Absolute Stereochemistry of Natural 3,4-Dihydroxy-#-ionone Glycosides by the Cd Exciton Chirality Method

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ABSOLUTE STEREOCHEMISTRY OF NATURAL 3,4-DIHYDROXY-β-IONONE GLYCOSIDES BY THE CD EXCITON CHIRALITY METHOD

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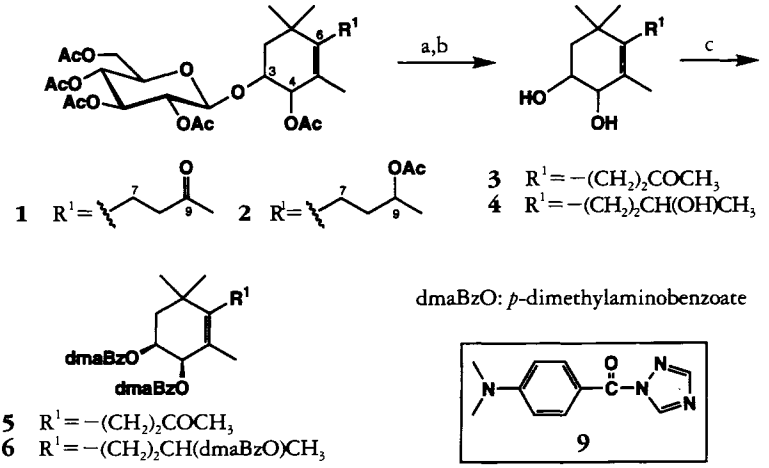
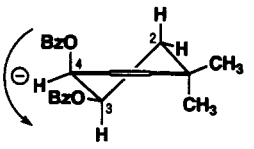
ABSTRACT.—Determination of the absolute stereochemistry of natural 3,4-dihydroxy-β-ionone glycosides by the bichromophoric exciton chirality method has established the configurations as being 3β,4β (3*S*,4*R*).

We have reported the isolation of 3,4-dihydroxy-7,8-dihydro β-ionone β-D-glucopyranoside [**1**] from red currant (*Ribes rubrum* L.) leaves and 3,4-dihydroxy-7,8-dihydro-β-ionol β-D-glucopyranoside [**2**] from gooseberry (*Ribes uva crispa* L.) leaves (1,2). These glycosides are natural precursors for important flavor compounds. We report herein the determination of the relative configuration by nmr and of the absolute stereochemistry by the exciton chirality method via a modified *p*-dimethylaminobenzoylation method which is adaptable to μg-scale derivatizations.

Irradiation of H-3 in both **1** and **2** led to a 4% nOe at H-4, i.e., the hydrogens are cis. The coupling constants (1,2) of **1** and **2** (see **7**/**8** below) (**1**: $J_{2a,3}$=12.7 Hz, $J_{2b,3}$=2.6 Hz, $J_{3,4}$=3.4 Hz; **2**: $J_{2a,3}$=12.6 Hz, $J_{2b,3}$=3.7 Hz, $J_{3,4}$=3.4 Hz) show that H-3 is axial and H-4 is equatorial. Accordingly, if the configurations of the hydroxyl groups are 3β,4β (3*S*,4*R*), **1** and **2** adopt the half-chair conformation **7** (see below, OH instead of OBz) whereas if they are 3α,4α (3*R*,4*S*), they adopt the half-chair conformation **8**. Computer analysis of the starting glycosides and benzoates with MacroModel 4.5 using the modified Allinger MM2 force field method also showed that the half-chair conformation **7** (or **8**) is the most preferred. Deacetylation of glycosides **1** and **2** with sodium methoxide, followed by hydrolysis with β-glucosidase (emulsin) (1,2) yielded the free aglycones **3** and **4**, which were submitted to cd analysis (Scheme 1).



SCHEME 1. Reagents: (a) NaOMe, MeOH; (b) β-glucosidase, buffer (pH=5); (c) *p*-dimethylaminobenzoyltriazole [**9**], DBU, MeCN.



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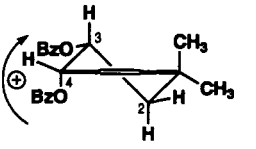

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The exciton chirality method is a microscale procedure for determination of the absolute configuration and conformation of a variety of molecules containing two or more chromophores (3,4). Hydroxyls are converted into various para-substituted benzoates and other chromophores (5), which may or may not be identical. When two identical benzoate chromophores interact through space, they give rise to bisignate cd curves, the signs of which are defined nonempirically by the absolute twists between the coupled chromophoric electric transition moments. One of the most widely used chromophores for hydroxy groups is *p*-methoxycinnamate, λ max (EtOH) 306 nm (ε 23,400) (6,7). However, it was noticed recently that the *p*-methoxycinnamate group is light-sensitive and undergoes cis/trans isomerization when left in the light for an extended period. *p*-Dimethylaminobenzoate, λ max (EtOH) 311 nm (ε 30,400), is another widely used chromophore; however, cases have been encountered in which the conventional derivatization via the chloride (3) does not give satisfactory yields in microscale reactions. This difficulty can be overcome by using its triazole derivative: it can be stored and used whenever needed, and the yield is ca. 90%. The sufficiently red-shifted maximum assures that coupling with the existing allylic double bond (8) is minimal.

Treatment of approximately 1 mg of **3** and **4** with *p*-dimethylaminobenzoyltriazole [9] gave the bis-(*p*-dimethylaminobenzoates) **5** and **6**. The similarity in *J* values for glycoside **1** and bisbenzoate **5** showed that the conformations should be the same. The cd and uv spectra of compounds **5** and **6** are shown in Figure 1. The negative split cd curves of **5** and **6** clearly show that the chirality between the 3,4 substituents is negative (counter-

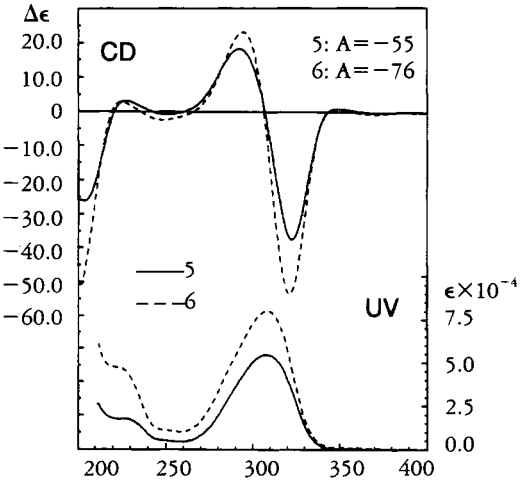

FIGURE 1. Cd and uv spectra of bis-(*p*-dimethylaminobenzoates) **5** (solid line) and **6** (dashed line) in MeCN.

clockwise). The cd curve of the bis-(*p*-dimethylaminobenzoate) **5** shows a strong negative split cd band with extrema at 321 nm (−37) and 292 nm (+18), amplitude *A* of −55, whereas **6** shows a similar negative split cd curve, with a larger *A* value of −76. The increased amplitude is probably due to the additional spatial coupling between the 9-benzoate and the 3 and/or 4 benzoates, but the flexibility of the side-chain does not allow determination of the C-9 configuration. The strong negative couplings observed for **5** and **6** led to a negative chirality between the La (long axis) transitions of the 3- and 4-benzoates.

The nmr data led to either the half-chair configuration **7** with a negative chirality, or configuration **8** with a positive chirality between the 3,4-bisbenzoates. Thus, as in the case of the nemadectins (9), the negative split cd curves (Figure 1), coupled with nmr data, establishes the configuration of glycosides **1** and **2** as 3*S*,4*R*, or 3β,4β as in **5** and **6**.

EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—^{1}H-Nmr spectra were recorded on a Varian 400 MHz instrument (Varian VXR400). Mass spectra were measured on a JEOL JMS-DX303HF mass spectrometer. Uv/vis and cd spectra were recorded as MeCN solutions on a Perkin-Elmer Lambda 4B uv/vis spectrometer and Jasco J-720 spectropolarimeter, respectively.

DEACETYLATION AND ENZYMATIC HYDROLYSIS OF **1** AND **2**.—After the addition of 3 mg of glycoside to a solution of 5 mg of sodium methoxide in 2 ml of MeOH and stirring overnight, 50 mg of Dowex 50-WX8 (20–50 mesh, H^+ form) was added. After 30 min, the exchanger was filtered off, the solvent removed under reduced pressure to dryness, and the residue was dissolved in 25 ml of 0.2 M citrate-phosphate buffer (pH 5.0) (yield 95%). The solution was incubated with 10 mg β-glucosidase (emulsin, Serva) at 37° for 12 h and the liberated aglycone extracted with Et_2O. The organic phase was dried over anhydrous Na_2SO_4 and carefully concentrated to dryness (yield 90% for **3**, 70% for **4**).

PREPARATION OF *p*-DIMETHYLAMINOBENZOYLTRIAZIOLE **9**.—A solution of *p*-dimethylamino benzoic acid (100 mg, 0.61 mmol) and 1,1′-carbonyl-*bis*(1,2,4-triazole) (109 mg, 0.67 mmol) in 2 ml MeCN was stirred overnight at room temperature. The reaction mixture, after concentration, was passed quickly through a short Si gel column (Et_2O-pentane, 9:1), yield 95%. **9**: ^{1}H nmr ($CDCl_3$) δ 9.02 (1H, s), 8.27 (2H, d, *J*=8.9 Hz), 8.09 (1H, s), 6.71 (2H, d, *J*=8.9 Hz), 3.12 (6H, s).

PROCEDURE FOR THE PREPARATION OF CHROMOPHORIC DERIVATIVES **5** AND **6**.—To a solution of **3** (1 mg, 4.4 μmol) in 0.5 ml dry MeCN was added dimethylaminobenzoyltriazole [**9**] (9.7 μmol) and distilled DBU (10.5 μmol). The mixture was stirred at room temperature for 3 h, quenched by addition of 1 drop H_2O, concentrated, and purified by flash chromatography to afford **5**, yield 95%. **5**: ^{1}H nmr ($CDCl_3$) δ 7.94 (2H, d, *J*=8.8 Hz), 7.69 (2H, d, *J*=9.2 Hz), 6.73 (2H, d, *J*=8.8 Hz), 6.58 (2H, d, *J*=9.2 Hz), 5.69 (1H, d, *J*=3.5 Hz, H-4), 5.31 (1H, ddd, *J*=13.0, 4.1, and 3.5 Hz, H-3), 3.05, 2.96 (12H, 2s), 2.4 (4H, m), 2.15 (3H, s), 1.95 (2H, dd, *J*=13.0 Hz), 1.75 (dd, *J*=13.0 and 4.1 Hz), 1.68 (3H, s), 1.1 (6H, 2s); fabms *m/z* 520 (M^+); uv λ max (MeCN) 306 nm (ε/dm^3 mol^{-1} cm^{-1} 54000); cd λ (MeCN) 292 nm (Δε +18), 321 (−37).

Chromophoric derivative **6** was prepared from **4** (1 mg, 4.4 μmol) using 14.5 μmol dimethylaminobenzoyltriazole [**9**] and 15.8 μmol DBU following the procedure above. **6**: Fabms *m/z* 669 (M^+); uv λ max (MeCN) 308 nm (ε/dm^3 mol^{-1} cm^{-1} 81000); cd λ (MeCN) 292 nm (Δε +23), 320 (−53).

ACKNOWLEDGMENTS

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ERRATUM

For the paper by Su *et al.* entitled "Lobocalone: a Novel Secondary Metabolite from the Soft Coral *Lobophytum caledonense*," *J. Nat. Prod.*, **56**, 279 (1993), the title compound is not novel. It has been reported previously by Gopichand and Schmitz, *Tetrahedron Lett.*, 3641 (1978). The authors accordingly wish to withdraw the name "lobocalone" from the literature.