Novel Dienediyne Systems Related to the Neocarzinostatin Chromophore: Molecular Design, Chemical Synthesis, and Evaluation

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Abstract: The molecular design and chemical synthesis of novel dienediyne systems related to the neocarzinostatin chromophore (**1**), which is the labile heart of an antitumor antibiotic, neocarzinostatin, and their chemical and DNA cleaving properties are described. The monocyclic dienediynes **6**−**8**, which are high simplified analogs of the neocarzinostatin chromophore (**1**), were effectively synthesized from xylitol (**13**) in a short step. The synthesis includes the conversion of the ketoaldehyde **24** into the highly strained 10-membered ring keto-enediyne **25** by a simple intramolecular aldol condensation using lithium hydroxide as the key step. The dienediyne **8** possessing acetoxy groups as leaving groups at propargylic positions was smoothly cycloaromatized by methyl thioglycolate in the presence of triethylamine in methanol to give two benzenoides **28** and **29** through radical pathways. The addition of pyrrolidine to **8** in ethanol also afforded the benzenoid **38** via a pathway similar to that for **29**. Furthermore, it was clearly found that the dienediyne **8** effectively cleaved DNA without any additive and the DNA cleaving activities significantly increased in the presence of the thiol, methyl thioglycolate.

Introduction

Neocarzinostatin (NCS) is a naturally occurring antitumor antibiotic isolated from *Streptomyces carzinostaticus* var. F-41 by Ishida and co-workers in 1965 as an original member of enediyne antibiotics[2] and has been used for the treatment of pancreatic cancer, gastric cancer, and leukemia in humans.[3] The clinically used agent consists of a structurally unprecedented non-protein chromophore (neocarzinostatin chromophore, NCS-C (**1**)) and its separable carrier apoprotein (apo-NCS). Goldberg et al. reported that the NCS-C (**1**) was essentially responsible for the biological activity of NCS and exhibits potent cytotoxicity and DNA cleaving activity.[4] The NCS-C (**1**) has three main structural subunits, a substituted naphthoate moiety, an amino sugar, and a highly strained bicyclo[7.3.0]dodecadienediyne epoxide unit.[5] **1** is exceedingly unstable, undergoing rapid decomposition at elevated pH or upon exposure to air or ambient light. The DNA cleavage is now recognized to be initiated by the nucleophilic addition of a thiol to the C12 of **1** followed by a rapid cycloaromatization reaction (Myers cyclization)[6] of an enyne-cumulene **2**, leading to the formation of a benzenoid diradical **3**, which is capable of cleaving DNA via hydrogen abstraction from the DNA sugar backbone with a high degree of base selectivity (T > A ≫ C ~ G)[7] (Figure 1). With the stimulant chemical and biological backgrounds, great effort has been devoted to the synthesis of the core units or the development of new analogs of **1**.[8−25] Novel DNA cleaving molecules, particularly those with high efficiency and sequence specificity, have considerable potential in chemistry, biology, and medicine.[26] In this context, we started a series of studies directed

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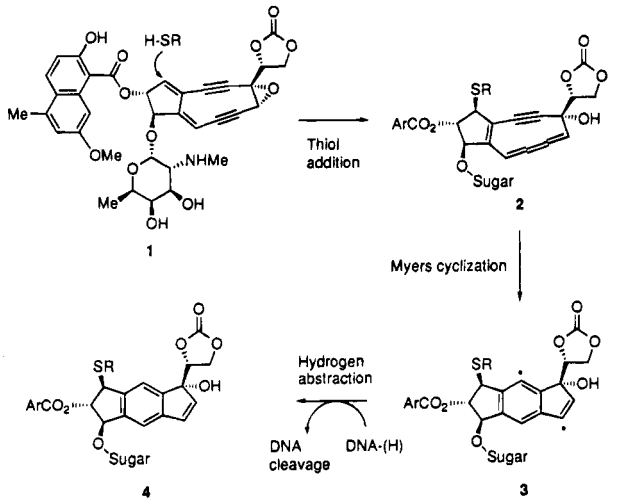

Figure 1. Mechanism of DNA-cleaving action of neocarzinostatin chromophore (**1**).

toward the design, synthesis, and investigation of novel DNA-cleaving molecules reminiscent of both the chemistry and biological action of **1**.[27] As a part of our studies, we designed the 10-membered monocyclic dienediyne system **5**, which is a highly simplified analog of the NCS-C (**1**).[28] Thus far, chemical synthesis, chemical properties, and DNA-cleaving activities of

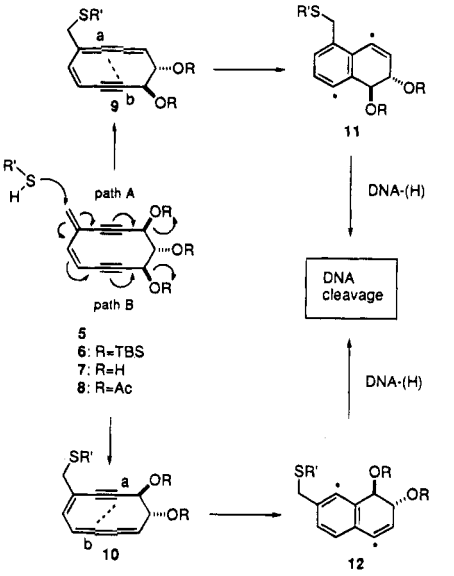

Figure 2. Designed dienediyne systems and their presumed mechanism of DNA-cleaving action.

a monocyclic system containing a conjugated dienediyne function have not been studied. In this paper, we report the full account of the molecular design, chemical synthesis, cycloaromatizations for the diradical formation, and DNA-cleaving activities of the novel dienediyne molecules **6**–**8** related to the NCS-C (**1**).

Results and Discussion

Design of Dienediyne Systems. The novel monocyclic dienediyne system **5** was designed on the basis of the following plan and expectation (Figure 2). (1) The 10-membered ring system[29] of **5** would have far increased stability as compared with that of the NCS-C (**1**) possessing the nine-membered ring system, and (2) the dienediyne system, which has good leaving groups at the propargylic positions (C6 and C8 positions), like **8** would smoothly cycloaromatize in the presence of a thiol through two enyne-cumulene intermediates **9** (path A) and **10** (path B) to produce two diradical species **11** and **12**, respectively, both of which would be capable of cleaving DNA. Furthermore, the monocyclic dienediyne system was expected to answer the question as to what is the minimum structure needed for such a novel molecular transformation generating diradical species and DNA cleavage. According to Nicolaou's report, the distances *ab* of the enyne-cumulenes **9** and **10** must be within ca. 3.3 Å for spontaneous aromatization at ambient temperature.[30] Molecular calculations indicated that the distances *ab* of the enyne-cumulenes **9** (R = H, R′ = Me) and **10** (R = H, R′ = Me) were 3.06 Å (by AM1) or 308 Å (by PM3) and 3.04 Å (by AM1) or 3.06 Å (by PM3), respectively.[31] Considering these points and the mechanism of DNA cleavage by the NCS-C (**1**), the novel monocyclic dienediyne system, especially **8** possessing good leaving groups at suitable positions, was expected to have a simple and indispensable structure and chemical properties for effective DNA cleavage.

Synthesis of Dienediynes 6–8. The straightforward synthesis of the novel dienediynes **6**–**8** is summarized in Scheme 1. In this synthesis, xylitol (**13**) was chosen as a cheap and

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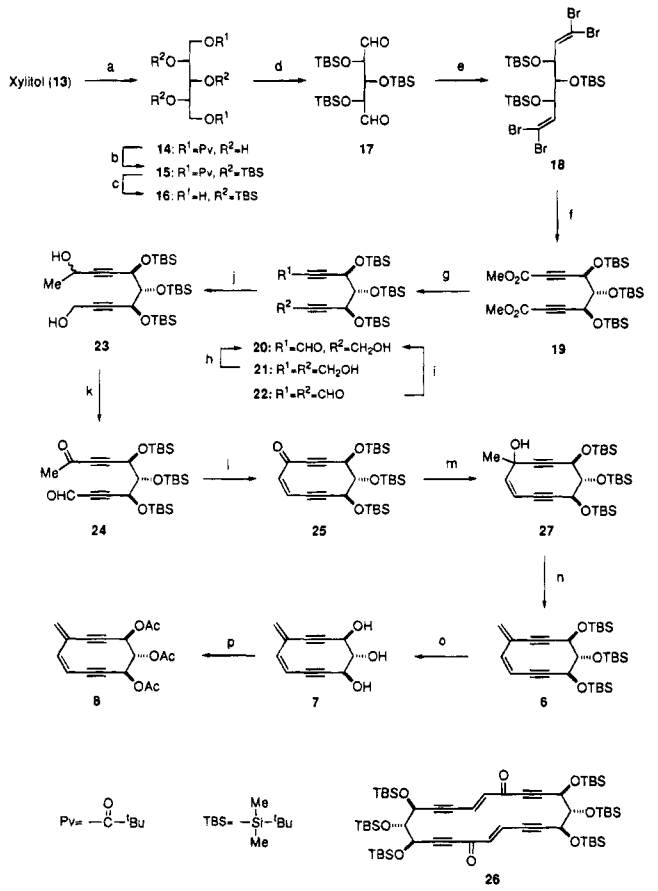
Scheme 1[a]



[a] Synthesis of designed dienediynes **6**–**8**. Reagents and conditions: (a) PvCl, Py, 26 °C, 15 h, 69%; (b) TBSCl, imidazole, DMF, 80 °C, 12 h, 95%; (c) DIBALH, PhMe, −78 °C, 40 min, 96%; (d) $(COCl)_2$, DMSO, Et_3N, CH_2Cl_2, −78 → 0 °C, 1.5 h, 100%; (e) CBr_4, PPh_3, CH_2Cl_2, 0 °C, 0.5 h, 97%; (f) *n*-BuLi, Et_2O, −78 °C, 0.5 h then $ClCO_2Me$, −78 °C, 1 h, 86%; (g) DIBALH, PhMe, −78 °C, 10 min then 0 °C, 20 min, 45%; (h) Dess–Martin periodinane, CH_2Cl_2, 0 °C, 0.5 h, 53%; (i) $NaBH_4$, MeOH, 0 °C, 0.5 h, 41%; (j) MeMgBr, Et_2O, 25 °C, 10 min, 95%; (k) Dess–Martin periodinane, CH_2Cl_2, 26 °C, 45 min, 99%; (l) LiOH, EtOH–H_2O (0.004 M for **24**), 26 °C, 2 h, 45%; (m) MeLi, Et_2O, 0 °C, 10 min, 88%; (n) MsCl, Et_3N, CH_2Cl_2, 0 °C, 20 min, 73%; (o) TBAF, THF, 0 °C, 0.5 h; (p) Ac_2O, Et_3N, 0 °C, 1 h, 48% from **6**.

readily available starting material. Our synthetic approach began with the selective conversion of **13** into the suitably protected diol **16** in three steps. Thus, the primary alcohols of **13** were selectively protected with pivaloyl groups using 2.5 equiv of pivaloyl chloride in pyridine to give the triol **14** in 69% yield. The triol **14** was then totally silylated with *tert*-butyldimethylsilyl groups to afford **15** in 95% yield. Subsequent reduction of the pivaloyl esters of **15** using diisobutylaluminum hydride (DIBALH) in toluene at −78 °C gave the diol **16** in 96% yield. The Swern oxidation of **16**, followed by bromo-olefination of the resulting crude dialdehyde **17** by Corey's method[32] using carbon tetrabromide and triphenylphosphine in CH_2Cl_2 gave the tetrabromide **18** in 97% overall yield. The bromide **18** was treated with 6.0 equiv of *n*-butyllithium/hexane in ether at −78 °C for 30 min and then 10 equiv of methyl chloroformate at −78 → 0 °C for 1 h to give the diyne **19** in 86% yield. Selective reduction of the methyl esters of **19** with 3.0 equiv of DIBALH in toluene at −78 → 0 °C for 30 min afforded the monoaldehyde **20** in 45% yield along with the diol **21** (38%) and the dialdehyde **22** (12%). Both **21** and **22** could be selectively converted into the desired **20** by the Dess–Martin oxidation[33] and reduction

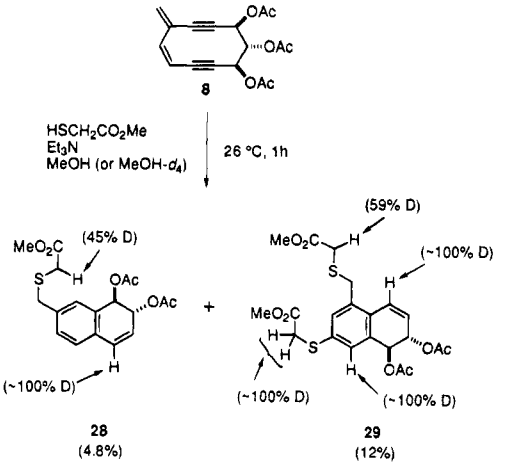


Figure 3. Cycloaromatization of **8** by methyl thioglycolate.

using sodium borohydride, respectively. The Grignard reaction of **20** using 4.5 equiv of methylmagnesium bromide in ether gave the alcohol **23**, which was subjected to the Dess–Martin oxidation to give the key intermediate, ketoaldehyde **24**, in 94% overall yield. The one-step conversion of **24** into the highly strained 10-membered ring keto-enediyne system was best effected by using 2.0 equiv of lithium hydroxide in ethanol–H_2O at 26 °C for 2 h under high dilution conditions (0.004 M for **24**) to afford the monocyclic product **25** and the dimer **26** in 45% and 34% yields, respectively. Notably, the 10-membered ring keto-enediyne **25** was found to be quite stable when handled at room temperature in air or ambient light. We next tried the Wittig reaction using methylenetriphenylphosphorane and the Horner–Wadsworth–Emmons reaction using dimethyl methylphosphonate and a suitable base to introduce an olefinic function onto **25**. Unfortunately, both attempts failed because of the low reactivity of the highly conjugated ketone of **25**. However, the desired dienediyne system was obtained in the following two steps. Thus, nucleophilic addition of methyl lithium to **25**, followed by dehydration of the resulting tertiary alcohol of **27** via methanesulfonylation using methanesulfonyl chloride and triethylamine gave the dienediyne **6** in 57% overall yield. Although the dienediyne triol **7** was obtained by standard desilylation using tetra-*n*-butylammonium fluoride (TBAF) in THF, **7** was found to be extremely unstable when handled at room temperature. Therefore, the dienediyne **8** possessing acetoxy groups as leaving groups at propargylic positions was synthesized in 48% yield by desilylation using TBAF following in situ acetylation using acetic anhydride and triethylamine in THF without isolation of the unstable dienediyne triol **7**. Although the protected dienediynes **6** and **8** were more stable than the free dienediyne **7** and could be handled at room temperaure in air or ambient light, **6** and **8** were considerably unstable when stored neat.

Cycloaromatizations of Dienediyne 8. Our attention next turned to the mode of cycloaromatization of the novel dienediyne system in the presence of a thiol. The addition of 3.0 equiv of methyl thioglycolate to the dienediyne **8** in the presence of 1.0 equiv of triethylamine in MeOH at 26 °C for 1 h gave the benzenoid products **28** and **29** in 4.8% and 12% yields, respectively. A similar experiment conducted in deuteriated solvent, MeOH-d_4, afforded **28** and **29** with the indicated levels of deuterium incorporation (Figure 3). Furthermore, when 0.3 equiv of methyl thioglycolate was used in the aromatization reaction, a significant decrease in the yield of **29** was observed and **28** and **29** were isolated in 4.5% and 3.5% yields,

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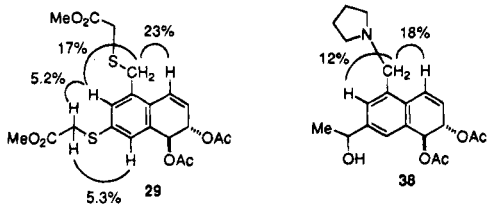

Figure 4. Selected NOE data of **29** and **38**.

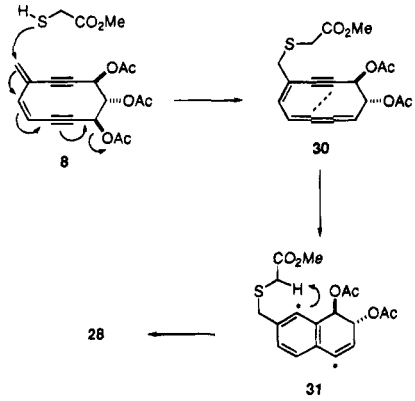

Figure 5. Mode of cycloaromatization of **8**.

respectively. The structure of **29** was ascertained by the observation of NOE experiments (Figure 4). From these results, the formation of **28** clearly indicates that the monocyclic dienediynes **8** undergoes the addition of methyl thioglycolate to produce the enyne-cumulene **30**, which proceeds the cycloaromatization leading to the benzenoid diradical **31**. The diradical intermediate **31** then undergoes a particularly effective intramolecular hydrogen atom transfer from the methylene group of the methyl thioglycolate moiety[34] as shown in Figure 5. On the other hand, the formation of **29** strongly suggests the following mechanism for another cycloaromatization pathway as shown in Figure 6. First, the addition of methyl thioglycolate to **8** generates the enyne-cumulene **32**,[35] which produces the diradical **33** by cycloaromatization. The benzenoid diradical **33** is equivalent to the allenic diradical **34**.[36] Under the aerobic conditions, the thiyl radical formed from the thiol and oxygen[37] might attack the less hindered C7 position of **34** to produce the adduct **35**. Furthermore, **35** would be converted into the diradical **36**, which is equivalent to the benzenoid diradical **37**, by abstraction of the hydrogen atom. Finally, the diradical **37** traps the hydrogen atoms from the solvent, methanol. In this case, the deuterium incorporation at the methylene groups of the two thioglycolate residues as indicated in Figure 3 would arise from the intermolecular hydrogen atom abstraction by deuteriated methanol radicals, which resulted from the benzenoid diradical, followed by trapping deuteriums from deuteriated

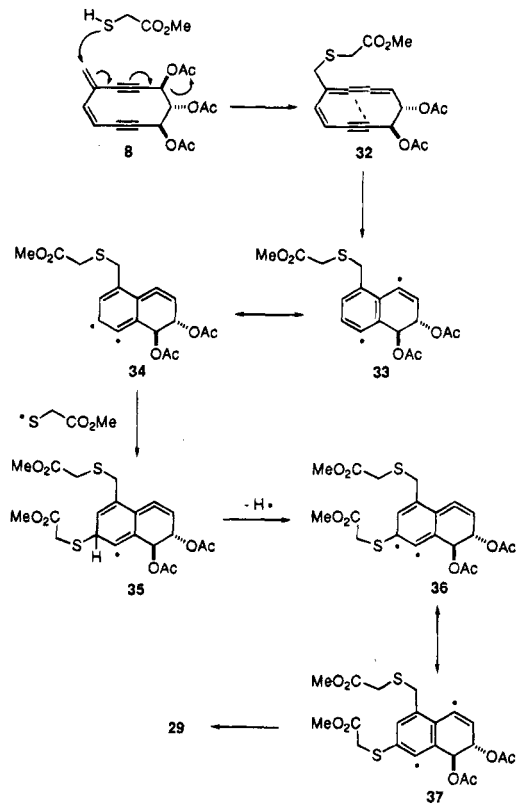

Figure 6. Mode of cycloaromatization of **8**.

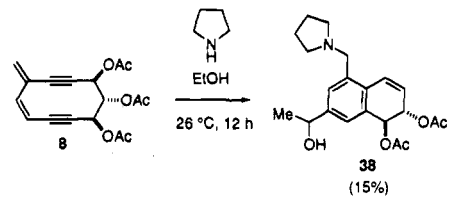

Figure 7. Cycloaromatization of **8** by pyrrolidine.

methanol. We also found that the addition of pyrrolidine instead of thiol to **8** in ethanol at 26 °C for 12 h afforded the benzenoid product **38** in 15% yield as shown in Figure 7. The structure of **38**, which was confirmed by NOE experiments (Figure 4), indicated that the benzenoid **38** was produced via a pathway similar to that for **29** by addition of a radical generated from ethanol. These results disclose that the dienediyne **8** is smoothly cycloaromatized not only by a thiol but also by an amine to produce the diradical species.[38]

DNA Cleavage with Dienediynes. The DNA-cleaving properties of the novel dienediynes **6** and **8**, except for the unstable **7**, were assayed using double-stranded supercoiled ΦX174 DNA. As expected from the mode of the cycloaromatizations, only the dienediyne **8** possessing good leaving groups at the propargylic positions was found to cleave DNA. Thus, aerobic incubations of **8** with the covalently closed supercoiled DNA (form I) at pH 6.5, 7.0, and 8.5, each in concentration of 10 000, 1000, and 100 μM at 37 °C for 24 h without any additive, caused a single-strand break leading to the nicked open circular DNA (form II) as shown in Figure 8. It is interesting to note that the production of form II DNA slightly increases as the pH decreases.[39] Furthermore, signifi-

(34) For related observations, see: (a) References 8c, 9g, and 10d. (b) Chin, D.-H.; Goldberg, I. H. *J. Am. Chem. Soc.* **1992**, *114*, 1914.

(35) For related observations, see: (a) Reference 9b. (b) Sugiyama, H.; Yamashita, K.; Nishi, M.; Saito, I. *Tetrahedron Lett.* **1992**, *33*, 515. (c) Sugiyama, H.; Fujiwara, T.; Kawabata, H.; Yoda, N.; Hirayama, N.; Saito, I. *J. Am. Chem. Soc.* **1992**, *114*, 5578.

(36) (a) Dilon, P. W.; Underwood, G. R. *J. Am. Chem. Soc.* **1974**, *96*, 779. (b) Dykstra, C. E.; *J. Am. Chem. Soc.* **1977**, *99*, 2060. (c) Seeger, R.; Krishnan, R.; Pople, J. A.; Schleyer, P. R. *J. Am. Chem. Soc.* **1977**, *99*, 7103. (d) Schmidt, M. W.; Angus, R. O., Jr.; Johnson, R. P. *J. Am. Chem. Soc.* **1982**, *104*, 6838. (e) Krogh-Jespersen, K. *J. Comput. Chem.* **1982**, *3*, 571. (f) Lam, B.; Johnson, R. P. *J. Am. Chem. Soc.* **1983**, *105*, 7479. (g) Angus, R. O., Jr.; Schmidt, M. W.; Johnson, R. P. *J. Am. Chem. Soc.* **1985**, *107*, 532. (h) Johnson, R. P. *Chem. Rev.* **1989**, *89*, 1111. (i) Danheiser, R. L.; Gould, A. E.; Pradilla, R. F.; Helgason, A. L. *J. Org. Chem.* **1994**, *59*, 5514.

(37) (a) Fava, A.; Reichenbach, G.; Peron, U. *J. Am. Chem. Soc.* **1967**, *89*, 6696. (b) Reference 9b.

(38) Intramolecular oxygen and carbon addition of naphthoate esters have been recently reported, see: (a) Lamothe, M.; Fuchs, P. L. *J. Am. Chem. Soc.* **1993**, *115*, 4483. (b) Hensens, O. D.; Helms, G.; Zink, D. L.; Chin, D.-H.; Kappen, L. S.; Goldberg, I. H. *J. Am. Chem. Soc.* **1993**, *115*, 11030.

(39) Similar tendency was observed in DNA damage by the NCS-C (**1**), see: Chin, D.-H.; Goldberg, I. H. *Biochemistry* **1986**, *25*, 1009.

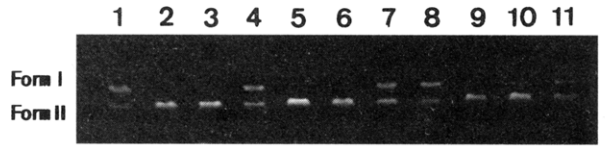

Figure 8. DNA cleavage with **8**. ΦX174 form I DNA (50 μM/base pair) was incubated at 37 °C for 24 h with **8** in 20% ethanol in various pH buffers and analyzed by electrophoresis (1% agarose gel, ethidium bromide stain): lane 1, DNA alone at pH 6.5; lane 2, **8** (10 000) at pH 6.5; lane 3, **8** (1000) at pH 6.5; lane 4, **8** (100) at pH 6.5; lane 5, **8** (10 000) at pH 7.0; lane 6, **8** (1000) at pH 7.0; lane 7, **8** (100) at pH 7.0; lane 8, DNA alone at pH 8.5; lane 9, **8** (10 000) at pH 8.5; lane 10, **8** (1000) at pH 8.5; lane 11, **8** (100 μM) at pH 8.5.

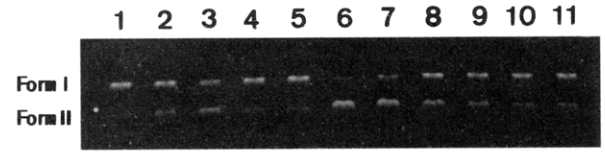

Figure 9. DNA cleavage with **8**. ΦX174 form I DNA (50 μM/base pair) was incubated at 37 °C for 24 h with **8** in 20% ethanol in Tris-HCl buffer (pH 7.0, 50 mM) and analyzed by electrophoresis (1% agarose gel, ethidium bromide stain): lane 1, DNA alone; lane 2, methyl thioglycolate (100); lane 3, **8** (100); lane 4, **8** (10); lane 5, **8** (1); lane 6, **8** (100) + methyl thioglycolate; lane 7, **8** (10) + methyl thioglycolate; lane 8, **8** (1) + methyl thioglycolate; lane 9, **8** (0.1) + methyl thioglycolate; lane 10, **8** (0.01) + methyl thioglycolate; lane 11, **8** (0.001 μM) + methyl thioglycolate.

cant enhancement of the DNA-cleaving activity of **8** was observed with the addition of thiol. Thus, the DNA-cleaving activity of **8** in the presence of methyl thioglycolate was 1000 times higher than that in the absence of methyl thioglycolate, and **8** cleaved DNA even at 0.1 μM and 37 °C (Figure 9). Remarkably, the potency of **8** was quite outstanding compared to that of the reported dienediyne-based nonnatural systems and very similar to that of the NCS-C (**1**).[40]

Conclusions

The present work shows not only the molecular design and chemical synthesis of novel dienediyne systems related to the neocarzinostatin chromophore but also their modes of cycloaromatizations and DNA-cleaving activities. It was clarified that even a designed simple molecule had strong DNA-cleaving activities at 37 °C and its activity could significantly increase in the presence of a thiol. The described chemistry and biological evaluation provided significant information about the molecular design of novel and simple DNA-cleaving agents based on the dienediyne system.

Experimental Section

General Methods. Melting points were determined on a micro hot-stage Yanaco MP-S3. ^{1}H-NMR spectra were obtained on a JEOL GSX270 spectrometer in $CDCl_3$ using TMS as the internal standard unless otherwise noted. High-resolution mass spectra (HRMS) were recorded on a JEOL LMS-DX302 mass spectrometer under electron impact (EI) conditions. Silica gel TLC and column chromatography were performed on Merck TLC 60F−254 (0.25 mm) and Merck Kieselgel 60 or Fuji-Davison BW-820MH, respectively. Preparative thin-layer chromatography was performed on 0.5 mm × 20 cm × 20 cm Merck silica gel plates (60F-254). Air- and/or moisture-sensitive reactions were carried out under an atmosphere of argon with oven-dried glassware. In general, organic solvents were purified and dried by the appropriate procedure, and evaporation and concentration were carried out under reduced pressure below 30 °C, unless otherwise noted.

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1,5-Di-*O*-pivaloylxylitol (14). To an ice-cold solution of xylitol (**13**) (10.4 g, 0.0686 mol) in dry pyridine (160 mL) was added dropwise pivaloyl chloride (21.1 mL, 0.171 mol) over 15 min with stirring. After the reaction mixture was stirred at 26 °C for 15 h, the mixture was concentrated *in vacuo*. H_2O (100 mL) was added to the residue, and then the resulting mixture was extracted with ethyl acetate (50 mL × 5). The extracts were washed with saturated aqueous NaCl (100 mL), dried over anhydrous Na_2SO_4, and concentrated *in vacuo*. Purification of the residue by flash column chromatography (500 g of silica gel, 3:1 toluene−acetone) gave **14** (15.2 g, 69%) as white crystals: R_f 0.25 (3:1 toluene−acetone); mp 75.5−76.0 °C (diethyl ether/*n*-hexane); ^{1}H-NMR δ 1.21 (18H, s, *t*-Bu of Pv), 3.00 (1H, d, J = 7.6 Hz, OH), 3.17 (2H, d, J = 4.0 Hz, OH), 3.56 (1H, dt, J = 7.6 and 6.0 Hz, H-3), 3.93−4.05 (2H, m, H-2 and 4), 4.22 (2H, d, J = 6.0 Hz, H-1 and 5); HRMS (EI) *m/z* 321.1949 (321.1913 calcd for $C_{15}H_{29}O_7$, M + H$^+$).

2,3,4-Tri-*O*-(*tert*-butyldimethylsilyl)-1,5-di-*O*-pivaloylxylitol (15). To an ice-cold solution of **14** (1.93 g, 6.02 mmol) in dry DMF (39 mL) were added imidazole (2.05 g, 30.1 mmol) and *tert*-butyldimethylsilyl chloride (4.08 g, 27.1 mmol) with stirring. After the resulting solution was stirred at 80 °C for 12 h, the reaction was quenched with H_2O (100 mL) and then the resulting mixture was extracted with *n*-hexane (50 mL × 3). The extracts were washed with saturated aqueous NaCl (100 mL), dried over anhydrous Na_2SO_4, and concentrated *in vacuo*. Purification of the residue by flash column chromatography (120 g of silica gel, 30:1 *n*-hexane−diethyl ether) gave **15** (3.79 g, 95%) as white crystals: R_f 0.24 (30:1 *n*-hexane−diethyl ether); mp 109.5−110.0 °C (methanol); ^{1}H-NMR δ 0.06 (6H, s, Me of TBS), 0.11 (6H, s, Me of TBS), 0.13 (6H, s, Me of TBS), 0.88 (9H, s, *t*-Bu of TBS), 0.90 (9H, s, *t*-Bu of TBS), 1.20 (9H, s, *t*-Bu of TBS), 3.73 (1H, t, J = 3.6 Hz, H-3), 4.02−4.23 (2H, m, H-2 and 4), 4.12 (2H, d, J = 11.0 Hz, H-1 and 5), 4.18 (2H, dd, J = 11.0 and 6.0 Hz, H-1 and 5); HRMS (EI) *m/z* 662.4431 (662.4429 calcd for $C_{33}H_{70}O_7Si_3$, M$^+$).

2,3,4-Tri-*O*-(*tert*-butyldimethylsilyl)xylitol (16). To a stirred solution of **15** (3.52 g, 5.31 mmol) in dry toluene (53 mL) at −78 °C was added dropwise 1.02 M diisobutylaluminum hydride (1.02 M = 1.02 mol dm^{-3}) in toluene (22.4 mL, 22.8 mmol). After the resulting solution was stirred at −78 °C for 40 min, the reaction was quenched with 1.64 M aqueous potassium sodium tartrate tetrahydrate (70 mL). The resulting mixture was stirred at 25 °C for 2 h and then extracted with ethyl acetate (30 mL × 3). The extracts were washed with saturated aqueous NaCl (50 mL), dried over anhydrous Na_2SO_4, and concentrated *in vacuo*. Purification of the residue by flash column chromatography (100 g of silica gel, 8:1 *n*-hexane−diethyl ether) gave **16** (2.53 g, 96%) as white crystals: R_f 0.30 (10:1 *n*-hexane−ethyl acetate); mp 108−109 °C (diethyl ether); ^{1}H-NMR δ 0.12 (18H, s, Me of TBS), 0.91 (27H, s, *t*-Bu of TBS), 2.30 (2H, t, J = 6.0 Hz, OH), 3.67 (2H, ddd, J = 17.0, 6.0 and 5.0 Hz, H-1 and 5), 3.74 (2H, ddd, J = 17.0, 6.0 and 5.2 Hz, H-1 and 5), 3.80 (1H, t, J = 4.8 Hz, H-3), 3.93 (2H, ddd, J = 5.2, 5.0 and 4.8 Hz, H-2 and 4); HRMS (EI) *m/z* 495.3352 (495.3358 calcd for $C_{23}H_{55}O_5Si_3$, M + H$^+$).

2,3,4-Tri-*O*-(*tert*-butyldimethylsilyl)-*xylo*-pentodialdose (17). To a stirred solution of oxalyl chloride (0.30 mL, 3.39 mmol) in dry CH_2Cl_2 (8.1 mL) at −78 °C was added dropwise a solution of dimethyl sulfoxide (0.33 mL, 4.52 mmol) in dry CH_2Cl_2 (0.8 mL). After the resulting solution was stirred at −78 °C for 10 min, a solution of **16** (0.56 g, 1.13 mmol) in dry CH_2Cl_2 (3.4 mL) was added to the reaction mixture at −78 °C. After the resulting solution was stirred at −78 °C for 25 min, triethylamine (1.58 mL, 11.3 mmol) was added to the reaction mixture. After 10 min, the resulting mixture was allowed to warm to 0 °C over 1.5 h with stirring. The reaction was quenched with H_2O (10 mL), and then the resulting mixture was extracted with *n*-hexane (20 mL × 3). The extracts were washed with saturated aqueous NaCl (10 mL), dried over anhydrous Na_2SO_4, and concentrated *in vacuo* to give **17** (0.557 g, 100%) as white crystals: R_f 0.80 (10:1 *n*-hexane−ethyl acetate); mp 91.0−91.5 °C (methanol); ^{1}H-NMR δ 0.03 (6H, s, Me of TBS), 0.05 (6H, s, Me of TBS), 0.07 (6H, s, Me of TBS), 0.88 (9H, s, *t*-Bu of TBS), 0.92 (18H, s, *t*-Bu of TBS), 4.05 (2H, dd, J = 4.0 and 1.0 Hz, H-2 and 4), 4.25 (1H, t, J = 4.0 Hz, H-3), 9.78 (2H, d, J = 1.0 Hz, CHO); HRMS (EI) *m/z* 491.2997 (491.3044 calcd for $C_{23}H_{51}O_5Si_3$, M + H$^+$).

(3*R*,5*S*)-1,1,7,7-Tetrabromo-3,4,5-tris[(*tert*-butyldimethylsilyl)-oxy]-1,6-heptadiene (18). To an ice-cold solution of carbon tetrabro-

mide (1.50 g, 5.42 mmol) in dry CH_2Cl_2 (9.0 mL) was added triphenylphosphine (2.40 g, 9.04 mmol) with stirring. After 3 min, a solution of **17** (0.555 g, 1.13 mmol) in dry CH_2Cl_2 (8.3 mL) was added dropwise to the reaction mixture and then the resulting mixture was stirred for 30 min under ice-cooling. The reaction was quenched with saturated aqueous $NaHCO_3$ (20 mL), and then the resulting mixture was extracted with chloroform (10 mL × 3). The extracts were washed with saturated aqueous NaCl (20 mL), dried over anhydrous Na_2SO_4, and concentrated *in vacuo*. Purification of the residue by flash column chromatography (40 g of silica gel, 20:1 *n*-hexane−diethyl ether) gave **18** (0.880 g, 97%) as a colorless oil: R_f 0.95 (20:1 *n*-hexane−diethyl ether); ^{1}H-NMR δ 0.07 (6H, s, Me of TBS), 0.08 (6H, s, Me of TBS), 0.11 (6H, s, Me of TBS), 0.90 (27H, s, *t*-Bu of TBS), 3.59 (1H, t, J = 4.0 Hz, H-4), 4.57 (2H, dd, J = 9.2 and 4.0 Hz, H-3 and 5), 6.74 (2H, d, J = 9.2 Hz, H-2 and 6).

Dimethyl (4*R*,6*S*)-4,5,6-Tris[(*tert*-butyldimethylsilyl)oxy]-2,7-nonadiyne-1,9-dioate (19). To a stirred solution of **18** (7.10 g, 8.85 mmol) in dry diethyl ether (70 mL) at −78 °C was added dropwise 1.63 M *n*-butyllithium/*n*-hexane (27.2 mL, 44.3 mmol). After 30 min, methyl chloroformate (6.80 mL, 88.5 mmol) was added dropwise to the reaction mixture. The resulting mixture was stirred at −78 °C for 30 min and then allowed to warm to 0 °C over 30 min. The reaction was quenched with saturated aqueous NH_4Cl (100 mL), and then the resulting mixture was extracted with ethyl acetate (70 mL × 3). The extracts were washed with saturated aqueous NaCl (100 mL), dried over anhydrous Na_2SO_4, and concentrated *in vacuo*. Purification of the residue by flash column chromatography (200 g of silica gel, 30:1 *n*-hexane−ethyl acetate) gave **19** (4.56 g, 86%) as a pale yellow oil: R_f 0.36 (10:1 *n*-hexane−diethyl ether); ^{1}H-NMR δ 0.12 (6H, s, Me of TBS), 0.14 (6H, s, Me of TBS), 0.17 (6H, s, Me of TBS), 0.92 (18H, s, *t*-Bu of TBS), 0.93 (9H, s, *t*-Bu of TBS), 3.76 (6H, s, OMe), 3.78 (1H, t, J = 4.8 Hz, H-5), 4.73 (2H, d, J = 4.8 Hz, H-4 and 6); HRMS (EI) *m/z* 598.3195 (598.3178 calcd for $C_{29}H_{54}O_7Si_3$, M^+).

***dl*-(4*R*,5*R*,6*R*)-4,5,6-Tris[(*tert*-butyldimethylsilyl)oxy]-9-hydroxy-2,7-nonadiyn-1-al (20).** To a stirred solution of **19** (57.6 mg, 0.0962 mmol) in dry toluene (1.2 mL) at −78 °C was added dropwise 1.02 M diisobutylaluminum hydride in toluene (0.283 mL, 0.289 mmol). After 10 min, the reaction mixture was further stirred for 20 min under ice-cooling. The reaction was quenched with 1.64 M aqueous potassium sodium tartrate tetrahydrate (0.88 mL, 1.45 mmol). The resulting mixture was stirred at 25 °C for 1 h and then extracted with ethyl acetate (5 mL × 3). The extracts were washed with saturated aqueous NaCl (10 mL), dried over anhydrous Na_2SO_4, and concentrated *in vacuo*. Purification of the residue by flash column chromatography (3.0 g of silica gel, 20:1 → 5:1 → 2:1 *n*-hexane−ethyl acetate) gave **20** (23.3 mg, 45%), **21** (19.6 mg, 38%), and **22** (6.7 mg, 12%) as a colorless oil, respectively. **20**: R_f 0.38 (5:1 *n*-hexane−ethyl acetate); ^{1}H-NMR δ 0.12 (3H, s, Me of TBS), 0.13 (6H, s, Me of TBS), 0.14 (3H, s, Me of TBS), 0.15 (3H, s, Me of TBS), 0.17 (3H, s, Me of TBS), 0.92 (9H, s, *t*-Bu of TBS), 0.925 (9H, s, *t*-Bu of TBS), 0.93 (9H, s, *t*-Bu of TBS), 1.44 (1H, d, J = 6.6 Hz, OH), 3.75 (1H, dd, J = 5.4 and 5.0 Hz, H-5), 4.28 (2H, dd, J = 6.6 and 2.0 Hz, H-9), 4.64 (1H, dd, J = 5.4 and 2.0 Hz, H-6), 4.80 (1H, d, J = 5.0 Hz, H-4), 9.21 (1H, s, CHO); HRMS (EI) *m/z* 540.3118 (540.3123 calcd for $C_{27}H_{52}O_5Si_3$, M^+). **21**: R_f 0.13 (5:1 *n*-hexane−ethyl acetate); ^{1}H-NMR δ 0.12 (12H, s, Me of TBS), 0.15 (6H, s, Me of TBS), 0.92 (27H, s, *t*-Bu of TBS), 1.47 (2H, t, J = 6.0 Hz, OH), 3.68 (1H, t, J = 5.0 Hz, H-5), 4.28 (4H, dd, J = 6.0 and 1.8 Hz, H-1 and 9), 4.66 (2H, dt, J = 5.0 and 1.8 Hz, H-4 and 6); HRMS (EI) *m/z* 543.3362 (543.3357 calcd for $C_{27}H_{55}O_5Si_3$, M + H^+). **22**: R_f 0.65 (5:1 *n*-hexane−ethyl acetate); ^{1}H-NMR δ 0.12 (6H, s, Me of TBS), 0.14 (6H, s, Me of TBS), 0.17 (6H, s, Me of TBS), 0.92 (18H, s, *t*-Bu of TBS), 0.93 (9H, s, *t*-Bu of TBS), 3.81 (1H, t, J = 4.6 Hz, H-5), 4.78 (2H, d, J = 4.6 Hz, H-4 and 6), 9.22 (2H, s, CHO); HRMS (EI) *m/z* 538.2954 (538.2966 calcd for $C_{27}H_{50}O_5Si_3$, M^+).

Oxidation of 21 into 20. To an ice-cold solution of **21** (3.51 g, 6.46 mmol) in dry CH_2Cl_2 (70 mL) was added Dess−Martin periodinane (2.73 g, 6.42 mmol). After the reaction mixture was stirred for 30 min under ice-cooling, diethyl ether (70 mL) and a mixture (160 mL) of 7:1 saturated aqueous $Na_2S_2O_3$ and saturated aqueous $NaHCO_3$ (20 mL) were added to the reaction mixture. The resulting mixture was stirred for 10 min and then extracted with diethyl ether (70 mL × 3). The extracts were washed with saturated aqueous NaCl (140 mL), dried over anhydrous Na_2SO_4, and concentrated *in vacuo*. Purification of the residue by flash column chromatography (130 g of silica gel, 10:1 → 5:1 → 2:1 *n*-hexane−ethyl acetate) gave **20** (1.85 g, 53%), **22** (0.49 g, 14%), and **21** (1.16 g, 33%).

Reduction of 22 into 20. To an ice-cold solution of **22** (1.34 g, 2.49 mmol) in methanol (25 mL) was added $NaBH_4$ (45.4 mg, 1.20 mmol) with stirring. After the resulting mixture was stirred for 30 min under ice-cooling, the reaction mixture was made neutral with ion exchange resin, CG-50. The resin was filtered off and washed with methanol (10 mL × 5), and then the filtrates were concentrated *in vacuo*. Purification of the residue by flash column chromatography (70 g of silica gel, 10:1 → 5:1 → 2:1 *n*-hexane−ethyl acetate) gave **20** (0.687 g, 51%), **21** (0.473 g, 35%), and **22** (0.134 g, 10%).

***dl*-(4*R*,5*R*,6*S*)-4,5,6-Tris[(*tert*-butyldimethylsilyl)oxy]-2,7-decadiyne-1,9-diol (23).** To an ice-cold solution of **20** (23.3 mg, 0.0431 mmol) in dry diethyl ether (0.35 mL) with stirring was added dropwise 2.98 M MeMgBr in diethyl ether (65.0 μL, 0.194 mmol). After the reaction mixture was stirred at 25 °C for 10 min, the resulting solution was poured into ice-cold and saturated aqueous NH_4Cl (0.4 mL) and then the resulting mixture was extracted with diethyl ether (0.5 mL × 3). The extracts were washed with saturated aqueous NaCl (0.4 mL), dried over anhydrous Na_2SO_4, and concentrated *in vacuo*. Purification of the residue by flash column chromatography (2.5 g of silica gel, 4:1 *n*-hexane−ethyl acetate) gave **23** (22.9 mg, 95%) as white crystals: R_f 0.27 (4:1 *n*-hexane−ethyl acetate); mp 77.5−78.5 °C (acetone/*n*-hexane); ^{1}H-NMR δ 0.12 (12H, s, Me of TBS), 0.15 (6H, s, Me of TBS), 0.92 (27H, s, *t*-Bu of TBS), 1.43 (1H, d, J = 6.4 Hz, OH), 1.44 (3H, d, J = 6.4 Hz, H-10), 1.68 (1H, t, J = 6.0 Hz, OH), 3.67 (1H, dd, J = 5.6 and 5.4 Hz, H-5), 4.28 (2H, dd, J = 6.0 and 1.8 Hz, H-1), 4.53 (1H, ddq, J = 6.4, 6.4 and 1.6 Hz, H-9), 4.60−4.70 (2H, m, H-4 and 6); HRMS (EI) *m/z* 556.3452 (556.3436 calcd for $C_{28}H_{56}O_5Si_3$, M^+).

***dl*-(4*R*,5*R*,6*S*)-4,5,6-Tris[(*tert*-butyldimethylsilyl)oxy]-9-oxo-2,7-decadiyn-1-al (24).** To a stirred solution of **23** (1.87 g, 3.36 mmol) in dry CH_2Cl_2 (37 mL) at 26 °C was added Dess−Martin periodinane (4.56 g, 10.8 mmol). After the reaction mixture was stirred for 45 min, diethyl ether (37 mL) and a mixture (80 mL) of 7:1 saturated aqueous $Na_2S_2O_3$ and saturated aqueous $NaHCO_3$ (20 mL) were added to the reaction mixture. The resulting mixture was stirred for 10 min and then extracted with diethyl ether (30 mL × 3). The extracts were washed with saturated aqueous NaCl (70 mL), dried over anhydrous Na_2SO_4, and concentrated *in vacuo*. Purification of the residue by flash column chromatography (90 g of silica gel, 10:1 *n*-hexane−diethyl ether) gave **24** (1.84 g, 99%) as a colorless oil: R_f 0.75 (4:1 *n*-hexane−ethyl acetate); ^{1}H-NMR δ 0.13 (6H, s, Me of TBS), 0.14 (6H, s, Me of TBS), 0.18 (6H, s, Me of TBS), 0.92 (9H, s, *t*-Bu of TBS), 0.925 (9H, s, *t*-Bu of TBS), 0.93 (9H, s, *t*-Bu of TBS), 2.32 (3H, s, H-10), 3.80 (1H, dd, J = 4.4 and 4.4 Hz, H-5), 4.77 (2H, d, J = 4.4 Hz, H-4 and 6), 9.21 (1H, s, CHO); HRMS (EI) *m/z* 552.3103 (552.3123 calcd for $C_{28}H_{52}O_5Si_3$, M^+).

***dl*-(2*Z*,6*R*,7*R*,8*S*)-6,7,8-Tris[(*tert*-butyldimethylsilyl)oxy]-2-cyclodecene-4,9-diyn-1-one (25).** To a stirred ethanol (167 mL) at 25 °C were added dropwise together a solution of **24** (1.84 g, 3.33 mmol) in ethanol (333 mL) and a solution of lithium hydroxide (0.160 g, 6.65 mmol) in 98% ethanol−H_2O (340 mL) over 2 h. After the reaction mixture was further stirred for 10 min, the resulting solution was made neutral with ion exchange resin, CG-50. The resin was filtered off and washed with ethanol (20 mL × 5) and then the filtrates were concentrated *in vacuo*. Purification of the residue by flash column chromatography (90 g of silica gel, 40:1 *n*-hexane−ethyl acetate) gave **25** (0.80 g, 45%) and dimer **26** (0.61 g, 34%) as a pale yellow oil, respectively. **25**: R_f 0.70 (10:1 *n*-hexane−ethyl acetate); ^{1}H-NMR δ 0.10 (6H, s, Me of TBS), 0.14 (6H, s, Me of TBS), 0.15 (6H, s, Me of TBS), 0.89 (9H, s, *t*-Bu of TBS), 0.93 (18H, s, *t*-Bu of TBS), 3.79 (1H, dd, J = 5.0 and 5.0 Hz, H-7), 4.56 (1H, d, J = 5.0 Hz, H-8), 4.57 (1H, dd, J = 5.0 and 2.0 Hz, H-6), 6.33 (1H, dd, J = 12.0 and 2.0 Hz, H-3), 6.41 (1H, d, J = 12.0 Hz, H-2); HRMS (EI) *m/z* 534.3011 (534.3016 calcd for $C_{28}H_{50}O_4Si_3$, M^+). **26**: R_f = 0.80 (10:1 *n*-hexane−ethyl acetate); ^{1}H-NMR δ 0.06−0.15 (36H, m, Me of TBS), 0.87 (18H, s, *t*-Bu of TBS), 0.89 (18H, s, *t*-Bu of TBS), 0.91 (18H, s, *t*-Bu of TBS), 3.75 (2H, dd, J = 5.9 and 4.8 Hz), 4.55 (2H, d, J = 4.8 Hz),

4.64 (2H, dd, J = 5.9 and 2.1 Hz), 6.49 (2H, d, J = 16.0 Hz), 7.32 (2H, dd, J = 16.0 and 2.1 Hz); MS (CI) m/z 1069 ($M + H^+$).

***dl*-(2*Z*,6*R*,7*R*,8*S*)-6,7,8-Tris[(*tert*-butyldimethylsilyl)oxy]-1-methyl-2-cyclodecene-4,9-diyn-1-ol (27).** To an ice-cold solution of **26** (17.0 mg, 0.0317 mmol) in dry diethyl ether (1.0 mL) with stirring was added dropwise 1.16 M methyl lithium in diethyl ether (60.0 μL, 0.0697 mmol). After the reaction mixture was stirred for 10 min under ice-cooling, the resulting solution was poured into ice-cold and saturated aqueous NH_4Cl (2 mL) and then the resulting mixture was extracted with diethyl ether (1 mL × 3). The extracts were washed with saturated aqueous NaCl (1 mL), dried over anhydrous Na_2SO_4, and concentrated *in vacuo*. Purification of the residue by flash column chromatography (1.5 g of silica gel, 10:1 *n*-hexane−ethyl acetate) gave **27** (15.0 mg, 88%) as a pale yellow oil: R_f 0.34 (10:1 *n*-hexane−ethyl acetate); ^{1}H-NMR δ 0.09 (6H, s, Me of TBS), 0.13 (6H, s, Me of TBS), 0.15 (6H, s, Me of TBS), 0.91 (9H, s, *t*-Bu of TBS), 0.92 (9H, s, *t*-Bu of TBS), 0.93 (9H, s, *t*-Bu of TBS), 1.52 ($^3/_2$H, s, Me), 1.54 ($^3/_2$H, s, Me), 2.18 ($^1/_2$H, s, OH), 2.21 ($^1/_2$H, s, OH), 3.73 ($^1/_2$H, dd, J = 5.2 and 5.2 Hz, H-7), 3.75 ($^1/_2$H, dd, J = 6.0 and 4.0 Hz, H-7), 4.38−4.46 (2H, m, H-6 and 8), 5.50 (1H, dd, J = 12.0 and 1.0 Hz, H-3), 5.83 (1H, d, J = 12.0, H-2); HRMS (EI) m/z 550.3330 (550.3330 calcd for $C_{29}H_{54}O_4Si_3$, M^+).

***dl*-(2*Z*,6*R*,7*R*,8*S*)-6,7,8-Tris[(*tert*-butyldimethylsilyl)oxy]-1,1-methylene-2-cyclodecene-4,9-diyne (6).** To an ice-cold solution of **27** (63.5 mg, 0.115 mmol) in dry CH_2Cl_2 (1.3 mL) with stirring were added triethylamine (0.129 mL, 0.992 mmol) and methanesulfonyl chloride (0.0357 mL, 0.461 mmol). After the reaction mixture was stirred for 20 min under ice-cooling, the resulting solution was poured into ice-cold H_2O (2 mL) and then the resulting mixture was extracted with *n*-hexane (1 mL × 3). The extracts were washed with saturated aqueous NaCl (2 mL), dried over anhydrous Na_2SO_4, and concentrated *in vacuo*. Purification of the residue by flash column chromatography (3 g of silica gel, 50:1 *n*-hexane−ethyl acetate) gave **6** (44.6 mg, 73%) as a pale yellow oil: R_f 0.85 (10:1 *n*-hexane−ethyl acetate); ^{1}H-NMR δ 0.10 (6H, s, Me of TBS), 0.13 (3H, s, Me of TBS), 0.14 (3H, s, Me of TBS), 0.15 (3H, s, Me of TBS), 0.16 (3H, s, Me of TBS), 0.90 (9H, s, *t*-Bu of TBS), 0.94 (9H, s, *t*-Bu of TBS), 3.79 (1H, dd, J = 6.0 and 6.0 Hz, H-7), 4.44 (1H, dd, J = 6.0 and 2.0 Hz, H-6), 4.46 (1H, d, J = 6.0 Hz, H-8), 5.37 (1H, dull s), 5.38 (1H, dull s), 5.46 (1H, dull d, J = 12.0 Hz, H-3), 6.26 (1H, d, J = 12.0 Hz, H-2); HRMS (EI) m/z 533.3282 (533.3302 calcd for $C_{29}H_{53}O_3Si_3$, $M + H^+$).

***dl*-(2*Z*,6*R*,7*R*,8*S*)-6,7,8-Triacetoxy-1,1-methylene-2-cyclodecene-4,9-diyne (8).** To an ice-cold solution of **6** (18.6 mg, 0.0348 mmol) in dry THF (0.4 mL) with stirring was added 1 M tetra-*n*-butylammonium fluoride−THF (0.115 mL, 0.115 mmol). After the reaction mixture was stirred for 30 min under ice-cooling, triethylamine (0.0388 mL) and acetic anhydride (0.0198 mL, 0.208 mmol) were added to the reaction mixture. After 1 h, the resulting solution was poured into H_2O (1 mL) and then the resulting mixture was extracted with *n*-hexane (0.5 mL × 3). The extracts were washed with saturated aqueous NaCl (0.5 mL), dried over anhydrous Na_2SO_4, and concentrated *in vacuo*. Purification of the residue by flash column chromatography (1.5 g of silica gel, 2:1 *n*-hexane−ethyl acetate) gave **8** (5.3 mg, 48%) as a pale yellow oil: R_f 0.41 (2:1 *n*-hexane−ethylene acetate); ^{1}H-NMR δ 2.05 (3H, s, OAc), 2.07 (3H, s, OAc), 2.09 (3H, s, OAc), 5.46 (1H, dd, J = 9.0 and 9.0 Hz, H-7), 5.49 (1H, dull d, J = 12.0 Hz, H-2 or 3), 5.51 (1H, dull s), 5.55 (1H, dull s), 5.67 (1H, d, J = 9.0 Hz, H-6 or 8), 5.72 (1H, d, J = 9.0 Hz, H-6 or 8), 6.34 (1H, d, J = 12.0 Hz, H-2 or 3); HRMS (EI) m/z 317.1021 (317.1025 calcd for $C_{17}H_{17}O_6$, $M + H^+$).

Cycloaromatization of 8 with Methyl Thioglycolate. To a stirred solution of **8** (21.6 mg, 0.0686 mmol) in methanol (0.68 mL) were added triethylamine (9.6 μL, 0.0686 mmol) and methyl thioglycolate (18.4 μL, 0.206 mmol). After the resulting solution was stirred at 25 °C for 1 h, the reaction mixture was concentrated *in vacuo*. Purification of the residue by preparative thin-layer chromatography (1:1 → 2:1 *n*-hexane−ethyl acetate) gave **28** (1.2 mg, 4.8%) and **29** (3.8 mg, 11.8%) as a colorless oil, respectively. **28**: R_f 0.41 (2:1 *n*-hexane−ethyl acetate); ^{1}H-NMR ($CDCl_3$) δ 2.06 (3H, s, OAc), 2.12 (3H, s, OAc), 3.10 (2H, s, $-CH_2SCH_2CO_2Me$), 3.73 (3H, s, OMe), 3.81 (2H, s, $-CH_2SCH_2CO_2Me$), 5.57 (1H, ddd, J = 6.0, 4.0 and 1.0 Hz, H-2), 5.97 (1H, dd, J = 10.0 and 4.0 Hz, H-3), 6.15 (1H, d, J = 6.0 Hz, H-1), 6.61 (1H, dd, J = 10.0 and 1.0 Hz, H-4), 7.12 (1H, d, J = 7.9 Hz, H-5), 7.26 (1H, d, J = 1.8 Hz, H-8), 7.28 (1H, dd, J = 7.9 and 1.8 Hz, H-6); ^{1}H-NMR (acetone-d_6) δ 3.17 (2H, s, $-CH_2SCH_2CO_2Me$), 3.67 (3H, s, OMe), 3.86 (2H, s, $-CH_2SCH_2CO_2Me$), 5.54 (1H, ddd, J = 6.4, 4.0 and 1.2 Hz, H-2), 5.98 (1H, dd, J = 10.0 and 4.0 Hz, H-3), 6.11 (1H, d, J = 6.4 Hz, H−1), 6.71 (1H, dull d, J = 10.0 Hz, H-4), 7.23 (1H, d, J = 7.9 Hz, H-5), 7.33 (1H, d, J = 1.8 Hz, H-8), 7.35 (1H, dd, J = 7.9 and 1.8 Hz, H-6); HRMS (EI) m/z 364.0967 (364.0981 calcd for $C_{18}H_{20}O_6S$, M^+). **29**: R_f 0.25 (2:1 *n*-hexane−ethyl acetate); ^{1}H-NMR ($CDCl_3$) δ 2.06 (3H, s, OAc), 2.09 (3H, s, OAc), 3.13 (2H, s, $-CH_2SCH_2CO_2Me$), 3.67 (2H, s, $-SCH_2CO_2Me$), 3.74 (3H, s, OMe), 3.76 (3H, s, OMe), 3.88 (2H, s, $-CH_2SCH_2CO_2Me$), 5.58 (1H, ddd, J = 7.2, 4.0 and 1.6 Hz, H-2), 6.03 (1H, dd, J = 10.0 and 4.0 Hz, H-3), 6.13 (1H, d, J = 7.2 Hz, H-1), 6.88 (1H, dull d, J = 10.0 Hz, H-4), 7.24 (1H, dull s, H-8), 7.27 (1H, dull s, H-6); ^{1}H-NMR (acetone-d_6) δ 3.26 (2H, s, $-CH_2SCH_2CO_2Me$), 3.69 (3H, s, OMe), 3.70 (3H, s, OMe), 3.84 (2H, s, $-SCH_2CO_2Me$), 3.98 (2H, s, $-CH_2SCH_2CO_2Me$), 5.55 (1H, ddd, J = 7.0, 3.9 and 1.6 Hz, H-2), 6.04 (1H, dd, J = 10.0 and 3.9 Hz, H-3), 6.08 (1H, d, J = 7.0 Hz, H-1), 7.02 (1H, dd, J = 10.0 and 1.6 Hz, H-4), 7.24 (1H, d, J = 1.9 Hz, H-8), 7.33 (1H, d, J = 1.9 Hz, H-6); HRMS (EI) m/z 468.0895 (468.0912 calcd for $C_{21}H_{24}O_8S_2$, M^+).

Cycloaromatization of 8 with Pyrrolidine. To a stirred solution of **8** (18.0 mg, 0.0570 mmol) in ethanol (2.0 mL) was added pyrrolidine (40.0 μL, 0.479 mmol). After the resulting solution was stirred at 25 °C for 12 h, the reaction mixture was concentrated *in vacuo*. Purification of the residue by flash column chromatography (2.5 g of silica gel, 5:1 chloroform−methanol) gave **38** (3.1 mg, 14.6%) as a pale yellow oil: R_f 0.50 (4:1 chloroform−methanol); ^{1}H-NMR δ 1.49 (3H, d, J = 6.2 Hz, Me), 1.72−1.83 (4H, m, $(-NCH_2CH_2)_2$), 2.06 (3H, s, OAc), 2.13 (3H, s, OAc), 2.45−2.55 (4H, m, $(-NCH_2CH_2)_2$), 3.65 (2H, s, $-CH_2N-$), 4.87 (1H, q, J = 6.2 Hz, $-CH(OH)Me$), 5.55 (1H, dd, J = 6.4 and 4.0 Hz, H-2), 5.97 (1H, dd, J = 10.0 and 4.0 Hz, H-3), 6.15 (1H, d, J = 6.4 Hz, H-1), 7.07 (1H, d, J = 10.0 Hz, H-4), 7.18 (1H, dull, s, H-8), 7.30 (1H, dull s, H-6); HRMS (EI) m/z 373.1889 (373.1889 calcd for $C_{21}H_{27}NO_5$, M^+).

DNA Cleavage Studies. All DNA cleavage experiments were performed with ΦX174 DNA (50 μM/base pair) in a volume of 10 μL containing 20% ethanol in buffer at 37 °C for 24 h. The DNA-sample levels were varied as indicated in the figure captions. The results were analyzed using 1% agarose gel electrophoresis and detection with ethidium bromide fluorescence. The electrophoresis gels were immediately visualized on a UV transilluminator and photographed using black and white instant film. Figures 8 and 9 show the pictures of the agarose gel electrophoresis results.

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