction occurred under agitation for 15 h. After filtration using a noncharcoal filter, the extract was concentrated down to one-tenth of its original volume under vacuum at 36 °C.

* To whom correspondence should be sent. Telephone: (33) 04 99 61 24 83. Fax: (33) 04 99 61 26 83. E-mail: puechjl@ensam.inra.fr.

10.1021/jf0002950 CCC: $19.00
Published on Web 08/17/2000

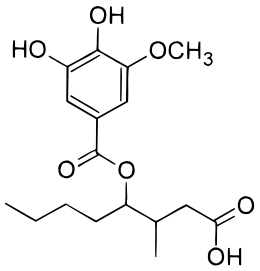

Figure 2. Structure of the precursor of β-methyl-γ-octalactone proposed by Otsuka et al. (1980a,b).

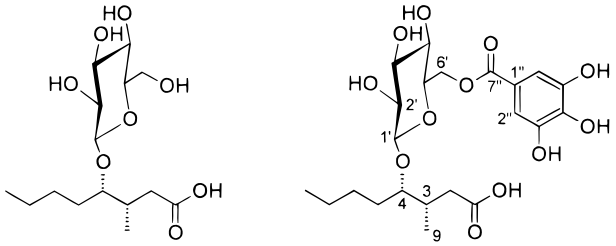

Figure 3. Structure of precursors of β-methyl-γ-octalactone proposed by Tanaka et Kouno (1996).

Semipreparative High Performance Liquid Chromatography (Semipreparative HPLC). The chromatograph consisted of two Gilson pumps (models 305 and 306), a Gilson 811B mixing chamber, a Gilson 806 manometric module, a Rheodyne 7125 (500 μL injection ring) manual injector, a UV−visible JASCO 875-UV detector, and Hewlett-Packard 3396A data collection and integration software. Separation was carried out on a Microsorb "Fast PCLC" C18 type column (50 mm × 21.4 mm, 3 μm) (Rainin). The elution gradient was [solvent A, H_2O/HCO_2H (98:2); solvent B, CH_3OH/H_2O (50:50)] 60−80% of solvent B during 5 min, 80−100% in 10 min, and then 100% for 5 min. The flow was 5 mL/min and detection occurred at 272 nm.

Liquid Chromatography Coupled with Mass Spectrometry (LC-MS). The liquid chromatograph consisted of a Applied Biosystems 140B pump and 785A detector. The Lichrospher RP 18 column (250 mm × 2 mm; 5 μm) (Merck, Darmstadt, Germany) was maintained at 32 °C. The elution gradient was [solvent A, H_2O/HCO_2H (98:2); solvent B, CH_3-$CN/H_2O/HCO_2H$ (80:18:2)] 0−20% of solvent B in 10 min, 20% maintained for 10 min, and then 20−100% in 60 min. A flow of 280 μL/min and an injection volume of 20 μL were used with detection at 280 nm.

An API I-Plus (Sciex, Thornhill, Ontario, Canada) mass spectrometer was used. Ionization was performed by using an electrospray source, and the ions were collected by a simple quadrupole covering a mass range of 0−1000 amu (atomic mass units). Detection was carried out in steps of 0.25 amu with a dwell time of 0.75 ms. The negative ionization mode was used with an opening potential of −60 or −150 V (fragmentation).

Gas Chromatography Coupled with Mass Spectrometry (GC-MS). The Hewlett-Packard 6890 series chromatograph was equipped with a DB WAX capillary column (30 m × 0.25 mm, 0.5 μm, J&W Scientific Inc., Folsom, CA) and an "on column" injector (30 to 250 °C at 180 °C/min). The helium gas vector flow was maintained at 1 mL/min throughout the analysis. Oven temperature followed the program: 30 to 100 °C at a ramp rate of 70 °C/min, constant for 2 min; 100 to 200 °C at a rate of 3 °C/min; 200 to 245 °C at 45 °C/min and then constant for 30 min.

A Hewlett-Packard 5973 mass spectrometer was used. Ionization was achieved under electron impact mode (ionization energy of 70 eV); the source, transmission line, and quadrupole temperatures were 250, 230, and 106 °C, respectively. Detection was carried out in scan mode (1.32 scan per second) covering a mass range (*m*/*z*) of 25−600 amu. The identification of the cis and trans isomers of β-methyl-γ-octalactone was confirmed by injection of pure standards.

Chiral Separation of the Stereoisomers of β-Methyl-γ-octalactone by Chiral Gas Chromatography. The stereodifferentiation of the lactone was realized with a Hewlett-Packard 6890 series gas chromatograph equipped with a FS-Lipodex E capillary column (25 m × 0.25 mm, Macherey & Nagel). Operating parameters were as follows: splitless injection (200 °C), FID detection (240 °C), hydrogen as carrier gas (1.9 mL/min), and oven temperature program: 110 °C for 20 min and then 110 to 140 °C at a rate of 2 °C/min.

The order of elution of the four stereoisomers of a commercial β-methyl-γ-octalactone from Aldrich on the Lipodex E column, (3*S*,4*R*), (3*R*,4*S*), (3*R*,4*R*), (3*S*,4*S*), was determined as previously described (Guichard et al., 1995).

NMR Spectroscopy. The structural analysis of the precursor was carried out from NMR spectra 1D ^1H, 2D (^1H) COSY, 2D (^1H,^{13}C) HSQC, and HMBC obtained with a Varian UNITY INOVA 500 MHz spectrometer. The frequencies used were 500 MHz for the proton spectra and 125.75 MHz for ^{13}C. Measurements were made at 25 °C in the solvent (acetone-d_6) used by Tanaka and Kouno (1996). The signals at 2.05 ppm (^1H) and 29.5 ppm (^{13}C) of the solvent were used as reference signals.

Elemental Composition. Elemental composition was determined by HR−FABMS in negative mode. Data were measured with a JEOL MStation JMS−SX 102 A instrument with glycerin as the matrix: *m*/*z* 487.1912 $[M - H]^-$ error mmu = +9.6 for $C_{22}H_{31}O_{12}$.

Specific Rotation. The specific rotation (29 °C, ray D) of the precursor was determined by using a polarimeter Perkin-Elmer 241 from a methanol solution of the precursor at a concentration of 5 g/L: $[\alpha]^{29}_D = -14°$ (*c* 0.005, MeOH). This value is close to that determined by Tanaka and Kouno (1996): $[\alpha]^{29}_D = -13.7°$ (*c* 0.6, MeOH).

Lactonization of the Precursor. To 4 mL of an aqueous tannase solution (4 g/L) was added 1 mL of an aqueous solution of the precursor (0.2 g/L). After 1 h of agitation, 10 mL of 24 *N* sulfuric acid was added and the mixture once again agitated for 2 h. Two extractions were carried out using 20 and 10 mL of a pentane/dichloromethane (2:1) solution (Carlo-Erba, RS quality). The organic phases were combined and dried with anhydrous sodium sulfate. The extract was subsequently concentrated down to about 1 mL on a Vigreux type column (water bath temperature of 36 °C).

RESULTS AND DISCUSSION

Detection of the Proposed Precursor(s) of β-Methyl-γ-octalactone. The methanol extract, obtained from fresh wood powder rich in β-methyl-γ-octalactone (Masson, 1999), was analyzed by HPLC-MS in the negative-ion mode to look for several molecular masses of interest. The precursor proposed by Otsuka et al. (1980a,b) (Figure 2) has a molecular mass of 340. The use of methanol as the extraction solvent may produce an ester of mass 354 as reported by Otsuka et al. (1980a,b). The compounds identified by Tanaka and Kouno (1996) (Figure 3) have molecular masses of 336 and 488 for the gallate form, with their methylated esters having masses of 350 and 502.

Of these six masses only a single peak was found, corresponding to a mass of 488. The fragmentation of this compound, named X, in negative ionization mode (opening potential, 150 V) gives two signals at *m*/*z* of 169 and 335. The first signal corresponds to gallic acid and the second to a nongallate compound isolated by Tanaka and Kouno (1996). Compound X would appear therefore to be the 6′-O-gallate derivative of 4-β-D-glucopyranosyloxy-3-methyloctanoic acid.

Purification of Compound X. Compound X was purified by the use of semipreparative HPLC. After lyophilization, about 5 mg of the product (a white

powder) was obtained. Analysis by HPLC-MS of the purified product found two peaks, each corresponding to the same molecular mass of 488 with one peak much larger than the other. The fragmentation of these two compounds in negative ionization mode gives two signals at *m*/*z* of 169 and 335 as described previously. Compound X is therefore present in the wood of Sessile oak in the form of two diastereoisomers.

The supposed structure of compound X, 6′-O-gallate derivative of 4-*β*-D-glucopyranosyloxy-3-methyloctanoic acid, was consistent with its molecular formula, $C_{22}H_{32}O_{12}$, determined by HRFAB-MS measurements (*m*/*z* 487.1912 $[M - H]^-$ error mmu = +9.6 for $C_{22}H_{31}O_{12}$).

NMR Structural Analysis. An NMR study was carried out to confirm whether the isolated product was the 6′-O-gallate derivative of 4-*β*-D-glucopyranosyloxy-3-methyloctanoic acid. The analysis of 1D ^{1}H and HSQC (^{1}H,^{13}C) spectra identified two CH_3 groups, five CH_2 groups, seven CH groups, and a correlation corresponding to two protons (H-2″ and H-6″) of the galloyl group. Two spin systems were identified from the COSY (^{1}H) spectrum. The first system, which notably involved two methyl groups, corresponded to a carbon chain. From the CH_3 signal at 0.74 ppm, which is found as a triplet in the 1D spectrum, it is possible to assign each proton from successive correlations between protons. The second system corresponds to a sugar. The correlations were determined from the characteristic protons H-6′a and H-6′b (δ ^{1}H = 4.52 and 4.36 ppm). An initial correlation was observed between these protons and the H-5′ proton, and from this the other protons were determined from successive correlations. The anomeric configuration was established from the coupling constant $J_{1',2'}$ = 8 Hz, which was characteristic of the anomer *β* and is confirmed by the high value (δ ^{13}C = 103,7 ppm) of the chemical shift of the carbon anomer C-1′ (Strack and Wray, 1989). The HMBC (^{1}H,^{13}C) spectrum allows the confirmation of the bonds linking the glucose, the carbon chain, and the galloyl group. The presence of correlations between the protons H-6′ and the carbon C-7″ indicates a bond between *β*-glucopyranose and the galloyl group at the location of the C-6′ carbon. The correlations [H-4/C-1′] and [H-1′/C-4] confirm the bond between the sugar and the carbon chain at the position of carbons C-4 and C-1′. Finally, the presence of a methyl group at carbon C-3 and the bond between carbon C-2 and a carboxyl group are corroborated by correlations [H-9/C-3 and H-3/C-9] and [H-2/C-1], respectively.

The δ ^{1}H and δ ^{13}C chemical shifts and the proton coupling constants, determined from 1D and 2D spectra, are shown in Table 1. The values are very close to those of Tanaka and Kouno (1996) (Table 2). Overall, these results allow us to confirm that the compound isolated from the wood of Sessile oak is one of the stereoisomers of the 6′-O-gallate derivative of 4-*β*-D-glucopyranosyloxy-3-methyloctanoic acid. The signals of the other minor stereoisomer were too weak for such analysis.

Lactonization. The formation of *β*-methyl-*γ*-octalactone from the 6′-O-gallate derivative of 4-*β*-D-glucopyranosyloxy-3-methyloctanoic acid was verified by enzymatic hydrolysis of the galloyl group followed by hydrolysis and cyclization in a concentrated acid medium.

The hydrolysis was carried out by the addition of tannase to an aqueous solution of the precursor. The solution was subsequently studied by HPLC-MS in negative-ion mode. Four peaks were found at *m*/*z* of 487 (two peaks), 335, and 169. Enzymatic hydrolysis would appear to result therefore in the formation of 4-*β*-D-glucopyranosyloxy-3-methyloctanoic acid (molecular mass of 336) and gallic acid (molecular mass of 170) in addition to the original compounds.

Lactonization is produced by the addition of 24 *N* sulfuric acid to the aqueous tannase and precursor solution. Analysis by GC-MS confirms the formation of *cis*- (98%) and *trans*-*β*-methyl-*γ*-octalactone (2%) from the purified precursor. This shows that the major *β*-methyl-*γ*-octalactone precursor has primarily an *erythro* configuration, either (3*R*,4*R*), (3*S*,4*S*), or a mixture

Table 1. ^{1}H and ^{13}C Chemical Shift Data[a] for the 6′-*O*-Gallate Derivative of 4-*β*-D-Glucopyranosyloxy-3-methyloctanoic Acid at 25 °C

position	δ ^{1}H, ppm	multiplicity	δ ^{13}C, ppm
1			174.4
2	2.20, 2.62	m	36.9
3	2.21	m	33.5
4	3.61	m	82.6
5a	1.42	m	31.3
5b	1.52		
6a	1.24	m	27.9
6b	1.35		
7a	1.15	m	22.6
7b	1.22		
8	0.74	t, $J_{8,7}$ = 7 Hz	13.7
9	0.92	d, $J_{9,3}$ = 7 Hz	14.3
1′	4.39	d, $J_{1',2'}$ = 8 Hz	103.7
2′	3.22	dd, $J_{2',1'}$ = 8 Hz, $J_{2',3'}$ = 9 Hz	74.8
3′	3.44	m	77.6
4′	3.43	m	70.8
5′	3.58	m	74.2
6′a	4.36	dd, $J_{6'a,5'}$ = 6 Hz, $J_{6'a,6'b}$ = 12 Hz	64.3
6′b	4.52	dd, $J_{6'b,5'}$ = 2 Hz, $J_{6'b,6'a}$ = 12 Hz	
1″			121.1
2″/6″	7.15	s	109.3
3″/5″			145.3
4″			138.1
7″			166.3

[a] The signals at 2.05 ppm (^{1}H) and 29.5 ppm (^{13}C) of the solvent (acetone-d_6) were used as reference signals.

Table 2. ^{1}H and ^{13}C Chemical Shift Data[a] Reported by Tanaka and Kouno (1996) for the 6′-*O*-Gallate Derivative of 4-*β*-D-Glucopyranosyloxy-3-methyloctanoic Acid at 25 °C

position	δ ^{1}H, ppm	multiplicity	δ ^{13}C, ppm
1	-		175.4
2	2.21, 2.62	m	37.4
3	2.21	m	34.0
4	3.60	m	83.0
5			31.7
6	1.15−1.56	m	28.5
7			23.0
8	0.76	t, $J_{8,7}$ = 7 Hz	14.2
9	0.93	d, $J_{9,3}$ = 7 Hz	14.8
1′	4.40	d, $J_{1',2'}$ = 8 Hz	104.0
2′	3.23	dd, $J_{2',1'}$ = 8 Hz, $J_{2',3'}$ = 9 Hz	74.6
3′	3.44	m	77.7
4′	3.44	m	71.3
5′	3.60	m	74.9
6′a	4.38	dd, $J_{6'a,5'}$ = 6 Hz, $J_{6'a,6'b}$ = 12 Hz	64.6
6′b	4.53	dd, $J_{6'b,5'}$ = 2 Hz, $J_{6'b,6'a}$ = 12 Hz	
1″			121.6
2″/6″	7.16	s	109.7
3″/5″			145.9
4″			138.6
7″			166.9

[a] The signals of the solvent (acetone-d_6) were used as reference signals.

of these two enantiomers. The minor precursor peak would therefore correspond to the *threo* form.

Absolute Configuration. The absolute configuration of the precursor isolated by Tanaka and Kouno (1996) is (3*S*,4*S*). β-Glucosylation generally leads to variations in the chemical shift data, by an order of −2 to −4 ppm, for the carbons at positions β and β′ of the glucoside aglycon (carbons C-3 and C-5 in our numeration) with respect to those of the free aglycon; these variations are dependent on the α-carbon configuration (C-4 in our numeration) (Tori et al., 1977; Seo et al., 1978). As the chemical shift data acquired for carbons C-3 and C-5 are as close to the values determined by Tanaka and Kouno (1996) (difference lower than 0.5 ppm) as those for the other carbons (Tables 1 and 2), we can therefore conclude that the main compound isolated has the same (3*S*,4*S*) configuration. That was consistent with its specific rotation, $[\alpha]^{29}_{D} = -14°$ (*c* 0.005, MeOH), close to that determined by Tanaka and Kouno (1996).

The absolute configuration of the precursor is also confirmed by analysis of the β-methyl-γ-octalactone liberated from the precursor by gas chromatography on a chiral fused silica capillary column as previously reported (Guichard et al., 1995). The major liberated β-methyl-γ-octalactone showed only one peak corresponding to the (3*S*,4*S*) stereoisomer. As the reactions of hydrolysis of the precursor and of cyclization of the aglycon involved no inversion of configuration, the major precursor of β-methyl-γ-octalactone isolated from the wood of Sessile oak is characterized as (3*S*,4*S*)-4-β-D-glucopyranosyloxy-3-methyloctanoic acid.

ABBREVIATIONS USED

HPLC, high-performance liquid chromatography; LC, liquid chromatography; MS, mass spectrometry; GC, gas chromatography; HR−FABMS, high-resolution fast atom bombardment mass spectrometry; NMR: nuclear magnetic resonance; COSY, correlation spectroscopy; HSQC, heteronuclear single quantum correlation; HMBC, heteronuclear multiple bond connectivity.

LITERATURE CITED

Abbott, N.; Puech, J.-L.; Bayonove, C.; Baumes, R. Determination of the aroma threshold of the cis and trans racemic forms of β-methyl-γ-octalactone by gas chromatography-sniffing analysis. *Am. J. Enol. Vitic.* **1995**, *46*, 292−294.

Dupouey, J.-L.; Badeau, V. Morphological variability of oaks (*Quercus robur* L., *Quercus petraea* (Matt.) Liebl., *Quercus pubescens* Willd.) in northeastern France: prelimimary results. *Ann. Sci. For.* **1993**, *50*, 35s−40s.

Guichard, E.; Fournier, N.; Masson, G.; Puech, J.-L. Stereoisomers of β-methyl-γ-octalactone. I. Quantification in brandies as a function of wood origin and treatment of the barrels. *Am. J. Enol. Vitic.* **1995**, *46*, 419−423.

Günther, C.; Mosandl, A. Stereoisomere Aromastoffe. XV. Chirospezifische Analyse natürlicher aromastoffe: 3-Methyl-4-octanolid − "Quercus-Whiskylacton". *Z. Lebensm.-Unters.-Forsch.* **1987**, *185*, 1−4.

Guymon, J. F.; Crowell, E. A. GC-separated brandy components derived from French and American oaks. *Am. J. Enol. Vitic.* **1972**, *23*, 114−120.

Kepner, R. E.; Webb, D. A.; Muller, C. J. Identification of 4-hydroxy-3-methyloctanoic acid gamma-lactone (5-butyl-4-methyldihydro-2-(3H)-furanone) as a volatile component of oak-wood-aged wines of *Vitis vinifera* var. "cabernet sauvignon". *Am. J. Enol. Vitic.* **1972**, *23*, 103−105.

Masson, E. Incidence du mode de séchage sur les caractéristiques physico-chimiques du bois de chêne de tonnellerie (*Quercus petraea* (Matt.) Liebl.). Thèse de Doctorat, Université H. Poincaré − Nancy 1, 1999, 277 pp.

Masson, G.; Guichard, E.; Fournier, N.; Puech, J.-L. Stereoisomers of β-methyl-γ-octalactone. II. Contents in the wood of French (*Quercus robur* and *Quercus petraea*) and American (*Quercus alba*) oaks. *Am. J. Enol. Vitic.* **1995**, *46*, 424−428.

Masuda, M.; Nishimura, K. Branched nonalactones from some *Quercus* species. *Phytochemistry* **1971**, *10*, 1401−1402.

Masuda, M.; Nishimura, K. Absolute configurations of *Quercus* lactones, (3*S*,4*R*)- and (3*S*,4*S*)-3-methyl-4-octanolide, from oak wood and chiroptical properties of mono-cyclic γ-lactones. *Chem. Lett.* **1981**, 1333−1336.

Otsuka, K.; Zenibayashi, Y.; Itoh, M.; Totsuka, A. Presence and significance of two diastereomers of β-methyl-γ-octalactone in aged distilled liquors. *Agric. Biol. Chem.* **1974**, *38*, 485−490.

Otsuka, K.; Sato, K.; Kawamatsu, M. Purification and properties of a precursor of β-methyl-γ-octalactone, an aging flavor compound of distilled liquors. *Hakkokogaku* **1980a**, *58*, 369−375.

Otsuka, K.-I.; Sato, K.; Yamashita, T. Structure of a precursor of β-methyl-γ-octalactone, an aging flavor compound of distilled liquors. *J. Ferment. Technol.* **1980b**, *58*, 395−398.

Seo, S.; Tomita, Y.; Tori, K.; Yoshimura, Y. Determination of the absolute configuration of a secondary hydroxy group in a chiral secondary alcohol using glycosidation shifts in carbon-13 nuclear magnetic resonance spectroscopy. *J. Am. Chem. Soc.* **1978**, *100*, 3331−3339.

Strack, D.; Wray, V. Anthocyanins. In *Methods in Plant Biochemistry; Dey, P. M., Harborne, J. B., Eds.; Academic Press: London, England, 1989; Vol. I, Plant phenolics, pp 325−356.*

Suomalainen, H.; Nykänen, L. Composition of whisky flavour. *Process Biochem.* **1970**, *5*, 13−18.

Tanaka, T.; Kouno, I. Whisky lactone precursors from the wood of *Platycarya strobilacea*. *J. Nat. Prod.* **1996**, *59*, 997−999.

Tori, K.; Seo, S.; Yoshimura, Y.; Arita, H.; Tomita, Y. Glycosidation shifts in carbon-13 NMR spectroscopy: Carbon-13 signal shifts from aglycon and glucose to glucoside. *Tetrahedron Lett.* **1977**, 179−182.

Received for review March 6, 2000. Revised manuscript received June 21, 2000. Accepted June 23, 2000. This study was financed by MARTELL & Co. (Cognac, France).

JF0002950