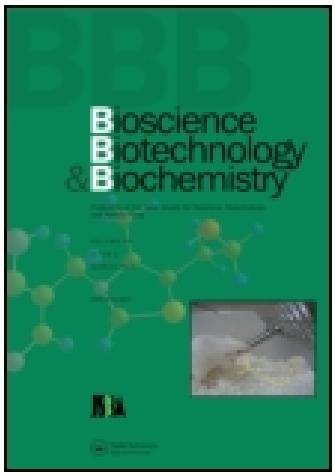


Bioscience, Biotechnology, and Biochemistry
Publication details, including instructions for authors and subscription information:
http://www.tandfonline.com/loi/tbbb20

Enantiomeric Resolution of a Germacrene-D Derivative by Chiral High-performance Liquid Chromatography

Yoshinori Nishii[a], Taichi Yoshida[a] & Yoo Tanabe[a]
[a] School of Science, Kwansei Gakuin University, 1-1-155 Uegahara, Nishinomiya, Hyogo 662, Japan
Published online: 12 Jun 2014.

To cite this article: Yoshinori Nishii, Taichi Yoshida & Yoo Tanabe (1997) Enantiomeric Resolution of a Germacrene-D Derivative by Chiral High-performance Liquid Chromatography, Bioscience, Biotechnology, and Biochemistry, 61:3, 547-548, DOI: 10.1271/bbb.61.547

To link to this article: http://dx.doi.org/10.1271/bbb.61.547

Biosci. Biotech. Biochem., **61** (3), 547–548, 1997

Note

Enantiomeric Resolution of a Germacrene-D Derivative by Chiral High-performance Liquid Chromatography

Yoshinori NISHII, Taichi YOSHIDA, and Yoo TANABE†

School of Science, Kwansei Gakuin University, 1–1–155 Uegahara, Nishinomiya, Hyogo 662, Japan

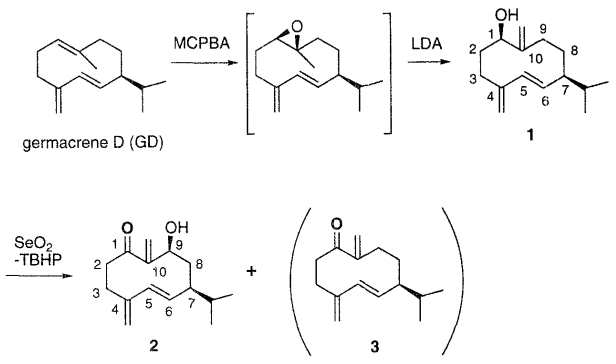
Received August 28, 1996

Direct enantiomeric resolution of the germacrene-D derivative, (5*E*,7*S,9*S**)-9-hydroxy-7-isopropyl-4,10-bis(methylene)-5-cyclodecen-1-one, was carried out by HPLC with a chiral stationary phase. This method showed that germacrene-D contained in *ylang ylang* oil was 98% *e.e.*, whereas germacrene-D extracted from *Solidago altissima* L. was almost racemic.**

Key words: germacrene-D; resolution; optical purity; chiral HPLC

Germacrene-D (GD) is one of the representative sesquiterpenes possessing a ten-membered ring structure.[1] Since its first isolation from *Pseudotsuga japonica*,[2] many studies have been devoted to GD for the following reasons: (a) its chemical ecology,[3] (b) its biomimetic behavior[2,4]; (c) the biological activity of its pheromone[5]; (d) its synthesis[6]; and (e) as the starting compound for the total syntheses of pheromone-active periplanones.[7] GD is present in many plants with varying optical purity. Although the optical rotation of GD has been estimated,[8] the enantiomeric purity of optically active GD has not been accurately determined by reliable high-performance liquid chromatography (HPLC) or gas chromatography. In connection with our interest in studying insecticidally active compounds,[9] we report here the first enantiomeric resolution of the GD derivative, (5*E*,7*S**,9*S**)-9-hydroxy-7-isopropyl-4,10-bis(methylene)-5-cyclodecen-1-one (**2**), by HPLC with a chiral stationary phase by using two samples of the GD derivative.

GD is a very labile and lipophilic compound, so we planned to transform it into more stable and polar derivatives while maintaining the germacrane skeleton with the expectation of easier separation by HPLC. To acheive this, we applied the Yamamura and Shizuri route for the total synthesis of periplanones,[7] since a family of oxidative derivatives of GD would be obtained without any loss of optical purity. We used two samples of GD, namely, optically active GD [(7*S*)-GD] contained in *ylang ylang* oil made in Madagascar and almost-racemic GD [(7*S*,*R*)-GD] extracted from *Solidago altissima* L. (Seitaka-awadachiso) in Japan.



We first tested HPLC analyses (using Chiralcel OE and OD columns) of 1-hydroxy derivative (1*R**,7*S**)-**1** which was afforded by an epoxydation of (7*S*,*R*)-GD and subsequent treatment with lithium diisopropylamide (LDA). However, these analyses showed no separation by HPLC, probably because there are three carbons between the inherent chiral center (7-position) and the anchor carbon (1-position) with a hydroxyl function in **1**.

We next examined the application of 9-hydroxy derivative (7*S**,9*S**)-**2** which was obtained by the further SeO_2/*tert*-butyl hydroperoxide (TBHP) oxidation from (1*R**,7*S**)-**1**. Enhancement of the favored molecular interaction by HPLC was expected, because derivative **2** possesses one carbon fewer between the two chiral centers. This experiment gave a successful result, by clear separation of the two enantiomers of (7*S**,9*S**)-**2** being obtained by HPLC with a Chiralcel OD column (Fig.).

The optical purity of (7*S*,9*S*)-**2** derived from (7*S*)-GD in *ylang ylang* oil was 98% *e.e.*, whereas (7*S**,9*S**)-**2** derived from (7*S*,*R*)-GD in *Solidago altissima* L. was almost racemic [(7*S*,9*S*):(7*R*,9*R*)= 51 : 49]. This HPLC analysis gave results in good accordance with the optical rotation values for corresponding compounds **2**. This clear contrast in the optical purity values would have been due to the chemotaxonomic difference from the GD origins. The absolute configurations of **1** and **2** were rationally deduced from the literature.[7a,10] It should be noted that (a) the two conversion sequences were highly stereoselective, because no other diastereomers of either **1** or **2** were detected by careful ^{1}H-NMR measurements; and (b) no substantial epimerization at the 7-position occurred during the conversion ($\leq$1%), because (7*S*,9*S*)-**2** showed 98% *e.e.* Therefore, these *e.e.* values would correspond to those of the original GD samples.

In conclusion, we achieved the first accurate determination of

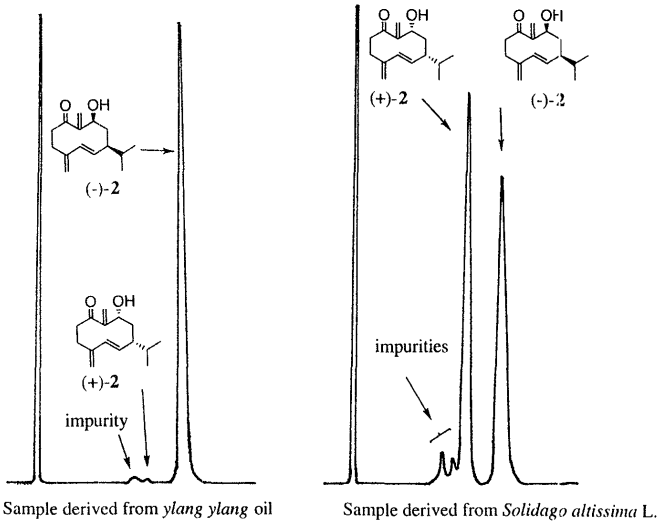


Fig. Direct Resolution of GD Derivative **2** by HPLC with a Chiral Stationary Phase (Chiralcel OD Column).

† To whom correspondence should be addressed.

the optical purity of GD derivative by using a reliable HPLC analysis, although a few conversion steps were required. By utilizing the result, we are now studying a total synthesis of optically active periplanone species which will be reported in the near future.

Experimental

Apparatus. Melting point (mp) data were determined with hot-stage microscope apparatus (Yanagimoto) and are uncorrected. ^{1}H-NMR spectra were recorded with a JEOL α (400 MHz) spectrometer, using TMS as an internal standard, while IR spectra were recorded with a JASCO FT/IR-8000 spectrophotometer. Analytical GLC was performed on a Shimadzu GC 9A with a DB-1701 wide-bore column (30 m × 0.545 mm).

High-performance liquid chromatography. Analyses were carried out by a Shimadzu LC-10A instrument fitted with a UV-VIS detector. The standard chromatographic conditions were a Chiralcel OB or OD column [Daicel, 4.6ϕ × 250 mm, column temperature of 24.0°C, UV detection at 255 nm, and injected amount of 1.5 mg/ml in CH_2Cl_2 (15 μl)].

Purification of (7S)-GD. The *ylang ylang* oil [containing about 10% (−)-GD] was purchased from Ogawa Koryo Co., Ltd. and purified up to about 40% by fractional distillation (68–75°C/0.2–1.0 mmHg). This fraction was further purified (~60%) by a SiO_2 flash column chromatography (Merck Art. 7734, ×10 wt; hexane eluent). Chemical purity was determined by analytical GLC, and this material was used for the subsequent conversion.

Extraction and purification of (7S,R)-GD. Dried leaves and stems (0.75 kg) of *Solidago altissima* L. (Seitaka-awadachiso) collected at Nishinomiya (Hyogo prefecture) in August 1995 were cut into small pieces and extracted with hexane. The extract was concentrated *in vacuo* to give 12.0 g of a green syrup. This syrup was subjected to silica-gel flash column chromatography (Merck Art. 7734, 120 g; hexane eluent) to give 0.76 g of (7*S*,*R*)-GD (about 80% content by GLC analysis and ^{1}H-NMR measurement).

Synthesis of (1R,5E,7S)- and (1R,5E,7S*)-7-isopropyl-4,10-bis(methylene)-5-cyclodecen-1-ol* [*(1R.7S)-1 and (1R*,7S*)-1*].[7a,10] According to the reported method for preparing racemic compound **1**,[7a] *m*-chloroperoxybenzoic acid (28.8 g, 0.17 mol) in ether (100 ml) was added to a stirred solution of (7*S*)-GD (34.0 g, *ca.* 0.10 mol) in ether (300 ml) at 0–10°C during 1 h, and the mixture was stirred at 0–5°C for 1 h. The mixture was then poured into an aqueous sat. $NaHCO_3$ solution (300 ml), which was extracted with ether (150 ml × 2). The combined organic phase was sequentially washed with water, and brine, dried (Na_2SO_4), and concentrated to give a crude monoepoxide of (7*S*)-GD (36.0 g). LDA (1.5 M in cyclohexane; 190 ml) was added to a stirred solution of this monoepoxide (36.0 g) in ether (300 ml) at −60°C under a nitrogen atmosphere. The mixture was allowed to warm and was then kept at room temp. for 10 h. Water was next added to the mixture, which was extracted with ether (500 ml × 2). The combined organic phase was sequentially washed with water and brine, dried (Na_2SO_4), concentrated, and purified by silica-gel chromatography (hexane/EtOAc = 7 : 1) to give 14.8 g of (1*R*,7*S*)-**1** [67% yield based on net (7*S*)-GD] as colorless crystals; mp 43–45°C; $[\alpha]_D^{18}$ −184.5° (*c* 0.2, $CHCl_3$); IR (KBr): 3418, 1460, 1038 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ: 0.80 (3H, d, *J* = 6.0 Hz), 0.89 (3H, d, *J* = 6.0 Hz), 1.10–1.80 (5H, m), 1.98–2.10 (2H, m), 2.14–2.24 (1H, m), 2.38–2.48 (1H, m), 2.58–2.68 (1H, m), 3.78 (1H, dd, *J* = 4.5 Hz and 7.3 Hz), 4.86 (1H, s), 4.94 (1H, s). 5.02 (1H, s), 5.29 (1H, s), 5.44 (1H, dd, *J* = 5.1 Hz and 9.6 Hz), 6.00 (1H, d, *J* = 10.9 Hz).

A similar procedure using (7*S*,*R*)-GD gave (1*R**,7*S**)-**1**[7a] as a colorless oil; $[\alpha]_D^{18}$ −2.8° (*c* 0.2, $CHCl_3$). An HPLC analysis was attempted for the resolution of this sample (using Chiralcel OB or OD and changing the mobile phase of hexane : 2-propanol = 40 : 1 to 200 : 1). However, the analysis showed only one peak.

Syntheses of (5E,7S,9S)- and (5E,7S,9S*)-9-hydroxy-7-isopropyl-4,10-bis(methylene)-5-cyclodecen-1-one* [*(7S,9S)-2 and (7S*,9S*)-2*].[7a] According to the reported method for preparing racemic compound **2**,[7a] TBHP (a 70% aqueous solution; 17.0 g, 0.132 mol) was added to a stirred solution of selenium (IV) oxide (1.50 g, 13.5 mmol) in CH_2Cl_2 (90 ml) at room temp. To the resulting suspension was added alcohol (1*R*,7*S*)-**1** (14.8 g, 67 mmol) in CH_2Cl_2 (45 ml) at 30–35°C under a nitrogen atmosphere, and the mixture was stirred for 24 h at that temperature. Then, 10% aqueous KOH was added to the mixture to adjust the mixture to pH value to *ca.* 7.0. Dimethyl sulfide (12.5 g) and acetic acid (8.0 g) were successively added to the mixture at 0–5°C, before neutralizing with K_2CO_3 (pH *ca.* 7.0) and extracting with CH_2Cl_2 (80 ml × 2). The combined organic phase was sequentially washed with water and brine, dried (Na_2SO_4), concentrated, and purified by silica-gel chromatography (hexane/EtOAc = 3 : 1 to 5 : 2) to give 3.30 g of (7*S*,9*S*)-**2** (21%) and 5.69 g of ketone (7*S*)-**3** (39%). (7*S*,9*S*)-**2**: pale yellow crystals; mp 37–41°C; $[\alpha]_D^{18}$ −167.0° (*c* 0.2, $CHCl_3$); IR (neat): 3447, 1672, 1026 cm^{-1}; ^{1}H-NMR (C_6D_6) δ: 0.67 (3H, d, *J* = 6.0 Hz), 0.77 (3H, d, *J* = 6.0 Hz), 1.17–1.36 (1H, m), 1.58–1.67 (2H, m), 2.03–2.17 (2H, m), 2.17–2.26 (1H, m), 2.37–2.57 (2H, m), 4.53 (1H, br. s), 4.70–4.76 (2H, m), 4.97–5.07 (1H, m), 5.29 (1H, s), 5.53 (1H, s), 5.60 (1H, s). Ketone (7*S*)-**3**: pale yellow oil; $[\alpha]_D^{25}$ −198.0° (*c* 0.50, $CHCl_3$); IR (neat): 1676, 1447, 976 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ: 0.80 (3H, d, *J* = 6.8 Hz), 0.87 (3H, d, *J* = 6.8 Hz), 1.46–1.58 (2H, m), 1.76–1.82 (1H, m), 1.93–2.00 (1H, m), 2.28–2.41 (3H, m), 2.47–2.53 (1H, m), 2.74–2.78 (1H, m), 3.02–3.08 (1H, m), 4.82 (1H, s), 4.91 (1H, s), 5.25 (1H, dd, *J* = 10.0 Hz and 16.0 Hz), 5.51 (1H, s), 5.73 (1H, s), 5.75 (1H, d, *J* = 16.0 Hz).

A similar procedure with (1*R**,7*S**)-**1** gave (7*S**,9*S**)-**2**[7a] as a colorless oil; $[\alpha]_D^{18}$ −2.4° (*c* 0.2, $CHCl_3$). This sample was separated by HPLC with Chiralcel OD (mobile phase of hexane : 2-propanol = 40 : 1); (7*R*,9*R*)-**2** retention time = 15.91 min; (7*S*,9*S*)-**2** retention time = 19.71 min).

Acknowledgment. We thank Professor Haruhisa Shirahama of Kwansei Gakuin Univ. for his helpful discussions and also thank Sumitomo Chemical Co., Ltd. for supplying the *ylang ylang* oil. This work was partially supported by a Grant-in-Aid for Scientific Research from the Ministry of Education, Science, and Culture of Japan.

References

1) "Dictionary of Natural Products," ed. by J. Buckingham, Chapman & Hall, London, 1990, pp. 2535–2536.
2) K. Yoshihara, Y. Ohta, T. Sakaki, and Y. Hirose, *Tetrahedron Lett.*, **1969**, 2263–2264.
3) a) M. Yoshikawa, S. Hatakeyama, N. Tanaka, T. Matsuoka, J. Yamahara, and N. Murakami, *Chem. Pharm. Bull.*, **41**, 2109–2112 (1993); b) S. Derosa, A. D. Giulio, C. Iodice, and N. Zavodink, *Phytochemistry*, **37**, 1327–1330 (1994).
4) a) Y. Shizuri, S. Yamaguchi, Y. Terada, and S. Yamamura, *Tetrahedron Lett.*, **27**, 57–60 (1986); b) D. P. Piet, R. Schrijvers, M. C. R. Franssen, and A. Degroot, *Tetrahedron*, **51**, 6303–6314 (1995).
5) S. Tahara, M. Yoshida, J. Mizutani, C. Kitamura, and S. Takahashi, *Agric. Biol. Chem.*, **39**, 1517–1521 (1975).
6) T. Kitahara and K. Mori, *J. Org. Chem.*, **49**, 3281–3285 (1984).
7) a) Y. Shizuri, S. Yamagichi, S. Yamamura, M. Ishihara, S. Ohba, Y. Saito, M. Niwa, Y. Terada, and M. Miyazaki, *Tetrahedron Lett.*, **28**, 3831–3834 (1987); b) Y. Shizuri, K. Matsunaga, K. Tamaki, S. Yamaguchi, and S. Yamamura, *Tetrahedron Lett.*, **29**, 1971–1974 (1988).
8) M. Niwa, M. Iguchi, and S. Yamamura, *Chem. Pharm. Bull.*, **28**, 997–999 (1980).
9) a) Y. Nishii, K. Wakimura, T. Tsuchiya, S. Nakamura, and Y. Tanabe, *J. Chem. Soc., Perkin Trans. 1*, **1996**, 1243–1249; b) Y. Nishii, H. Matsumura, Y. Muroya, T. Tsuchiya, and Y. Tanabe, *Biosci. Biotech. Biochem.*, **59**, 1355–1357 (1995); c) Y. Sanemitsu, Y. Nakayama, Y. Tanabe, and T. Yano, *Agric. Biol. Chem.*, **53**, 2843–2845 (1989).
10) a) E. Fattorusso, S. Magno, L. Mayol, V. Amico, G. Oriente, M. Piattelli, and C. Tringali, *Tetrahedron Lett.*, **1978**, 4149–4152; b) F. Nagashima, M. Toyota, and Y. Asakawa, *Phytochemistry*, **29**, 2169–2174 (1990).