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Synthetic Studies on Duocarmycin. 1. Total Synthesis of *dl*-Duocarmycin A and Its 2-Epimer[1]

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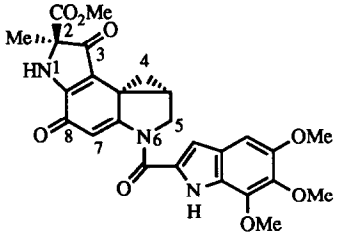
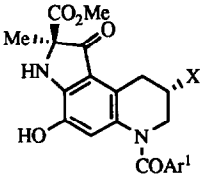
Key Words: dl-duocarmycin A, dl-2-epi-duocarmycin A, total synthesis, antitumor antibiotic, cytotoxicity

Abstract: The title synthesis was first achieved by employing novel methoxycarbonylation of the C_4-position of the 5-aminoindoline by way of the isatin and subsequent Dieckmann cyclization to the methyl 2-methylindoxyl-2-carboxylate as key steps. *In vitro* cytotoxicity assay against P388 murine leukemia obviously disclosed that cytotoxicities of the synthesized compounds are comparable and almost half of that of natural (+)-duocarmycin A.

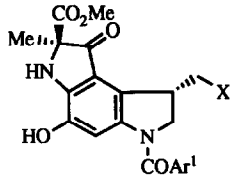
Duocarmycin A (**1**)[3] isolated from *Streptomyces sp.* by a research group at Kyowa Hakko, is an extremely potent antitumor antibiotic which is effective against various strains of experimental cancer cell lines, and represents one of the novel classes of antitumor compounds, the duocarmycin family involving duocarmycins C_1 (**2**), C_2 (**3**), B_1 (**4**) and B_2 (**5**) isolated along with **1**.[4] It was found that the antibiotics, pyrindamycin A and B,[5] independently isolated at almost the same time by workers at Meiji Seika from the culture broth of *Streptomyces sp.* were identical with **3** and **2**, respectively.[3c] Recently, duocarmycin SA (**6**) showing improved stability and antibacterial as well as cytotoxic activity at lower concentrations than those for **1** has been also reported.[6]



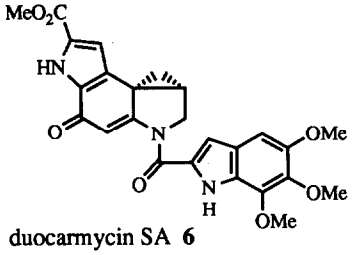
duocarmycin A **1**



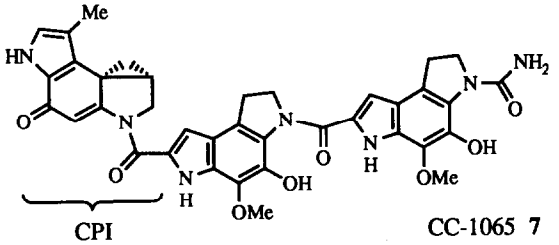
duocarmycin C_1 **2** : X = Cl
(pyrindamycin B)
duocarmycin B_1 **4** : X = Br



duocarmycin C_2 **3** : X = Cl
(pyrindamycin A)
duocarmycin B_2 **5** : X = Br



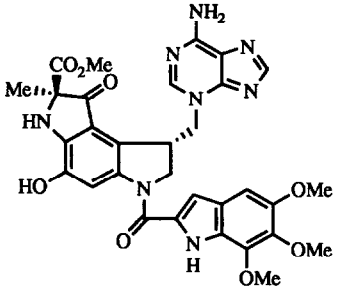
duocarmycin SA **6**



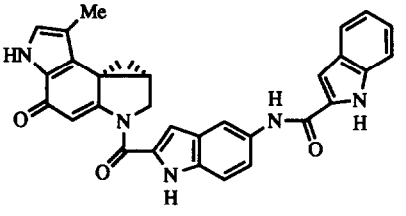
CPI

CC-1065 **7**

The striking structural feature of **1** is the close resemblance of its upper half to the left hand segment, so called cyclopropapyrroloindole (CPI), of the potent antitumor antibiotic, (+)-CC-1065 (**7**).[7] This CPI unit has been recognized to be responsible for the biological activities of **7** and demonstrated to be the site of nucleophilic attack by adenine N3 in DNA.[8] Therefore, the biological activities of **1** were also supposed to be deeply related to the upper half segment involving an electrophilic cyclopropadienone system.[8c] In fact, this speculation has been rigorously supported by recent studies on the covalent alkylation of DNA with **1** resulting in the isolation of duocarmycin A - adenine adduct (**8**).[9]

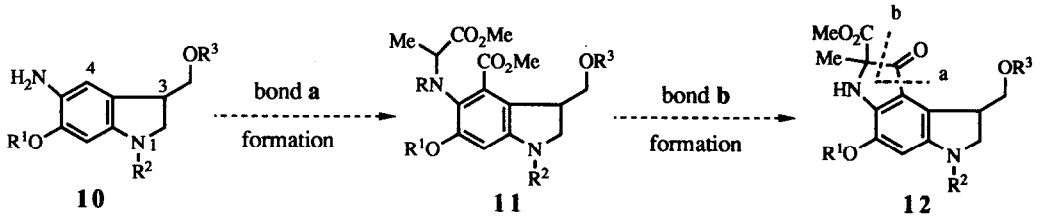


duocarmycin A - adenine adduct **8**



U-71,184 **9**

In spite of its extremely potent antitumor activity, **7** could not be developed as an anticancer agent because of its unusual delayed lethality.[10] Extensive investigations, however, culminated in development of the less toxic analogue of **7**, U-71,184 (**9**),[11] consisting of CPI and modified middle and right hand segments. This successful example clearly suggests the possibility to control antitumor activity as well as toxicity of duocarmycin analogues by modification of the lower half segment. With these notable aspects in mind, we embarked on the synthesis of **1** and its analogues to explore prominent anticancer agents. We, herein, disclose the details of our recent accomplishment, the first total synthesis of *dl*-duocarmycin A (*dl*-**1**) and its 2-epimer [*dl*-2-epiduocarmycin A (*dl*-2-epi-**1**)].[1] The synthetic scheme developed in these studies was also applied to the successful synthesis of optically active (+)-**1** and its three possible stereoisomers [(-)-**1**, (+)-2-epi-**1**, and (-)-2-epi-**1**]. This is the subject of the accompanying paper.[12,13]



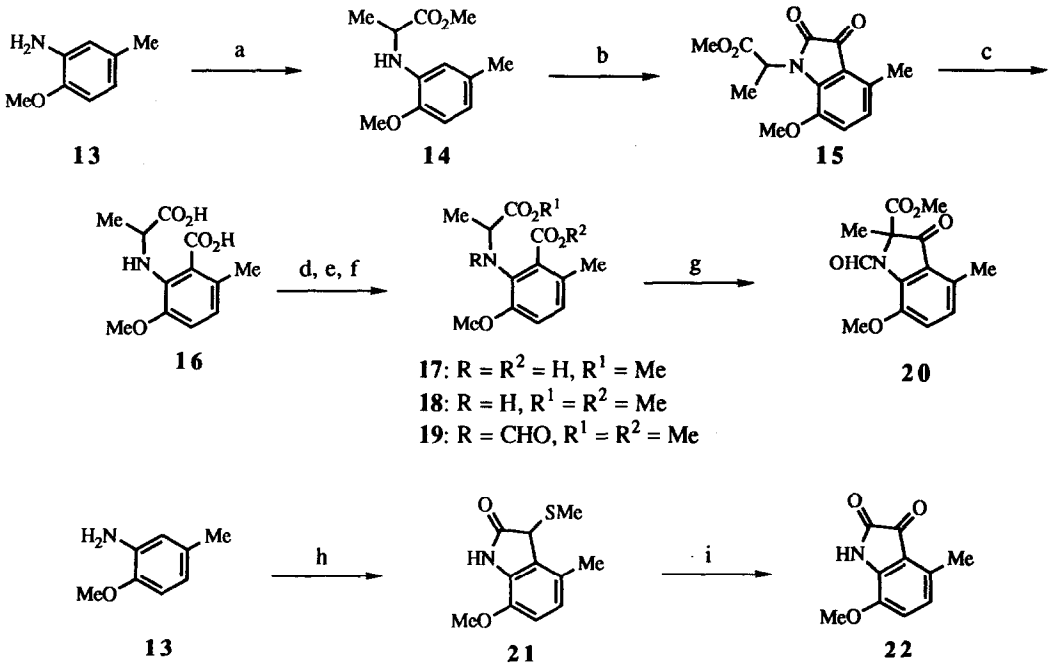
10 **11** **12**

Scheme 1

Taking into account potential applications of the explored synthetic route to preparation of various analogues of **1**, the synthetic scheme was designed in which amide formation between the upper half of **1** with 5,6,7-trimethoxyindole-2-carboxylic acid (**40**) is examined after functionalizations required for crucial Wierenga-Kelly-Winstein Ar-3' cyclization have been completed.[14] Since construction of the 5-aminoindoline derivative such as **10** has been well investigated in the previous synthetic studies on **7**,[15] our synthetic efforts were primarily focused on elaboration of the 2-methylindoxyl-2-carboxylate (**12**) system characterizing the structure of **1**. To construct **12** from **10**, we examined the synthetic scheme involving initial introduction of one carbon unit into the C_4-position by Friedel-Crafts-type reaction (by bond **a** formation) followed by the Dieckmann cyclization of resulting diester (**11**) (by bond **b** formation) as shown in **Scheme 1**.

Model studies for the upper half of 1

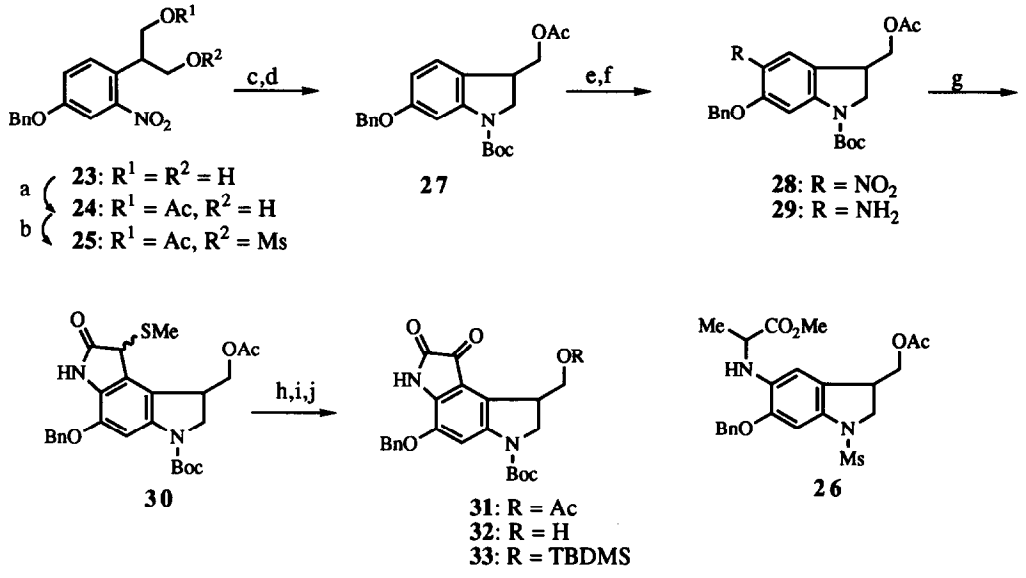
To explore feasibility of the synthetic plan, we first examined model studies using commercially available 6-methoxy-3-methylaniline (**13**). After experimentation, it was found that introduction of one carbon unit can be successfully achieved by way of isatin derivative (**15**) obtainable by the Friedel-Crafts reaction of *N*-alkyl derivative (**14**) with oxalyl chloride (**Scheme 2**).



Scheme 2
Conditions: a) Methyl 2-bromopropionate, Et_2NPh, 95% b) $(COCl)_2$, $TiCl_4$, 58% c) KOH, H_2O_2 d) $SOCl_2$, MeOH e) Me_2SO_4, K_2CO_3, DMF, 71% 3 steps from **15** f) HCO_2H, Ac_2O, 98% g) LDA, THF, -78 °C, 40% h) 1. SO_2Cl_2, $CH_3SCH_2CO_2Me$, CH_2Cl_2, then **13**, 1,8-bis(dimethylamino)naphthalene, -78 °C, 3.5h 2. Et_3N, -78 °C, 2h, rt, 21h 3. AcOH, rt, 1h, 81% i) NCS, $CuCl_2$, CuO, acetone, 83%.

Thus, monoalkylation of **13** with methyl 2-bromopropionate followed by treatment of resulting **14** with oxalyl chloride in the presence of $TiCl_4$ gave rise to **15**. Oxidative cleavage of the dicarbonyl moiety of **15** afforded diacid (**16**), of which two carboxyl groups were sequentially methylated to afford dimethyl ester (**18**). Prior to subsequent Dieckmann cyclization, the secondary amino group in **18** was protected with a formyl group, yielding the formamide (**19**). Although possible decarbomethoxylation was anticipated for the resulting non-enolizable β-keto ester (**20**),[16] treatment of **19** with LDA in THF at -78 °C smoothly underwent the Dieckmann cyclization to give a reasonable yield of **20** involving the 2-methylindoxyl-2-carboxylate system of **1**.

On the other hand, isatin derivative (**22**) could be also prepared by featuring the Gassman's oxindole synthesis[17] followed by oxidation of the resulting 3-methylthiooxindole.[18] Thus, the reaction of **13** with the chlorosulfonium salt of methyl (methylthio)acetate followed by base-promoted [2,3]-sigmatropic rearrangement and acid-catalyzed ring closure cleanly produced the 3-methylthiooxindole (**21**). Oxidation of **21** to **22** was effectively achieved by chlorination of **21** with *N*-chlorosuccinimide followed by treatment with a combination of cupric chloride and cupric oxide.[18]



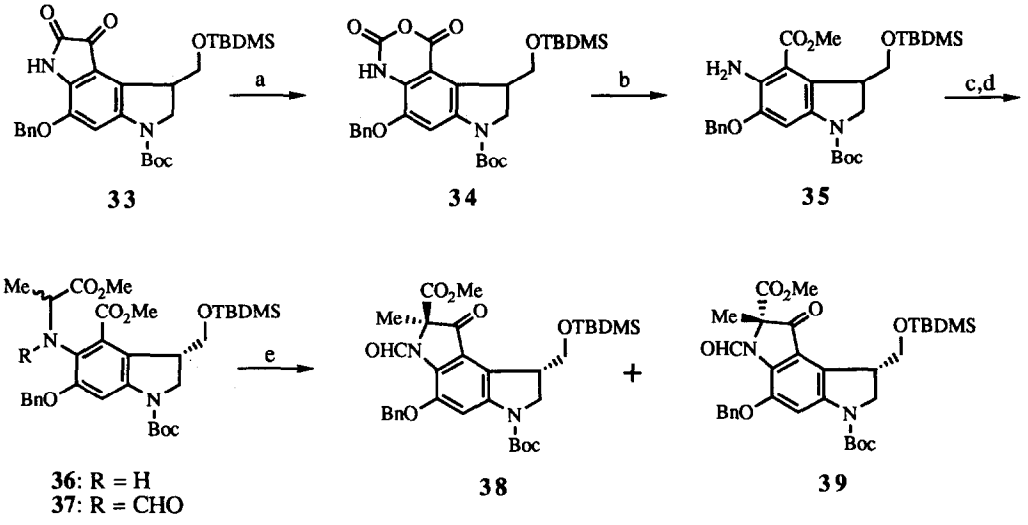
Scheme 3
Conditions: a) Ac_2O, Et_3N, CH_2Cl_2, 0 °C, 2.5h, 74% b) MsCl, Et_3N, CH_2Cl_2, 0 °C, 0.5h, 100% c) H_2 (3 atm), PtO_2, Et_3N, THF, rt, 20min d) Boc_2O, CH_2Cl_2, rt, 12h, 95% e) HNO_3, Ac_2O, then **27**, CH_3NO_2, -20 °C, 3h, 77% f) H_2 (3 atm), PtO_2, THF, rt, 15min 95% g) 1. SO_2Cl_2, $CH_3SCH_2CO_2Me$, CH_2Cl_2, then **29**, 1,8-bis(dimethylamino)naphthalene, -78 °C, 3.5h 2. Et_3N, -78 °C, 2h, rt, 21h 3. AcOH, rt, 1h h) $CuCl_2$, CuO, acetone, rt, 1.5h, 77% from **29** i) K_2CO_3, MeOH, rt, 2.5h j) TBDMSCl, imidazole, DMF, rt, 8h, 90% from **31**.

Synthesis of the upper half of 1

Having established the synthetic scheme to construct the 2-methylindoxyl-2-carboxylate system involved in **1**, we then initiated our synthesis of *dl*-**1** from the known diol (**23**).[11a] Thus, monoacetylation of the symmetrical diol moiety of **23** and mesylation of the remaining alcohol provided mesylate (**25**). Since, in other

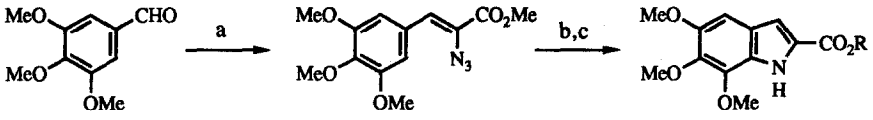
experiments, we experienced the unexpected difficulties to obtain the isatin derivative by the Friedel-Crafts reaction of the 1-mesylindoline (**26**) with oxalyl chloride which had been established in the model studies (e.g. **14** → **15**), we decided to use a t-Boc group to protect the indoline nitrogen and to employ the Gassman's protocol for further synthetic elaboration. Reduction of the nitro group in **25** simultaneously effected the indoline formation, affording the 1-t-butoxycarbonylindoline (**27**) after *in situ* protection of the generated secondary amino group. Nitration of **27** with acetyl nitrate cleanly occurred at the C_5-position (the indoline numbering) to give nitro compound (**28**), which in turn was reduced to the 5-aminoindoline (**29**). Treatment of **29** with the chlorosulfonium salt of methyl (methylthio)acetate and subsequent manipulations described in model studies produced the 3-methylthiooxindole (**30**). Being different from the model studies, oxidation of **30** to isatin derivative (**31**) was found to be effected by treating directly with a combination of cupric chloride and cupric oxide. The acetyl group of **31** was removed by methanolysis and subsequent protection of the resulting primary alcohol with a t-butyldimethylsilyl (TBDMS) group afforded silyl ether (**33**) (**Scheme 3**).

With **33** in hand, we then examined its transformation into dimethyl ester (**37**) by alkylation at the N_1-nitrogen in **33** followed by oxidative opening of the isatin ring. Although alkylation of **33** with methyl 2-bromopropionate proceeded only in a low yield due to its instability under basic conditions, desired **37** could be obtained by way of the anthranilic acid ester (**35**). Thus, oxidation of **33** with m-chloroperbenzoic acid (m-CPBA) cleanly produced the isatoic anhydride (**34**), which without separation was transformed into **35** by methanolysis. After extensive investigations on alkylation conditions, it was finally found that the amino group of **35** could be alkylated with methyl 2-bromopropionate in the presence of 1,8-bis(dimethylamino)naphthalene in *N,N*-dimethylacetamide (DMAC) at 70 °C for 2 days to give the alkylation product (**36**) in a good yield.



Scheme 4
Conditions: a) m-CPBA, $NaHCO_3$, CH_2Cl_2, -15 °C, 2h b) K_2CO_3, MeOH, 10 °C, 1h, 94% from **33** c) $CH_3CHBrCO_2Me$, 1,8-bis(dimethylamino)naphthalene, DMAC, 70 °C, 48h, 88% d) HCO_2H, Ac_2O, then **36**, rt, 9h, 93% e) LDA, THF, -78 °C, 5.5h, more polar isomer **38** 28%, less polar isomer **39** 28%.

The resulting secondary amino group was then protected with a formyl group to produce the formamide (**37**). The Dieckmann cyclization of **37** was successfully carried out under similar conditions to those described in the model studies, affording a diastereomeric mixture of β-keto esters (**38** and **39**), which were separable by preparative TLC to give more polar **38** and less polar **39**, respectively (**Scheme 4**). Although their relative stereochemistries could not be assigned at this stage, they were later definitely confirmed by successful elaboration of more polar **38** to *dl*-**1** as shown in **Scheme 6**.



41 **42** **43**: R = Me
40: R = H

Scheme 5
Conditions: a) $N_3CH_2CO_2Me$, NaOMe, MeOH, 0 °C, 5.5h, 97% b) xylene reflux, 1h, 94% c) 40% KOH, MeOH, 50 °C, 0.5h, 96%.

Having completed construction of the upper half of **1**, we, then, focused our efforts on coupling of **38** and **39** with 5,6,7-trimethoxyindole-2-carboxylic acid (**40**) and subsequent formation of the cyclopropadienone systems to accomplish the projected synthesis of *dl*-**1** and *dl*-2-epi-**1**. 5,6,7-Trimethoxyindole-2-carboxylic acid (**40**) was prepared from 3,4,5-trimethoxybenzaldehyde (**41**) in a straightforward fashion as follows.[19,20] Thus, condensation of **41** with methyl 2-azidoacetate produced unsaturated azido ester (**42**). Upon heating **42** in refluxing xylene, nitrene formation and subsequent C-H bond insertion took place, producing the indole (**43**), hydrolysis of the ester moiety of which afforded **40** in a good yield (**Scheme 5**).

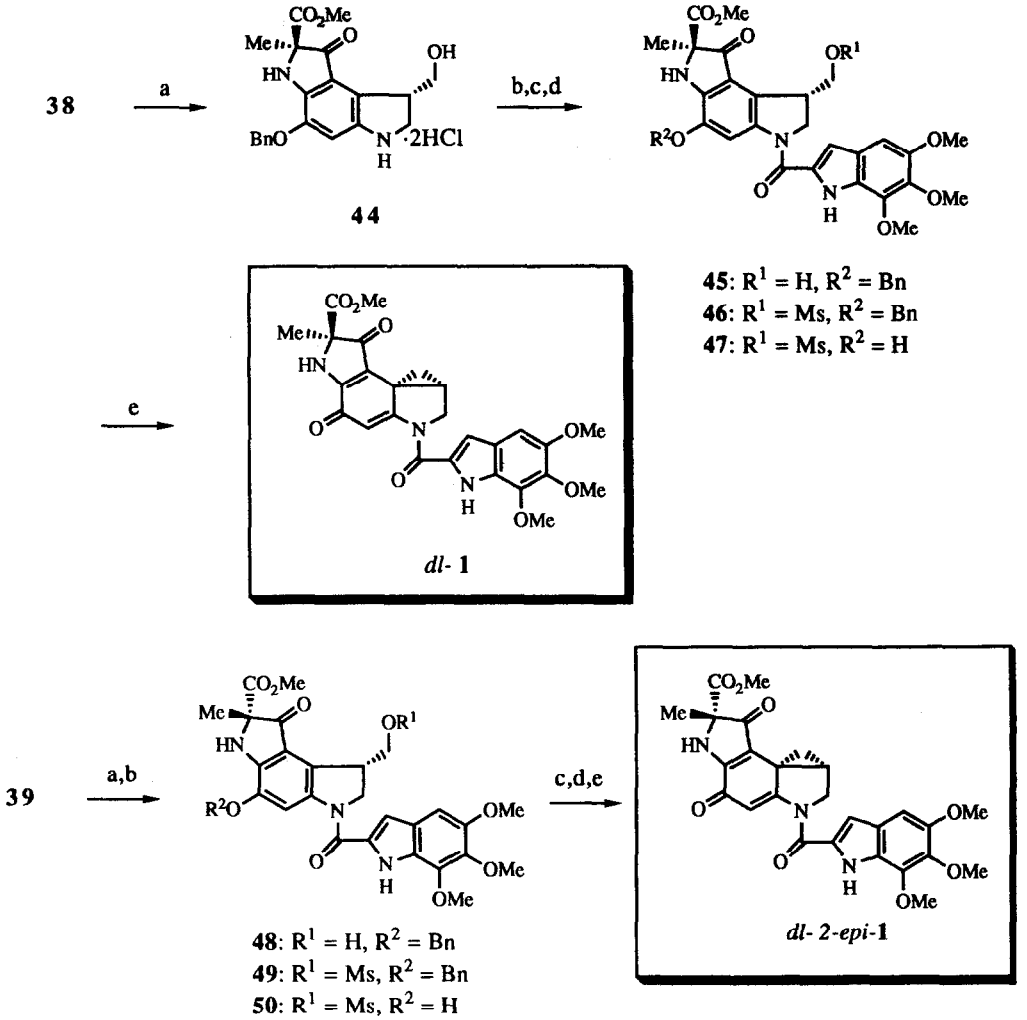
As expected, coupling of diamino alcohol dihydrochloride (**44**) prepared by acid hydrolysis of **38**, with **40** in the presence of 1-[3-(dimethylamino)propyl]-3-ethylcarbodiimide hydrochloride occurred at the less hindered nitrogen, giving monoamide (**45**) as a sole product. After mesylation of the primary alcohol of **45**, deprotection of the benzyl ether of resulting mesylate (**46**) gave rise to the phenol (**47**). Finally, formation of the cyclopropadienone system was effected by treatment of **47** with NaH to furnish *dl*-**1** (**Scheme 6**). Similarly, the less polar β-keto ester (**39**) was transformed into *dl*-2-*epi*-**1** *via* alcohol (**48**).

With *dl*-**1** and *dl*-2-*epi*-**1** in hand, they were subjected to *in vitro* cytotoxicity assay against P388 murine leukemia, providing the following IC_{50} values (ng/ml); *dl*-**1**, 0.25; *dl*-2-epi-**1**, 0.17; natural (+)-**1**, 0.079. These results clearly indicate that relative stereochemistry between the C_2-position and the cyclopropane ring has a little effect on the cytotoxicity of *dl*-**1** and *dl*-2-epi-**1**. On the other hand, the observation that *dl*-**1** exhibits the cytotoxicity being almost half of that of natural (+)-**1**, obviously suggests that there could be a significant difference between the cytotoxicity of natural (+)-**1** and unnatural (-)-**1**.

Conclusion

As described above, we have succeeded in the first total synthesis of *dl*-**1** and *dl*-2-epi-**1** by employing novel methoxycarbonylation of the C_4-position of **29** by way of **31** - **34** and subsequent Dieckmann cyclization of **37** as key steps. By comparing cytotoxicity of *dl*-**1** and *dl*-2-epi-**1**, it appeared that relative stereochemistries between the C_2-position and the cyclopropane ring are not important factor for the cytotoxicity. From viewpoint of molecular recognition by DNA, however, the absolute configuration of the cyclopropane moiety seems to

cause significant differentiation between each enantiomer of duocarmycin A [(+)- and (-)-**1**]. It is, thus, considered that the absolute configuration of cyclopropane ring in **1** might play an important factor to control its conformation to bind with DNA minor groove. In the accompanying paper,[12] we disclose the effects of absolute configuration of cyclopropane ring on the cytotoxicity by successful synthesis of (+)-**1** and its three possible stereoisomers [(-)-**1**, (+)-2-epi-**1**, and (-)-2-epi-**1**].



Scheme 6
Conditions: a) HCl in MeOH, rt, 11h, 100% from **38** and **39** b) 1-[3-(dimethylamino)propyl]-3-ethylcarbodiimide hydrochloride, $NaHCO_3$, DMF, rt, 20h, 57% for **45**, 62% for **48** c) MsCl, Et_3N, CH_2Cl_2, 0 °C, 0.5h, 99% for **46** and **49** d) H_2 (1 atm), 10% Pd/C, THF, 85% for **47**, 83% for **50** e) NaH, THF, rt, 3.5h, 60% for *dl*-**1**, 56% for *dl*-2-epi-**1**.

Experimental Section

All melting points were determined with a Yamato MP-21 melting point apparatus and were uncorrected. Optical rotations were measured with a Horiba SEPA-200 automatic digital polarimeter. Infrared (IR) spectra were recorded on a JASCO A-202 spectrometer. ^{1}H-NMR spectra were measured with a Hitachi R-90H (90 MHz) and a Brucker AM 400 (400MHz) spectrometer. The chemical shifts were expressed in parts per million downfield from tetramethylsilane, using tetramethylsilane (δ=0) and/or residual solvents such as chloroform (δ=7.26) and benzene (δ=7.20) as internal standards. Splitting pattern were indicated as s, singlet; d, doublet; t, triplet; q, quartet; m, multiplet; br, broad peak. Mass spectra were taken with a Hitachi RMU-6MG, a Hitachi M-80A and a Hitachi M-80B mass spectrometer. Unless otherwise noted, all experiments were carried out using anhydrous solvents under an atmosphere of dry argon. Especially, tetrahydrofuran and diethyl ether (ether) were distilled from sodium benzophenone ketyl. Throughout this work, Merck pre-coated TLC plates (silica gel 60 F_{254}, 0.25 mm, Art 5715) were used for thin layer chromatographic (TLC) analyses. Wako Gel C-200 and C-300 were used as an adsorbent for flash column chromatography.

1-Methoxy-2-(1-methoxycarbonylethyl)amino-4-methylbenzene (14)

A solution of commercially available **13** (686 mg, 5.0 mmol), methyl 2-bromopropionate (0.67 ml, 1.00 g, 6.0 mmol) and *N,N*-diethylaniline (0.95 ml, 895 mg, 6.0 mmol) in benzene (5 ml) was refluxed for 24 hr. After cooling and dilution with benzene, the resulting mixture was washed with H_2O and brine, dried over anhydrous $MgSO_4$, filtered, then concentrated *in vacuo*. Flash chromatography (SiO_2; hexane:EtOAc=10:1) of the residue gave **14** as a pale yellow oil (1.06 g, 95%). ^{1}H-NMR ($CDCl_3$): δ = 1.50 (d, 3H, J=7.0 Hz, -NCH*CH*$_3$), 2.23 (s, 3H, Ar-*CH*$_3$), 3.73 (s, 3H, -CO_2*CH*$_3$), 3.82 (s, 3H, Ar-O*CH*$_3$), 4.14 (q, 1H, J=7.2 Hz, -N*CH*CH_3), 4.63 (br d, 1H, J=7.7 Hz, NH), 6.33 (d, 1H, J=1.9 Hz), 6.48 (ddd, 1H, J=8.0, 1.9, 0.6 Hz), 6.66 (d, 1H, J=8.0 Hz). IR ($CHCl_3$): 3340, 1740, 1520, 1225, 1185 cm^{-1}. MS (m/e) (%): 223 (M^+) (24), 164 (100). HRMS for $C_{12}H_{17}NO_3$ (M^+): Calcd. 223.1029. Found 223.1024.

2,3-Dihydro-2,3-dioxo-7-methoxy-1-(1-methoxycarbonylethyl)-4-methyl-*1H*-indole (15)

To a solution of oxalyl chloride (0.18 ml, 261 mg, 2.1 mmol) in CH_2Cl_2 (0.5 ml) was added a solution of **14** (288 mg, 1.3 mmol) in CH_2Cl_2 (1.5 ml) at 0 °C, and the mixture was stirred for 2 hr at room temperature. After concentration *in vacuo*, the residue was diluted with CH_2Cl_2 (1.5 ml) and treated with $TiCl_4$ (1M CH_2Cl_2 solution, 5.2 ml, 5.2 mmol) at -10 °C. The resulting mixture was stirred for overnight at 12 °C. The reaction was quenched by addition of ice and the resulting mixture was extracted with CH_2Cl_2. The organic layer was dried over anhydrous $MgSO_4$, filtered, then concentrated *in vacuo*. Flash chromatography (SiO_2; CH_2Cl_2) of the residue gave **15** as red needles (206 mg, 58%), mp. 131.5-132.5 °C. ^{1}H-NMR ($CDCl_3$): δ = 1.63 (d, 3H, J=7.1 Hz), 2.51 (s, 3H, Ar-*CH*$_3$), 3.71 (s, 3H, -CO_2*CH*$_3$), 3.79 (s, 3H, Ar-O*CH*$_3$), 5.34 (q, 1H, J=7.1 Hz), 6.84 (dd, 1H, J=8.5, 0.6 Hz), 7.06 (d, 1H, J=8.4 Hz). IR (KBr): 1725, 1620, 1595, 1280, 1260, 1240, 1100 cm^{-1}. MS (m/e) (%): 277 (M^+) (27), 190 (100). *Anal.* Calcd. for $C_{14}H_{15}NO_5$: C, 60.65; H, 5.45; N, 5.05%. Found: C, 60.61; H, 5.26; N, 5.02%.

Methyl 3-methoxy-2-(1-methoxycarbonylethyl)amino-6-methylbenzoate (18)

To a solution of **15** (194 mg, 0.7 mmol) in 40% KOH (0.8 ml) was added 35% H_2O_2 (0.21 ml, 2.1 mmol) at room temperature, and the mixture was stirred for 2 hr. After concentration of the mixture *in vacuo*, the residue was suspended to MeOH (3 ml). Thionyl chloride (0.51 ml, 7.0 mmol) was then added at 0 °C, and the mixture was stirred for 5 hr. After neutralization (pH 4 - 5) with addition of 1N NaOH, the resulting mixture was extracted with $CHCl_3$ and washed with brine. The organic layer was dried over anhydrous $MgSO_4$, filtered, then concentrated *in vacuo* to give crude 3-methoxy-2-(1-methoxycarbonylethyl)amino-6-methylbenzoic acid (**17**). This crude product was used for the next step without purification. An analytical sample of **17**, however, was obtained by flash chromatography (SiO_2, $CHCl_3$:MeOH:c-NH_4OH = 40:8:1) of crude **17** prepared by another experiment, followed by recrystallization from benzene, mp. 77-78 °C. ^{1}H-NMR ($CDCl_3$): δ = 1.37 (d, 3H, J=6.8 Hz, -CH*CH*$_3$), 2.55 (s, 3H, Ar-*CH*$_3$), 3.76 (s, 3H, -CO_2*CH*$_3$), 3.85 (s, 3H, Ar-O*CH*$_3$), 3.98 (m, 1H), 6.92 (d, 1H, J=8.3 Hz), 7.03 (d, 1H, J=9.3 Hz), 7.36 (s, 1H), 9.13 (br, 1H). IR (KBr): 3350, 1745, 1720, 1580, 1470, 1240 cm^{-1}. MS (m/e) (%): 267 (M^+) (19), 208 (38), 190 (100), 147 (6), 91 (5). *Anal.* Calcd. for $C_{13}H_{17}NO_5$: C, 58.42; H, 6.41; N, 5.24%. Found: C, 58.67; H, 6.53, N; 4.96%. Potassium carbonate (102 mg 0.74 mmol) and dimethyl sulfate (70 µl, 0.74 mmol) was added to crude **17** in DMF (2.7 ml) at 0 °C, and the mixture was stirred for 1 hr. After dilution with $CHCl_3$, the resulting mixture was washed with H_2O, brine, dried over anhydrous $MgSO_4$, filtered, then concentrated *in vacuo*. Flash chromatography (SiO_2; hexane:EtOAc = 5:1) of the residue gave **18** as a colorless oil (141 mg, 71% from **15**). ^{1}H-NMR ($CDCl_3$): δ = 1.38 (d, 3H, J=7.0 Hz, -CH*CH*$_3$), 2.27 (s, 3H, Ar-*CH*$_3$), 3.66 (s, 3H, -CO_2*CH*$_3$), 3.80 (s, 3H, -CO_2*CH*$_3$), 3.92 (s, 3H, Ar-O*CH*$_3$), 4.27 (m, 1H), 6.70 (d, 1H, J=8.4 Hz), 6.78 (d, 1H, J=8.4 Hz), 8.10 (br, 1H). IR ($CHCl_3$): 3350, 1720, 1440, 1395, 1290, 1195, 1065 cm^{-1}. MS (m/e) (%): 281 (M^+) (4), 190 (100). HRMS for $C_{14}H_{19}NO_5$ (M^+): Calcd. 281.1264. Found 281.1267.

Methyl 2-[*N*-formyl-*N*-(1-methoxycarbonylethyl)]amino-3-methoxy-6-methylbenzoate (19)

To a solution of the mixed anhydride prepared from formic acid (40 μl, 1.1 mmol) and acetic anhydride (100 μl, 1.1 mmol) was added **18** (135 mg, 0.48 mmol) in CH_2Cl_2 (0.3 ml), and the mixture was stirred overnight at room temperature. After dilution with CH_2Cl_2, the resulting mixture was washed with H_2O and brine, dried over anhydrous $MgSO_4$, filtered, then concentrated *in vacuo*, giving **19** as a mixture of four rotamers and as a colorless caramel (146 mg, 98%). ^{1}H-NMR ($CDCl_3$): δ = 1.14 (d, 3H, J=7.3 Hz, for rotamer A), 1.39 (d, 3H, J=7.3 Hz, for rotamer B), 3.76 (s, 3H, for rotamer A), 3.77 (s, 3H, for rotamer B), 3.79 (s, 3H, for rotamer B), 3.80 (s, 3H, for rotamer A), 3.89 (s, 3H, for rotamer B), 3.94, (s, 3H, for rotamer A), 4.14 (q, 1H, J=7.8 Hz, for rotamer D), 4.24 (q, 1H, J=7.3 Hz, for rotamer C), 4.44 (q, 1H, J=7.3 Hz), 4.53 (q, 1H, J=7.3 Hz), 6.91 (t, 1H, J=8.8 Hz), 7.22 (t, 1H, J=8.3 Hz), 8.06 (s, 1H, for rotamer B), 8.08 (s, 1H, for rotamer A), 8.51 (s, 1H, for rotamer C), 8.58 (s, 1H, for rotamer D). Rotamer A : B : C : D = 1 : 0.710 : 0.066 : 0.040. IR ($CHCl_3$): 1740, 1680, 1300, 1075 cm^{-1}. MS (m/e) (%): 309 (M^+) (8), 222 (30), 190 (100). HRMS for $C_{15}H_{19}NO_6$ (M^+): Calcd 309.1213. Found 309.1212.

Methyl 2,3-dihydro-2,4-dimethyl-1-formyl-7-methoxy-3-oxo-*1H*-indole-2-carboxylate (20)

To a solution of LDA prepared from diisopropylamine (5.1 μl, 0.037 mmol) and n-BuLi (1.6M hexane solution, 23 μl, 0.037 mmol) in THF (50 μl) was added **19** (10.3 mg, 0.033 mmol) in THF (50 μl) at -78 °C, and the mixture was stirred for 4.5 hr. After quenching the reaction with addition of sat. NH_4Cl and dilution with $CHCl_3$, the chloroform layer was dried over anhydrous $MgSO_4$, filtered, then concentrated *in vacuo*. Flash chromatography (SiO_2; hexane:EtOAc = 2:1) of the residue gave **20** as colorless crystals (3.7 mg, 40%), mp. 140-141 °C. ^{1}H-NMR ($CDCl_3$): δ = 1.82 (s, 3H, Ar-CH_3), 2.55 (s, 3H, Ar-CH_3), 3.73 (s, 3H, CO_2CH_3), 3.97 (s, 3H, Ar-OCH_3), 6.93 (dd, 1H, J=8.2, 0.8 Hz), 7.13 (d, 1H, J=8.2 Hz), 9.57 (s, 1H, CHO). IR ($CHCl_3$): 1755, 1705, 1665, 1325, 1255 cm^{-1}. MS (m/e) (%): 277 (M^+) (13), 190 (100), 99 (8). HRMS for $C_{14}H_{15}NO_5$ (M^+); Calcd 277.0951. Found 277.0935.

2,3-Dihydro-7-methoxy-4-methyl-3-methylthio-*1H*-indol-2-one (21)

To a solution of methyl methylthioacetate (63.1 mg, 0.53 mmol) in CH_2Cl_2 (5 ml) was added sulfuryl chloride (42 μl, 0.53 mmol) at -78 °C, and the mixture was stirred for 15 min. A solution of **13** (68.6 mg, 0.5 mmol) and 1,8-bis(dimethylamino)naphthalene (110 mg, 0.52 mmol) in CH_2Cl_2 (3 ml) was added to the dichloromethane solution of S-chloro-S-methyl-S-methoxycarbonylmethylsulfonium chloride prepared above at -78 °C, and the mixture was stirred for 3 hr at the same temperature. Triethylamine (72 μl, 0.52 mmol) was then added, and the stirring was continued for 15 min at -78 °C and for 20 hr at room temperature. Acetic acid (0.5 ml) was added, and the mixture was further stirred for 1 hr at room temperature. The resulting mixture was washed with H_2O and brine. The organic layer was dried over anhydrous $MgSO_4$, filtered, then concentrated *in vacuo*. Flash chromatography (SiO_2; CH_2Cl_2:EtOAc = 20:1) of the residue gave **21** as pale yellow crystals (90.6 mg, 81%), mp. 156-157 °C. ^{1}H-NMR ($CDCl_3$): δ = 2.01 (s, 3H, -SCH_3), 2.36 (s, 3H, Ar-CH_3), 3.84 (s, 3H, -OCH_3), 4.20 (s, 1H), 6.75 (d, 1H, J=8.5 Hz), 6.79 (dd, 1H, J=8.4, 0.5 Hz), 7.53 (br s, 1H). IR (KBr): 3150, 1700, 1630, 1510, 1280, 1250 cm^{-1}. MS (m/e) (%): 223 (M^+) (39), 176 (100). *Anal.* Calcd. for $C_{11}H_{13}N_2S$: C, 59.17; H, 5.87; N, 6.27%. Found: C, 59.01; H, 5.90; N, 6.33%.

2,3-Dihydro-2,3-dioxo-7-methoxy-4-methyl-*1H*-indole (22)

To a solution of **21** (11.2 mg, 0.05 mmol) in CH_2Cl_2 (1 ml) was added *N*-chlorosuccinimide (97.4 mg, 0.06 mmol) at 0 °C, and the mixture was stirred at room temperature for 1 hr. After concentration *in vacuo*, the residue was diluted with acetone (5 ml) and treated with $CuCl_2$ (8.1 mg, 0.06 mmol) and CuO (4.8 mg, 0.06 mmol) at room temperature for 1 hr. After filtration and concentration *in vacuo*, the residue was diluted with CH_2Cl_2 and washed with 1% $NaHCO_3$ and brine. The organic layer was dried over anhydrous $MgSO_4$, filtered, then concentrated *in vacuo*. Flash chromatography (SiO_2; CH_2Cl_2:EtOAc = 10:1) of the residue gave **22** as red crystals (8.0 mg, 83%), mp. 240-241 °C. ^{1}H-NMR ($CDCl_3$): δ = 2.49 (d, 3H, J=0.3 Hz, Ar-CH_3), 3.88 (s, 3H, -OCH_3), 6.81 (dd, 1H, J=8.4, 0.7 Hz), 7.00 (d, 1H, J=8.4 Hz), 7.64 (br s, 1H). IR ($CHCl_3$): 3425, 1760, 1740, 1600, 1285, 1270 cm^{-1}. MS (m/e) (%): 191 (M^+) (100), 163 (54), 135 (83). HRMS for $C_{10}H_9NO_3$ (M^+); Calcd 191.0583. Found 191.0565.

3-Acetoxy-2-(4-benzyloxy-2-nitrophenyl)propan-1-ol (24)

To a solution of **23**[11a] (1.82 g, 6.0 mmol) and Et_3N (0.84 ml, 0.61 g, 6.0 mmol) in CH_2Cl_2 (20 ml) was added acetic anhydride (0.85 ml, 0.92 g, 9.0 mmol) at 0 °C, and the mixture was stirred for 2.5 hr at the same temperature. The resulting mixture was successively washed with H_2O, saturated $NaHCO_3$ solution and brine, dried over anhydrous $MgSO_4$, filtered, then concentrated *in vacuo*. Flash chromatography (SiO_2; CH_2Cl_2:EtOAc=20:1) of the residue gave **24** as an oil (1.54 g, 74%). ^{1}H-NMR ($CDCl_3$): δ = 2.04 (s, 3H, -$COCH_3$), 3.65 (m, 1H, -CH), 3.87 (dd, 1H, J=11.3, 5.5 Hz, -CHOH), 3.90 (dd, 1H, J=11.3, 5.8 Hz, -CHOH), 4.32 (dd, 1H, J=11.1, 6.2 Hz, -CHOAc), 4.48 (dd, 1H, J=11.1, 6.8 Hz, -CHOAc), 5.09 (s, 2H,

-O*CH*$_2$Ph), 7.17 (dd, 1H, J=8.7, 2.7 Hz, *C*$_5$'-*H*), 7.33-7.41 (6H, *C*$_6$*H*$_5$ and *C*$_3$'-*H*), 7.44 (d, 1H, J=8.8 Hz, *C*$_6$'-*H*). IR ($CHCl_3$): 3600, 3450, 1730, 1530, 1230 cm^{-1}. MS (m/e) (%): 345 (M^+), 91 (100). HRMS for $C_{16}H_{15}NO_4$ [(M-AcOH)$^+$]; Calcd 285.1002. Found 285.1002.

3-Acetoxy-2-(4-benzyloxy-2-nitrophenyl)propan-1-yl methanesulfonate (25)

To a solution of **24** (1.53 g, 4.4 mmol) and Et_3N (0.68 ml, 0.49 g, 4.9 mmol) in CH_2Cl_2 (15 ml) was added methanesulfonyl chloride (0.38 ml, 0.56 g, 4.9 mmol) at 0 °C, and the mixture was stirred for 30 min at the same temperature. The resulting mixture was successively washed with H_2O and brine, dried over anhydrous $MgSO_4$, filtered, then concentrated *in vacuo*. Flash chromatography (SiO_2; CH_2Cl_2:EtOAc=50:1) of the residue gave **25** as an oil (1.86 g, 100%). ^{1}H-NMR ($CDCl_3$): δ = 2.04 (s, 3H, -CO*CH*$_3$), 2.95 (s, 3H, -OSO$_2$*CH*$_3$), 3.94 (m, 1H, -*CH*), 4.36 (dd, 1H, J=11.3, 6.0 Hz, -*CH*OAc), 4.44 (dd, 1H, J=11.2, 6.8 Hz, -*CH*OAc), 4.50 (dd, 1H, J=11.4, 5.7 Hz, -*CH*OMs), 4.53 (dd, 1H, J=11.5, 5.9 Hz, -*CH*OMs), 5.11 (s, 2H, -O*CH*$_2$Ph), 7.20 (dd, 1H, J=8.8, 2.7 Hz, *C*$_5$'-*H*), 7.34-7.42 (6H, *C*$_6$*H*$_5$ and *C*$_6$'-*H*), 7.48 (d, 1H, J=2.7 Hz, *C*$_3$'-*H*). IR ($CHCl_3$): 1740, 1530, 1360, 1230, 1180 cm^{-1}. MS (m/e) (%): 423 (M^+), 363 (1), 91 (100). HRMS for $C_{17}H_{17}NO_6S$ [(M-AcOH)$^+$]; Calcd 363.0777. Found 363.0796.

3-Acetoxymethyl-6-benzyloxy-1-t-butoxycarbonyl-2,3-dihydro-*1H*-indole (27)

A solution of **25** (1.85 g, 4.4 mmol), Et_3N (0.61 ml, 0.44 g, 4.4 mmol) and PtO_2 (139 mg, 0.61 mmol) in THF (20 ml) was hydrogenated at 3 atm for 20 min at room temperature. After filtration of the catalyst, the filtrate was diluted with CH_2Cl_2. The resulting solution was successively washed with H_2O and brine, dried over anhydrous $MgSO_4$, filtered, then concentrated *in vacuo*. Crude 3-acetoxymethyl-6-benzyloxy-2,3-dihydro-*1H*-indole thus obtained was dissolved in CH_2Cl_2 and treated with di-t-butyl dicarbonate (1.05 g, 4.8 mmol) at room temperature for 12 hr. After concentration of the resulting solution *in vacuo*, the crude product was purified with flash chromatography (SiO_2; benzene:EtOAc=20:1) to give **27** as colorless prisms (1.65 g, 95%). An analytical sample was obtained by recrystallization from hexane, mp. 75-76.5 °C. ^{1}H-NMR ($CDCl_3$): δ = 1.56 (s, 9H, -*t*-*Bu*), 2.08 (s, 3H, -CO*CH*$_3$), 3.56 (m, 1H, *C*$_3$-*H*), 3.80 (m, 1H, -*CH*OAc), 4.02-4.10 (2H, *C*$_2$-*H* and -*CH*OAc), 4.24 (dd, 1H, J=10.9, 5.8 Hz, *C*$_2$-*H*), 5.06 (s, 2H, -O*CH*$_2$Ph), 6.57 (dd, 1H, J=8.2, 2.4 Hz, *C*$_5$-*H*), 7.06 (dd, 1H, J=8.2, 0.5 Hz, *C*$_4$-*H*), 7.28-7.45 (5H, *C*$_6$*H*$_5$), 7.66 (br, 1H, *C*$_7$-*H*). IR ($CHCl_3$): 1735, 1690, 1395, 1220 cm^{-1}. MS (m/e) (%): 397 (M^+) (3), 281 (23), 91 (100). *Anal.* Calcd. for $C_{23}H_{27}NO_5$: C, 69.50; H, 6.85; N, 3.52%. Found: C, 69.41; H, 6.92; N, 3.50%.

3-Acetoxymethyl-6-benzyloxy-1-t-butoxycarbonyl-2,3-dihydro-5-nitro-*1H*-indole (28)

To a solution of **27** (1.65 g, 4.2 mmol) in nitromethane (80 ml) was added acetyl nitrate prepared from acetic anhydride (2.4 ml, 2.55 g, 25 mmol) and nitric acid (90%, 0.26 ml, 6.2 mmol) at -20 °C, and the mixture was stirred for 3 hr at the same temperature. After dilution with benzene, the resulting mixture was washed with sat. $NaHCO_3$ and brine, dried over anhydrous $MgSO_4$, filtered, then concentrated *in vacuo*. Flash chromatography (SiO_2; benzene:EtOAc=20:1) of the residue gave **28** as pale yellow crystals (1.41 g, 77%). An analytical sample was obtained by recrystallization from methanol, mp. 134.5-135.5 °C. ^{1}H-NMR ($CDCl_3$): δ = 1.58 (s, 9H, -*t*-*Bu*), 2.10 (s, 3H, -CO*CH*$_3$), 3.62 (m, 1H, *C*$_3$-*H*), 3.82 (dd, 1H, J=11.2, 4.7 Hz -*CH*OAc), 4.11-4.17 (2H, *C*$_2$-*H* and -*CH*OAc), 4.22 (dd, 1H, J=11.0, 6.5 Hz, *C*$_2$-*H*), 5.25 (s, 2H, -O*CH*$_2$Ph), 7.30-7.56 (5H, *C*$_6$*H*$_5$), 7.81 (br, 1H, *C*$_7$-*H*), 7.88 (d, 1H, J=1.0 Hz, *C*$_4$-*H*). IR (KBr): 1715, 1310, 1240 cm^{-1}. MS (m/e) (%): 442 (M^+) (38), 326 (16), 282 (5), 205 (6), 91 (100). *Anal.* Calcd. for $C_{23}H_{26}N_2O_7$: C, 62.43; H, 5.92; N, 6.33%. Found: C, 62.29; H, 5.80; N, 6.26%.

3-Acetoxymethyl-5-amino-6-benzyloxy-1-t-butoxycarbonyl-2,3-dihydro-*1H*-indole (29)

A solution of **28** (443 mg, 1.0 mmol) in THF (5 ml) was hydrogenated in the presence of PtO_2 (22.7 mg, 0.10 mmol) at 3 atm at room temperature for 15 min. After filtration of the catalyst, the filtrate was concentrated *in vacuo*. Flash chromatography (SiO_2; benzene:EtOAc=20:1) of the residue gave **29** as colorless crystals (390 mg, 95%), mp. 86.5-87.5 °C. ^{1}H-NMR ($CDCl_3$): δ = 1.55 (s, 9H, -*t*-*Bu*), 2.09 (s, 3H, -CO*CH*$_3$), 3.52 (m, 1H, *C*$_3$-*H*), 3.74 (m, 1H, -*CH*OAc), 4.01-4.06 (2H, *C*$_2$-*H* and -*CH*OAc), 4.24 (dd, 1H, J=10.7, 5.5 Hz, *C*$_2$-*H*), 5.09 (s, 2H, -O*CH*$_2$Ph), 6.63 (s, 1H, *C*$_4$-*H*), 7.31-7.47 (5H, *C*$_6$*H*$_5$), 7.65 (br, 1H, *C*$_7$-*H*). IR ($CHCl_3$): 3440, 3370, 1735, 1685, 1500, 1240 cm^{-1}. MS (m/e) (%): 412 (M^+) (10), 356 (24), 205 (100), 161 (19), 91 (36). *Anal.* Calcd. for $C_{23}H_{28}N_2O_5$: C, 66.97; H, 6.84; N, 6.79%. Found: C, 67.04; H, 6.90; N, 6.81%.

8-Acetoxymethyl-4-benzyloxy-6-t-butoxycarbonyl-1,2,3,6,7,8-hexahydro-1,2-dioxobenzo [1,2-b;4,3-b']dipyrrole (31)

To a solution of methyl methylthioacetate (137 mg, 1.1 mmol) in CH_2Cl_2 (9 ml) was added sulfuryl chloride (84 μl, 1.1 mmol) at -78 °C, and the mixture was stirred for 20 min. A solution of **29** (390 mg, 1.0 mmol) and 1,8-bis(dimethylamino)naphthalene (224 mg, 1.1 mmol) in CH_2Cl_2 (6 ml) was added to the resulting solution of S-chloro-S-methyl-S-methoxycarbonylmethylsulfonium chloride at -78 °C over 20 min, and the mixture was

stirred for 3.5 hr at the same temperature. Triethylamine (0.15 ml, 1.1 mmol) was then added, and the stirring was continued for 2 hr at -78 °C and 21 hr at room temperature. Acetic acid (3 ml) was added, and the mixture was further stirred for 1 hr at room temperature. The resulting mixture was washed with H_2O and brine. The organic layer was dried over anhydrous $MgSO_4$, filtered, then concentrated *in vacuo*, to give crude 8-acetoxymethyl-4-benzyloxy-6-t-butoxycarbonyl-1,2,3,6,7,8-hexahydro-1-methylthio-1,2-dioxo-benzo[1,2-b;4,3-b']dipyrrole (**30**). This crude material was diluted with acetone (20 ml) and treated with $CuCl_2$ (128 mg, 1.0 mmol) and CuO (113 mg, 1.4 mmol) at 0 °C for 1.5 hr. After filtration and dilution with CH_2Cl_2, the resulting mixture was washed with sat. NH_4Cl and brine, dried over anhydrous $MgSO_4$, filtered, then concentrated *in vacuo*. Flash chromatography (SiO_2; CH_2Cl_2:EtOAc=10:1) of the residue gave **31** as purple crystals (340 mg, 77%). An analytical sample was obtained by recrystallization from ethanol, mp. 178-179 °C. ^{1}H-NMR ($CDCl_3$): δ = 1.56 (s, 9H, -*tBu*), 2.03 (s, 3H, -CO*CH*$_3$), 3.83-3.91 (2H, *C*$_8$-*H* and -*CH*OAc), 4.05 (dd, 1H, J=11.6, 9.7 Hz, *C*$_7$-*H*), 4.25 (m, 1H, -*CH*OAc), 4.34 (dd, 1H, J=11.2, 4.4 Hz, *C*$_7$-*H*), 5.14 (s, 2H, -O*CH*$_2$Ph), 7.40 (5H, *C*$_6$*H*$_5$), 7.63 (br, 1H, *NH*), 8.00 (br s, 1H, *C*$_5$-*H*). IR (KBr): 3550, 3200, 1740, 1700 cm^{-1}. MS (m/e) (%): 466 (M^+), 350, 306, 249, 205 (5), 91 (100). *Anal.* Calcd. for $C_{25}H_{26}N_2O_7$: C, 64.37; H, 5.62; N, 6.01%. Found: C, 64.37; H, 5.51; N, 5.96%.

4-Benzyloxy-6-t-butoxycarbonyl-8-(t-butyldimethylsilyloxy)methyl-1,2,3,6,7,8-hexahydro-1,2-dioxobenzo[1,2-b;4,3-b']dipyrrole (33)

To a MeOH (7 ml) solution of **31** (162 mg, 0.35 mmol) was added K_2CO_3 (96.7 mg, 0.7 mmol) at 0 °C, and the mixture was stirred for 2.5 hr at room temperature. The reaction was quenched with addition of AcOH (70 μl), and the mixture was diluted with CH_2Cl_2. The organic layer was washed with brine, dried over anhydrous Na_2SO_4, filtered, then concentrated *in vacuo*, to give crude 4-benzyloxy-6-t-butoxycarbonyl-8-hydroxymethyl-1,2,3,6,7,8-hexahydro-1,2-dioxo-benzo[1,2-b;4,3-b']dipyrrole (**32**) as purple crystals. Without purification, this was dissolved in DMF (1.5 ml) and treated with t-butyldimethylchlorosilane (106 mg, 0.7 mmol) and imidazole (47.7 mg, 0.7 mmol) at room temperature for 8 hr. After dilution with benzene, the mixture was washed with sat. NH_4Cl and brine, dried over anhydrous $MgSO_4$, filtered, then concentrated *in vacuo*. Flash chromatography (SiO_2; benzene:EtOAc=8:1) of the residue gave **33** as reddish purple crystals (169 mg, 90%). An analytical sample was obtained by recrystallization from ethanol, mp. 197-198 °C. ^{1}H-NMR ($CDCl_3$): δ = -0.05, 0.04 (sx2, each 3H, -Si*Me*$_2$), 0.82 (s, 9H, -Si-*tBu*), 1.55 (s, 9H, -*tBu*), 3.60-3.90 (2H), 3.94-4.00 (2H), 4.09 (m, 1H), 5.13 (s, 2H, -O*CH*$_2$Ph), 7.36-7.41 (5H, *C*$_6$*H*$_5$), 7.76 (br, 1H, *NH*), 7.98 (br s, 1H, *C*$_5$-*H*). IR (KBr): 3200, 1740, 1715 cm^{-1}. MS (m/e) (%): 538 (M^+), 482, 424, 381, 304, 260, 91 (100). *Anal.* Calcd. for $C_{29}H_{38}N_2O_6Si$: C, 64.66; H, 7.11; N, 5.20%. Found: C, 64.61; H, 7.17; N, 5.19%.

Methyl 5-amino-6-benzyloxy-1-t-butoxycarbonyl-3-(t-butyldimethylsilyloxy)methyl-2,3-dihydro-*1H*-indole-4-carboxylate (35)

To a solution of **33** (53.9 mg, 0.10 mmol) in CH_2Cl_2 (1 ml) was added m-CPBA (31.1 mg, 0.18 mmol) and $NaHCO_3$ (16.8 mg, 0.20 mmol) at -15 °C, and the mixture was stirred for 2 hr at the same temperature. After dilution with CH_2Cl_2, the reaction mixture was washed with 1% $NaHSO_3$, sat. $NaHCO_3$, and brine, dried over anhydrous Na_2SO_4, filtered, then concentrated *in vacuo*. Crude isatoic anhydride derivative (**34**) was dissolved in MeOH (1 ml), then treated with K_2CO_3 (16.6 mg, 0.12 mmol) at 0 °C for 1 hr. The reaction was quenched with addition of AcOH (0.07 ml), and the resulting mixture was diluted with brine and extracted with benzene. The organic phase was dried over anhydrous Na_2SO_4, filtered, then concentrated *in vacuo*. Flash chromatography (SiO_2; benzene:EtOAc=50:1 to 30:1) of the residue gave **35** as pale yellow crystals (50.9 mg, 94%). An analytical sample was obtained by recrystallization from methanol, mp. 120.5-121 °C. ^{1}H-NMR ($CDCl_3$): δ = -0.01, 0.02 (sx2, each 3H, -Si*Me*$_2$), 0.81 (s, 9H, -Si-*tBu*), 1.55 (s, 9H, -*tBu*), 1.55 (br, 1H, *NH*), 3.66 (dd, 1H, J=9.3, 3.9 Hz), 3.70-3.82 (2H), 3.89 (s, 3H, -O*CH*$_3$), 4.11 (d, 1H, J=10.3 Hz), 5.10 (s, 2H, -O*CH*$_2$Ph), 5.78 (br, 1H, *NH*), 7.34-7.43 (5H, *C*$_6$*H*$_5$), 7.89 (br s, 1H, *C*$_7$-*H*). IR (KBr): 3550, 3545, 1700 cm^{-1}. MS (m/e) (%): 542 (M^+) (33), 486 (36), 429 (3), 333 (9), 231 (100), 187 (34), 91 (83). *Anal.* Calcd. for $C_{29}H_{42}N_2O_6Si$: C, 64.18; H, 7.80; N, 5.16%. Found: C, 64.08; H, 7.85; N, 5.04%.

Methyl 6-benzyloxy-1-t-butoxycarbonyl-3-(t-butyldimethylsilyloxy)methyl-2,3-dihydro-5-(1-methoxycarbonylethyl)amino-*1H*-indole-4-carboxylate (36)

A solution of **35** (56.1 mg, 0.10 mmol), methyl 2-bromopropionate (58 μl, 86 mg, 0.52 mmol), and 1,8-bis(dimethylamino)naphthalene (44.3 mg, 0.21 mmol) in DMAC (0.5 ml) was stirred for 48 hr at 70 °C. After cooling, the reaction mixture was extracted with benzene. The organic phase was washed with H_2O and brine, dried over anhydrous Na_2SO_4, filtered, then concentrated *in vacuo*. Flash chromatography (SiO_2; benzene:EtOAc=20:1) of the residue gave **36** as a diastereomeric mixture and as a pale yellow caramel (57.4 mg, 88%). This material solidified upon standing at room temperature. ^{1}H-NMR ($CDCl_3$): δ = -0.08-0.40 (6H, -Si*Me*$_2$), 0.84, 0.86 (sx2, total 9H, -Si-*tBu* in each diastereomer), 1.28, 1.36 (dx2, total 3H, J=7.0, 7.2 Hz, in each diastereomer), 3.55, 3.58 (sx2, total 3H, -CO$_2$*CH*$_3$ in each diastereomer), 3.92-3.94 (s, 3H, Ar-CO$_2$*CH*$_3$

in each diastereomer), 3.30-4.12 (5H), 4.29-4.41 (m, 1H, -NC*H* in each diastereomer), 5.09 (br s, 2H, -OCH_2Ph), 7.30-7.48 (5H, C_6H_5), 7.86 (br s, 1H). IR ($CHCl_3$): 3350, 1730, 1690, 1460, 1145 cm^{-1}. MS (m/e) (%): 628 $(M)^+$, 572, 527, 481, 349, 289, 245. HRMS for $C_{33}H_{48}N_2O_8Si$ (M^+); Calcd 628.3181. Found 628.3158.

Methyl 6-benzyloxy-1-t-butoxycarbonyl-3-(t-butyldimethylsilyloxy)methyl-2,3-dihydro-5-[*N*-formyl-*N*-(1-methoxycarbonylethyl)]amino-1*H*-indole-4-carboxylate (37)

To a solution of the mixed anhydride prepared from formic acid (58.0 μl, 1.5 mmol) and acetic anhydride (132 μl, 1.4 mmol) was added **36** (43.9 mg, 0.07 mmol), and the mixture was stirred for 9 hr at room temperature. After dilution with benzene, the reaction mixture was washed with 10% $NaHCO_3$. The organic phase was dried over anhydrous Na_2SO_4, filtered, then concentrated *in vacuo*. Preparative thin layer chromatography (SiO_2; benzene:MeOH=20:1) of the residue gave **37** as a diastereomeric mixture and as a colorless caramel (42.7 mg, 93%). ^{1}H-NMR ($CDCl_3$): δ = -0.20-0.12 (6H, -Si*Me*$_2$), 0.69-0.96 (m, total 9H, -Si-^{t}*Bu* in each diastereomer), 1.06-1.49 (m, 3H, -*CH*$_3$ for two rotamers in each diastereomer), 3.53, 3.72 (br s, 3H, -CO_2*CH*$_3$ for two rotamers in each diastereomer), 3.84-4.01 (m, 3H, Ar-CO_2*CH*$_3$ for two rotamers in each diastereomer), 3.36-4.26 (5H), 4.96-5.16 (m, 1H, -NC*H* for two rotamers in each diastereomer), 5.12 (br s, 2H, -OCH_2Ph), 7.36 (br s, 5H, C_6H_5), 7.66 (br s, 1H), 7.96-8.10 (m, 1H, -*CHO* for two rotamers in each diastereomer). IR ($CHCl_3$): 1725, 1695, 1670, 1460, 1340, 1300, 1155, 840 cm^{-1}. MS (m/e) (%): 656 (M^+) (7), 599 (21), 543 (67), 499 (7), 367 (3), 323 (5), 245 (4), 91 (100). HRMS for $C_{34}H_{48}N_2O_9Si$ (M^+); Calcd 656.3130. Found 656.3150.

Methyl (2*R,8*S**)-4-benzyloxy-6-t-butoxycarbonyl-8-(t-butyldimethylsilyloxy)methyl-3-formyl-1,2,3,6,7,8-hexahydro-2-methyl-1-oxobenzo[1,2-b;4,3-b']dipyrrole-2-carboxylate (38) and Its (2*S**,8*S**)-Isomer (39)**

To a solution of **37** (38.5 mg, 0.06 mmol) in THF (2 ml) was added a solution of LDA (0.88 M THF solution, 0.10 ml, 0.88 mmol) at -78 °C, and the mixture was stirred for 5.5 hr at the same temperature. After quenching the reaction with addition of AcOH (2 drops) and dilution with benzene, the resulting mixture was washed with NH_4Cl, dried over anhydrous Na_2SO_4, filtered, then concentrated *in vacuo*. Preparative thin layer chromatography (SiO_2; hexane:Et_2O=2:1) of the residue gave **38** (10.4 mg, 28%) and **39** (10.4 mg, 28%) both as a pale yellow caramel. **38**: ^{1}H-NMR ($CDCl_3$): δ = -0.13, 0.01 (sx2, each 3H, Si*Me*$_2$), 0.79 (s, 9H, Si-^{t}*Bu*), 1.57 (s, 9H, -^{t}*Bu*), 1.81 (s, 3H, -*CH*$_3$), 3.72 (s, 3H, -CO_2*CH*$_3$), 3.64-3.76 (2H, -CH_2OTBS, C_8-*H*), 3.86 (dd, 1H, J=9.5, 3.3 Hz, C_7-*H*), 3.99 (t, 1H, J=10.3 Hz, -CH_2OTBS), 4.19 (d, 1H, J=10.9 Hz, C_7-*H*), 5.20 (d, 1H, J=10.9 Hz, -OCH_2Ph), 5.24 (d, 1H, J=11.3 Hz, -OCH_2Ph), 7.35-7.43 (5H, C_6H_5), 8.18 (br s, 1H, C_5-*H*), 9.47 (s, 1H, *CHO*). IR ($CHCl_3$): 1760, 1710, 1695, 1670, 1340, 1150 cm^{-1}. MS (m/e) (%): 624 (M^+) (2), 568 (9), 524 (21), 483 (15), 379 (8), 273 (13), 91 (100). HRMS for $C_{33}H_{44}N_2O_8Si$ (M^+); Calcd 624.2868. Found 624.2856. **39**: ^{1}H-NMR ($CDCl_3$): δ = -0.15, 0.00 (sx2, each 3H, Si*Me*$_2$), 0.79 (s, 9H, Si-^{t}*Bu*), 1.57 (s, 9H, -^{t}*Bu*), 1.80 (s, 3H, -*CH*$_3$), 3.71 (s, 3H, -CO_2*CH*$_3$), 3.68-3.75 (2H, -CH_2OTBS, C_8-*H*), 3.87 (m, 1H, C_7-*H*), 3.99 (t, 1H, J=10.7 Hz, -CH_2OTBS), 4.20 (d, 1H, J=10.4 Hz, C_7-*H*), 5.20 (d, 1H, J=10.5 Hz, -OCH_2Ph), 5.24 (d, 1H, J=10.9 Hz, -OCH_2Ph), 7.35-7.43 (5H, C_6H_5), 8.17 (br s, 1H, C_5-*H*), 9.46 (s, 1H, -*CHO*). IR ($CHCl_3$): 1760, 1710, 1695, 1670, 1340, 1150 cm^{-1}. MS (m/e) (%): 624 (M^+) (13), 568 (6), 511 (22), 483 (6), 379 (4), 273 (6), 91 (100). HRMS for $C_{33}H_{44}N_2O_8Si$ (M^+); Calcd 624.2868. Found 624.2868.

Methyl (*Z*)-1-azido-2-(3,4,5-trimethoxyphenyl)acrylate (42)

To a solution of NaOMe (1.6 M in methanol, 25 ml, 40 mmol) was added a solution of 3,4,5-trimethoxybenzaldehyde (981 mg, 5.0 mmol) and methyl azidoacetate (5.76 g, 50 mmol) in MeOH (8 ml) at -20 °C, and the mixture was stirred at 0 °C for 5.5 hr . After addition of cold water, the resulting precipitate was collected by filtration. The solid was washed with water and dried *in vacuo* to give **42** as pale yellow crystals (1.42 g, 97%), mp. 100.5-101.5 °C. ^{1}H-NMR ($CDCl_3$): δ = 3.891, 3.894, 3.897, 3.90 (sx4, each 3H, Ar-OCH_3x3 and -CO_2*CH*$_3$), 6.85 (s, 1H), 7.10 (s, 2H, *Ar*-*H*x2). IR (KBr): 2120, 1715, 1240, 1130 cm^{-1}. MS (m/e) (%): 293 (M^+) (1), 265 (100), 206 (88), 160 (20). *Anal.* Calcd. for $C_{13}H_{15}N_3O_5$: C, 53.24; H, 5.16; N, 14.33%. Found: C, 53.58; H, 5.10; N, 13.25%.

Methyl 4,5,6-trimethoxy-1*H*-indole-2-carboxylate (43)

To a refluxing xylene (5 ml) was added a xylene solution (5 ml) of **42** (293 mg, 1.0 mmol), and the mixture was refluxed for 1 hr. After cooling, the solvent was removed *in vacuo*. Flash chromatography (SiO_2; benzene:EtOAc=15:1) of the residue gave **43** as colorless crystals (250 mg, 94%). An analytical sample was obtained by recrystallization from methanol, mp. 106-107 °C. ^{1}H-NMR ($CDCl_3$): δ = 3.90, 3.926, 3.929 4.07 (sx4, each 3H, Ar-OCH_3x3 and -CO_2*CH*$_3$), 6.82 (s, 1H, C_4-*H*), 7.18 (d, 1H, J=2.3 Hz, C_3-*H*), 8.87 (br s,

1H, *NH*). IR (KBr): 3280, 1700 cm^{-1}. MS (m/e) (%): 265 (M^+) (100), 218 (44), 160 (18). *Anal.* Calcd. for $C_{13}H_{15}NO_5$: C, 58.86; H, 5.70; N, 5.28%. Found: C, 58.95; H, 5.59; N, 5.19%.

4,5,6-Trimethoxy-*1H*-indole-2-carboxylic acid (40)

A solution of **43** (50.2 mg, 0.20 mmol) in 40% KOH (0.5 ml) and MeOH (0.5 ml) was stirred at 50 °C for 30 min. After neutralization with addition of con. HCl, the resulting precipitates were collected by filtration, washed with water, and dried *in vacuo* to give **40** as colorless crystals (45.5 mg, 96%), mp. 215.5-216 °C. ^{1}H-NMR ($CDCl_3$): δ = 3.25 (br s, 2H, *NH* and CO_2H), 3.77, 3.79, 3.89 (sx3, each 3H, -OCH_3x3), 6.91 (s, 1H, C_4-*H*), 7.00 (d, 1H, J=1.5 Hz, C_3-*H*). IR (KBr): 3275, 2590, 1630, 1230 cm^{-1}. MS (m/e) (%): 251 (M^+) (100), 218 (32), 160 (15). *Anal.* Calcd. for $C_{12}H_{13}NO_5$: C, 57.37; H, 5.22; N, 5.58%. Found: C, 57.21; H, 5.10; N, 5.43%.

Methyl (*2R*,8S)-4-benzyloxy-1,2,3,6,7,8-hexahydro-8-hydroxymethyl-2-methyl-1-oxo-benzo[1,2-b;4,3-b']dipyrrole-2-carboxylate dihydrochloride (44) and Its (*2S*,8S**)-Isomer**

To a 10% HCl-MeOH solution (2 ml) was added **38** (10.3 mg, 0.02 mmol), and the mixture was allowed to stand at room temperature for 11 hr. After evaporation of the solvent *in vacuo*, crude **44** was obtained as pale yellow crystals (6.9 mg, 100%). This crude material was used for the next reaction without further purification. ^{1}H-NMR (DMSO-d_6): δ = 1.45 (s, 3H, C_2-*Me*), 3.65 (s, 3H, -CO_2CH_3), 3.40-3.95 (5H), 7.20-7.60 (6H), 7.80 (3H).

The same treatments of **39** (10.4 mg, 0.02 mmol) as described for **38** gave crude methyl (*2S*,8S**)-4-benzyloxy-1,2,3,6,7,8-hexahydro-8-hydroxymethyl-2-methyl-1-oxo-benzo[1,2-b;4,3-b']dipyrrole-2-carboxylate dihydrochloride as pale yellow crystals (7.0 mg, 100%), after concentration of the reaction mixture *in vacuo*. ^{1}H-NMR (DMSO-d_6) δ = 1.47 (s, 3H, C_2-CH_3), 3.62 (s, 3H, -CO_2CH_3), 3.40-3.95 (5H), 7.20-7.60 (6H), 7.79 (3H). Without further analysis this material was also subjected to the next reaction.

Methyl (*2R*,8S)-4-benzyloxy-1,2,3,6,7,8-hexahydro-8-hydroxymethyl-2-methyl-1-oxo-6-[(5,6,7-trimethoxy-*1H*-indole-2-yl)carbonyl]benzo[1,2-b;4,3-b']dipyrrole-2-carboxylate (45) and Its (*2S*,8S**)-Isomer (48)**

To a solution of **44** (6.9 mg, 0.02 mmol) in DMF (0.18 ml) was added **40** (4.2 mg, 0.02 mmol), 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride (9.6 mg, 0.05 mmol), and $NaHCO_3$ (7.0 mg, 0.08 mmol), and the mixture was stirred for 20 hr at room temperature. After dilution with EtOAc, the resulting mixture was washed with H_2O, dried over anhydrous Na_2SO_4, filtered, then concentrated *in vacuo*. Preparative thin layer chromatography (SiO_2; $CHCl_3$:acetone=4:1) of the residue gave **45** as a yellow caramel (5.8 mg, 57%). ^{1}H-NMR ($CDCl_3$): δ = 1.69 (s, 3H, 2-CH_3), 2.78 (br, 1H, -*OH*), 3.77 (s, 3H, -CO_2CH_3), 3.82 (m, 1H, C_8-*H*), 3.93-4.01 (2H, C_7-H_2), 3.91, 3.94, 4.07 (sx3, each 3H, -OCH_3x3) , 4.45 (dd, 1H, J=10.3, 3.9 Hz, -*CH*OH), 4.61 (t, 1H, J=10.0 Hz, -*CH*OH), 5.21 (s, 2H, -OCH_2Ph), 5.29 (s, 1H, C_3-*H*), 6.85 (s, 1H, C_4-*H*), 6.95 (s, 1H, $C_{3'}$-*H*), 7.36-7.51 (5H, -C_6H_5), 8.45 (s, 1H, C_5-*H*), 9.35 (br s, 1H, $C_{1'}$-*H*). IR ($CHCl_3$): 3420, 1740, 1730, 1700, 1610, 1500, 1305 cm^{-1}. MS (m/e) (%): 615 (M^+) (25), 525 (2), 382 (14), 291 (6), 234 (100), 201 (13), 91 (45). HRMS for $C_{33}H_{33}N_3O_9$ (M^+); Calcd 615.2218. Found 615.2204.

Treatments of the (*2S*,8S**)-isomer of **44** (7.0 mg, 0.02 mmol) in the same manner as described for **44** gave **48** as a yellow powder (6.2 mg, 62%) after separation with preparative thin layer chromatography. ^{1}H-NMR ($CDCl_3$): δ = 1.67 (s, 3H, 2-CH_3), 2.55 (br, 1H, -*OH*), 3.78 (s, 3H, -CO_2CH_3), 3.79 (dd, 1H, J=10.7, 5.9 Hz, C_8-*H*), 3.96-4.02 (2H, C_7-H_2), 3.91, 3.94, 4.07 (sx3, each 3H, -OCH_3x3), 4.49 (dd, 1H, J=10.7, 4.4 Hz, -*CH*OH), 4.61 (t, 1H, J=10.3 Hz, -*CH*OH), 5.20 (s, 2H, -OCH_2Ph), 5.25 (s, 1H, C_3-*H*), 6.85 (s, 1H, C_4-*H*), 6.96 (d, 1H, J=2.4 Hz, $C_{3'}$-*H*), 7.36-7.50 (5H, -C_6H_5), 8.45 (s, 1H, C_5-*H*), 9.37 (br s, 1H, $C_{1'}$-*H*). IR ($CHCl_3$): 3445, 1740, 1700, 1615, 1500, 1305 cm^{-1}. MS (m/e) (%): 615 (M^+) (25), 382 (14), 234 (100), 201 (12), 91 (45). HRMS for $C_{33}H_{33}N_3O_9$ (M^+); Calcd 615.2218. Found 615.2233.

Methyl (*2R*,8S)-4-benzyloxy-1,2,3,6,7,8-hexahydro-8-(methanesulfonyloxy)methyl-2-methyl-1-oxo-6-[(5,6,7-trimethoxy-*1H*-indole-2-yl)carbonyl]benzo[1,2-b;4,3-b']dipyrrole-2-carboxylate (46) and Its (*2S*,8S**)-Isomer (49)**

To a solution of **45** (3.4 mg, 6 μmol) in CH_2Cl_2 (0.6 ml) was added methanesulfonyl chloride (0.7 μl, 8.4 μmol) and Et_3N (1.6 μl, 11 μmol) at 0 °C, and the mixture was stirred for 30 min. The reaction mixture was washed with 1% $NaHCO_3$ and brine, dried over anhydrous Na_2SO_4, filtered, then concentrated *in vacuo*. Preparative thin layer chromatography (SiO_2; CH_2Cl_2:EtOAc=7:2) of the residue gave **46** as a yellow caramel (3.8 mg, 99%). ^{1}H-NMR ($CDCl_3$): δ = 1.69 (s, 3H, 2-CH_3), 3.04 (s, 3H, -SO_2CH_3), 3.77 (s, 3H, -CO_2CH_3), 3.92, 3.95, 4.08 (sx3, each 3H, -OCH_3x3), 4.15 (m, 1H, C_8-*H*), 4.39 (t, 1H, J=8.3 Hz, -*CH*OMs), 4.58-4.67 (2H, C_7-H_2), 4.80 (dd, 1H, J=10.3, 3.4 Hz), 5.21 (s, 2H, -OCH_2Ph), 6.87 (s, 1H, C_4-*H*), 6,96 (d, 1H, J=2.3 Hz, $C_{3'}$-*H*), 7.39-7.51 (5H, -C_6H_5), 8.42 (s, 1H, C_5-*H*), 9.33 (br s, 1H, $C_{1'}$-*H*). IR ($CHCl_3$): 3445, 1735,

1700, 1615, 1500, 1365, 1305, 1170 cm^{-1}. MS (m/e) (%): 693 (M^+) (4), 597 (40), 506 (3), 364 (26), 303 (15), 234 (80), 91 (100). HRMS for $C_{34}H_{35}N_3O_{11}S$ (M^+); Calcd 693.1994. Found 693.1972.

The same treatments of **48** (6.2 mg, 0.01 mmol) as described for **45** gave **49** as a yellow caramel (6.9 mg, 99%) after purification with preparative thin layer chromatography. ^{1}H-NMR ($CDCl_3$): δ = 1.67 (s, 3H, 2-CH_3), 2.98 (s, 3H, -SO_2CH_3), 3.79 (s, 3H, -CO_2CH_3), 3.92, 3.95, 4.08 (sx3, each 3H, -OCH_3x3), 4.18 (m, 1H, C_8-H), 4.46 (dd, 1H, J=9.8, 7.8 Hz, -CHOMs), 4.57-4.68 (2H, C_7-H_2), 4.75 (dd, 1H, J=9.8, 3.4 Hz), 5.21 (s, 2H, -OCH_2Ph), 5.26(s, 1H, 3-NH), 6.87 (s, 1H, C_4'-H), 6,96 (d, 1H, J=2.4 Hz, C_3'-H), 7.39-7.51 (5H, -C_6H_5), 8.43 (s, 1H, C_5-H), 9.34 (br s, 1H, C_1'-H). IR ($CHCl_3$): 3445, 1735, 1695, 1610, 1490, 1360, 1300, 1170 cm^{-1}. MS (m/e) (%): 693 (M^+) (2), 597 (25), 364 (16), 273 (17), 234 (54), 91 (100). HRMS for $C_{34}H_{35}N_3O_{11}S$ (M^+); Calcd 693.1994. Found 693.1994.

Methyl (*2R*,8S)-1,2,3,6,7,8-hexahydro-4-hydroxy-8-(methanesulfonyloxy)methyl-2-methyl-1-oxo-6-[(5,6,7-trimethoxy-*1H*-indole-2-yl)carbonyl]benzo[1,2-b;4,3-b']dipyrrole-2-carboxylate (47) and Its (*2S*,8S**)-Isomer (50)**

A solution of **46** (3.8 mg, 5.4 μmol) in THF (0.5 ml) was hydrogenated in the presence of 10% Pd/C (2 mg) at 1 atm. After filtration of the catalyst, the filtrate was concentrated *in vacuo*. Preparative thin layer chromatography (SiO_2; $CHCl_3$:acetone=2:1) of the residue gave **47** (2.8 mg, 85%) as yellow crystals. ^{1}H-NMR ($CDCl_3$): δ = 1.69 (s, 3H, 2-CH_3), 3.03 (s, 3H, -SO_2CH_3), 3.69 (s, 3H, -CO_2CH_3), 3.91, 3.95, 4.11 (sx3, each 3H, -OCH_3x3), 4.15 (m,1H, C_8-H), 4.38 (t, 1H, J=9.8 Hz, -CHOMs), 4.58-4.59 (2H, C_7-H_2), 4.78 (dd, 1H, J=9.8, 2.9 Hz, -CHOMs), 5.53(br s, 1H), 6.83 (s, 1H, C_4'-H), 6,98 (d, 1H, J=2.0 Hz, C_3'-H), 8.52 (s, 1H, C_5-H), 9.58 (br s, 1H, C_3-H), 9.73 (br s, 1H, C_1'-H). IR ($CHCl_3$): 3355, 2255, 1700, 1585, 1460, 1380, 1300, 1100, 890 cm^{-1}. SIMS: 604 $(M+H)^+$. HRMS for $C_{26}H_{25}N_3O_8$ [$(M\text{-}MsOH)^+$]; Calcd 507.1643. Found 507.1644.

Treatments of **49** (6.9 mg, 9.9 μmol) in the same manner as described for **46** gave **50** as yellow crystals (5.0 mg, 83%) after purification with preparative thin layer chromatography. ^{1}H-NMR ($CDCl_3$): δ = 1.69 (s, 3H, 2-CH_3), 2.98 (s, 3H, -SO_2CH_3), 3.77 (s, 3H, -CO_2CH_3), 3.91, 3.96, 4.13 (sx3, each 3H, -OCH_3x3), 4.15 (m,1H, C_8-H), 4.47 (dd, 1H, J=9.8, 7.8 Hz, -CHOMs), 4.57-4.66 (2H, C_7-H_2), 4.75 (dd, 1H, J=9.8, 2.9 Hz, -CHOMs), 5.41(s, 1H), 6.85 (s, 1H, C_4'-H), 6,99 (d, 1H, J=2.0 Hz, C_3'-H), 8.50 (s, 1H, C_5-H), 9.51 (br s, 1H, C_1'-H), 9.67(br s, 1H). IR ($CHCl_3$): 3360, 2270, 1690, 1590, 1460, 1370, 1305, 1100, 900 cm^{-1}. SIMS: 604 $(M+H)^+$. HRMS for $C_{26}H_{25}N_3O_8$ [$(M\text{-}MsOH)^+$]; Calcd 507.1643. Found 507.1632.

***dl*-Duocarmycin A (*dl*-1) and *dl*-2-Epiduocarmycin A (*dl*-2-epi-1)**

To a solution of **47** (2.8 mg, 4.6 μmol) in THF (1.5 ml) was added NaH (50% oil dispersion, 0.8 mg, 20 μmol), and the mixture was stirred for 3.5 hr at room temperature. After dilution with EtOAc, the reaction mixture was washed with 0.5 M KH_2PO_4 solution and brine, dried over anhydrous Na_2SO_4, filtered, then concentrated *in vacuo*. Flash chromatography (alumina; $CHCl_3$:acetone=5:1) of the residue gave *dl*-**1** as a pale yellow solid (1.4 mg, 60%). ^{1}H-NMR ($CDCl_3$): δ = 1.29 (t, 1H, J=4.4 Hz, C_4-H), 1.67 (s, 3H, 2-CH_3), 2.25 (dd, 1H, J=7.8, 3.9 Hz, C_4-H), 3.07 (m, 1H, C_{4a}-H), 3.75 (s, 3H, -CO_2CH_3), 3.89, 3.94, 4.08 (sx3, each 3H, -OCH_3x3), 4.42 (d, 1H, J=10.3 Hz), 4.45 (dd, 1H, J=10.7, 4.4 Hz), 6.01 (s, 1H, 1-NH), 6.78 (s, 1H, C_4'-H), 6.94 (d, 1H, J=2.4 Hz, C_3'-H), 7.18 (s, 1H, C_7-H), 9.24 (br s, 1H, $1'$-NH). IR ($CHCl_3$): 3430, 3280, 1735, 1680, 1630, 1380, 1300 cm^{-1}. MS (m/e) (%): 507 (M^+) (22), 448 (4), 274 (4), 234 (100). These spectral behaviors were identical with those of natural (+)-**1** measured in the same states. HRMS for $C_{26}H_{25}N_3O_8$ (M^+); Calcd 507.1643. Found 507.1638.

The same treatments of **50** (6.0 mg, 9.9 μmol) as described for **47** gave *dl*-2-epi-**1** as a pale yellow solid (2.4 mg, 56%) after purification with flash chromatography. ^{1}H-NMR ($CDCl_3$): δ = 1.33 (t, 1H, J=4.4 Hz, C_4-H), 1.65 (s, 3H, 2-CH_3), 2.24 (dd, 1H, J=7.8, 3.9 Hz, C_4-H), 3.09 (m, 1H, C_{4a}-H), 3.78 (s, 3H, -CO_2CH_3), 3.90, 3.94, 4.08 (sx3, each 3H, -OCH_3x3), 4.38-4.47 (m, 2H, C_5-H_2), 6.04 (s, 1H, 1-NH), 6.78 (s, 1H, C_4'-H), 6.94 (d, 1H, J=2.5 Hz, C_3'-H), 7.17 (s, 1H, C_7-H), 9.23 (br s, 1H, 1'-NH). This ^{1}H-NMR spectrum was obviously different from that of natural (+)-**1** measured in the same state. IR ($CHCl_3$): 3430, 1730, 1690, 1605, 1370, 1300 cm^{-1}. MS (m/e) (%): 507 (17), 448 (3), 274 (3), 234 (100). HRMS for $C_{26}H_{25}N_3O_8$ (M^+); Calcd 507.1643. Found 507.1627.

General Procedure for Cytotoxicity Assay

P388 murine leukemia cells ($7.5x10^4$/ml) were seeded in dishes containing the RPMI-1640 medium which involves fetal bovin serum in a 10% concentration. Compounds to be tested were added in graded concentrations. After cultivation for 72 hr at 37°C, the tumor cells were counted by MTT method,[21] and the IC_{50} value was determined by means of the growth curve.

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