Pergamon

Tetrahedron Vol. 50, No. 25, pp. 7385-7400, 1994

0040-4020/94 $7.00+0.00

0040-4020(94)E0354-V

Didemnilactones A and B and Neodidemnilactone, Three New Fatty Acid Metabolites Isolated from the Tunicate *Didemnum moseleyi* (Herdman)

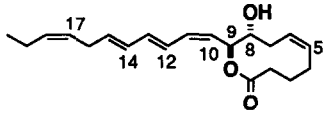
Haruki Niwa,* Masaru Watanabe, Hideaki Inagaki, and Kiyoyuki Yamada*

Department of Chemistry, Faculty of Science, Nagoya University, Chikusa, Nagoya 464, Japan

Abstract: Three new fatty acid metabolites didemnilactones A (1) and B (2) and neodidemnilactone (3) were isolated from the tunicate *Didemnum moseleyi* (Herdman). Their structures including absolute stereochemistry were established on the basis of spectral studies and chemical synthesis. Didemnilactones exhibited inhibitory activity against lipoxygenase and weak binding activity to leukotriene B_4 receptors.

INTRODUCTION

Tunicates have yielded a variety of structurally novel and pharmacologically interesting compounds.[1] In connection with our research on the isolation of biologically active marine natural products, we have examined the constituents of the colonial tunicate *Didemnum moseleyi* (Herdman) collected in Ago Bay, Mie Prefecture, Japan and isolated three new fatty acid metabolites didemnilactones A (1) and B (2) together with neodidemnilactone (3), all possessing a 10-membered lactone and a conjugated triene system as the common structural part. Described is a full account of the isolation of these fatty acid metabolites and their structure determination by means of spectral analysis and chemical synthesis.[2]



didemnilactone A (1)

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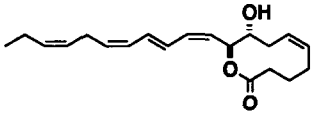
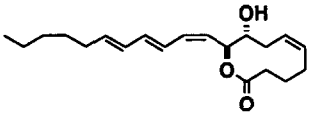
("didemnilactone")[2]



didemnilactone B (2)



neodidemnilactone (3)

RESULTS AND DISCUSSION

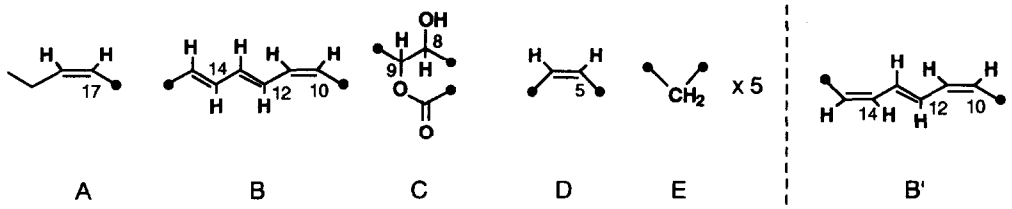
Isolation of Didemnilactones and Neodidemnilactone

The EtOAc-soluble material obtained from the MeOH extract of the tunicate *D. moseleyi* was subjected to repeated chromatography on silica gel and alumina followed by reversed-phase HPLC, affording

didemnilactones A (**1**) (1.1 x 10^{-5}% wet weight), B (**2**) (4.6 x 10^{-6}% wet weight), and neodidemnilactone (**3**) (1.8 x 10^{-6}% wet weight).

Structures of Didemnilactones and Neodidemnilactone

Didemnilactone A (1). The molecular formula of didemnilactone A (**1**), $C_{20}H_{28}O_3$, was determined from the ^{13}C NMR spectrum (Table **2**) and the high-resolution EIMS measurement [*m/z* 316.2048 (M^+), Δ 1.0 mmu]. The IR and ^{13}C NMR (Table **2**) spectra indicated that **1** has an OH group (ν_{max} 3570 and 3450) and a lactone (or ester) group [ν_{max} 1730 cm^{-1}; δ_C 172.5 (s, C–1)]. The detailed analysis of ^1H (Table **1**) and ^{13}C NMR spectral data indicated the presence of the following groups in **1**: 1 x $-CO_2CH-$, 5 x $-CH=CH-$, 1 x $-CHOH$, 6 x $-CH_2-$, 1 x $-CH_3$. The UV spectrum of **1** showed the absorptions at λ_{max} 261 (ε 24,600), 271 (30,500), and 279 nm (25,400), suggesting the presence of a conjugated triene system in **1**,[13] which was also implied from the proton signals [δ_H 5.22 (dd, *J* = 11.5, 9.1 Hz, H–10), 6.17 (dd, *J* = 11.5, 11.5 Hz, H–11), 7.03 (dd, *J* = 14.7, 11.5 Hz, H–12), 6.18 (dd, *J* = 14.7, 11.2 Hz, H–13), 6.02 (ddt, *J* = 14.7, 11.2, 1.0 Hz, H–14), and 5.57 (dt, *J* = 14.7, 6.6 Hz, H–15)] and the carbon signals [δ_C 127.0 (d, C–10), 134.9 (d, C–11), 126.7 (d, C–12), 136.5 (d, C–13), 131.2 (d, C–14), and 134.6 (d, C–15)]. The oxymethine proton at δ_H 3.65 (H–8) was coupled to the vicinal oxymethine proton at δ_H 5.75 (H–9) with a coupling constant of 9.1 Hz. The methyl protons at δ_H 0.88 (t, *J* = 7.6 Hz, H–20) were coupled to the methylene protons at δ_H 1.95 (m, H–19), which were in turn coupled to the olefinic proton at δ_H 5.41 (m) (H–18), indicating the existence of a vinyl ethyl moiety in **1**. The presence of a di-π-methane system in **1** was deduced from the methylene proton signal at δ_H 2.69 (td, *J* = 6.6, 1.0 Hz, H–16) and the carbon signal at δ_C 30.8 (t, C-16). Further detailed interpretation of the ^1H and ^{13}C NMR spectral data revealed the presence of partial structures A–E in **1**. The stereochemistry of the double bonds in the partial structures was assigned as 5*Z*, 10*Z*, 12*E*, 14*E*, and 17*Z* from the spin coupling constants of the corresponding olefinic protons [$J_{5,6}$ = 10.8 Hz, $J_{10,11}$ = 11.5 Hz, $J_{12,13}$ = $J_{14,15}$ = 14.7 Hz, and $J_{17,18}$ = ca. 11.5 Hz (determined by decoupling experiments)]. The partial structures A–E accounted for all carbons as well as protons in **1**. The extensive analysis of ^1H NMR, ^1H–^1H COSY and HMQC spectra of **1** defined the assignments of all carbons and protons, and the connectivity of these partial structures, elucidating the gross structure of didemnilactone A to be as depicted in the formula **1**. The presence of a 10-membered lactone in **1** was deduced from the chemical shifts of H–8 (δ_H 3.65) and H–9 (δ_H 5.75).



Didemnilactone B (2). The molecular formula of didemnilactone B (**2**), $C_{20}H_{28}O_3$, identical with that of **1** was determined from the ^{13}C NMR spectrum (Table **2**) in conjunction with the high-resolution EIMS measurement [*m/z* 316.2026 (M^+), Δ –1.2 mmu]. Comparison of the spectral properties of **2** with those of **1**

Table 1. ^{1}H NMR Spectral Data of Didemnilactones A (1) and B (2) and Neodidemnilactone (3)[a]

No.	1	2	3
2	1.85–2.10 (m, 2 H)[b]	1.85–2.04 (m, 2 H)[d]	1.85–2.10 (m, 2 H)[g]
3a	1.32 (m, 1 H)[c]	1.31 (m, 1 H)[e]	1.10–1.40 (m, 1 H)[h]
3b	1.63 (m, 1 H)	1.61 (m, 1 H)	1.63 (m, 1 H)
4	1.85–2.10 (m, 2 H)[b]	1.85–2.04 (m, 2 H)[d]	1.85–2.10 (m, 2 H)[g]
5	5.35–5.40 (m, 1 H)	5.18–5.43 (m, 1 H)[f]	5.37 (m, 1 H)
6	5.75 (m, 1 H)	5.74 (m, 1 H)	5.77 (m, 1 H)
7a	2.19 (m, 1 H)	2.19 (m, 1 H)	2.20 (m, 1 H)
7b	2.62 (m, 1 H)	2.61 (m, 1 H)	2.62 (m, 1 H)
8	3.65 (ddd, 1 H) (9.1, 3.9, 3.9)	3.56 (ddd 1 H) (9.6, 4.6, 4.6)	3.65 (ddd, 1 H) (9.1, 3.9, 3.9)
9	5.75 (dd, 1 H) (9.1, 9.1)	5.74 (dd, 1 H) (9.6, 9.6)	5.74 (dd, 1 H) (9.1, 9.1)
10	5.22 (dd, 1 H) (11.5, 9.1)	5.24 (dd, 1 H) (11.2, 9.6)	5.22 (dd, 1 H) (11.5, 9.1)
11	6.17 (dd, 1 H) (11.5, 11.5)	6.22 (dd, 1 H) (11.2, 11.2)	6.19 (dd, 1 H) (11.5, 11.5)
12	7.03 (dd, 1 H) (14.7, 11.5)	7.10 (dd, 1 H) (14.8, 11.5)	7.03 (dd, 1 H) (14.7, 11.5)
13	6.18 (dd, 1 H) (14.7, 11.2)	6.58 (dd, 1 H) (14.8, 11.2)	6.20 (dd, 1 H) (14.7, 11.2)
14	6.02 (ddt, 1 H) (14.7, 11.2, 1.0)	6.02 (dd, 1 H) (11.2, 11.2)	5.99 (ddt, 1 H) (14.7, 11.2, 1.0)
15	5.57 (dt, 1 H) (14.7, 6.6)	5.43 (dt, 1 H) (11.2, 6.6)	5.62 (dt, 1 H) (14.7, 6.6)
16	2.69 (td, 2 H) (6.6, 1.0)	2.85 (t, 2 H) (6.6)	1.85–2.10 (m, 2 H)[g]
17	5.36 (m, 1 H)	5.18–5.43 (m, 1 H)[f]	1.10–1.40 (m, 2 H)[h]
18	5.41 (m, 1 H)	5.18–5.43 (m, 1 H)[f]	1.10–1.40 (m, 2 H)[h]
19	1.95 (m, 2 H)	1.85–2.04 (m, 2 H)[d]	1.10–1.40 (m, 2 H)[h]
20	0.88 (t, 3 H) (7.6)	0.88 (t, 3 H) (7.6)	0.86 (t, 3 H) (7.6)
OH	1.32 (1 H)[c]	1.31 (1 H)[e]	1.10–1.40 (1 H)[h]

a) Spectra were taken in C_6D_6 at 270 MHz. Chemical shifts are in δ values from internal TMS and coupling constants (lower parentheses) in Hz.
b–h) Signals with identical superscripts are overlapped.

indicated that their structures were closely related and **2** was supposed to be a geometrical isomer of **1**. The presence of a conjugated triene system **B'** in **2** was implied from the proton signals [δ_H 5.24 (dd, J = 11.2, 9.6 Hz, H–10), 6.22 (dd, J = 11.2, 11.2 Hz, H–11), 7.10 (dd, J = 14.8, 11.5 Hz, H–12), 6.58 (dd, J = 14.8, 11.2 Hz, H–13), 6.02 (dd, J = 11.2, 11.2 Hz, H–14), and 5.43 (dt, J = 11.2, 6.6 Hz, H–15)]. The stereochemistry of the double bonds in the partial structure **B'** was assigned as 10*Z*, 12*E*, and 14*Z* from the spin coupling constants of the corresponding olefinic protons ($J_{10,11}$ = 11.2 Hz, $J_{12,13}$ = 14.8 Hz, and $J_{14,15}$ = 11.2 Hz). The intensive studies of ^{1}H NMR, ^{13}C NMR, ^{1}H–^{1}H COSY, and ^{1}H–^{13}C COSY spectra of **2** defined the assignments of all carbons and protons, and the connectivity of all carbons, elucidating the gross structure of didemnilactone B to be as depicted in the formula **2**. The stereochemistry of a double bond between C–17 and C–18 was assigned as *Z* from the spin coupling constant of the corresponding olefinic protons ($J_{17,18}$ =

Table 2. ^{13}C NMR Spectral Data of Didemnilactones A (**1**) B (**2**) and Neodidemnilactone C (**3**)[a]

No.	1	2	3
1	172.5 (s)	172.5 (s)	172.4 (s)
2	34.7 (t)	34.7 (t)	34.7 (t)
3	25.4 (t)	25.4 (t)	25.4 (t)
4	26.3 (t)	26.3 (t)	26.3 (t)
5	131.7 (d)	131.8 (d)	131.7 (d)
6	125.4 (d)	125.4 (d)	125.5 (d)
7	32.6 (t)	32.6 (t)	32.7 (t)
8	72.6 (d)	72.5 (d)	72.6 (d)
9	72.4 (d)	72.3 (d)	72.4 (d)
10	127.0 (d)	127.8 (d)	126.8 (d)
11	134.9 (d)	134.8 (d)	136.8 (d)
12	126.7 (d)	128.6 (d)	126.3 (d)
13	136.5 (d)	131.2 (d)	136.7 (d)
14	131.2 (d)	129.1 (d)	131.1 (d)
15	134.6 (d)	132.4 (d)	135.0 (d)
16	30.8 (t)	26.4 (t)	29.2 (t)
17	126.3 (d)	126.9 (d)	31.7 (t)
18	132.9 (d)	133.0 (d)	33.0 (t)
19	20.8 (t)	20.8 (t)	22.8 (t)
20	14.3 (q)	14.3 (q)	14.2 (q)

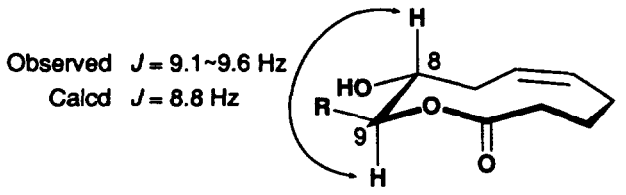
a) Spectra were taken in C_6D_6 at 67.8 MHz. Chemical shifts are in δ values from internal TMS and multiplicities in parentheses.

ca. 11.5 Hz) determined by decoupling experiments. The presence of a 10-membered lactone in **2** was deduced from the chemical shifts of H–8 (δ_H 3.56) and H–9 (δ_H 5.74).

Neodidemnilactone (3). Neodidemnilactone (**3**) has the molecular formula, $C_{20}H_{30}O_3$, which was determined from the ^{13}C NMR spectrum (Table 2) coupled with the high-resolution EIMS measurement [*m/z* 318.2191 (M^+), Δ –0.3 mmu]. Spectral properties of **3** suggested that the structure of **3** was very similar to that of **1**, and **3** was supposed to be a dihydro compound of **1**. The ^1H NMR spectrum indicated that neither vinyl ethyl nor di-π-methane groups were present in **3**, indicating **3** to be the 17,18-dihydro compound of **1**. The intensive studies of ^1H NMR, ^{13}C NMR, ^1H–^1H COSY, and ^1H–^{13}C COSY spectra of **3** defined the assignments of all carbon and proton signals, and the connectivity of all carbons, elucidating the gross structure of neodidemnilactone to be as depicted in the formula **3**. The presence of a 10-membered lactone in **2** was deduced from the chemical shifts of H–8 (δ_H 3.65) and H–9 (δ_H 5.74).

Relative Stereochemistry of Didemnilactones and Neodidemnilactone

The relative configurations at C–8 and C–9 in **1–3** were deduced to be 8*R**,9*S** (erythro configuration) on the basis of the coupling constants between H–8 and H–9 (**1**, $J_{8,9}$ = 9.1 Hz; **2**, $J_{8,9}$ = 9.6 Hz; **3**, $J_{8,9}$ = 9.1 Hz) coupled with conformational consideration using molecular models: the dihedral angles between H–8 and H–9 in (8*R**,9*S**)-**1**, **2**, and **3** are nearly 180° in a likely ring conformation **F**. Molecular mechanics calculations using Still's MacroModel (MULTIC and BATCHMIN) predicted the conformation **F** (R = vinyl) to be the global minimum.[3] The calculated coupling constant between H–8 and H–9 in the conformation **F** (R = vinyl) was 8.8 Hz, being consistent with the observed ones for **1**, **2**, and **3**.



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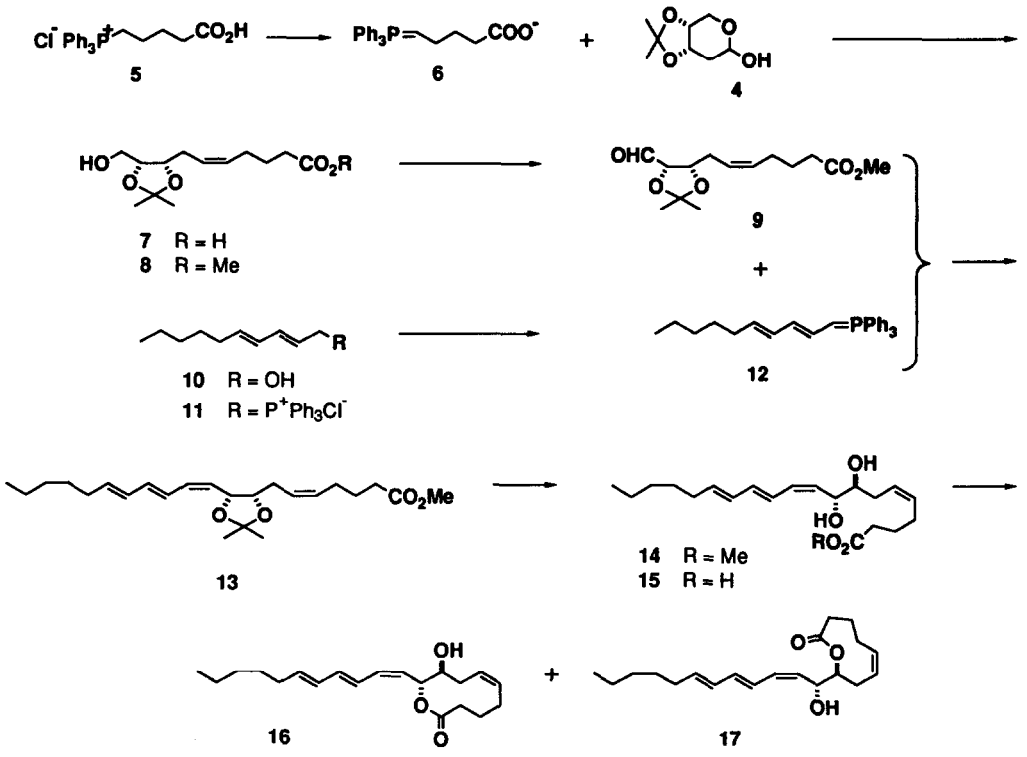
Synthesis of Didemnilactones and Neodidemnilactone.

In order to establish the structures of didemnilactones A and B and neodidemnilactone including the absolute stereochemistry, the unambiguous synthesis of the compounds having the structures **16**, **27**, and **34**, enantiomers of natural neodidemnilactone (**3**), didemnilactone A (**1**), and didemnilactone B (**2**), respectively, was performed.

The synthesis of (8*S*,9*R*)-neodidemnilactone (**16**) started with 3,4-*O*-isopropylidene-2-deoxy-D-ribose (**4**)[4] (Scheme 1). The Wittig reaction of **4** with ylide **6** generated from (4-carboxybutyl)triphenylphosphonium bromide (**5**) with $NaN(SiMe_3)_2$ in THF afforded stereoselectively (*Z*)-olefinic acid **7**, which was converted into methyl ester **8** with diazomethane (58% overall). Swern oxidation of **8** provided aldehyde **9** (88%). The next Wittig reaction required [(*E*,*E*)-(2,4-decadienyl)]triphenylphosphonium chloride (**11**),

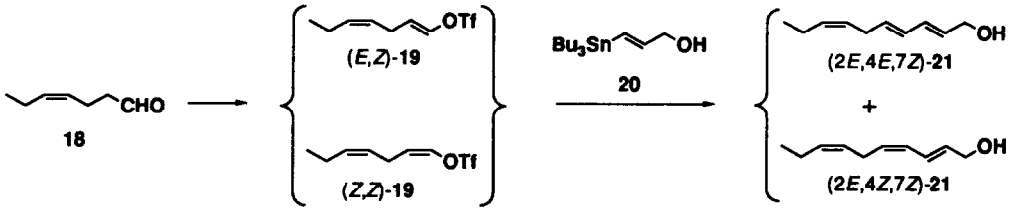
Scheme 1.



which was prepared in 65% yield from (*E*,*E*)-2,4-decadien-1-ol (**10**) by reaction with Ph_3P and HCl in MeOH.[5] Wittig reaction[6] of **9** with ylide **12** generated from **11** with $NaN(SiMe_3)_2$ in THF at –78 °C afforded stereoselectively (5*Z*,10*Z*,12*E*,14*E*)-tetraenoic acid methyl ester **13** in 85% yield. Acidic hydrolysis (AcOH–H_2O) of the acetonide group in **13** and subsequent basic hydrolysis (LiOH, MeOH–H_2O) of the ester group in the resulting dihydroxy ester **14** yielded seco acid **15** (65% overall). Lactonization of **15** by means of Yamaguchi's method[7] furnished dextrorotatory (8*S*,9*R*)-neodidemnilactone (**16**) [$[\alpha]_D^{18}$ +218° (*c* 0.054, MeOH)] in 53% yield together with the isomeric 9-membered lactone **17** (18%). Except for the sign of the specific rotation, spectral and chromatographic properties of synthetic **16** were identical with those of natural levorotatory neodidemnilactone (**3**) [$[\alpha]_D^{22}$ –200° (*c* 0.17, MeOH)] in all respects. Thus the absolute stereochemistry of natural neodidemnilactone (**3**) was established to be 8*R*,9*S*.

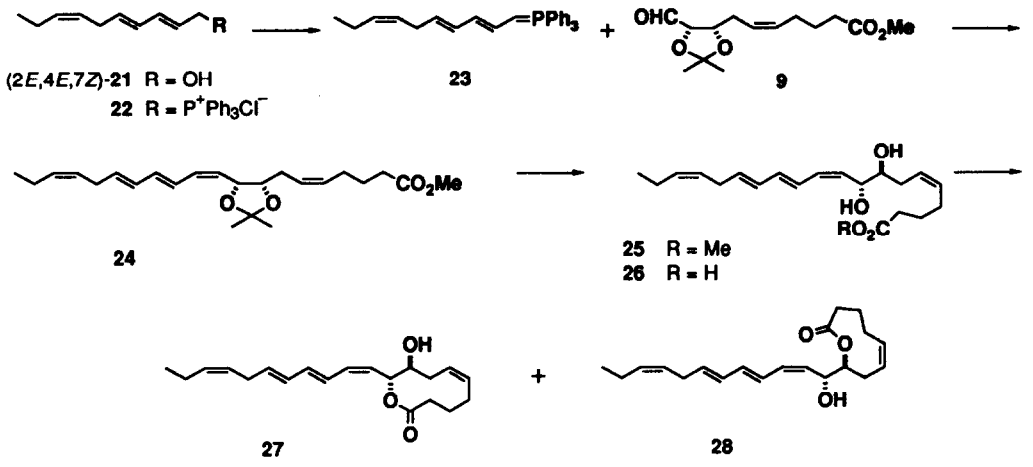
The synthesis of (8*S*,9*R*)-didemnilactone A (**27**) and (8*S*,9*R*)-didemnilactone B (**34**) required triene alcohols (2*E*,4*E*,7*Z*)-**21** and (2*E*,4*Z*,7*Z*)-**21**, respectively, which were prepared starting with (*Z*)-4-heptenal (**18**) (Scheme 2). Thus, the reaction of **18** with trifluoromethanesulfonic anhydride and 2,6-di-*t*-butyl-4-methylpyridine in 1,2-dichloroethane at reflux temperature[8] gave an inseparable 1:4 mixture of enol triflates (*E*,*Z*)-**19** and (*Z*,*Z*)-**19** in 57% combined yield. The Stille coupling reaction[9] of the 1:4 mixture of (*E*,*Z*)-**19** and (*Z*,*Z*)-**19** with [(*E*)-3-hydroxy-1-propenyl]tributyltin] (**20**)[10] was effected with $(PPh_3)_4Pd$ and LiCl in THF, providing the desired triene alcohols (2*E*,4*E*,7*Z*)-**21** (11%) and (2*E*,4*Z*,7*Z*)-**21** (47%) after chromatographic separation.

Scheme 2.



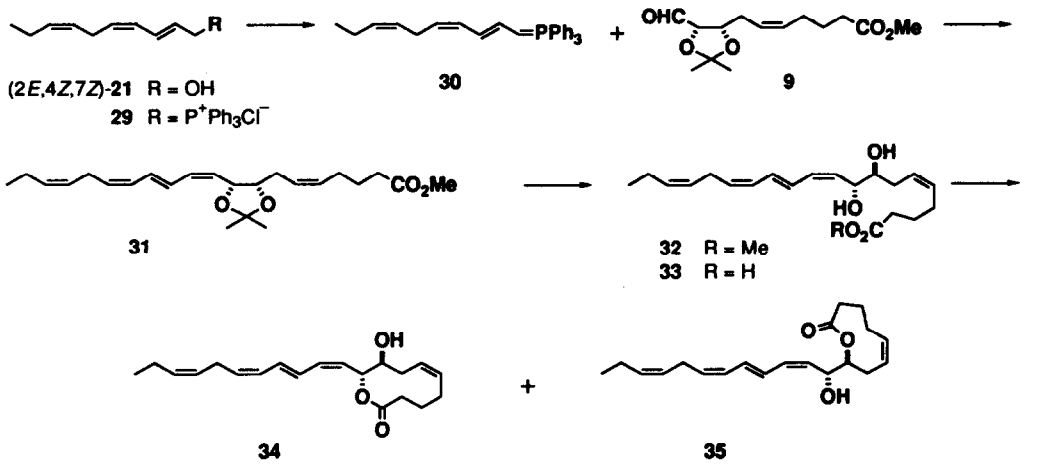
The synthesis of (8*S*,9*R*)-**27** started with (2*E*,4*E*,7*Z*)-**21** (Scheme 3). Thus, (2*E*,4*E*,7*Z*)-**21** was converted into phosphonium salt **22** (68% overall) by reaction with *N*-chlorosuccinimide and dimethyl sulfide[11] in CH_2Cl_2 followed by treatment of the resulting chloride with triphenylphosphine in acetonitrile. Wittig reaction[6] of **9** with ylide **23** generated from **22** with $NaN(SiMe_3)_2$ in THF at –78 °C afforded stereoselectively (5*Z*,10*Z*,12*E*,14*E*,17*Z*)-pentaenoic acid methyl ester **24** in 73% yield. Acidic hydrolysis (AcOH–H_2O) of the acetonide group in **24** and subsequent basic hydrolysis (LiOH, MeOH–H_2O) of the ester group in the resulting dihydroxy ester **25** yielded seco acid **26** (64% overall). Lactonization of **26** by means of Yamaguchi's method[7] furnished dextrorotatory (8*S*,9*R*)-didemnilactone A (**27**) [$[\alpha]_D^{22}$ +193° (*c* 0.180, MeOH)] in 42% yield together with the isomeric 9-memebered lactone **28** (17%). Except for the sign of the specific rotation, spectral and chromatographic properties of synthetic **27** were identical with those of natural levorotatory didemnilactone A (**1**) [$[\alpha]_D^{22}$ –190° (*c* 0.18, MeOH)] in all respects. Thus the absolute stereochemistry of natural didemnilactone A (**1**) was established to be 8*R*,9*S*.

Scheme 3.



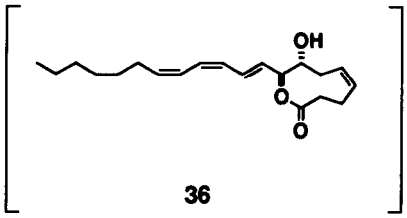
According to the same reaction sequence for the synthesis of (8*S*,9*R*)-**27** from (2*E*,4*E*,7*Z*)-**21**, the dextrorotatory (8*S*,9*R*)-didemnilactone B (**34**) [$[\alpha]_D^{26}$ +281° (*c* 0.280, MeOH)] was synthesized starting with (2*E*,4*Z*,7*Z*)-**21** and **9** (Scheme 4). Except for the sign of the specific rotation, spectral and chromatographic properties of synthetic **34** were identical with those of natural levorotatory didemnilactone B (**2**) [$[\alpha]_D^{22}$ +378° (*c* 0.005, MeOH)][12] in all respects. Thus, the absolute stereochemistry of natural didemnilactone B (**2**) was established to be 8*R*,9*S*.

Scheme 4.



CONCLUSION

Didemnilactones A (**1**) and B (**2**) and neodidemnilactone (**3**) are unsaturated fatty acid metabolites possessing a 10-membered lactone and a conjugated triene. Although a number of fatty acid metabolites have been isolated from various marine organisms, [1] the occurrence of the metabolites having a 10-membered lactone is rather rare. Previously, there was reported the isolation of a series of the related compounds named ascidiatrienolides having a 9-membered lactone and a conjugated triene, from the tunicate *Didemnum candidum*.[13] Ascidiatrienolide A is a member of the ascidiatrienolides reported and the structure was originally assigned as depicted in the formula **36**. Recently, Holmes and co-workers have synthesized the compound having structure **36**.[14] However, the spectroscopic and chromatographic properties of synthetic **36** was significantly different from natural ascidiatrienolide A. Subsequently, on the basis of unambiguous synthesis, Holmes and co-workers revised the structure of ascidiatrienolide A to be as depicted in the formula **37**.[14] Thus, ascidiatrienolide A (**37**) is a geometrical isomer of neodidemnilactone (**3**) with the same absolute stereochemistry.



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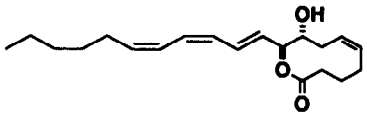

ascidiatrienolide A (**37**)

Didemnilactones exhibited moderate inhibitory activities against 5-lipoxygenase and 15-lipoxygenase of human polymorphonuclear leukocytes (Table 3). Didemnilactone A (**1**) and neodidemnilactone (**3**) showed weak binding activity to leukotriene B_4 receptors of human polymorphonuclear leukocyte membrane fractions. IC_{50} values of **1** and **3** were as follows; **1**, 38 μM; **3**, 50 μM.

Table 3. Inhibitory Activities of Didemnilactones and the Related Compounds against 5-Lipoxygenase and 15-Lipoxygenase of Human PMNL

Compound	IC_{50} (μM)	
	5-lipoxygenase	15-lipoxygenase
didemnilactone A (**1**)	9.4	41
didemnilactone B (**2**)	8.5	*
(8*S*,9*R*)-didemnilactone A (**16**)	2.9	4.9
(8*S*,9*R*)-didemnilactone B (**27**)	6.1	12
(8*S*,9*R*)-neodidemnilactone (**34**)	>10	>10

* Activity was not measured.

EXPERIMENTAL

IR spectra were taken on a JASCO IR-810 spectrophotometer. ^{1}H NMR spectra were recorded on either JEOL JNM EX-270 (270 MHz) or JEOL JNM-C675 (270 MHz) spectrometer : Chemical shifts (δ) are reported in ppm downfield from internal tetramethylsilane, and coupling constants in Hz. ^{13}C NMR spectra were recorded on either JEOL JNM EX-270 (67.8 MHz) or JEOL JNM-C675 (67.8 MHz) spectrometer in C_6D_6: Chemical shifts (δ) are reported in ppm downfield from internal tetramethylsilane, and coupling constants in Hz. Low-resolution (EIMS and CIMS) and high-resolution mass spectra (HREIMS) were measured on a JEOL JMS-LG2000 instrument. Fuji-Davison silica gel BW-820MH and Merck aluminum oxide 90 (activity II–III, Art. 1097) (alumina) were used for column chromatography. Merck precoated silica gel 60 F_{254} plates, 0.25 mm thickness were used for analytical thin layer chromatography (TLC) and for preparative TLC. Tetrahydrofuran (THF) was distilled from sodium-benzophenone ketyl under nitrogen. Toluene was distilled from sodium under nitrogen. Dichloromethane (CH_2Cl_2), acetonitrile, pyridine, and triethylamine (Et_3N) were distilled from calcium hydride (CaH_2) under nitrogen. Dimethyl sulfoxide was distilled from CaH_2 under reduced pressure. Unless otherwise stated, the organic solutions obtained by extractive workup were washed with saturated aqueous NaCl solution, dried over anhydrous sodium sulfate (Na_2SO_4) and concentrated under reduced pressure by a rotary evaporator.

Extraction and Isolation Procedure. Specimens of *Didemnum moseleyi* (Herdman) were collected in Ago Bay, Mie Prefecture, Japan in May 1993. Specimens (wet weight 10.9 kg) were homogenized in MeOH using a blender. The homogenized specimens were soaked in MeOH (20 l) at room temperature for 2 days. The mixture was filtered with suction. The filtration residue was washed with MeOH (5 l). The combined filtrate and washings were concentrated under reduced pressure to a volume of 0.8 l. The resulting aqueous mixture was extracted with EtOAc (4 x 0.9 l). The combined EtOAc extracts were dried over anhydrous Na_2SO_4 and concentrated under reduced pressure to give a dark brown oil (14.5 g), which was subjected to column chromatography on silica gel (260 g) using benzene (1 l), 4:1 benzene–EtOAc (1 l), and MeOH (1 l) as eluent. The fraction **A** (4.3 g) eluted with 4:1 benzene–EtOAc was further chromatographed on alumina (170 g) with MeOH (500 ml) and 10:1 MeOH–AcOH (500 ml) as eluent. The fraction **B** (1.7 g) eluted with MeOH was suspended in a small amount of MeOH (3 ml) and insoluble materials were removed by filtration to give the MeOH-soluble fraction **C** (1.3 g), which was further separated by column chromatography on silica gel (130 g) using benzene (3.3 l), 30:1 benzene–EtOAc (500 ml), and EtOAc (520 ml) as eluent. The fraction **D** (346 mg) eluted with 30:1 benzene–EtOAc was dissolved in CH_3CN. The solution was loaded on a sample pretreatment cartridge TOYOPAK ODS-M and the cartridge was eluted with CH_3CN (ca. 5 ml). The fraction **E** (127 mg) eluted with CH_3CN was further separated by preparative TLC on silica gel [(200 mm x 200 mm x 0.25 mm) x 5 plates, 3:1 hexane–EtOAc] to give the fraction **F** (35 mg) containing didemnilactones A (**1**) and B (**2**) and neodidemnilactone (**3**). Further separation of the fraction **F** by repeated HPLC [Develosil ODS-HG-5 (250 mm x 10 mm i.d.), 75:25 CH_3CN–H_2O, flow rate 2 ml/min, UV detection at 254 nm] provided didemnilactone A (**1**) (1.2 mg, 1.1 x 10^{-5}% wet weight), didemnilactone B (**2**) (0.5 mg, 4.6 x 10^{-6}% wet weight), and neodidemnilactone (**3**) (0.2 mg, 1.8 x 10^{-6}% wet weight).

Didemnilactone A (1): $C_{20}H_{28}O_3$; a colorless oil; $[\alpha]_D^{22}$ –190° (*c* 0.18, MeOH); UV (MeOH) λ_{max} 261 (ε 24,600), 271 (30,500), and 279 nm (25,400); IR ($CHCl_3$) 3570, 3450 (br), 1730, 1440, 1140, and 990 cm^{-1}; ^{1}H NMR, Table **1**; ^{13}C NMR, Table **2**; EIMS *m/z* (relative intensity) 316 (M^+, 40), 298 (18), and 160 (100); HREIMS calcd for $C_{20}H_{28}O_3$ (M^+) 316.2038, found 316.2048.

Didemnilactone B (2): $C_{20}H_{28}O_3$; a colorless oil; $[\alpha]_D^{25}$ –378° (*c* 0.005, MeOH);[12] UV (MeOH) λ_{max} 267 (ε 36,400), 274 (44,900), and 284 nm (36,100); IR ($CHCl_3$) 3600, 3400 (br), 1730, 1440, 1140, and 990 cm^{-1};

^{1}H NMR, Table **1**; ^{13}C NMR, Table **2**; EIMS *m/z* (relative intensity) 316 (M^+, 100), 298 (21), 279 (10), and 160 (95); HREIMS calcd for $C_{20}H_{28}O_3$ (M^+) 316.2038, found 316.2026.

Neodidemnilactone (3): $C_{20}H_{30}O_3$; a colorless oil; $[\alpha]_D^{22}$ –200° (*c* 0.17, MeOH); UV (MeOH) λ_{max} 261 (ε 22,900), 271 (29,100), and 279 nm (24,000); IR ($CHCl_3$) 3570, 3450 (br), 1725, 1440, 1350, 1140, and 990 cm^{-1}; ^{1}H NMR, Table **1**; ^{13}C NMR, Table **2**; EIMS *m/z* (relative intensity) 318 (M^+, 49), 300 (12), 179 (33), and 162 (100); HREIMS calcd for $C_{20}H_{30}O_3$ (M^+) 318.2194, found 318.2191.

Methyl (8*S*,9*R*,5*Z*)-10-Hydroxy-8,9-isopropylidenedioxy-5-decenoate (8). To a stirred suspension of 4-(carboxybutyl)triphenylphosphonium bromide (**5**) (6.79 g, 15.3 mmol, dried in vacuo at 80 °C for 1 h just prior to use) in THF (90 ml) under nitrogen was added dropwise 1.0 M $NaN(SiMe_3)_2$ in THF (30 ml, 30 mmol), and the mixture was stirred at room temperature for 30 min to give an orange solution of ylide **6**. The ylide solution was cooled to –78 °C, and a solution of 3,4-*O*-isopropylidene-2-deoxy-D-ribose (**4**)[4] (748 mg, 4.30 mmol) in THF (3 ml) was added dropwise. The reaction mixture was stirred at –78 °C for 30 min and then at room temperature for 2 h. The reaction was quenched by addition of H_2O (90 ml). The aqueous mixture was washed with ether (100 ml). The aqueous layer was acidified to pH 3 with 6 M HCl and extracted with EtOAc (5 x 100 ml). The combined extracts were concentrated under reduced pressure to give crude acid **7**, which was dissolved in ether (30 ml). To the ethereal solution of crude **7** was added an ethereal solution of diazomethane until the yellow color persisted. The reaction mixture was concentrated under reduced pressure to give an oily residue, which was purified by column chromatography on silica gel (200 g, 1:1→1:2 hexane–ether), affording **8** (677 mg, 58% overall) as a colorless oil: $[\alpha]_D^{26}$ +7.70° (*c* 1.74, $CHCl_3$); IR ($CHCl_3$) 3590, 3500 (br), 1730, 1420, 1380, 1370, 1160, and 1030 cm^{-1}; ^{1}H NMR (270 MHz, $CDCl_3$) δ 1.37 (s, 3 H), 1.48 (s, 3 H), 1.70 (br s, 1 H, OH), 1.71 (tt, *J* = 7.3, 7.3 Hz, 2 H), 2.10 (m, 2 H), 2.30 (t, *J* = 7.3 Hz, 2 H), 2.31 (m, 2 H), 3.64 (m, 2 H), 3.67 (s, 3 H), 4.14–4.26 (m, 2 H), and 5.39–5.56 (m, 2 H); EIMS *m/z* (relative intensity) 272 (M^+, 6), 257 (89), 241 (71), and 113 (100); HRCIMS calcd for $C_{14}H_{25}O_5$ [$(M+H)^+$] 273.1702, found 273.1690.

Methyl (8*S*,9*S*,5*Z*)-10-Oxo-8,9-isopropylidenedioxy-5-decenoate (9). To a cooled (–78 °C), stirred solution of oxalyl chloride (0.023 ml, 0.26 mmol) in CH_2Cl_2 (1 ml) under nitrogen was added dimethyl sulfoxide (0.025 ml, 0.35 mmol). After the mixture was stirred at –78 °C for 3 min, a solution of **8** (47.1 mg, 0.17 mmol) in CH_2Cl_2 (0.8 ml) was added dropwise. After the mixture was stirred at –78 °C for 15 min, Et_3N (0.12 ml, 0.86 mmol) was added. The mixture was warmed to 0 °C and stirred for an additional 15 min at 0 °C. The reaction was quenched by addition of saturated NH_4Cl solution (3 ml), and the aqueous mixture was extracted with CH_2Cl_2 (3 x 4 ml). The organic layers were combined, washed, dried, and concentrated under reduced pressure. The oily residue was purified by column chromatography on silica gel (4 g, 1:1 hexane–ether), affording **9** (41.1 mg, 88%) as a colorless oil: $[\alpha]_D^{27}$ –9.55° (*c* 1.04, $CHCl_3$); IR ($CHCl_3$) 1730, 1420, 1380, 1370, 1220, 1160, and 1060 cm^{-1}; ^{1}H NMR (270 MHz, $CDCl_3$) δ 1.41 (s, 3 H), 1.60 (s, 3 H), 1.70 (tt, *J* = 7.3, 7.3 Hz, 2 H), 2.06 (td, *J* = 7.3, 6.3 Hz, 2 H), 2.31 (m, 2 H), 2.32 (t, *J* = 7.3 Hz, 2 H), 3.67 (s, 3 H), 4.31 (dd, *J* = 7.3, 3.0 Hz, 1 H), 4.39 (dt, *J* = 7.3, 7.3 Hz, 1 H), 5.45 (m, 1 H), 5.51 (m, 1 H), and 9.67 (d, *J* = 3.0 Hz, 1 H); EIMS *m/z* (relative intensity) 270 (M^+, 6), 255 (89), 241 (49), and 101 (100); HRCIMS calcd for $C_{14}H_{23}O_5$ [$(M+H)^+$] 271.1546, found 271.1570.

[(*E*,*E*)-2,4-Decadienyl]triphenylphosphonium Chloride (11). Triphenylphosphine (4.80 g, 18.3 mmol) was dissolved in 1.04 M dry HCl in MeOH (18 ml) under nitrogen. To the solution was added dropwise a solution of (*E*,*E*)-decadien-1-ol (**10**) (2.83 g, 18.3 mmol) in MeOH (6 ml). The mixture was stirred at room temperature for 39 h and concentrated under reduced pressure. The oily residue was purified by column

chromatography on silica gel (70 g, 4:1→0:1 EtOAc–MeOH), affording **11** (5.15 g, 65%) as a colorless resin: ^{1}H NMR (270 MHz, CD_3OD) δ 0.89 (t, *J* = 6.6 Hz, 3 H), 1.20–1.55 (m, 6 H), 2.05 (m, 2 H), 4.34 (dd, *J* = 15.8, 7.3 Hz, 2 H), 5.45 (m, 1 H), 5.66 (dtd, *J* = 14.8, 7.3, 2.6 Hz, 1 H), 6.01 (dd, *J* = 14.8, 10.4 Hz, 1 H), and 6.22 (ddd, *J* = 14.8, 10.4, 5.3 Hz, 1 H). This material was azeotropically dried several times with toluene just prior to use for the next Wittig reaction.

Methyl (8*S*,9*R*,5*Z*,10*Z*,12*E*,14*E*)-8,9-Isopropylidenedioxy-5,10,12,14-eicosatetraenoate (13). To a stirred suspension of the azeotropically dried **11** (217 mg, 0.50 mmol) in THF (3.5 ml) under nitrogen was added dropwise 1.0 M $NaN(SiMe_3)_2$ in THF (0.5 ml, 0.5 mmol), and the mixture was stirred at room temperature for 15 min to give an orange solution of ylide **12**. To the cooled (–78 °C), stirred solution of **12** was added dropwise a solution of **9** (64.3 mg, 0.238 mmol) in THF (3 ml). After the reaction mixture was stirred at –78 °C for 1 h, the reaction was quenched by addition of saturated NH_4Cl solution (6 ml). The aqueous mixture was extracted with EtOAc (5 x 6 ml). The combined extracts were washed, dried, and concentrated under reduced pressure. The oily residue was purified by column chromatography on silica gel (14 g, 15:1 hexane–EtOAc), affording **13** (79.4 mg, 85%) as a colorless oil: $[\alpha]_D^{26}$ –76.6° (*c* 0.851, $CHCl_3$); UV (MeOH) λ_{max} 262 (ε 28800), 271 (33900), and 281 nm (29100); IR ($CHCl_3$) 1730, 1440, 1380, 1365, 1160, and 990 cm^{-1}; ^{1}H NMR (270 MHz, $CDCl_3$) δ 0.89 (t, *J* = 6.6 Hz, 3 H), 1.24–1.44 (m, 6 H), 1.38 (s, 3 H), 1.49 (s, 3 H), 1.68 (tt, *J* = 7.3, 7.3 Hz, 2 H), 2.01–2.31 (m, 6 H), 2.30 (t, *J* = 7.3 Hz, 2 H), 3.66 (s, 3 H), 4.12 (dt, *J* = 9.9, 5.9 Hz, 1 H), 5.05 (dd, *J* = 9.9, 5.9 Hz, 1 H), 5.36–5.48 (m, 3 H), 5.78 (dt, *J* = 14.2, 7.3 Hz, 1 H), 6.20 (t, *J* = 9.9 Hz, 1 H), and 6.05–6.41 (m, 3 H); EIMS *m/z* (relative intensity) 390 (M^+, 100), 375 (15), 332 (80), 315 (84), and 220 (96); HREIMS calcd for $C_{23}H_{35}O_4$ [$(M–Me)^+$] 375.2535, found 375.2513.

Methyl (8*S*,9*R*,5*Z*,10*Z*,12*E*,14*E*)-8,9-Dihydroxy-5,10,12,14-eicosatetraenoate (14). A solution of **13** (17.1 mg, 0.044 mmol) in degassed 4:1 AcOH–H_2O (1.6 ml) under argon was stirred at room temperature for 4 h and concentrated under reduced pressure. The oily residue was purified by column chromatography on silica gel (4 g, 3:1 hexane–EtOAc), affording **14** (10.2 mg, 66%) as a colorless oil: $[\alpha]_D^{26.5}$ –32.9° (*c* 0.377, MeOH); UV (MeOH) λ_{max} 261 (ε 37500), 270 (45500), and 280 nm (37000); IR ($CHCl_3$) 3560, 3440 (br), 1730, 1480, 1440, 1370, 1150, and 1000 cm^{-1}; ^{1}H NMR (270 MHz, C_6D_6) δ 0.87 (t, *J* = 6.3 Hz, 3 H), 1.15–1.35 (m, 6 H), 1.56 (tt, *J* = 7.3, 7.3 Hz, 2 H), 1.74 (br.s, 1 H, OH), 1.90–2.10 (m, 6 H), 2.23 (m, 1 H), 2.33 (m, 1 H), 3.34 (m, 3 H), 3.63 (ddd, J = 8.6, 4.3, 4.3 Hz, 1 H), 4.51 (dd, *J* = 8.6, 4.3 Hz), 5.30–5.57 (m, 3 H), 5.63 (dt, *J* = 14.2, 7.3 Hz, 1 H), 6.07 (m, 1 H), 6.12 (dd, *J* = 10.9, 10.9 Hz, 1 H), 6.18 (dd, *J* = 14.2, 10.9 Hz, 1 H), and 6.47 (dd, *J* = 14.4, 10.9 Hz, 1 H); EIMS *m/z* (relative intensity) 350 (M^+, 6), 332 (3), 316 (6), 301 (4), and 171 (100); HREIMS calcd for $C_{21}H_{34}O_4$ (M^+) 350.2457, found 350.2482.

(8*S*,9*R*,5*Z*,10*Z*,12*E*,14*E*)-8,9-Dihydroxy-5,10,12,14-eicosatetraenoic Acid (15). To a solution of **14** (8.6 mg, 0.025 mmol) in degassed 3:1 MeOH–H_2O (3 ml) under argon was added LiOH (10.3 mg). The mixture was stirred at room temperature for 22 h and concentrated under reduced pressure. The residue was diluted with saturated NaCl solution (4 ml). The aqueous mixture was acidified to pH 4 with 1 M HCl and extracted with EtOAc (3 x 5 ml). The combined extracts were washed, dried, and concentrated under reduced pressure. The oily residue was purified by column chromatography on silica gel (1 g, 2:1 EtOAc–hexane), affording **15** (8.2 mg, 99%) as a colorless oil : $[\alpha]_D^{19}$ –23.4° (*c* 0.20, MeOH); UV (MeOH) λ_{max} 261 (ε 27500), 270 (32800), and 280 nm (27300); IR ($CHCl_3$) 3600–2400, 1700, 1460, 1410, and 990 cm^{-1}; ^{1}H NMR (270 MHz, C_6D_6) δ 0.88 (t, *J* = 6.3 Hz, 3 H), 1.15–1.40 (m, 6 H), 1.53 (tt, *J* = 7.3, 7.3 Hz, 2 H), 1.87–2.35 (m, 8 H), 2.24 (m, 1 H), 2.41 (m, 1 H), 3.76 (ddd, J = 8.6, 4.3, 4.3 Hz, 1 H), 4.67 (dd, *J* = 8.6, 4.3 Hz), 5.37–5.60 (m, 3 H), 5.67 (dt, *J* = 14.5, 7.3 Hz, 1 H), 6.14 (dd, *J* = 14.5, 10.2 Hz, 1 H), 6.21 (m, 1 H), 6.25 (dd, *J* = 14.5, 10.2 Hz, 1

H), and 6.55 (dd, J = 14.2, 10.2 Hz, 1 H); EIMS *m/z* (relative intensity) 336 (M^+, 6), 318 (2), 278 (100), and 256 (12); HREIMS calcd for $C_{20}H_{32}O_4$ (M^+) 336.2301, found 336.2272.

(8*S*,9*R*)-Neodidemnilactone (16). To a solution of **15** (8.2 mg, 0.024 mmol) in THF (1.2 ml) under nitrogen were added a solution of Et_3N (2.7 mg, 0.024 mmol) in THF (0.07 ml) and a solution of 2,4,6-trichlorobenzoyl chloride (6.0 mg, 0.024 mmol) in THF (0.07 ml). The mixture was stirred at room temperature for 3 h and then diluted with toluene (12 ml). The diluted mixture was added dropwise over a 1.3-h period to a refluxing toluene (2.5 ml) solution containing 4-(dimethylamino)pyridine (59.6 mg, 0.48 mmol) under nitrogen. After the addition, the mixture was cooled to room temperature and diluted with ether (20 ml). The diluted mixture was successively washed with 1 M HCl (2 ml), saturated $NaHCO_3$ solution (2 ml), and saturated NaCl solution (2 ml), dried, and concentrated under reduced pressure. The oily residue was purified by column chromatography on silica gel (3 g, 50:1→20:1 benzene–EtOAc), affording desired **16** (4.1 mg, 53%) and the isomer **17** (1.4 mg, 18%), as a colorless oil, respectively.
16: $[\alpha]_D^{18}$ +218° (*c* 0.054, MeOH); HREIMS calcd for $C_{20}H_{30}O_3$ (M^+) 318.2194, found 318.2197. The spectral and chromatographic properties of synthetic **16** were identical with those of natural **3** in all respects except for the sign of the specific rotation.
17: IR ($CHCl_3$) 3600, 3450 (br), 1730, 1480, 1350, 1260, 1140, and 990 cm^{-1}; ^1H NMR (270 MHz, C_6D_6) δ 0.87 (t, J = 6.6 Hz, 3 H), 1.21–1.55 (m, 8 H), 1.72 (m, 1 H), 1.89–2.09 (m, 6 H), 2.35 (m, 2 H), 4.50 (m, 1 H), 4.93 (ddd, J = 10.9, 5.3, 1.3 Hz, 1 H), 5.31–5.45 (m, 1 H), 5.64 (dt, J = 14.2, 6.9 Hz, 1 H), 6.04–6.10 (m, 1 H), 6.18 (dd, J = 13.2, 10.9 Hz), and 6.46 (dd, J = 13.2, 11.9 Hz, 1 H); EIMS *m/z* (relative intensity) 318 (M^+, 98), 300 (10), 278 (12), 216 (18), and 192 (100); HREIMS calcd for $C_{20}H_{30}O_3$ (M^+) 318.2194, found 318.2194.

(1*E*,4*Z*)-1,4-Heptadienyl Trifluoromethanesulfonate [(*E*,*Z*)-19] and (1*Z*,4*Z*)-1,4-Heptadienyl Trifluoromethanesulfonate [(*Z*,*Z*)-19]. To a stirred solution of (*Z*)-4-heptenal (**18**) (2.06 g, 18.4 mmol) in 1,2-dichloroethane (40 ml) under nitrogen was added a solution of 2,6-di-*t*-butyl-4-methylpyridine (4.15 g, 20.2 mmol) in 1,2-dichloroethane (10 ml) and subsequently trifluoromethanesulfonic anhydride (3.40 ml, 20.2 mmol) was added. The mixture was heated under reflux for 2 h, cooled to room temperature, and diluted with pentane (150 ml). The mixture was washed successively with 1 M HCl (500 ml) and saturated $NaHCO_3$ (50 ml). The organic layer was dried and concentrated under reduced pressure. The oily residue was purified by column chromatography on silica gel (90 g, hexane), affording an inseparable 1:4 mixture of (*E*,*Z*)-**19** and (*Z*,*Z*)-**19** (2.57 g, 57%) as a colorless oil: IR ($CHCl_3$) 1420, 1140, 1220, 1020, and 960 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) for (*E*,*Z*)-**19**, δ 0.98 (t, J = 7.6 Hz, 3 H), 2.07 (qt, J = 7.6, 5.6 Hz, 2 H), 2.80 (dd, J = 7.1, 6.9 Hz, 2 H), 5.27 (m, 1 H), 5.49 (dt, J = 10.7, 5.6 Hz, 1 H), 5.78 (dt, J = 11.9, 7.1 Hz, 1 H), 6.51 (d, J = 11.9 Hz, 1 H); ^1H NMR (270 MHz, $CDCl_3$) for (*Z*,*Z*)-**19**, δ 0.98 (t, J = 7.6 Hz, 3 H), 2.07 (qt, J = 7.6, 5.6 Hz, 2 H), 2.94 (dd, J = 7.4, 7.4 Hz, 2 H), 5.24 (dt, J = 5.3, 7.4 Hz, 1 H), 5.27 (m, 1 H), 5.49 (dt, J = 10.7, 5.6 Hz, 1 H), and 6.54 (d, J = 5.3 Hz, 1 H); EIMS *m/z* (relative intensity) 244 (M^+, 10), 190 (100), and 111 (5); HREIMS calcd for $C_8H_{11}O_3F_3S$ (M^+) 244.0381, found 244.0378.

(2*E*,4*E*,7*Z*)-2,4,7-Decatrien-1-ol [(2*E*,4*E*,7*Z*)-21] and (2*E*,4*Z*,7*Z*)-2,4,7-Decatrien-1-ol [(2*E*,4*Z*,7*Z*)-21]. To a mixture of tetrakis(triphenylphosphine)palladium(0) (48.7 mg, 0.04 mmol) and LiCl (179 mg, 4.22 mmol) in THF (3 ml) under nitrogen were added dropwise a solution of the 1:4 mixture of (*E*,*Z*)-**19** and (*Z*,*Z*)-**19** (343 mg, 1.41 mmol) in THF (5 ml) and a solution of [(*E*)-3-hydroxy-1-propenyl]tributyltin **10** (585 mg, 1.69 mmol) in THF (5 ml). The mixture was heated under reflux for 3 h. After cooling, the reaction mixture was diluted with hexane (15 ml) and washed successively with saturated $NaHCO_3$ solution (15 ml), 10%

aqueous KF solution (5 ml), and saturated NaCl (5 ml) solution. The organic layer was dried and concentrated under reduced pressure. The oily residue was purified by medium pressure liquid chromatography [Micro Bead Silica Gel B–(30–70) μ, 250 mm x 20 mm i.d., 2:1 hexane–ether, flow rate 15 ml/min, UV detection 230 nm], affording (2*E*,4*E*,7*Z*)-**21** (23.1 mg, 11%) and (2*E*,4*Z*, 7*Z*)-**21** (100 mg, 47%) as a colorless oil, respectively.

(2*E*,4*E*,7*Z*)-**21**: IR ($CHCl_3$) 3600, 1460, 1380, 1080, and 990 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 0.97 (t, *J* = 7.6 Hz, 3 H), 2.05 (qd, *J* = 7.6, 7.3 Hz, 2 H), 2.83 (m, 2 H), 4.16 (d, *J* = 5.4 Hz, 2 H), 5.35 (dt, *J* = 10.8, 6.9 Hz, 1 H), 5.47 (dt, *J* = 10.8, 7.3 Hz, 1 H), 5.70 (dt, *J* = 14.4, 6.9 Hz, 1 H), 5.75 (dt, *J* = 14.4, 5.4 Hz, 1 H), 6.07 (dd, *J* = 14.4, 10.8 Hz, 1 H), and 6.25 (dd, *J* = 14.4, 10.8 Hz, 1 H); EIMS *m/z* (relative intensity) 152 (M^+, 3), 134 (15), 121 (42), and 79 (100); HREIMS calcd for $C_{10}H_{14}$ [$(M–H_2O)^+$] 134.1096, found 134.1078.

(2*E*,4*Z*,7*Z*)-(**21**): IR ($CHCl_3$) 3600, 1460, 1380, 1080, and 980 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 0.98 (t, *J* = 7.6 Hz, 3 H), 2.10 (qd, *J* = 7.6, 7.3 Hz, 2 H), 2.94 (m, 2 H), 4.22 (d, *J* = 5.4 Hz, 2 H), 5.32 (dt, *J* = 10.8, 7.2 Hz, 1 H), 5.44 (m, 2 H), 5.85 (dt, *J* = 15.3, 5.4 Hz, 1 H), 6.03 (dd, *J* = 10.8, 10.8 Hz, 1 H), and 6.56 (dd, *J* = 15.3, 10.8 Hz, 1 H); EIMS *m/z* (relative intensity) 152 (M^+, 51), 134 (10), 121 (28), and 79 (100); HREIMS calcd for $C_{10}H_{14}$ [$(M–H_2O)^+$] 134.1096, found 134.1089.

[(2*E*,4*E*,7*Z*)-2,4,7-Decatrienyl]triphenylphosphnium Chloride (22). To a cooled (–48 °C), stirred solution of *N*-chlorosuccinimide (112 mg, 0.842 mmol) in CH_2Cl_2 (1.8 ml) under nitrogen was added dropwise dimethyl sulfide (0.065 ml, 0.884 mmol). The mixture was stirred at 0 °C for 5 min, and recooled to –48 °C. To the mixture was added dropwise a solution of (2*E*,4*E*,7*Z*)-**21** (64.0 mg, 0.421 mmol) in CH_2Cl_2 (1 ml). The reaction mixture was stirred at 0 °C for 2 h and then at room temperature for 15 min, and concentrated under reduced pressure. The residue was dissolved in dry CH_3CN (2 ml) under nitrogen and triphenylphosphine (331 mg, 1.26 mmol) was added to the solution. The mixture was stirred at room temperature for 66 h and concentrated under reduced pressure. The oily residue was purified by column chromatography on silica gel (30 g, 4:1 EtOAc–MeOH), affording **22** (123 mg, 68%) as a colorless resin :^1H NMR (270 MHz, $CDCl_3$) δ 0.93 (t, *J* = 7.6 Hz, 3 H), 2.00 (qd, *J* = 7.6, 7.3 Hz, 2 H), 2.76 (m, 2 H), 4.78 (dd, *J* = 15.2, 7.6 Hz, 2 H), 5.27 (dt, *J* = 10.6, 7.3 Hz, 1 H), 5.30 (m, 1 H), 5.43 (dt, *J* = 10.6, 7.3 Hz, 1 H), 5.63 (dtd, *J* = 15.2, 6.3, 2.3 Hz, 1 H), 5.90 (dd, *J* = 15.2, 10.6 Hz, 1 H), and 6.34, (dtd, *J* = 15.2, 10.6, 5.3 Hz, 1 H). This material was azeotropically dried several times with toluene just prior to use for the next Wittig reaction.

Methyl (8*S*,9*R*,5*Z*,10*Z*,12*E*,14*E*,17*Z*)-8,9-Isopropylidenedioxy-5,10,12,14,17-eicosapentaenoate (24). To a stirred suspension of azeotropically dried **22** (120 mg, 0.278 mmol) in THF (5 ml) under nitrogen was added dropwise 1.0 M $NaN(SiMe_3)_2$ in THF (0.28 ml, 0.28 mmol), and the mixture was stirred at room temperature for 30 min to give an orange solution of ylide **23**. To the cooled (–78 °C), stirred solution of **23** was added dropwise a solution of **9** (50.9 mg, 0.188 mmol) in THF (1 ml). After the reaction mixture was stirred at –78 °C for 30 min and then at –48 °C for 2 h, the reaction was quenched by addition of saturated NH_4Cl solution (8 ml). The aqueous mixture was extracted with EtOAc (3 x 10 ml). The combined extracts were washed, dried, and concentrated under reduced pressure. The oily residue was purified by column chromatography on silica gel (10 g, 15:1→8:1 hexane–EtOAc), affording **24** (53.2 mg, 73%) as a colorless oil: $[\alpha]_D^{26}$ –77.4° (*c* 0.430, $CHCl_3$); UV (MeOH) λ_{max} 265 (ε 18900), 273 (23900), and 282 nm (19400); IR ($CHCl_3$) 1730, 1440, 1380, 1365, 1160, and 990 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 0.97 (t, *J* = 7.6 Hz, 3 H), 1.38 (s, 3 H), 1.49 (s, 3 H), 1.68 (tt, *J* = 7.6, 7.6 Hz, 2 H), 2.03–2.28 (m, 6 H), 2.30 (t, *J* = 7.6 Hz, 2 H), 2.86 (m, 2 H), 3.66 (s, 3 H), 4.18 (dt, *J* = 8.6, 5.9 Hz, 1 H), 5.04 (dd, *J* = 8.6, 5.9 Hz, 1 H), 5.30–5.53 (m, 5 H), 5.75 (dt, *J* = 14.9, 6.3 Hz, 1 H), 6.07–6.20 (m, 2 H), 6.24 (dd, *J* = 14.9, 10.9 Hz, 1 H), and 6.38 (dd, *J* = 14.9, 10.9 Hz, 1 H); EIMS

m/z (relative intensity) 388 (M^+, 13), 373 (3), 330 (20), 313 (20), and 110 (100); HREIMS calcd for $C_{24}H_{36}O_4$ (M^+) 388.2614, found 388.2586.

Methyl (8*S*,9*R*,5*Z*,10*Z*,12*E*,14*E*,17*Z*)-8,9-Dihydroxy-5,10,12,14,17-eicosapentaenoate (25). A solution of **24** (65.2 mg, 0.168 mmol) in degassed 4:1 AcOH–H_2O (7 ml) under argon was stirred at 10 °C for 21 h and concentrated under reduced pressure. The oily residue was purified by column chromatography on silica gel (16 g, 3:1→3:2 hexane–EtOAc), affording **25** (42.6 mg, 73%) as a colorless oil: $[\alpha]_D^{26}$ –37.2° (*c* 0.230, MeOH); UV (MeOH) λ_{max} 265 (ε 34700), 273 (41700), and 282 nm (34300); IR ($CHCl_3$) 3560, 3440 (br), 1730, 1440, 1380, 1150, and 1000 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 0.97 (t, *J* = 7.6 Hz, 3 H), 1.69 (tt, *J* = 7.6, 7.6 Hz, 2 H), 2.00–2.35 (m, 8 H), 2.31 (t, *J* = 7.6 Hz, 2 H), 2.86 (t, *J* = 6.9 Hz, 2 H), 3.67 (s, 3 H), 3.73 (m, 1 H), 4.61 (m, 1 H), 5.30–5.56 (m, 5 H), 5.76 (dt, *J* = 14.9, 6.9 Hz, 1 H), 6.12 (dd, *J* = 14.9, 10.0 Hz, 1 H), 6.17 (m, 1 H), 6.26 (dd, *J* = 14.9, 10.0 Hz, 1 H), and 6.40 (dd, *J* = 14.9, 10.0 Hz, 1 H); EIMS *m/z* (relative intensity) 348 (M^+, 5), 330 (35), 312 (25), 278 (11), and 109 (100); HREIMS calcd for $C_{21}H_{32}O_4$ (M^+) 348.2301, found 348.2313.

(8*S*,9*R*,5*Z*,10*Z*,12*E*,14*E*,17*Z*)-8,9-Dihydroxy-5,10,12,14,17-eicosapentaenoic Acid (26). To a solution of **25** (12.4 mg, 0.0356 mmol) in degassed 3:1 MeOH–H_2O (6 ml) under argon was added LiOH (15 mg). The mixture was stirred at room temperature for 23 h and concentrated under reduced pressure. The residue was diluted with saturated NaCl solution (6 ml). The aqueous mixture was acidified to pH 3 with 1 M HCl and extracted with EtOAc (3 x 6 ml). The combined extracts were washed, dried, and concentrated under reduced pressure. The oily residue was purified by column chromatography on silica gel (4 g, 3:2 EtOAc–hexane), affording **26** (10.3 mg, 87%) as a colorless oil: $[\alpha]_D^{25}$ –30.9° (*c* 0.370, MeOH); UV (MeOH) λ_{max} 264 (ε 23900), 270 (29200), and 280 nm (24400); IR ($CHCl_3$) 3600–2400, 1705, 1460, 1400, and 990 cm^{-1}; ^1H NMR (270 MHz, C_6D_6) δ 0.97 (t, *J* = 7.6 Hz, 3 H), 1.71 (tt, *J* = 7.6, 7.6 Hz, 2 H), 2.00–2.35 (m, 7 H), 2.35 (t, *J* = 7.6 Hz, 2 H), 2.86 (t, *J* = 6.9 Hz, 2 H), 3.73 (ddd, *J* = 8.6, 4.3, 4.3 Hz, 1 H), 4.61 (dd, *J* = 9.6, 4.3 Hz, 1 H), 5.34 (dt, *J* = 10.6, 6.9 Hz, 1 H), 5.44–5.56 (m, 4 H), 5.76 (dt, *J* = 14.8, 6.9 Hz, 1 H), 6.11 (dd, *J* = 14.8, 10.6 Hz, 1 H), 6.20 (m, 1 H), and 6.25 (dd, *J* = 14.8, 10.6 Hz, 1 H); EIMS *m/z* (relative intensity) 334 (M^+, 1), 316 (2), 178 (47), and 109 (100); HREIMS calcd for $C_{20}H_{30}O_4$ (M^+) 334.2144, found 334.2114.

(8*S*,9*R*)-Didemnilactone A (27). To a solution of **26** (16.8 mg, 0.0503 mmol) in THF (2.5 ml) under nitrogen were added a solution of Et_3N (5.6 mg, 0.0553 mmol) in THF (0.1 ml) and a solution of 2,4,6-trichlorobenzoyl chloride (12.3 mg, 0.0503 mmol) in THF (0.1 ml). The mixture was stirred at room temperature for 2 h and then diluted with toluene (24 ml). The diluted mixture was added dropwise over a 1.5-h period to a refluxing toluene (6 ml) solution containing 4-(dimethylamino)pyridine (123 mg, 1.01 mmol) under nitrogen. After the addition, the mixture was cooled to room temperature and diluted with ether (36 ml). The mixture was successively washed with 1 M HCl (2.5 ml), saturated $NaHCO_3$ solution (6 ml), and saturated NaCl solution (6 ml), dried, and concentrated under reduced pressure. The oily residue was purified by column chromatography on silica gel (8 g, 50:1→20:1 benzene–EtOAc), affording desired **27** (6.7 mg, 42%) and the isomer **28** (2.7 mg, 17%) as a colorless oil, respectively.

27: $[\alpha]_D^{22}$ +193° (*c* 0.180, MeOH); HREIMS calcd for $C_{20}H_{28}O_3$ (M^+) 316.2038, found 316.2053. The spectral and chromatographic properties of synthetic **27** were identical with those of natural **1** in all respects, except for the sign of the specific rotation.

28: IR ($CHCl_3$) 3600, 3450 (br), 1730, 1480, 1350, 1140, and 990 cm^{-1}; ^1H NMR (270 MHz, C_6D_6) δ 0.89 (t, *J* = 7.6 Hz, 3 H), 1.45–1.80 (m, 3 H), 1.90–2.15 (m, 6 H), 2.29–2.41 (m, 2 H), 2.73 (m, 2 H), 4.50 (m, 1 H), 4.93 (m, 1 H), 5.29–5.72 (m, 6 H), 6.08 (dd, *J* = 14.8, 10.6 Hz, 1 H), 6.14 (m, 1 H), 6.16 (dd, *J* = 14.8, 10.6

Hz, 1 H), and 7.08 (dd, J = 14.8, 11.9 Hz, 1 H); EIMS *m/z* (relative intensity) 316 (M^+, 69), 298 (5), 190 (65), and 55 (100); HREIMS calcd for $C_{20}H_{28}O_3$ (M^+) 316.2038, found 316.2036.

[(2*E*,4*Z*,7*Z*)-2,4,7-Decatrienyl]triphenylphosphnium Chloride (29). According to essentially the same procedure for the preparation of **22** from (2*E*,4*E*,7*Z*)-**21**, (2*E*,4*Z*,7*Z*)-**21** (245 mg, 1.61 mmol) was converted into **29** (596 mg, 87%): a colorless resin; ^{1}H NMR (270 MHz, CD_3OD) δ 0.94 (t, J = 7.6 Hz, 3 H), 2.01 (qd, J = 7.6, 7.3 Hz, 2 H), 2.78 (m, 2 H), 4.39 (dd, J = 16.2, 7.6 Hz, 2 H), 5.17 (dt, J = 10.5, 7.3 Hz, 1 H), 5.38 (dt, J = 10.5, 7.3 Hz, 1 H), 5.45 (m, 1 H), 5.60 (dt, J = 15.3, 7.6 Hz, 1 H), 5.95 (dd, J = 10.5, 10.5 Hz, 1 H), and 6.58 (dddd, J = 15.3, 10.5, 5.3, 1.3 Hz, 1 H).

Methyl (8*S*,9*R*,5*Z*,10*Z*,12*E*,14*Z*,17*Z*)-8,9-Isopropylidenedioxy-5,10,12,14,17-eicosapentaenoate (31). According to essentially the same procedure for the preparation of **24** from **9** and **22**, Wittig reaction of **9** (182 mg, 0.673 mmol) with ylide **30** generated from **29** (590 mg, 1.52 mmol) was performed, yielding **31** (194 mg, 74% from **9**) as a colorless oil: $[\alpha]_D^{27}$ –40.4° (*c* 0.161, $CHCl_3$); UV (MeOH) λ_{max} 270 (ε 35900), 275 (38900), and 284 nm (33200); IR ($CHCl_3$) 1730, 1440, 1380, 1365, 1160, and 990 cm^{-1}; ^{1}H NMR (270 MHz, $CDCl_3$) δ 0.98 (t, J = 7.6 Hz, 3 H), 1.38 (s, 3 H), 1.49 (s, 3 H), 1.68 (tt, J = 7.6, 7.6 Hz, 2 H), 2.06–2.32 (m, 6 H), 2.30 (t, J = 7.6 Hz, 2 H), 2.95 (m, 2 H), 3.66 (s, 3 H), 4.18 (dt, J = 8.6, 5.4 Hz, 1 H), 5.05 (dd, J = 8.6, 6.3 Hz, 1 H), 5.26–5.65 (m, 6 H), 6.06 (dd, J = 10.9, 10.9 Hz, 1 H), 6.26 (dd, J = 10.9, 10.9 Hz, 1 H), 6.45 (dd, J = 14.5, 10.9 Hz, 1 H), and 6.58 (dd, J = 14.5, 10.9 Hz, 1 H), ; EIMS *m/z* (relative intensity) 388 (M^+, 17), 330 (11), 299 (5), 218 (42), and 110 (100); HREIMS calcd for $C_{24}H_{36}O_4$ (M^+) 388.2614, found 388.2588.

Methyl (8*S*,9*R*,5*Z*,10*Z*,12*E*,14*Z*,17*Z*)-8,9-Dihydoxy-5,10,12,14,17-eicosapentaenoate (32). According to essentially the same procedure for the preparation of **25** from **24**, acidic hydrolysis of **31** (91.0 mg, 0.266 mmol) was performed, yielding **32** (67.7 mg, 83%) as a colorless oil: $[\alpha]_D^{19}$ +7.93° (*c* 0.580, MeOH); UV (MeOH) λ_{max} 265 (ε 27300), 274 (34800), and 284 nm (27800); IR ($CHCl_3$) 3560, 3440 (br), 1730, 1440, 1360, 1130, and 990 cm^{-1}; ^{1}H NMR (270 MHz, $CDCl_3$) δ 0.98 (t, J = 7.6 Hz, 3 H), 1.69 (tt, J = 7.6, 7.6 Hz, 2 H), 2.06–2.24 (m, 8 H), 2.31 (t, J = 7.6 Hz, 2 H), 2.95 (m, 2 H), 3.66 (s, 3 H), 3.73 (m, 1 H), 4.61 (m, 1 H), 5.26–5.57 (m, 6 H), 6.06 (dd, J = 10.9, 10.9 Hz, 1 H), 6.28 (dd, J = 10.9, 10.9 Hz, 1 H), 6.47 (dd, J = 14.5, 10.9 Hz, 1 H), and 6.59 (dd, J = 14.5, 10.9 Hz, 1 H); EIMS *m/z* (relative intensity) 348 (M^+, 2), 330 (5), 299 (4), and 171 (100); HREIMS calcd for $C_{21}H_{32}O_4$ (M^+) 348.2301, found 348.2309.

(8*S*,9*R*,5*Z*,10*Z*,12*E*,14*Z*,17*Z*)-8,9-Dihydroxy-5,10,12,14,17-eicosapentaenoic Acid (33). According to essentially the same procedure for the preparation of **26** from **25**, basic hydrolysis of **32** (67.7 mg, 0.195 mmol) was performed, yielding **33** (62.0 mg, 95%) as a colorless oil: $[\alpha]_D^{18}$ –8.12° (*c* 0.350, MeOH); UV (MeOH) λ_{max} 266 (ε 24500), 274 (32300), and 283 nm (25600); IR ($CHCl_3$) 3600–2400, 1705, 1450, 1400, 1130, and 990 cm^{-1}; ^{1}H NMR (270 MHz, $CDCl_3$) δ 0.98 (t, J = 7.6 Hz, 3 H), 1.68 (tt, J = 7.6, 7.6 Hz, 2 H), 2.05–2.32 (m, 8 H), 2.35 (t, J = 7.6 Hz, 2 H), 2.95 (m, 2 H), 3.74 (m, 1 H), 4.63 (dd, J = 8.9, 3.6 Hz, 1 H), 5.26–5.56 (m, 6 H), 6.06 (dd, J = 10.9, 10.9 Hz, 1 H), 6.27 (dd, J = 10.9, 10.9 Hz, 1 H), 6.46 (dd, J = 14.8, 10.9 Hz, 1 H), and 6.58 (dd, J = 14.8, 10.9 Hz, 1 H); EIMS *m/z* (relative intensity) 334 (M^+, 2), 316 (1), 300 (1), 178 (70), and 109 (100); HREIMS calcd for $C_{20}H_{30}O_4$ (M^+) 334.2144, found 334.2130.

(8*S*,9*R*)-Didemnilactone B (34). According to essentially the same procedure for the lactonization of **26**, **33** (28.4 mg, 0.085 mmol) was lactonized under Yamaguchi's conditions, yielding desired **34** (12.6 mg, 47%) and the isomer **35** (4.2 mg, 11%) as a colorless oil, respectively.

34: $[\alpha]_D^{26}$ +281° (*c* 0.280, MeOH);[12] HREIMS calcd for $C_{20}H_{28}O_3$ (M^+) 316.2038, found 316.2038. The spectral and chromatographic properties of synthetic **34** were identical with those of natural **2** in all respects, except for the sign of the specific rotation.

35: IR ($CHCl_3$) 3600, 3440 (br), 1740, 1450, 1350, 1140, and 990 cm^{-1}; ^1H NMR (270 MHz, C_6D_6) δ 0.89 (t, *J* = 7.6 Hz, 3 H), 1.45–1.54 (m, 2 H), 1.73 (m, 1 H), 1.89–2.07 (m, 6 H), 2.29–2.41 (m, 2 H), 2.88 (t, *J* = 6.9 Hz, 2 H), 4.66 (m, 1 H), 5.12 (ddd, *J* = 9.9, 5.3, 1.3 Hz, 1 H), 5.32–5.49 (m, 6 H), 6.08 (dd, *J* = 10.9, 10.9 Hz, 1 H), 6.12 (dd, *J* = 10.9, 10.9 Hz, 1 H), 6.74 (dd, *J* = 14.8, 10.9 Hz, 1 H), and 6.76 (dd, *J* = 14.8, 10.9 Hz, 1 H); EIMS *m/z* (relative intensity) 316 (M^+, 95), 298 (6), and 177 (100); HREIMS calcd for $C_{20}H_{28}O_3$ (M^+) 316.2038, found 316.2036.

Acknowledgements: We are grateful to Drs. Masaaki Toda and Nobuyuki Hamanaka, Ono Pharmaceutical Co., Ltd. for biological tests. This work was supported by Grant-in-aid for Scientific Research (No. 04640524) from the Ministry of Education, Science, and Culture.

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2. Part of this work was preliminarily published: Niwa, H.; Inagaki, H.; Yamada, K. *Tetrahedron Lett.* **1991**, *32*, 5127–5128. In this report we renamed the compound **1** didemnilactone A, which was originally called "didemnilactone" in the preliminary communication.
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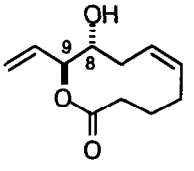

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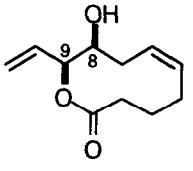

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(*Received in Japan* 15 *March* 1994; *accepted* 18 *April* 1994)