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Sesquiterpenes of Lauraceae Plants. IV.[1] Germacranolides from *Laurus nobilis* L.

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By virtue of correlation with pyrethrosin **6a**, the structure of laurenobiolide **1**, a component of *Laurus nobilis* L., was confirmed.

At least, two races of this plant exist, one of which contains **1** as a major component and the other costunolide **9**.

Although many investigations[3] have been conducted on the essential oil from leaves of *Laurus nobilis* L., none have been on the intact plant.

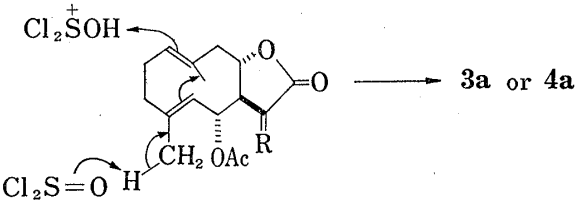
In connection with a chemosystematic study on components of lauraceous plants growing in Japan, we have extracted roots and leaves of *L. nobilis* L. From the roots, laurenobiolide was isolated as a major component whose structure was elucidated as (**1**)[4]

Reduction of laurenobiolide **1** with $NaBH_4$ yielded a mixture of C-11 epimers (**2a**) and (**2b**), the former was separated by fractional recrystallisation. The configuration of the C-13 methyl group in **2a** was suggested by the solvent shift[5] to be α-pseudoequatorial [$\Delta\delta(CDCl_3$–$C_6D_6)$+0.04]. The other epimer (**2b**) was not obtained in a pure state, but the proton signals of the C-13 methyl group could be clearly distinguished and the solvent shift suggested that the configuration is β-pseudoaxial ($\Delta\delta$+0.17). Hydrolysis of **2a** followed by acidification and acetylation caused regeneration of the original dihydrolactone **2a**, indicating that the lactone linkage in **2a** must be C(7)–C(8) and not C(7)–C(6).[6]

Treatment of **2a** with $SOCl_2$[7] afforded the expected cyclisation product (**3a**) as a major component (38%), whose proton magnetic resonance (PMR) spectrum showed a little contamination with Δ^3-isomer (**3b**).[8] Another product (24%) was the 6α-chloro compound (**3c**) whose structure was suggested by PMR (6-H: t, J=10 Hz; also indicates a little contamination with Δ^3-isomer); **3c** was not further investigated as it deteriorates rapidly even in an ice-box. The coupling constants of 6-H (t, J=10.2 Hz) and 8-H (ddd, J=12.0, 11.0, and 3.9 Hz) in **3a** indicate that the 5-, 6-, 7-, and 8-protons are *trans* diaxial. Similarly, **1** was cyclised by the same reagent to yield (**4a**) (**4b**) and (**4c**) but these products showed a tendency to deteriorate. The structures were suggested by only PMR, infrared (IR), and circular dichroism (CD) spectra, using freshly prepared samples. In this case, pseudolaurenobiolide (**4d**) was also isolated from the reaction product. One of the two methyl groups on the double bond in **1** was isomerised to *exo*-methylene in **4d**. The structure was determined by PMR; namely, olefinic 5-H appears as doublet and 6-H is found at the same chemical shift as in **1**.

1) Part III: K. Takeda, K. Sakurawi, and H. Ishii, *Tetrahedron*, **27**, 6049 (1971).
2) Location: *Fukushima-ku, Osaka, 553, Japan.*
3) *a*) J.W. Hogg, S.J. Terhune, and B.M. Lawrence, *Phytochem.*, **13**, 868 (1973), and references cited there in; *b*) J. Foussereau, C. Benezra, and G. Ourisson, *The Trans. St. John's Hospital Dermatol. Soc.*, **53**, 147 (1967).
4) H. Tada and K. Takeda, *Chem. Comm.*, **1971**, 1391.
5) C.R. Narayanan and N.K. Venkatasubramanian, *J. Org. Chem.*, **33**, 3156 (1968).
6) H. Yoshioka, W. Renold, and T.J. Mabry, *Chem. Comm.*, **1970**, 148.
7) R.W. Doskotch and F.S. El-Feraly, *J. Org. Chem.*, **35**, 1928 (1970).
8) In this case, the β-isomer ($\Delta^{4,(15)}$) was obtained and the amounts of α-(Δ^3) and γ-isomers (Δ^4) were very small, if any were at all present.

In the course of cyclisation, the role of $SOCl_2$ was speculated to be as follows:



According to this speculation, $POCl_3$ may be also used in the cyclisation reaction of **1** or **2a**, and the same product pattern as in the case of $SOCl_2$ was obtained.

Reversible Cope rearrangement of **2a** occurred on heating at 205°. The proton signals of 6-H (t, J=10.8 Hz) and 8-H (ddd, J=11.7, 11.1, and 4.0 Hz) in Cope product (**5**) show again that 5-, 6-, 7-, and 8-protons are *trans* diaxially disposed.

Since the relative configurations of **3a** and **5** were confirmed by NOE experiments,[4] and *trans* junction of C(5)–C(10) in **5** suggested that the double bonds in **1** must be *trans-trans*,[9] laurenobiolide **1** was treated with peracid. The oxidation product was apparently a mixture (almost 1: 1) of epoxy-lactone epimers according to PMR. Olefinic proton appears as a doublet and 6-H is found at the same chemical shift as in **1**, therefore the C(1)–C(10) double bond must have been oxidised. Epoxylaurenobiolide (**6a**) was obtained by fractional crystallisation and was directly compared with pyrethrosin.[10] Both compounds proved to be identical in all respects (mp, IR, PMR, CD, and $[\alpha]_D$). As the structure of pyrethrosin had already been confirmed byX-ray analysis,[11] the structure of laurenobiolide, with absolute configuration, was thus confirmed. Interestingly laurenobiolide **1** affords C(1)–C(10) epoxides by the action of peracid, while desacetyllaurenobiolide (**7**) has been reported to yield C(4)–C(5) epoxides (**8**),[12] presumably due to affinity of peracid with C-6 oxygen function and/or to steric hindrance.

Also, in several extractions of different sources of *L. nobilis* L., some materials contained costunolide (**9**) as a major component and only a small amount of **1**. From the other material, we isolated **1**, **2a**, **7**, and dihydrodesacetyllaurenobiolide (**10**). These differences are not due to i) locality of plant cultivation, ii) sex, and iii) seasonal variation; there are probably two races of *L. nobilis* L., even if they are morphologically same[13] (Table I).

Table I

Locality	Sex	Season	Laurenobiolide **1**	Costunolide **9**
Tokyo No. 1	unknown	May	###	–
No. 2	♂	Feb.	+	###
Kyoto No. 1	unknown	Apr.	+	###
No. 2	♂	Aug.	±	##
No. 3	♀	Aug.	+	+
Kobe No. 1	♀	Feb.	###	–
		Jun.	###	–

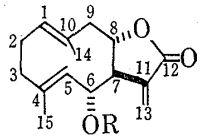
The ether extracts of leaves of some different sources of *L. nobilis* L., were also investigated. The major component was always desacetyllaurenobiolide **7**, and minor component was **1** or **9**. As a result, this plant could be classified as follows (Table II).

9) K. Takeda, I. Horibe, and H. Minato, *Chem. Comm.*, **1971**, 308.
10) S. Iriuchijima and S. Tamura, *Agr. Biol. Chem.*, **34**, 204 (1970).
11) E.J. Gabe, S. Neidle, D. Rogers, and C.E. Nordman, *Chem. Comm.*, **1971**, 559.
12) F. Shafizadeh and N.R. Bhadane, *Phytochem.*, **12**, 857 (1973).
13) Private communication from Prof. G. Ourisson. There is another similar observation that the linalool content is extremely different between the essential oil from leaves collected in Tunisia and that in Morocco.

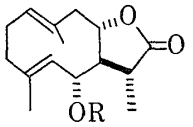
Table II

Material[a]	Components of roots		Components of leaves		
	1	2	7	1	2
A	##	−	##	+	−
B	+	##	##	−	+

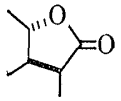
a) A: Tokyo No. 1 and Kobe No. 1 *etc.*
B: Tokyo No. 2 and Kyoto No. 1 *etc.*



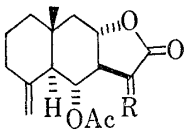
1 : R=Ac laurenobiolide
7 : R=H



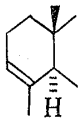
2a : R=Ac
10 : R=H



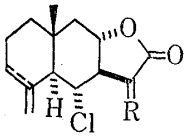
2b



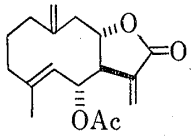
3a : R=α-Me, β-H
4a : R=CH_2



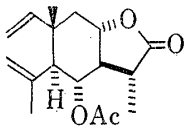
3b
4b



3c : R=α-Me, β-H
4c : R=CH_2



4d



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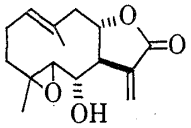

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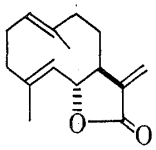

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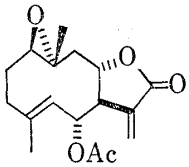
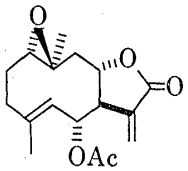


6a ≡ pyrethrosin



6b

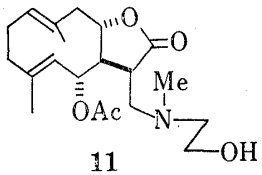


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Chart 1

Experimental

All melting points were measured on a hot plate and not corrected. Infrared spectra (IR) spectra were obtained in $CHCl_3$ solution with a Hitachi EPI-G3 spectrometer. CD spectra were obtained in MeOH solution with a JASCO ORD/UV 6. PMR spectra were determined in $CDCl_3$ solution with a Varian A56/60 spectrometer. Merck SiO_2 (0.05—0.2 mm) deactivated with additional 10% of water was used for column chromatography. Thin-layer chromatography (TLC) was developed with benzene–ethyl acetate (9: 1). Rotations were measured in EtOH solution (c=1) with a Perkin Elmer polarimeter 141.

Isolation of the Components——a) Dried and ground roots of *Laurus nobilis* L. (Tokyo No. 1, 2.8 kg) were extracted with ether at room temperature. The ether extract (26 g) was chromatographed on SiO_2 (200 g) and semisolid eluates (benzene to 5% EtOAc–benzene fractions) were triturated with hexane to yield crystals (6.2 g, 0.2%), which on recrystallisation from ether–hexane, gave laurenobiolide (**1**, 5.1 g); mp 101—103°; *Anal.* Calcd. for $C_{17}H_{22}O_4$: C, 70.32; H, 7.64. Found: C, 70.52; H, 7.61. Mass Spectrum *m/e*: 290 (M^+); $[\alpha]_D$ +17.1°. IR ν_{max} cm^{-1}: 1760, 1738, 1656. PMR ppm: 1.68 (s, two olefinic Me's), 2.05 (OAc), 5.88 and 6.35 (dd, J=3.0 and 1.0 Hz, *exo*-CH_2), 4.0 (m, 8-H), 4.5—5.3 (m, 1-H and 5-H), 5.33 (approx. t, 6-H). CD $[\theta]$ (nm): +43800 (208) (positive maximum) and −5660 (253) (negative maximum).

b) Another collection of roots (Kyoto No. 1, 1.7 kg) was extracted as above. On trituration with hexane, the extract (14.3 g) yielded costunolide (**2**, 2.25 g). The mother liquid was then chromatographed on SiO_2 (200 g). From the benzene eluates, further crystals of costunolide were obtained (3.1 g, total 5.35 g, 0.31%). The following eluates (2% and 5% EtOAc–benzene; 1.6 g) were rechromatographed on Al_2O_3 (Woelm neutral grade II, 100 g) to yield **1** (1.06 g, 0.06%) and phytosterols.

c) The ether extract (129 g) from roots of the third collection (Kobe No. 1, 7.1 kg) was chromatographed on SiO_2 (630 g) and the semisolid eluates (benzene to 5% EtOAc–benzene fractions) gave a mixture of **1** and **2a** (35.5 g). Further eluates (10% EtOAc–benzene and EtOAc fractions) were treated with Ac_2O followed by trituration with isopropyl ether to give additional mixture of **1** and **2a** (14.1 g, total 49.6 g, 0.7%).

d) The ether extract of leaves (470 g) was chromatographed on SiO_2, and an eluate (5% EtOAc–benzene) gave, after purification by PLC, desacetyllaurenobiolide **7** as an amorphous solid (27 mg): IR ν_{max} cm^{-1} 3575, 3455, 1755, 1655. PMR ppm: 1.1—1.2 (two olefinic Me's), 4.05 (m, 8-H), 4.25 (approx. t, 6-H), 4.6—5.3 (m, 1-H and 5-H), 6.18 and 6.35 (m, and dd, J=3.0 and 1.5 Hz, *exo*-CH_2). Acetylation of this solid yielded a crystalline acetate, which proved to be identical with laurenobiolide **1** in every respect.

Separation of 1 and 2a——a) A mixture of **1** and **2a** (3.0 g) was dissolved in EtOH (30 ml). The solution was stirred with N-methylethanolamine (3 ml) for 1 hr at room temperature. After evaporation of ethanol *in vacuo*, water was added then extracted with ether. The ether solution was treated with 1N HCl. From the neutral fraction, **2a** (1.1 g, mp 143—144°) was obtained, and the basic fraction afforded an adduct 2.25 g), which gave, on crystallisation from isopropyl ether, pure **11**, mp 136—137°. *Anal.* Calcd. for $C_{20}H_{31}O_5$: C, 65.73; H, 8.55; N, 3.83. Found: C, 65.58; H, 8.52; $[\alpha]_D$ +106.7°. IR ν_{max} cm^{-1}: 3470, 1763, 1735, 1667. PMR ppm: 1.62 (s, 14-Me), 1.67 (d, J=1 Hz, 15-Me), 2.04 (OAc), 2.32 (NMe), 4.15 (m, 8-H), 4.67 (m, 5-H), 4.95 (m, 1-H), 5.4 (m, 6-H).

b) A solution of the adduct **11** (365 mg) and methyl iodide (2 ml) in 10% MeOH· aq. (10 ml) was left in an ice box for 48 hr. After evaporation of the solvent, the residue was suspended in CH_2Cl_2-ether (1: 1) and treated with a saturated solution of $NaHCO_3$ for 1 hr at room temperature. The organic layer gave laurenobiolide (**1**, 167 mg, 58%) and recovered adduct **11** (140 mg, 38%) after treatment with 1N HCl.

$NaBH_4$ Reduction of Laurenobiolide 1——To a solution of **1** (1.45 g, 5 mmole) in MeOH (30 ml) was added $NaBH_4$ (190 mg, 5 mmole) at 0° and stirred for 30 min. Usual treatment gave a crude product which gave, on fractional recrystallisation, dihydro compound **2a** (558 mg, 38%): mp 142—144°; *Anal.* Calcd. for $C_{17}H_{24}O_4$: C, 69.83, H, 8.27. Found: C, 69.91, H, 8.37. Mass Spectrum *m/e*: 292 (M^+); $[\alpha]_D$ +120.3°; IR ν_{max} cm^{-1}: 1762, 1728, 1666. PMR ppm: 1.39 (d, J=7.0 Hz, 13-Me), 1.58 (br-s, 14-Me), 1.69 (d, J=1 Hz, 15-Me), 2.00 (OAc), 4.17 (br-t, 8-H), 4.65 (br-d, 5-H), 4.84 (m, 1-H), 5.45 (br-t, 6-H). CD [θ] (nm): +111000 (216) (positive maximum). PMR of other fractions showed additional signals [1.21 (13-Me), 1.50 (14-Me), 1.74 (15-Me), 2.03 (OAc), 4.53 (5-H), 5.38 (6-H)] due to epimer **2b**.

Cyclisation Reaction of Dihydrolaurenobiolide 2a——a) With $SOCl_2$: To a solution of **2a** (235 mg) in $CHCl_3$ (25 ml), was added $SOCl_2$ (0.65 ml). The reaction mixture was left standing for 30 min at room temperature then poured into water. Crude product (approximately two spots, 240 mg) were purified by PLC. The less polar product (51 mg, 24%) a labile oil even at 0°, gave a positive Beilstein test and reasonable spectra for chloro-lactone **3c**. IR ν_{max} cm^{-1}: 1774, 1648, 900. PMR ppm: 0.89 (s, 14-Me), 1.47 (d, 13-Me), 4.04 (t, J=10.5 Hz, 6-H), 4.65, 5.06 (=CH_2), [additional small signals due to *endo*-isomer: 0.93 (14-Me), 5.40 (m, 3-H)]. The polar product (86 mg, 38%) was recrystallised from ether–hexane to give acetoxy-lactone **3a**, mp 120—124°, *Anal.* Calcd. for $C_{17}H_{24}O_4$: C, 69.83; H, 8.27. Found: C, 70.07, H, 8.34. IR ν_{max} cm^{-1}: 1773, 1729, 1650, 902. PMR ppm: 0.90 (s, 14-Me), 1.17 (d, 13-Me), 2.02 (OAc), 4.12 (t-d, J=11.0 and 4.5 Hz, 8-H), 4.35, 4.84 (=CH_2), 5.27 (t, J=10.5 Hz, 6-H), small signals 0.96 (14-Me), 5.20 (t, J=10.5 Hz, 6-H), 5.38 (m, 3-H) were also observed in the spectrum, indicating the presence of Δ^3-isomer **4b** as a minor product.

b) With $POCl_3$: A mixture of **2a** (146 mg) in $CHCl_3$ (15 ml) and $POCl_3$ (0.4 ml) was left standing for 3 hr at room temperature. The crude product (approximately two spots, 146 mg) was purified by PLC to give **3c** (67 mg, 51%) and **3a** (44 mg, 30%).

Cyclisation Reaction of Laurenobiolide 1——a) With $SOCl_2$: Laurenobiolide (**1**, 290 mg) in $CHCl_3$ (30 ml) was allowed to react with $SOCl_2$ (0.8 ml) at room temperature for 7 hr. The crude product (300 mg) was purified by PLC to yield chloro-lactone **4c** (21 mg, 8%), acetoxy-lactone **4a** (74 mg, 26%), and pseudo-laurenobiolide **4d** (26 mg, 9%).

4a: labile oil, IR ν_{max} cm^{-1}: 1770, 1731, 1672, 1650, 906. PMR ppm: 0.89 (s, 14-Me), 2.05 (OAc), 4.08 (t-d, J=11.5 and 4.0 Hz, 8-H), 4.52, 4.83 (15-CH_2), 5.45 (t, J=10.5, 6-H), 5.43, 6.09 (d, J=3.0 Hz, 13-CH_2), small signals 0.95 (14-Me), 2.11 (OAc), 5.4 (m, 3-H) were also observed in the spectrum, indicating the presence of *endo*-isomer **4b** as a minor product.

4c: labile oil (positive Beilstein test), IR ν_{max} cm^{-1}: 1767, 1667, 1648, 893. PMR ppm: 0.88 (s, 14-Me), 4.15 (t, J=10.5 Hz, 6–H), 4.68, 5.07 (15–CH_2), 6.11, 6.21 (d, J=3.0 Hz, 13–CH_2).

4d: mp 157—159°, *Anal.* Calcd. for $C_{17}H_{22}O_4$: C, 70.32, H, 7.64. Found: C, 70.41, H, 7.60; IR ν_{max} cm^{-1}: 1760, 1737, 1657, 1648, 895. PMR ppm: 1.79 (d, J=1 Hz, 15-Me), 2.06 (OAc), 4.18 (ddd, J=10.5, 6.0, and 3.0 Hz, 8-H), 4.90, 5.02 (14-CH_2), 5.25 (t, J=10.5 Hz, 6–H), 5.84, 6.35 (dd, J=3.0 and 1.0 Hz, 13–CH_2).

b) With $POCl_3$: A mixture of **1** (145 mg) in $CHCl_3$ (15 ml) and $POCl_3$ (0.4 ml) was left standing at room temperature for 5 hr. The crude product (130 mg) was purified by PLC to yield **4c** (15 mg, 12%), **4a** (33 mg, 22%), and **4d** (20 mg, 14%).

Cope rearrangement: a) Dihydrolaurenobiolide (**2a**, 60 mg) was heated in a preheated bath (205°) for 5 min followed by purification on silica gel plate to give recovered **2a** (34 mg, 57%) and rearranged product (23 mg, 38%). The latter was recrystallised from ether–hexane to yield **5**, mp 138—140°, *Anal.* Calcd. for $C_{17}H_{24}O_4$: C, 69.83; H, 8.27. Found: C, 70.01; H, 8.32; $[\alpha]_D$ +12.2°, IR ν_{max} cm^{-1}: 3085, 1775, 1731, 1648, 920, 903. PMR ppm: 1.15 (s, 14-Me), 1.18 (d, 13-Me), 1.72 (15-Me), 2.00 (OAc), 4.12 (t-d, J=11.0 and 4.5 Hz, 8-H), 4.65, 5.00 (3–CH_2), 4.98, 5.00 (2–CH_2), 5.83 (dd, J=18.5 and 9.0 Hz, 1-H).

b) Rearranged product **5** (10 mg) was treated as above. Purification by PLC gave **2a** (4.3 mg) and **5** (3.3 mg).

Epoxidation of Laurenobiolide 1——A mixture of **1** (290 mg) in $CHCl_3$ (20 ml) and *m*-chloroperbenzoic acid (250 mg) in $CHCl_3$ (10 ml) was left standing overnight at 0°. Trituration of crude product (319 mg) with ether gave crystals (297 mg, mp 165—195°), whose PMR showed to be a mixture of epoxy-lactones **6a** and **6b**. Repeated fractional crystallisation of the mixture gave **6a** (50 mg, 16%, collapsed at 198—200°, $[\alpha]_D$ −31.2°), which proved to be identical with pyrethrosin in every respect.

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