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SYNTHETIC STUDIES ON NOGALAMYCIN CONGENERS [4][1,2] SYNTHESES AND ANTITUMOR ACTIVITY OF VARIOUS NOGALAMYCIN CONGENERS

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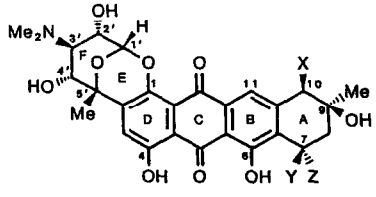
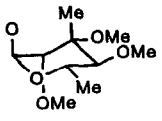
Abstract: Various structural types of nogalamycin congeners and their partial structures, which had been previously synthesized in the course of our synthetic studies on the total syntheses or were originally produced by employing the explored synthetic scheme, were subjected to *in vitro* cytotoxicity and *in vivo* antitumor activity assay against P388 murine leukemia. These studies obviously disclosed novel aspects of the structure-activity relationships of nogalamycin congeners.

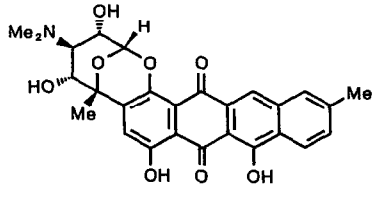
Nogalamycin (**1**) and its congeners, the notable anthracycline antibiotics, have attracted much attention in recent years due to their prominent anticancer activity and unique structures.[2] Especially, 7-con-*O*-methylnogarol (**2**), the most well-known semisynthetic derivative of **1**, has been selected for clinical trials because of its superior antitumor activity to that of the parent compound (**1**).[2]



X = CO_2Me, Y = H, Z =  **1**

X = H, Y = OMe, Z = H **2**

X = Y = Z = H **3**



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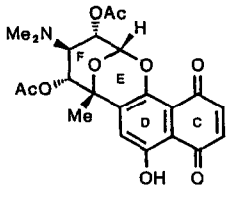

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As mentioned in the preceding papers,[1a-c,2] we have achieved the first total syntheses of nogalamycin congeners such as (+)-nogarene (**4**), (+)-7-deoxynogarol (**3**), and 7-con-*O*-methylnogarol (**2**). The explored synthetic route to **2**-**4** features the regioselective Diels-Alder reactions of the (+)-naphthoquinone (**5**), the CDEF-

ring system of nogalamycin congeners, with the dienes having the A-ring functionalities as key steps.[1a-c,2] With completion of the total synthesis, we became interested in the structure-activity relationships of nogalamycin congeners in light of prominent anticancer activity of **2**. Accordingly, various structural types of nogalamycin congeners and their partial structures, which had been previously synthesized in the course of our studies on the total synthesis, or were originally synthesized by employing the explored synthetic scheme, were subjected to *in vitro* cytotoxicity and *in vivo* antitumor activity assay against P388 murine leukemia. These studies obviously disclosed novel aspects of the structure-activity relationships, which should hold promise for designing the nogalamycin congeners which may exhibit characteristic antitumor activity. The fourth part of this series of papers details these studies on the structure-activity relationships.[1d]

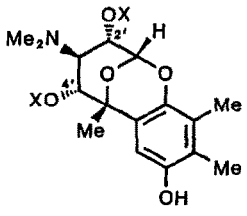
Results and Discussion

Syntheses and Antitumor Activity of the Partial Structures of Nogalamycin Congeners. Various partial structures of nogalamycin congeners such as the DEF-ring systems (**7**), the diastereomer of **7** (**9**), the CDEF-ring system (**5**), the compounds related to **5** (**10-12**), the BCDEF-ring systems (**15** and **18**), and some of their 2',4'-diacetates (**6**, **8**, **14**, and **17**) (nogalamycin numbering),[3] were first subjected to *in vitro* cytotoxicity assay against P388 murine leukemia. As reported in the preceding papers,[1a,b,2] the compounds (**5-12**) had already been prepared in the

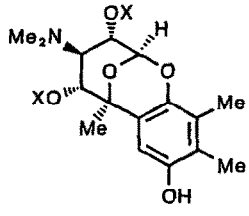
Table I. *In Vitro* Cytotoxicity of the Partial Structures of Nogalamycin Congeners against P388 Murine Leukemia Cells

Compounds Related to DEF-Ring System		Compounds Related to CDEF-Ring System		Compounds Related to BCDEF-Ring System	
Compound	IC_{50} (μg/ml)[a]	Compound	IC_{50} (μg/ml)[a]	Compound	IC_{50} (μg/ml)[a]
6	> 10	**10**	> 10	**14**	1.8
7	> 10	**11**	0.14	**15**	1.6
8	> 10	**12**	> 10	**17**	1.4
9	> 10	**5**	0.10	**18**	1.5

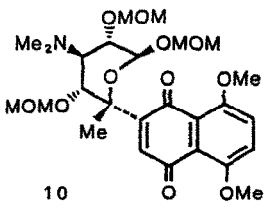
a) Concentration (μg/ml) necessary to inhibit cell growth (initial cell density: 5 x 10^4 cells/ml) at 50% after incubation for 48 h at 37 °C.



X = Ac **6**
X = H **7**



X = Ac **8**
X = H **9**



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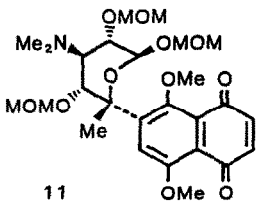

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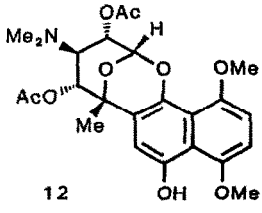

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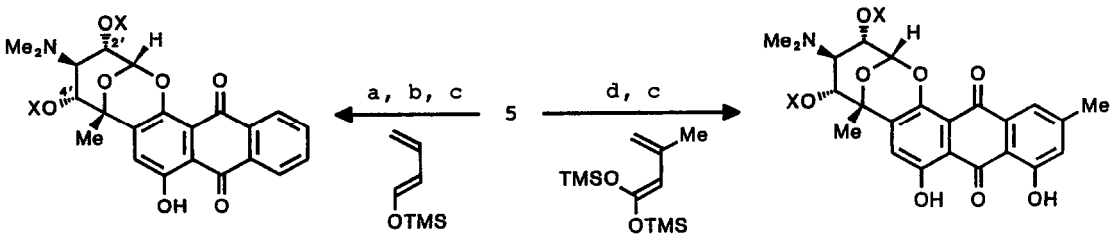
Scheme 1



a) 1) **13**, THF, 60 °C, 3 h 2) 3M HCl, 60 °C, 20 min b) Et_3N, $CHCl_3$, 40 °C, 30 min, 91% (2 steps, from 5) c) 1M HCl, 5 h, 88% (**15**), 82% (**18**) d) 1) **16**, THF, 60 °C, 40 min 2) 3M HCl, rt, 15 min, 64% (from 5).

Table II. *In Vivo* Antitumor Activity of the Partial Structures of Nogalamycin Congeners (**11** and **5**) against P388 Murine Leukemia Cells[a]

Compound	T/C[b]				
	Dose (mg/kg)[c]				
	100	50	25	12.5	6.25
11	0	0	102	95	-
5	-	-	0	94	108

a) Evaluated by the same method as that employed at Drug Evaluation Branch, National Cancer Institute (NCI), NIH, U.S.A.

b) Median survival time of test animals x 100 / median survival time of control animals.

c) P388 murine leukemia cells (10^6) were inocultated into CDF_1 mice (6 mice/group) intraperitoneally. Drugs were administrated, starting 24 h after inoculation, at day 1 and 5, intraperitoneally.

course of our total syntheses of **2-4**. Preparations of the BCDEF-ring systems (**15** and **18**) were examined as shown in **Scheme 1**. Thus, the regioselective Diels-Alder reactions of **5** with the linear dienes (**13** and **16**),[4] followed by deacetylation, gave rise to **15**, mp 254-264 °C (decomp.) and $[\alpha]_D^{20}$ +707° (c 0.230, $CHCl_3$), and **18**, mp 259-261 °C (decomp.) and $[\alpha]_D^{20}$ +820° (c 0.120, $CHCl_3$), by way of the 2',4'-diacetates (**14** and **17**), **14**: mp 259-264 °C (decomp.); $[\alpha]_D^{20}$ +315° (c 0.118, $CHCl_3$), and **17**: mp 281-283 °C (decomp.); $[\alpha]_D^{20}$ +425° (c 0.130, $CHCl_3$). Results summarized in **Table I** cleanly disclose that the partial structures (**6-10**, **12**, **14-18**) except for **11** and **5** showed no significant cytotoxicity. Although marginal cytotoxicity was observed for **11** and **5**, they showed no inhibitory activity against P388 murine leukemia *in vivo* as shown in **Table II**. Based on these results, it appeared evident that all the carbon framework (the ABCDEF-ring system) is indispensable for pronounced antitumor activity of the nogalamycin congeners.

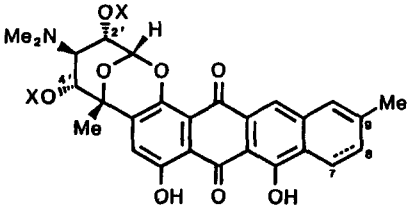
Antitumor Activity of (+)-Nogarene, (+)-7-Deoxynogarol, (+)-7-con-O-Methyl-

nogarol, and Their Related Compounds. Next, the nogalamycin congeners (**4**-**2**) and their 2',4'-diacetates (**19**, **22**, and **25**) were subjected to *in vitro* cytotoxicity assay against P388 murine leukemia along with their related compounds such as (+)-7,8-dihydronogarene (**21**), (+)-9-*epi*-7-deoxynogarol (**24**), (+)-7,9-di-*epi*-7-con-*O*-methylnogarol (**27**), and their 2',4'-diacetates (**20**, **23**, and **26**). The latter congeners (**20**, **21**, **23**, **24**, **26**, and **27**) were obtained for the first time in the course of our studies on the total syntheses of **2**-**4**.[1c,2] As shown in **Table III**, the 7-demethoxycongeners (**4**, **21**, **3**, and **24**) and their 2',4'-diacetates (**19**, **20**, **22**, and **23**) were found to exhibit marginal *in vitro* cytotoxicity. No significant *in vivo* antitumor activity against P388 murine leukemia was observed for **21** and **24** (**Table IV**). It had been reported that **3** and **4** exhibited no activity in P388 *in vivo* test.[5] In contrast with **2** and its C_7-epimer, 7-dis-*O*-methylnogarol (**28**), showing potent antitumor activity against P388 murine leukemia *in vivo*,[5] the marginal cytotoxicity was only observed for the 7,9-di-*epi*-congener (**27**) and its 2',4'-diacetate (**26**).

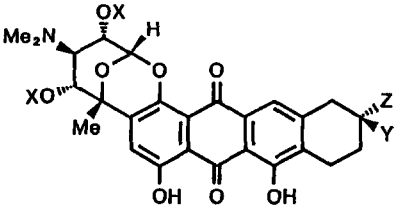
Table III. *In Vitro* Cytotoxicity of (+)- Nogarene, (+)-7-Deoxynogarol, (+)-7-con-*O*-Methylnogarol, and Their Related Compounds against P388 Murine Leukemia Cells

Nogalamycin Congeners		2',4'-Diacetates	
Compound	IC_{50} (μg/ml)[a]	Compound	IC_{50} (μg/ml)[a]
4	0.11	**19**	0.17
21	0.13	**20**	0.58
3	0.41	**22**	0.17
24	0.31	**23**	0.30
2 (synthetic)	0.003-0.0012	**25** (synthetic)	0.014
2 (natural)	0.006		
27	0.40	**26**	0.53

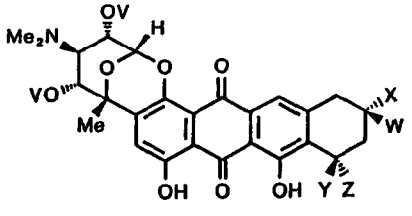
a) See the footnote a) in **Table I**.



X = Ac, $C_{7,8}$ = CH=CH **19**
X = Ac, $C_{7,8}$ = CH_2-CH_2 **20**
X = H, $C_{7,8}$ = CH_2-CH_2 **21**



X = Ac, Y = OH, Z = Me **22**
X = Ac, Y = Me, Z = OH **23**
X = H, Y = Me, Z = OH **24**



V = Ac, W = OH, X = Me, Y = OMe, Z = H **25**
V = Ac, W = Me, X = OH, Y = H, Z = OMe **26**
V = H, W = Me, X = OH, Y = H, Z = OMe **27**
V = H, W = OH, X = Me, Y = H, Z = OMe **28**

Table IV. *In Vivo* Antitumor Activity of (+)-7,8-Dihydronogarene (**21**) and (+)-9-*epi*-7-Deoxynogarol (**24**) against P388 Murine Leukemia Cells[a]

Compound	T/C[b]		
	Dose (mg/kg)[c]		
	10	5	2.5
21	109	95	93
24	109	109	109

a-c) See the footnotes a-c) in **Table II.**

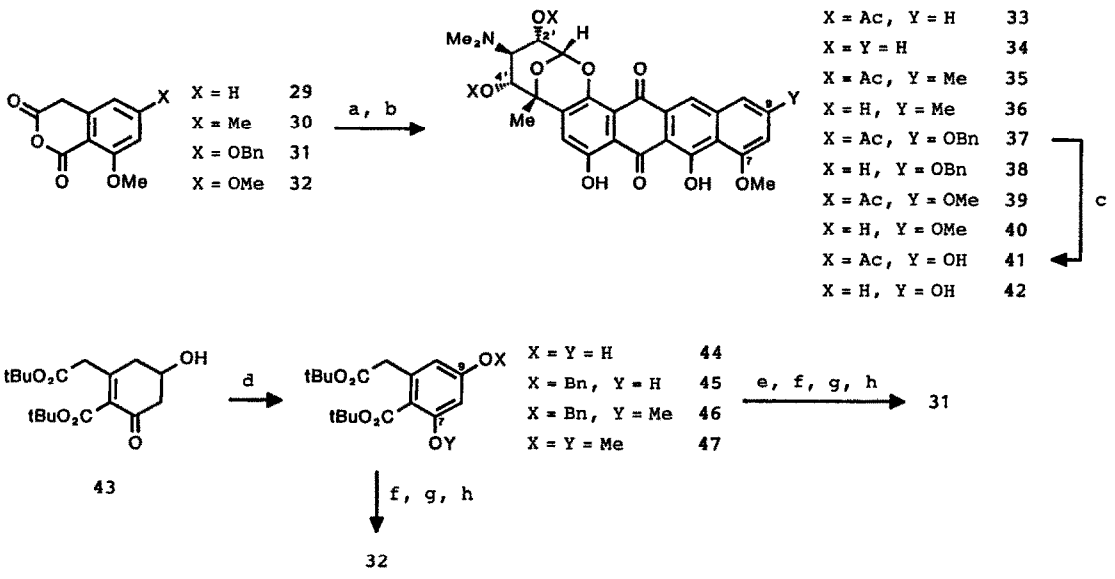
Accordingly, it became evident that in addition to the ABCDEF-ring system, the C_7-methoxy group is indispensable for potent antitumor activity of the nogalamycin congeners. Furthermore, taking into account the inhibitory activity of **2**, **27**, and **28**, absolute stereochemistry of the C_9-position is anticipated to play an additional important role.

Syntheses and Antitumor Activity of the Various 7-Methoxynogarene Derivatives. In light of the results described above, it was of interest to evaluate antitumor activity of the 7-methoxynogarene derivatives (**34**, **36**, **38**, **40**, and **42**) carrying various functionalities at the C_9-positions (**Scheme 2**). For the synthesis of these compounds, it was expected that their 11-deoxyanthracyclinone skeletons could be effectively constructed from **5** by employing the strong base induced cycloaddition of homophthalic anhydride explored by Tamura *et al.*[6] Although it had been established that the Diels-Alder reaction of **5** with a highly polarized diene under neutral condition cleanly occurred in a completely regioselective manner, the base induced cycloaddition with a homophthalic anhydride had not been examined on **5**.

Thus, the base induced cycloadditions of **5** with the well-known homophthalic anhydrides (**29** and **30**)[7] were first attempted. Similarly to the reported results,[6] the reactions of **5** with the sodium salts of **29** and **30** were found to take place smoothly in a completely regioselective manner, affording the (+)-2',4'-di-*O*-acetyl-9-demethyl-7-methoxynogarene (**33**), mp 175-178 °C and $[\alpha]_D^{20}$ +533° (c 0.030, $CHCl_3$), and (+)-2',4'-di-*O*-acetyl-7-methoxynogarene (**35**), mp 250-255 °C and $[\alpha]_D^{20}$ +545° (c 0.110, $CHCl_3$), after concomitant decarboxylation and air oxidation of the addition products during work-up. Both diacetates (**33** and **35**) were deprotected, giving (+)-9-demethyl-7-methoxynogarene (**34**), mp 230-235 °C and $[\alpha]_D^{20}$ +400° (c 0.030, $CHCl_3$), and (-)-7-methoxynogarene (**36**), mp 231-235 °C (decomp.) and $[\alpha]_D^{20}$ -900° (c 0.070, $CHCl_3$), respectively.

Next, syntheses of 7-methoxynogarenes (**38**, **40**, and **42**) carrying the oxygen functionalities at the C_9-positions were examined. Syntheses of the requisite homophthalic anhydrides (**46** and **47**) were preformed using the keto diester (**43**)[8] as the starting material. Thus, the keto diester (**43**) was first converted to the diphenol (**44**) by Jones oxidation. After selective benzylation[9] of the C_9-hydroxyl group of **44**, the remaining C_7-hydroxyl group of the benzyl ether (**45**) was methylated to yield the diether (**46**). Cleavage of the two *tert*-butyl esters, followed by dehydration of the resulting dicarboxylic acid with acetic anhydride, afforded **31**, mp 171-172 °C. On the other hand, after methylation of the two hydroxyl groups of **44**, similar sequential treatments converted the obtained dimethyl ether (**47**) into **32**, mp 167-168 °C.

Scheme 2



a) 1) NaH, THF, rt, 20 min 2) **5**, 0 °C → rt, 1 h, 62% (**33**, from **5**), 81% (**35**, from **5**), 100% (**37**, from **5**), 96% (**39**, from **5**) b) K_2CO_3, MeOH, 40 °C, 30 min (for **33** and **35**), MeOH-$CHCl_3$, rt, 5 h (for **37**, **39**, and **41**), 78% (**34**), 72% (**36**), 63% (**38**), 96% (**40**), 18% (**42**) c) H_2, 10% Pd-$BaSO_4$, MeOH, rt, 2 h, 79% d) Jones reagent, Me_2CO, 0 °C, 2 h, 89% e) BnBr, $K_2CO_3 \cdot 1.5H_2O$, Me_2CO, reflux, 3 h, 83% f) Me_2SO_4, K_2CO_3, reflux, 3 h, 95% (**46**), 98% (**47**) g) CF_3CO_2H, CH_2Cl_2, rt, 12 h h) Ac_2O, PhMe, 100 °C, 20 min, 99% (**31**, 2 steps), 98% (**32**, 2 steps).

The regioselective cycloadditions of **5** with sodium salts of **31** and **32**, followed by concomitant decarboxylation and air oxidation of the addition products during work-up, gave rise to (+)-2',4'-di-*O*-acetyl-9-benzyloxy-9-demethyl-7-methoxynogarene (**37**), mp 251-254 °C and $[\alpha]_D^{20}$ +706° (c 0.051, $CHCl_3$), and (+)-2',4'-di-*O*-acetyl-9-demethyl-7,9-dimethoxynogarene (**39**), mp 174-177 °C and $[\alpha]_D^{20}$ +649° (c 0.111, $CHCl_3$). Deacetylation of **37** and **39** readily produced (-)-9-benzyloxy-9-demethyl-7-methoxynogarene (**38**), mp >270 °C (decomp.) and $[\alpha]_D^{20}$ -1780° (c 0.009, $CHCl_3$), and (-)-9-demethyl-7,9-dimethoxynogarene (**40**), mp 228-231 °C and $[\alpha]_D^{20}$ -1040° (c 0.025, $CHCl_3$), respectively. Removal of the benzyl group of **37** by hydrogenation gave (-)-2',4'-di-*O*-acetyl-9-demethyl-9-hydroxy-7-methoxynogarene (**41**), mp >290 °C and $[\alpha]_D^{20}$ -2500° (c 0.020, $CHCl_3$), which on further deacetylation afforded 9-demethyl-9-hydroxy-7-methoxynogarene (**42**).[10]

These 7-methoxynogarenes (**34**, **36**, **38**, **40**, and **42**) and their 2',4'-diacetates (**33**, **35**, **37**, **39**, and **41**) were subjected to *in vitro* cytotoxicity assay against P388 murine leukemia. Results shown in **Table V** revealed that, among the tested samples, the 7-methoxynogarenes (**34**, **36**, **40**, and **42**) and the 2',4'-diacetates (**33** and **41**) exhibited prominent cytotoxicity. The 2',4'-diacetates (**33**, **35**, and **41**) being chemically more stable than the corresponding diols (**34**, **36**, and **42**) were subjected to *in vivo* test for antitumor activity against P388 murine leukemia. As shown in **Table VI**, **33** showed only marginal activity and no significant antitumor activity was observed for **35** and **41**. Thus, it became obvious that aromatization of the A-ring causes almost complete loss of inhibitory activity regardless of the presence of the C_7-methoxy and C_9-hydroxy groups.

Table V. *In Vitro* Cytotoxicity of 7-Methoxynogarenes and Their 2',4'-Diacetates against P388 Murine Leukemia Cells

7-Methoxynogarenes		2',4'-Diacetates	
Compound	IC_{50} (μg/ml)[a]	Compound	IC_{50} (μg/ml)[a]
34	0.040	**33**	0.038
36	0.057	**35**	0.13
38	0.13	**37**	0.18
40	0.043	**39**	0.11
42	0.032	**41**	0.016

a) See the footnote a) in **Table I.**

Table VI. *In Vivo* Antitumor Activity of (+)-2',4'-Di-*O*-acetyl-9-demethyl-7-methoxynogarene (**33**), (+)-2',4'-Di-*O*-acetyl-7-methoxynogarene (**35**), and (-)-2',4'-Di-*O*-acetyl-9-demethyl-9-hydroxy-7-methoxynogarene (**41**) against P388 Murine Leukemia Cells

Compound	T/C[b]				
	Dose (mg/kg)[c]				
	80	40	20	10	5
33	110	116	121	98	111
35	96	90	97	106	88
41	-	90	104	104	108

a-c) See the footnotes a-c) in **Table II.**

Conclusion

Summing up the results obtained above, it appears evident that (1) all the carbon framework (the ABCDEF-ring system) and the C_7-methoxy group are both indispensable for pronounced inhibitory activity, (2) absolute stereochemistry of the C_9-position also plays an important role in addition to the C_7-methoxy group, and (3) aromatization of the A-ring causes almost complete loss of activity regardless of the presence of the C_7-methoxy and C_9-hydroxy groups. These studies on the structure-activity relationships should hold promise for designing novel nogalamycin congeners which may exhibit characteristic antitumor activity.

Experimental

General. All melting points were determined with a Yamato MP-21 melting point apparatus and are uncorrected. Measurement of optical rotations were carried out using a Horiba SEPA-200 automatic digital polarimeter. IR spectra measurements were performed with a JASCO A-200 IR spectrometer. ^1H NMR spectra were measured with a Hitachi R-90H spectrometer (90 MHz) and a Bruker AM 400 spectrometer (400 MHz). All signals are expressed as ppm down field from tetramethylsilane used as an internal standard (δ value). The following abbreviations are used: singlet (s), doublet (d), triplet (t), multiplet (m), broad (br). Assignments of peaks are indicated according to the numbering of IUPAC nomenclature to avoid confusion. Mass spectra were taken with a Hitachi RMU-6MG mass spectrometer. Unless other-

wise noted, all reactions were performed using anhydrous solvents. Especially, tetrahydrofuran and ether freshly distilled from sodium benzophenone ketyl were used. Wako Gel C-200 was used as an adsorbent for column chromatography. The following abbrevations are used for solvents and reagents: acetic anhydride (Ac_2O), chloroform ($CHCl_3$), dichloromethane (CH_2Cl_2), ethanol (EtOH), ethyl acetate (AcOEt), methanol (MeOH), tetrahydrofuran (THF), trimethylsilyl chloride (TMSCl).

1-(Trimethylsilyloxy)-1,3-butadiene (13). Treatments of crotonaldehyde with $ZnCl_2$, triethylamine, and TMSCl following the reported procedure,[4] afforded pure **13** as a colorless oil, bp 63-64 °C/50 Torr [lit. bp 57-60 °C/50 Torr[4]]. ^{1}H NMR (90 MHz, $CDCl_3$) δ 0.22 (9H, s, $SiMe_3$), 4.83 (1H, dd, J = 2 and 10 Hz, C_{4-cis}-H), 4.99 (1H, dd, J = 2 and 18 Hz, $C_{4-trans}$-H), 5.72 (1H, t, J = 10 Hz, C_2-H), 6.25 (1H, dt, J = 18 and 10 Hz, C_3-H), 6.55 (1H, d, J = 10 Hz, C_1-H).

(2R,3S,4R,5R,6R)-(+)-3,5-Diacetoxy-4-dimethylamino-8-hydroxy-6-methyl-2,6-epoxy-3,4,5,6-tetrahydro-2H-anthraceno[1,2-b]oxocine-9,14-dione (14). A solution of **5** (45 mg, 0.10 mmol) and **13** (0.43 g, 3.0 mmol) in THF (1.0 ml) was heated at 60 °C for 3 h under an argon atmosphere. The reaction mixture was cooled to ambient temperature and concentrated *in vacuo*. The residue was dissolved in THF (5.0 ml) and 3M HCl (1.0 ml) was added to the tetrahydrofuran solution. The solution was heated at 40 °C for 20 min on exposure to air and diluted with AcOEt. The mixture was cooled in an ice bath and neutralized to pH 8 with saturated aqueous $NaHCO_3$. The upper ethyl acetate layer was separated, dried ($MgSO_4$), filtered, and concentrated *in vacuo*. The residual caramel was dissolved in $CHCl_3$ (10 ml) and triethylamine (1.5 g, 15 mmol) was added to the chloroform solution. The solution was heated at 40 °C for 30 min, cooled to room temperature, and concentrated *in vacuo*. The residue was chromatographed (SiO_2, AcOEt-hexane) to afford pure **14** as a yellow solid (45 mg, 91%). Recrystallization from EtOH-hexane gave an analytical sample of **14** as yellow crystals, mp 259-264 °C (decomp.) and $[\alpha]_D^{20}$ +315° (c 0.118, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.60 (3H, s, C_6-Me), 2.14 and 2.17 (6H, two s, COMe x 2), 2.28 (6H, s, NMe_2), 2.69 (1H, t, J = 10.8 Hz, C_4-H), 5.14 (1H, d, J = 10.8 Hz, C_5-H), 5.22 (1H, dd, J = 4.0 and 10.8 Hz, C_3-H), 5.90 (1H, d, J = 4.0 Hz, C_2-H), 7.07 (1H, s, C_7-H), 7.71-7.89 and 8.20-8.41 (4H, two m, C_{10}-H, C_{11}-H, C_{12}-H, C_{13}-H), 12.98 (1H, s, OH); IR (KBr) 3460, 1745, 1670, 1635, 1595, 1230, 1040 cm^{-1}; MS m/z 495 (M^+), 435. *Anal.* Calcd. for $C_{26}H_{25}NO_9 \cdot 0.5H_2O$: C, 61.90; H, 5.19; N, 2.78%. Found: C, 61.73; H, 5.12; N, 2.72%.

(2R,3S,4R,5R,6R)-(+)-4-Dimethylamino-3,5,8-trihydroxy-6-methyl-2,6-epoxy-3,4,5,6-tetrahydro-2H-anthraceno[1,2-b]oxocine-9,14-dione (15). A suspension of **14** (30 mg, 73 μmol) in 1M HCl (25 ml) was heated at reflux for 5 h and cooled in an ice bath. After addition of $CHCl_3$, the mixture was neutralized to pH 8 with saturated aqueous $NaHCO_3$. The lower chloroform layer was separated, dried (Na_2SO_4), filtered, and concentrated *in vacuo*. The residue was chromatographed (SiO_2, MeOH-$CHCl_3$) to give pure **15** as yellow solid (22 mg, 88%). Recrystallization from EtOH-hexane yielded an analytical sample of **15** as yellow crystals, mp 254-264 °C (decomp.) and $[\alpha]_D^{20}$ +707° (c 0.230, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$-D_2O) δ 1.74 (3H, s, C_6-Me), 2.51 (6H, s, NMe_2), 2.73 (1H, t, J = 10.7 Hz, C_4-H), 3.57 (1H, d, J = 10.7 Hz, C_5-H), 4.16 (1H, dd, J = 3.5 and 10.7 Hz, C_3-H), 5.89 (1H, d, J = 3.5 Hz, C_2-H), 7.26 (1H, s, C_7-H), 7.53-8.08 (4H, m, C_{10}-H, C_{11}-H, C_{12}-H, C_{13}-H); IR (KBr) 3470, 1665, 1620, 1570, 1230 cm^{-1}; MS m/z 411 (M^+), 324, 307. *Anal.* Calcd. for $C_{22}H_{21}NO_7 \cdot 0.3H_2O$: C, 63.39; H, 5.22; N, 3.36%. Found: C, 63.32; H, 5.25; N, 3.35%.

3-Methyl-1,1-bis(trimethylsilyloxy)-1,3-butadiene (16). Butyllithium (6.7 ml, 1.5M hexane solution, 10 mmol) was added to a solution of diisopropylamine (1.2 g,

12 mmol) in THF (10 ml) cooled at -78 °C under an argon atmosphere. After stirring for 20 min at the same temperature, a solution of 3-methylcrotonic acid (0.50 g, 5.0 mmol) in THF (5.0 ml) and TMSCl (2.2 g, 20 mmol) were successively added and the reaction mixture was warmed up to room temperature. After stirring for 1.5 h at room temperature, the mixture was concentrated *in vacuo*. The residue was diluted with hexane and the hexane suspension was kept standing at room temperature. A supernatant of the hexane suspension was taken out by a syringe and concentrated *in vacuo* to give **16** as a pale yellow caramel. Assignments of ^{1}H NMR spectrum (90 MHz, $CDCl_3$) could not be achieved probably due to high sensitivity of **16** to moisture. Thus, this diene was directly used for the next step without any further purification.

(2R,3S,4R,5R,6R)-(+)-3,5-Diacetoxy-4-dimethylamino-8,10-dihydroxy-6,12-dimethyl-2,6-epoxy-3,4,5,6-tetrahydro-2H-anthraceno[1,2-b]oxocine-9,14-dione (17). A solution of **5** (60 mg, 0.13 mmol) in THF (0.50 ml) was added to a solution of **16** [prepared from 3-methylcrotonic acid (0.10 g, 1.0 mmol) according to the procedure described above] in THF (0.50 ml) at room temperature under an argon atmosphere. The tetrahydrofuran solution was heated at 60 °C for 40 min and cooled to ambient temperature. 3M HCl (1.0 ml) was added to the reaction mixture and stirring was continued for 15 min on exposure to air. The mixture was neutralized to pH 8 with saturated aqueous $NaHCO_3$ and extracted with $CHCl_3$. The combined extracts were washed with brine, dried (Na_2SO_4), filtered, and concentrated *in vacuo*. The residue was chromatographed (SiO_2, AcOEt-$CHCl_3$) to give crude **17** as an orange solid. Recrystallization from EtOH-hexane afforded pure **17** as orange crystals (45 mg, 64%), 281-283 °C (decomp.) and $[\alpha]_D^{20}$ +425° (c 0.130, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.60 (3H, s, C_6-Me), 2.13 and 2.16 (6H, two s, COMe x 2), 2.28 (6H, s, NMe_2), 2.47 (3H, s, C_{12}-Me), 2.67 (1H, t, J = 10.7 Hz, C_4-H), 5.13 (1H, d, J = 10.7 Hz, C_5-H), 5.21 (1H, dd, J = 4.0 and 10.7 Hz, C_3-H), 5.89 (1H, d, J = 4.0 Hz, C_2-H), 7.02 (1H, s, C_7-H), 7.07 and 7.62 (2H, two d, each J = 1.5 Hz, C_{11}-H, C_{13}-H), 11.92 and 12.40 (2H, two s, OH x 2); IR (KBr) 3450, 1750, 1730, 1670, 1625, 1230, 1210, 1050 cm^{-1}; MS m/z 525 (M^+), 466, 465, 424. *Anal.* Calcd. for $C_{27}H_{27}NO_{10}$: C, 61.71; H, 5.18; N, 2.67%. Found: C, 61.44; H, 5.28; N, 2.56%.

(2R,3S,4R,5R,6R)-(+)-4-Dimethylamino-3,5,8,10-tetrahydroxy-6,12-dimethyl-2,6-epoxy-3,4,5,6-tetrahydro-2H-anthraceno[1,2-b]oxocine-9,14-dinone (18). The same acidic hydrolysis of **17** (26 mg, 49 μmol) as that described for **14**, followed by separation by column chromatography (SiO_2, EtOH-$CHCl_3$), gave crude **18** as an orange solid after concentration of the chloroform solution *in vacuo*. Recrystallization from $CHCl_3$-hexane afforded pure **18** as orange crystals, mp 259-261 °C (decomp.) and $[\alpha]_D^{20}$ +820° (c 0.120, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$-D_2O) δ 1.73 (3H, s, C_6-Me), 2.29 (3H, s, C_{12}-Me), 2.53 (6H, s, NMe_2), 2.83 (1H, t, J = 10.6 Hz, C_4-H), 3.57 (1H, d, J = 10.6 Hz, C_5-H), 4.16 (1H, dd, J = 3.5 and 10.6 Hz, C_3-H), 5.88 (1H, d, J = 3.5 Hz, C_2-H), 6.81 and 6.95 (2H, two brs, C_{11}-H, C_{13}-H), 7.22 (1H, s, C_7-H); IR (KBr) 3470, 1665, 1620, 1570, 1230 cm^{-1}; MS m/z 441 (M^+), 354; High-resolution MS (M^+) 411.1403 (411.1421 calcd. for $C_{23}H_{23}NO_8$).

8-Methoxyhomophthalic Anhydride (29). According to the reported method,[7,11] this compound was prepared by the sequence of (1) condensation of crotonaldehyde and ethyl acetoacetate (44%),[11] (2) aromatization of ethyl 6-methyl-2-oxo-3-cyclohexene-1-carboxlate (56%),[11] (3) methylation of ethyl 2-hydoxy-6-methylbenzoate,[11] (4) hydroylsis (86%, 2 steps),[11] (5) introduction of the carboxyl group into the C_6-methyl group of 2-methoxy-6-methylbenzoic acid (95%),[7] and (6) dehydration of 2-carboxy-3-methoxybenzene-1-acetic acid (96%).[7] The anhydride (**29**) was obtained as colorless crystals, mp 136-139 °C [lit. 133-140 °C[7]].

(2R,3S,4R,5R,6R)-(+)-3,5-Diacetoxy-4-dimethylamino-8,10-dihydroxy-11-methoxy-6-

methyl-2,6-epoxy-3,4,5,6-tetrahydro-2H-naphthaceno[1,2-b]oxocine-9,16-dione [(+)-2',4'-Di-*O*-acetyl-9-demethyl-7-methoxynogarene] (33). A solution of **29** (0.15 g, 0.68 mmol) in THF (2.0 ml) was added to a stirred suspension of NaH (24 mg, 50% dispersion in oil, 0.50 mmol) in THF (3.0 ml) at room temperature under an argon atmosphere. After stirring for 20 min, **5** (0.15 g, 0.34 mmol) was added to the reaction mixture cooled in an ice bath and stirring was continued at ambient temperature for 1 h. The mixture was neutralized to pH 8 with 1M HCl and extracted with AcOEt. The combined extracts were washed successively with saturated aqueous $NaHCO_3$ and brine, dried (Na_2SO_4), filtered, and concentrated *in vacuo*. Column chromatography (SiO_2, AcOEt-$CHCl_3$) of the residue gave pure **33** as a red solid (0.12 g, 62%, from **5**). Recrystallization from EtOH-hexane afforded an analytical sample of **33** as red crystals, mp 175-178 °C and $[\alpha]_D^{20}$ +533° (c 0.030, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.61 (3H, s, C_6-Me), 2.15 and 2.19 (6H, two s, COMe x 2), 2.28 (6H, s, NMe_2), 2.71 (1H, t, J = 10.7 Hz, C_4-H), 4.10 (3H, s, OMe), 5.14 (1H, d, J = 10.7 Hz, C_5-H), 5.28 (1H, dd, J = 4.0 and 10.7 Hz, C_3-H), 5.91 (1H, d, J = 4.0 Hz, C_2-H), 7.07 (1H, s, C_7-H or C_{15}-H), 7.06 and 7.56 (2H, two d, each J = 7.7 Hz, C_{12}-H, C_{14}-H), 7.66 (1H, t, C_{13}-H), 8.17 (1H, s, C_7-H or C_{15}-H), 12.64 and 14.79 (2H, two s, OH x 2); IR (KBr) 3450, 1750, 1670, 1635, 1595, 1230, 1040 cm^{-1}; MS m/z 495 (M^+), 435, 307. *Anal.* Calcd. for $C_{26}H_{25}NO_9 \cdot 0.5H_2O$: C, 61.90; H, 5.19; N, 2.78%. Found: C, 61.73; H, 5.12; N, 2.72%.

(2R,3S,4R,5R,6R)-(+)-4-Dimethylamino-3,5,8,10-tetrahydro-11-methoxy-6-methyl-2,6-epoxy-3,4,5,6-tetrahydro-2H-naphthaceno[1,2-b]oxocine-9,16-dione [(+)-9-Demethyl-7-methoxynogarene] (34). Anhydrous K_2CO_3 (50 mg, 0.36 mmol) was added to a solution of **33** (15 mg, 25 μmol) in MeOH (1.0 ml) at room temperature under an argon atmosphere. The reaction mixture was heated at 40 °C for 30 min and cooled to ambient temperature. Oxalic acid was added to the reaction mixture until the color changed from purple to orange. The mixture was diluted with $CHCl_3$ and the chloroform solution was washed successively with saturated aqueous $NaHCO_3$ and brine, dried (Na_2SO_4), and filtered. Concentration *in vacuo*, followed by separation by column chromatography (SiO_2, EtOH-$CHCl_3$), gave crude **34** as an orange solid. This was recrystalized from EtOH-hexane to give pure **34** as orange crystals (10 mg, 78%), mp 230-235 °C and $[\alpha]_D^{20}$ +400° (c 0.030, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.77 (3H, s, C_6-Me), 2.56 (6H, s, NMe_2), 2.92 (1H, t, J = 10.2 Hz, C_4-H), 3.61 (1H, d, J = 10.2 Hz, C_5-H), 4.12 (3H, s, OMe), 4.19 (1H, dd, J = 3.5 and 10.2 Hz, C_3-H), 5.96 (1H, d, J = 3.5 Hz, C_2-H), 6.88-7.32 (4H, m, C_7-H, C_{12}-H, C_{14}-H, C_{15}-H), 7.64 (1H, t, J = 8.0 Hz, C_{13}-H), 12.06 (1H, s, ArOH), 13.84 (1H, brs, ArOH); IR (KBr) 3450, 1660, 1615, 1590, 1570, 1450, 1270, 1230, 1100 cm^{-1}; MS m/z 507 (M^+), 420. *Anal.* Calcd. for $C_{27}H_{25}NO_9 \cdot 0.8H_2O$: C, 62.12; H, 5.14; N, 2.68%. Found: C, 61.83; H, 4.85; N, 2.61%.

8-Methoxy-6-methylhomophthalic Anhydride (30). According to the reported method,[7] this compound was prepared by the sequence of (1) cycloaddition of 6-methoxy-4-methyl-2-pyrone with dimethyl allene-1,3-dicarboxylate,[7] (2) hydrolysis of methyl 2-carbomethoxy-3-methoxybenzene-1-acetate,[7] and (3) dehydration of 2-carboxy-3-methoxy-5-methylbenzene-1-acetic acid (39%, 3 steps).[7] The anhydride (**30**) was obtained as colorless crystals, mp 169-172 °C [lit. 171-173 °C[7]].

(2R,3S,4R,5R,6R)-(+)-3,5-Diacetoxy-4-dimethylamino-8,10-dihydroxy-11-methoxy-6,13-dimethyl-2,6-epoxy-3,4,5,6-tetrahydro-2H-naphthaceno[1,2-b]oxocine-9,16-dione [(+)-2',4'-Di-*O*-acetyl-7-methoxynogarene] (35). A solution of **30** (62 mg, 0.20 mmol) in THF (1.0 ml) was added to a stirred suspension of NaH (12 mg, 50% dispersion in oil, 0.25 mmol) in THF (2.0 ml) at room temperature under an argon atmosphere. After stirring for 20 min, **5** (0.10 g, 0.22 mmol) was added to the reaction mixture cooled in an ice bath and stirring was continued at ambient tempera-

ture for 1 h. The mixture was worked up in the same manner as that described for **33** to give crude **35** after concentration of the combined ethyl acetate extracts *in vacuo*. Separation by column chromatography (SiO_2, AcOEt-$CHCl_3$) and recrystallization from EtOH-hexane gave pure **35** as red crystals (0.11 g, 81%, from **5**), mp 250-254 °C and $[\alpha]_D^{20}$ +545° (c 0.110, $CHCl_3$). ^1H NMR (400 MHz, $CDCl_3$) δ 1.60 (3H, s, C_6-Me), 2.14 and 2.18 (6H, two s, COMe x 2), 2.28 (6H, s, NMe_2), 2.53 (3H, s, C_{13}-Me), 2.70 (1H, t, J = 10.8 Hz, C_4-H), 4.08 (3H, s, OMe), 5.13 (1H, d, J = 10.8 Hz, C_5-H), 5.22 (1H, dd, J = 4.0 and 10.8 Hz, C_3-H), 5.90 (1H, d, J = 4.0 Hz, C_2-H), 6.87 (1H, brs, C_{12}-H or C_{14}-H), 7.04 (1H, s, C_7-H or C_{15}-H), 7.33 (1H, brs, C_{12}-H or C_{14}-H), 8.06 (1H, s, C_7-H or C_{15}-H), 12.59 and 14.69 (2H, two s, OH x 2); IR (KBr) 3460, 1750, 1670, 1615, 1590, 1450, 1380, 1370, 1355, 1220, 1120, 1040 cm^{-1}; MS m/z 605 (M^+), 546, 545. *Anal.* Calcd. for $C_{32}H_{31}NO_{11}\cdot 0.5H_2O$: C, 62.53; H, 5.25; N, 2.28%. Found: C, 62.50; H, 5.14; N, 2.18%.

(2R,3S,4R,5R,6R)-(-)-4-Dimethylamino-3,5,8,10-tetrahydroxy-11-methoxy-6,13-dimethyl-2,6-epoxy-3,4,5,6-tetrahydro-2H-naphthaceno[1,2-b]oxocine-9,16-dione [(-)-7-Methoxynogarene] (36). The same transesterification of **35** (20 mg, 33 μmol) as that described for **33**, afforded crude **36** after concentration of the chloroform solution *in vacuo*. Separation by column chromatography (SiO_2, EtOH-$CHCl_3$), followed by recrystallization from EtOH-hexane, gave pure **36** as red crystals (13 mg, 72%), mp 231-235 °C (decomp.) and $[\alpha]_D^{20}$ -900° (c 0.070, $CHCl_3$). ^1H NMR (400 MHz, $CDCl_3$-D_2O) δ 1.77 (3H, s, C_6-Me), 2.53 (6H, s, NMe_2), 2.54 (3H, s, C_{13}-Me), 2.85 (1H, t, J = 10.4 Hz, C_4-H), 3.60 (1H, d, J = 10.4 Hz, C_5-H), 4.09 (3H, s, OMe), 4.17 (1H, dd, J = 3.5 and 10.4 Hz, C_3-H), 5.95 (1H, d, J = 3.5 Hz, C_2-H), 6.86, 7.06, and 7.27 (4H, three s, C_7-H, C_{12}-H, C_{14}-H, C_{15}-H); IR (KBr) 3455, 1665, 1610, 1590, 1450, 1390, 1355, 1270, 1225, 1120 cm^{-1}; MS m/z 521 (M^+), 434. *Anal.* Calcd. for $C_{28}H_{27}NO_9\cdot 0.8H_2O$: C, 62.75; H, 5.27; N, 2.61%. Found: C, 62.78; H, 5.12; N, 2.54%.

dl-*tert*-Butyl 2-*tert*-Butoxycarbonyl-5-hydroxy-3-oxo-1-cyclohexene-1-acetate (43). *tert*-Butyl lithioacetate (0.69 g, 5.7 mmol)[12] was added to a stirred solution of diethyl 3-hydroxyglutarate (86 mg, 0.42 mmol) in THF (11 ml) in an ice bath under an argon atmosphere. After stirring for 30 min at 0 °C, the reaction mixture was allowed to warm up to room temperature and stirring was further continued for 30 min. The mixture was concentrated *in vacuo* and the residue was dissolved in AcOEt. The ethyl acetate solution was neutralized to pH 1 with 1M HCl. The upper organic layer was separated and the lower aqueous layer was further extracted with AcOEt. The combined extracts were washed with brine, dried (Na_2SO_4), filtered, and concentrated *in vacuo*. The residue was dissolved in MeOH (5.0 ml) and calcium acetate monohydrate (0.90 g, 5.1 mmol) was added to the methanol solution at room temperature. After stirring for 15 h, the reaction mixture was diluted with water and extracted with ether. The ethereal extracts were combined, washed with brine, and dried (Na_2SO_4). Filtration and concentration *in vacuo*, followed by separation by column chromatography (SiO_2, AcOEt-hexane), gave pure **43** as a colorless solid (0.12 g, 90%). Recrystallization from hexane afforded an analytical sample of **43** as colorless crystals, mp 83-84 °C. ^1H NMR (90 MHz, $CDCl_3$) δ 1.47 and 1.53 (18H, two s, $CO_2{}^tBu$ x 2), 1.87 (1H, brs, OH), 2.5-2.8 (4H, m, C_4-H_2, C_6-H_2), 3.30 (2H, CH_2CO_2), 4.2-4.5 (1H, m, C_5-H); IR (KBr) 3450, 1730, 1710, 1660, 1620 cm^{-1}; MS m/z 327 (M^+H), 270, 214. *Anal.* Calcd, for $C_{17}H_{26}O_6$: C, 62.56; H, 8.03%. Found: C, 62.59; H, 8.11%.

***tert*-Butyl 2-*tert*-Butoxycarbonyl-3,5-dihydroxybenzene-1-acetate (44).** Jones reagent was added to a solution of **43** (0.34 g, 1.1 mmol) in acetone (21 ml) in an ice bath until the red color persisted. After stirring for 2 h, the excess Jones reagent was destroyed by the addition of isopropyl alcohol. The mixture was

diluted with water and concentrated *in vacuo*. The residue was extracted with ether and the ethereal extracts were combined, washed successively with saturated aqueous $NaHCO_3$ and brine, and dried (Na_2SO_4). Filtration and concentration *in vacuo* afforded pure **44** as a colorless solid (0.30 g, 89%). Recrystallization from benzene-hexane gave an analytical sample of **44** as colorless crystals, mp 124-126 °C. ^{1}H NMR (90 MHz, $CDCl_3$) δ 1.43 and 1.60 (18H, two s, $CO_2{}^tBu$ x 2), 3.88 (2H, s, CH_2CO_2), 5.23 (1H, s, C_5-OH), 6.18 and 6.36 (2H, two d, each J = 3 Hz, C_4-H, C_6-H), 11.85 (1H, s, C_3-OH); IR (KBr) 3330, 1700, 1640, 1590 cm^{-1}; MS m/z 324 (M^+), 268. *Anal.* Calcd. for $C_{17}H_{24}O_6$; C, 62.95; H, 7.46%. Found: C, 62.83; H, 7.40%.

***tert*-Butyl 5-Benzyloxy-2-*tert*-butoxycarbonyl-3-hydroxybenzene-1-acetate (45).** A mixture of **44** (0.39 g, 1.2 mmol), benzyl bromide (0.29 g, 1.7 mmol), and $K_2CO_3 \cdot 1.5H_2O$ (1.4 g, 8.4 mmol) in acetone (50 ml) was heated at reflux for 3 h under an argon atmosphere. The reaction mixture was concentrated *in vacuo* and the residue was diluted with water. The mixture was extracted with ether and the ethereal extracts were combined, washed successively with water and brine, and dried ($MgSO_4$). Filtration and concentration *in vacuo*, followed by separation by column chromatography (SiO_2, AcOEt-hexane), gave pure **45** as a colorless solid (0.42 g, 83%). Recrystallization from hexane afforded an analytical sample of **45** as colorless crystals, mp 83-84 °C. ^{1}H NMR (90 MHz, $CDCl_3$) δ 1.45 and 1.60 (18H, two s, $CO_2{}^tBu$ x 2), 3.90 (2H, s, CH_2CO_2), 5.09 (2H, s, $C\underline{H}_2Ph$), 6.38 and 6.52 (2H, two d, each J = 3 Hz, C_4-H, C_6-H), 7.3-7.5 (5H, m, $CH_2\underline{Ph}$), 11.97 (1H, s, OH); IR (KBr) 3450, 1730, 1650, 1580 cm^{-1}; MS m/z 414 (M^+), 358, 302. *Anal.* Calcd. for $C_{24}H_{30}O_6$: C, 69.55; H, 7.30%. Found: C, 69.83; H, 7.51%.

***tert*-Butyl 5-Benzyloxy-2-*tert*-butoxycarbonyl-3-methoxybenzene-1-acetate (46).** A mixture of **45** (0.31 g, 0.74 mmol), dimethyl sulfate (0.16 g, 1.3 mmol), and anhydrous K_2CO_3 (0.61 g, 4.4 mmol) in acetone (18 ml) was heated at reflux for 3 h under an argon atmosphere. After cooling to room temperature, the reaction mixture was concentrated *in vacuo*. The residue was diluted with water, and the mixture was extracted with ether. The ethereal extracts were combined, washed successively with water and brine, and dried ($MgSO_4$). Filtration and concentration *in vacuo*, followed by purification by column chromatography (SiO_2, AcOEt-hexane), afforded pure **46** as a colorless solid (0.30 g, 95%). Recrystallization from hexane gave an analytical sample of **46** as colorless crystals, mp 80-81 °C. ^{1}H NMR (90 MHz, $CDCl_3$) δ 1.47 and 1.60 (18H, two s, $CO_2{}^tBu$ x 2), 3.62 (2H, s, CH_2CO_2), 3.87 (3H, s, OMe), 5.12 (2H, s, $C\underline{H}_2Ph$), 6.48 and 6.59 (2H, two d, each J = 3 Hz, C_4-H, C_6-H), 7.3-7.5 (5H, m, $CH_2\underline{Ph}$); IR (KBr) 1740, 1700, 1600 cm^{-1}; MS m/z 428 (M^+), 372, 316. *Anal.* Calcd. for $C_{25}H_{32}O_6$: C, 70.07; H, 7.53%. Found: C, 69.80; H, 7.72%.

6-Benzyloxy-8-methoxyhomophthalic Anhydride (31). Trifluoroacetic acid (1.3 g, 11 mmol) was added to a solution of **46** (0.29 g, 0.68 mmol) in CH_2Cl_2 (18 ml) in an ice bath under an argon atmosphere. After stirring at room temperature for 12 h, the reaction mixture was concentrated *in vacuo* and the residue was suspended in toluene (15 ml). Ac_2O (6.5 g, 60 mmol) was added to the toluene suspension at room temperature under an argon atmosphere. The mixture was heated at 100 °C for 20 min, and cooled to ambient temperature. Concentration *in vacuo* gave pure **31** as a colorless solid (0.20 g, 99%). Recrystallization from benzene afforded an analytical sample of **31** as colorless crystals, mp 171-172 °C. ^{1}H NMR (90MHz, $CDCl_3$) δ 3.97 (3H, s, OMe), 4.00 (2H, s, C_3-H_2), 5.20 (2H, s, $C\underline{H}_2Ph$), 6.47 and 6.60 (2H, two d, each J = 3 Hz, C_5-H, C_7-H), 7.45 (5H, s, $CH_2\underline{Ph}$); IR (KBr) 1800, 1740, 1600 cm^{-1}; MS m/z 298 (M^+), 254. *Anal.* Calcd. for $C_{17}H_{14}O_5$: C, 68.45; H, 4.73%. Found: C, 68.26; H, 4.80%.

***tert*-Butyl 2-*tert*-Butoxycarbonyl-3,5-dimethoxybenzene-1-acetate (47).** A mix-

ture of **44** (71 mg, 0.22 mmol), dimethyl sulfate (93 mg, 0.74 mmol), and anhydrous K_2CO_3 (0.36 g, 2.6 mmol) in acetone (6.0 ml) was heated at reflux under an argon atmosphere for 3 h, and cooled to ambient temperature. The mixture was worked up in the same manner as that described for **46**, afforded crude **47** after concentration of the combined ethereal extracts *in vacuo*. This was purified by column chromatography (SiO_2, AcOEt-hexane) to give pure **47** as a colorless solid (76 mg, 98%). Recrystallization from hexane gave an analytical sample of **47** as colorless crystals, mp 65-66 °C. ^{1}H NMR (90 MHz, $CDCl_3$) δ 1.43 and 1.58 (18H, two s, $CO_2{}^tBu$ x 2), 3.60 (2H, s, CH_2CO_2), 3.80 (6H, s, OMe x 2), 6.43 (2H, s, C_4-H, C_6-H); IR (KBr) 1720, 1610, 1590 cm^{-1}; MS m/z 352 (M^+), 296, 240. *Anal.* Calcd. for $C_{19}H_{28}O_6$: C, 64.75; H, 8.01%. Found: C, 64.95; H, 8.14%.

6,8-Dimethoxyhomophthalic Anhydride (32). The successive treatments of **47** (0.12 g, 0.34 mmol) in the same manner as that described for **46** gave crude **32** as a pale yellow solid after concentration of the reaction mixture in toluene *in vacuo*. Trituration of this solid with hexane afforded pure **32** as a colorless solid (75 mg, 98%). Recrystallization from benzene gave an analytical sample of **32** as colorless crystals, mp 167-168 °C. ^{1}H NMR (90 MHz, $CDCl_3$) δ 3.90 (2H, s, C_3-H_2), 4.00 (6H, s, OMe x 2), 6.39 and 6.50 (2H, two d, each J = 3 Hz, C_5-H, C_7-H); IR (KBr) 1780, 1740, 1600, 1580 cm^{-1}; MS m/z 222 (M^+), 194. *Anal.* Calcd. for $C_{11}H_{10}O_5$: C, 59.46; H, 4.54%. Found: C, 59.71; H, 4.64%.

(2R,3S,4R,5R,6R)-(+)-3,5-Diacetoxy-13-benzyloxy-4-dimethylamino-8,10-dihydroxy-11-methoxy-6-methyl-2,6-epoxy-3,4,5,6-tetrahydro-2H-naphthaceno[1,2-b]oxocine-9,16-dione [(+)-2',4'-Di-*O*-acetyl-9-benzyloxy-9-demethyl-7-methoxynogarene] (37). A solution of **31** (0.12 g, 0.40 mmol) in THF (2.0 ml) was added to a stirred suspension of NaH (16 mg, 50% dispersion in oil, 0.33 mmol) in THF (2.0 ml) at room temperature under an argon atmosphere. After stirring for 20 min, **5** (78 mg, 0.18 mmol) was added to the reaction mixture cooled in an ice bath and stirring was continued for 1 h at ambient temperature. The mixture was worked up in the same manner as that described for the preparation of **33**, to give crude **37** after concentration of the combined ethyl acetate extracts *in vacuo*. This was chromatographed (SiO_2, AcOEt-toluene) to afford pure **37** as a red solid (0.12 g, 100%, from **5**). Trituration with EtOH gave an analytical sample of **37** as red crystals, mp 251-254 °C and $[\alpha]_D^{20}$ +706° (c 0.051, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.60 (3H, s, C_6-Me), 2.15 and 2.19 (6H, two s, COMe x 2), 2.71 (6H, s, NMe_2), 2.71 (1H, t, J = 10.7 Hz, C_4-H), 4.04 (3H, s, OMe), 5.14 (1H, d, J = 10.7 Hz, C_5-H), 5.22 (1H, dd, J = 4.0 and 10.7 Hz, C_3-H), 5.22 (2H, s, $C\underline{H}_2Ph$), 5.90 (1H, d, J = 4.0 Hz, C_2-H), 6.72 and 6.97 (2H, two d, each J = 2.2 Hz, C_{12}-H, C_{14}-H), 7.04 (1H, s, C_7-H or C_{15}-H), 7.4-7.5 (5H, m, $CH_2\underline{Ph}$), 8.00 (1H, s, C_7-H or C_{15}-H), 12.64 and 14.71 (2H, two s, OH x 2); IR (KBr) 3450, 1750, 1600 cm^{-1}; MS m/z 697 (M^+), 683, 638. *Anal.* Calcd. for $C_{38}H_{35}NO_{12}\cdot 1.25H_2O$: C, 63.37; H, 5.07; N, 1.94%. Found: C, 63.18; H, 4.90; H, 1.87%.

(2R,3S,4R,5R,6R)-(-)-13-Benzyloxy-4-dimethylamino-3,5,8,10-tetrahydroxy-11-methoxy-6-methyl-2,6-epoxy-3,4,5,6-tetrahydro-2H-naphthaceno[1,2-b]oxocine-9,16-dione [(-)-9-Benzyloxy-9-demethyl-7-methoxynogarene] (38). Anhydrous K_2CO_3 (41 mg, 0.29 mmol) was added to a solution of **37** (15 mg, 20 μmol) in a mixture of $CHCl_3$ (1.0 ml) and MeOH (2.0 ml) at room temperature under an argon atmosphere. After stirring for 5 h, oxalic acid was added to the reaction mixture until the color changed from purple to red. The mixture was diluted with water and extracted with $CHCl_3$. The combined extracts were washed with brine, dried (Na_2SO_4), filtered, and concentrated *in vacuo*. The residue was chromatographed (SiO_2, EtOH-$CHCl_3$) to give pure **38** as a red solid (8.0 mg, 63%). Trituration with EtOH afforded red crystals, mp >270 °C (decomp.) and $[\alpha]_D^{20}$ -1780° (0.009, $CHCl_3$). ^{1}H

NMR (400 MHz, $CDCl_3$) δ 1.77 (3H, s, C_6-Me), 2.53 (6H, s, NMe_2), 2.84 (1H, t, J = 10.7 Hz, C_4-H), 3.59 (1H, d, J = 10.7 Hz, C_5-H), 4.05 (3H, s, OMe), 4.17 (1H, dd, J = 3.5 and 10.7 Hz, C_3-H), 5.18 and 5.37 (2H, two d, each J = 11.2 Hz, $C\underline{H}_2Ph$), 5.92 (1H, d, J = 3.5 Hz, C_2-H), 6.55, 6.71, 7.10, and 7.25 (4H, four s, C_7-H, C_{12}-H, C_{14}-H, C_{15}-H), 7.41-7.58 (5H, m, $CH_2\underline{Ph}$), 12.32 and 13.90 (2H, two s, OH x 2); IR (KBr) 3450, 1600, 1450 cm^{-1}; MS m/z 613 (M^+), 599, 522; High-resolution MS (M^+) 612.1953 (612.1946 calcd. for $C_{34}H_{31}NO_{10}$).

(2R,3S,4R,5R,6R)-(+)-3,5-Diacetoxy-4-dimethylamino-8,10-dihydroxy-11,13-dimethoxy-6-methyl-2,6-epoxy-3,4,5,6-tetrahydro-2H-naphthaceno[1,2-b]oxocine-9,16-dione [(+)-2',4'-Di-*O*-acetyl-9-demethyl-7,9-dimethoxynogarene] (39). A solution of **32** (23 mg, 0.10 mmol) in THF (2.0 ml) was added to a stirred suspension of NaH (4.0 mg, 50% dispersion in oil, 80 μmol) in THF (2.0 ml) at room temperature under an argon atmosphere. After stirring for 20 min at the same temperature, **5** (10 mg, 20 μmol) was added to the reaction mixture cooled in an ice bath and the stirring was continued for 1 h at ambient temperature. The same work-up as that described for the preparation of **33** gave crude **39** after concentration of the combined ethyl acetate extracts *in vacuo*. Purification by column chromatography (SiO_2, AcOEt-toluene) afforded pure **39** as a red solid (13 mg, 96%, from **5**). Trituration with EtOH gave an analytical sample of **39** as red crystals, 174-177 °C and $[\alpha]_D^{20}$ +649° (c 0.111, $CHCl_3$). ^1H NMR (400 MHz, $CDCl_3$) δ 1.60 (3H, s, C_6-Me), 2.15 and 2.18 (6H, two s, COMe x 2), 2.28 (6H, s, NMe_2), 2.70 (1H, t, J = 10.7 Hz, C_4-H), 3.97 and 4.04 (6H, two s, OMe x 2), 5.14 (1H, d, J = 10.7 Hz, C_5-H), 5.22 (1H, dd, J = 4.0 and 10.7 Hz, C_3-H), 5.91 (1H, d, J = 4.0 Hz, C_2-H), 6.61 and 6.87 (2H two d, each J = 2.2 Hz, C_{12}-H, C_{14}-H), 7.04 and 8.00 (2H, two s, C_7-H, C_{15}-H), 12.65 and 14.70 (2H, two s, OH x 2); IR (KBr) 3450, 1750, 1610 cm^{-1}; MS m/z 621 (M^+), 561. *Anal.* Calcd. for $C_{32}H_{31}NO_{12}\cdot 0.5H_2O$: C, 60.95; H, 5.11; N, 2.22%. Found: C, 60.97; H, 5.02; N, 2.20%.

(2R,3S,4R,5R,6R)-(-)-4-Dimethylamino-3,5,8,10-tetrahydroxy-11,13-dimethoxy-6-methyl-2,6-epoxy-3,4,5,6-tetrahydro-2H-naphthaceno[1,2-b]oxocine-9,16-dione [(-)-9-demethyl-7,9-Dimethoxynogarene] (40). The same transesterification of **39** (36 mg, 60 μmol) as that described for **37** gave crude **40** after concentration of the combined chloroform extracts *in vacuo*. Separation by column chromatography (SiO_2, EtOH-$CHCl_3$) afforded pure **40** as a red solid (20 mg, 63%). Trituration with EtOH gave an analytical sample of **40** as red crystals, mp 228-231 °C and $[\alpha]_D^{20}$ -1040° (c 0.025, $CHCl_3$). ^1H NMR (400 MHz, $CDCl_3$) δ 1.76 (3H, s, C_6-Me), 2.53 (6H, s, NMe_2), 2.85 (1H, t, J = 10.5 Hz, C_4-H), 3.59 (1H, d, J = 10.5 Hz, C_5-H), 4.01 and 4.06 (6H, two s, OMe x 2), 4.17 (1H, dd, J = 3.5 and 10.5 Hz, C_3-H), 5.92 (1H, d, J = 3.5 Hz, C_2-H), 6.47, 6.63, 7.10, and 7.22 (4H, four brs, C_7-H, C_{12}-H, C_{14}-H, C_{15}-H), 12.24 and 13.93 (2H, two s, ArOH x 2); IR (KBr) 3450, 1610, 1590 cm^{-1}; MS m/z 537 (M^+), 523, 508. *Anal.* Calcd. for $C_{28}H_{27}NO_{10}\cdot 0.75H_2O$: C, 61.03; H, 5.21; N, 2.54%. Found: C, 61.08; H, 4.97; N, 2.48%.

(2R,3S,4R,5R,6R)-(-)-3,5-Diacetoxy-4-dimethylamino-8,10,13-trihydroxy-11-methoxy-6-methyl-2,6-epoxy-3,4,5,6-tetrahydro-2H-naphthaceno[1,2-b]oxocine-9,16-dione [(-)-2',4'-Di-*O*-acetyl-9-demethyl-9-hydroxy-7-methoxynogarene] (41). A mixture of **37** (0.12 g, 0.18 mmol) and 10% Pd-$BaSO_4$ (0.10 g) in MeOH (16 ml) was stirred for 2 h at room temperature under a hydrogen atmosphere. The catalyst was filtered off and washed with $CHCl_3$. The combined filtrates were concentrated *in vacuo*. The residue was separated by column chromatography (SiO_2, AcOEt-toluene) to give pure **41** as a red solid (86 mg, 79%). Trituration with EtOH gave an analytical sample of **41** as red crystals, mp >290 °C and $[\alpha]_D^{20}$ -2500° (c 0.020, $CHCl_3$). ^1H NMR (400 MHz, $CDCl_3$) δ 1.80 (3H, s, C_6-Me), 1.93 and 2.13 (6H, two s, COMe x 2), 2.24 (6H, s, NMe_2), 2.61 (1H, t, J = 10.7 Hz, C_4-H), 4.10 (3H, s, OMe), 5.17

(1H, d, J = 10.7 Hz, C_5-H), 5.29 (1H, dd, J = 3.7 and 10.7 Hz, C_3-H), 5.87 (1H, d, J = 3.7 Hz, C_2-H), 6.15, 7.09, 7.78, and 7.96 (4H, four s, C_7-H, C_{12}-H, C_{14}-H, C_{15}-H), 8.86 (1H, brs, C_{13}-OH), 13.63 and 14.45 (2H, two s, C_8-OH, C_{10}-OH); IR (KBr) 3450, 1750, 1600 cm^{-1}; MS m/z 607 (M^+), 548, 506. *Anal.* Calcd. for $C_{31}H_{29}NO_{12}\cdot 0.5H_2O$: C, 60.38; H, 4.90; N, 2.27%. Found: C, 60.21; H, 4.67; N, 2.25%.

(2R,3S,4R,5R,6R)-4-Dimethylamino-3,5,8,10,13-pentahydroxy-11-methoxy-6-methyl-2,6-epoxy-3,4,5,6-tetrahydro-2H-naphthaceno[1,2-b]oxocine-9,16-dione [9-Demethyl-9-hydroxy-7-methoxynogarene] (42). Transesterification of **41** (13 mg, 20 μmol) in the same manner as that described for **37** gave crude **42** after concentration of the combined chloroform extracts *in vacuo*. This was separated by column chromatography (SiO_2, EtOH-$CHCl_3$) to give **42** as a red solid (2.1 mg, 18%). ^{1}H NMR (400 MHz, CD_3OD) δ 1.72 (3H, s, C_6-Me), 2.71 (6H, s, NMe_2), 3.04 (1H, brs, C_4-H), 3.80 (1H, d, J = 10.7 Hz, C_5-H), 3.96 (3H, s, OMe), 4.13 (1H, brd, J = 10.7 Hz, C_3-H), 5.83 (1H, brs, C_2-H), 6.47, 6.57, 7.03, and 7.15 (4H, four s, C_7-H, C_{12}-H, C_{14}-H, C_{15}-H); IR (KBr) 3450, 1600, 1580 cm^{-1}; MS m/z 523 (M^+), 509; High-resolution MS (M^+) 523.1467 (523.1476 calcd. for $C_{27}H_{25}NO_{10}$). Since this compound (**42**) was found to be unstable, further purification could not be attempted. Since **42** was sufficiently pure (*ca.* 90% by ^{1}H NMR), it was directly subjected to *in vitro* cytotoxicity assay.

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References and Notes

1) Parts of this series of reports have been the subjects of four preliminary communications: a) M. Kawasaki, F. Matsuda, and S. Terashima, *Tetrahedron Lett.*, **26**, 2693 (1985); b) *Idem*, *ibid.*, **27**, 2145 (1986); c) *Idem*, *ibid.*, **29**, 791 (1988); d) F. Matsuda, M. Kawasaki, M. Ohsaki, K. Yamada, and S. Terashima, *Chem. Lett.*, 653 (1988).
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