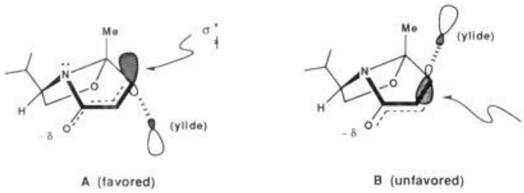

A (favored) B (unfavored)

available in the formation of the $\sigma^*_\ddagger$ orbital. Thus, the idea of stabilization of $\sigma^*_\ddagger$ orbitals in the transition state may prove to be a useful guide to understanding some unexpected stereoelectronic effects. These and others will be reported in more detail in the future.

Acknowledgment. We are grateful to the National Institutes of Health for generous financial support. A NIH-NRSA Postdoctoral Fellowship (to S.A.F.) is also gratefully acknowledged.

DNA Cleavage by a Synthetic Mimic of the Calicheamicin–Esperamicin Class of Antibiotics

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DNA cleavage is currently a topic of intense research investigations.[1] Both naturally occurring[2] and synthetic[3] compounds have demonstrated the ability to cleave DNA under appropriate conditions which often include metal ions, thiols, photolysis and/or oxygen as cofactors. The recently reported calicheamicin[4] (represented by calicheamicin $\gamma_{1\alpha}{}^{\mathrm{I}}$) and esperamicin[5] class of antibiotics have shown striking capacities to induce DNA scission[6,7] via a proposed mechanism that involves hydrogen abstraction from the phosphate backbone of DNA by benzenoid diradicals.[4-6] According to this mechanistic proposal, a cascade reaction sequence, triggered upon DNA binding of the molecules, generates the reactive diradical species from the cyclodecaenediyne moiety present in these complex structures. Inspired by this fascinating hypothesis, we recently initiated a program directed toward the design, synthesis, and evaluation of simple structures that might mimic the biological action of these natural products. In this communication, we report the first synthetic mimic of the calicheamicin–esperamicin class of antibiotics and its DNA-cleaving properties.

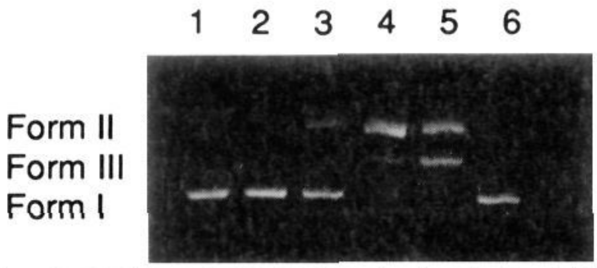


Figure 1. ΦX174 Form I DNA (50 μM per base) was incubated with compound **1** in Tris-acetate buffer (pH 8.5, 50 mM) at 37 °C for 12 h and analyzed by agarose gel electrophoresis. Lane 1, DNA alone; lanes 2–5, DNA + **1** at 1.0, 10, 100, and 500 μM, respectively; lane 6, DNA + **3** at 2 mM.

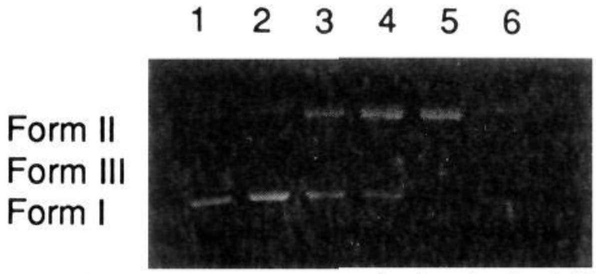
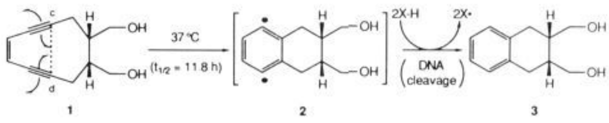
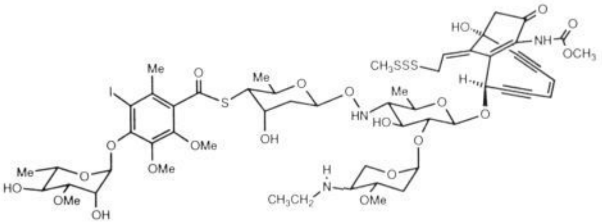


Figure 2. ΦX174 Form I DNA (50 μM per base) was incubated with compound **1** (20 μM) for various times at 37 °C and analyzed as described under Figure 1. Lanes 1–5, 0, 6, 12, 24, and 48 h, respectively; lane 6, DNA alone.

Scheme I[a]



[a] Presumed mechanism of DNA cleaving action of compound **1**.



calicheamicin $\gamma_{1\alpha}{}^{\mathrm{I}}$

On the basis of previous calculations and experimental results from these laboratories,[8] the conjugated cyclodecaenediyne diol **1** (Scheme I) was designed as a potential DNA-cleaving molecule. The crucial expectation was that **1** would be sufficiently stable at ambient temperatures to allow its isolation and handling, but that it would undergo Bergman cyclization[9] at 37 °C (body

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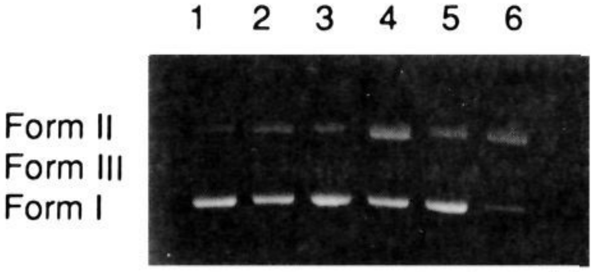


Figure 3. ΦX174 Form I DNA (50 μM per base) was incubated with compound **1** (20 μM) at various temperatures and times and analyzed as described under Figure 1. Lane 1, DNA alone 22 °C, 48 h; lane 2, 22 °C, 48 h; lane 3, DNA alone 37 °C, 24 h; lane 4, 37 °C, 24 h; lane 5, DNA alone, 45 °C, 24 h; lane 6, 45 °C, 24 h.

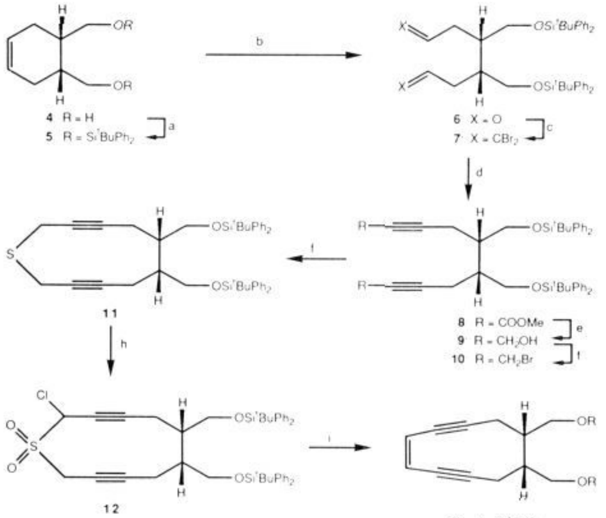
temperature) to benzenoid diradical **2** (and thence to aromatic compound **3**, Scheme I) at useful rates to cause DNA scission. More specifically, this expectation was based on a calculated distance (cd) (Scheme I) of 3.20 Å and an estimated energy of activation (E_a) of 23.6 kcal/mol[10] for the transformation **1** → **2**. The hydroxy groups were included in the designed structure (**1**) both for solubility reasons and for attaching further functionality for recognition purposes at a later stage of the program.

Starting with diol **4**[11] and following our recently developed strategy for the construction of cyclic conjugated enediynes,[8] the designed compound (**1**) was synthesized as summarized in Scheme II.[12]

Compound **1** was indeed sufficiently stable for isolation and handling at ambient temperatures. At 37 °C, however, **1** smoothly cyclized with a half life ($t_{1/2}$) of 11.8 h (benzene solution, excess 1,4-cyclohexadiene, estimated E_a = 23.6 kcal/mol[10]) leading to compound **3**[13] (Scheme I) via presumed diradical **2**, heightening expectations for **1** showing DNA-cleaving properties. *Indeed, compound **1** caused clean scission of double-stranded DNA in the absence of any additives.*[14] Thus, incubation of **1** (1.0–500 μM) with ΦX174 Form I DNA aerobically at 37 °C produced cleanly Form II DNA and, finally, Form III DNA as shown by gel electrophoresis analysis. The extent of DNA cleavage was shown to be dependent on (a) the concentration of **1** (Figure 1), (b) the incubation time (Figure 2), and (c) the temperature (Figure 3). As expected, incubation of **3** with DNA did not cause any cleavage (Figure 1).

These results provide support for the proposed[4-6] mechanism of action of the calicheamicins and esperamicins and confirm our recent hypothesis[8] that simple cyclic conjugated enediynes should spontaneously cleave DNA[15] in the absence of any cofactors and, therefore, serve as "warheads" for designed systems against selected targets such as specific DNA segments, oncogenes, and tumor cells. It is expected that incorporation of **1** or similar structures into molecular assemblies carrying suitable delivery and/or site-specific moieties[16] may result in powerful biotechnology tools and possibly useful therapeutic agents. These goals are currently being pursued in these laboratories.

Scheme II[a]



[a] Synthesis of compound **1**: (a) 2.0 equiv of t-BuPh$_2$SiCl, imidazole, DMF, 12 h, 71%; (b) O$_3$, EtOAc, MeOH (1:1), −78 °C, then 2.0 equiv of (MeO)$_3$P, −78 → 25 °C, 10 h; (c) 2.5 equiv of CBr$_4$, 5.4 equiv of PPh$_3$, CH$_2$Cl$_2$, 0 → 25 °C, 8 h, 57%, overall from **5**; (d) 4.5 equiv of n-BuLi, THF, −78 °C, 0.5 h, then 10 equiv of ClCOOMe, 0 °C, 45 min, 74%; (e) 2.0 equiv of DIBAL, CH$_2$Cl$_2$, −78 °C, 0.5 h, 97%; (f) 3.0 equiv of P(oct)$_3$, 2.2 equiv of CBr$_4$, Et$_2$O, 0 °C, 1 h, 100%; (g) 7.0 equiv of Na$_2$S·9H$_2$O, EtOH, H$_2$O (5:1), 78 °C, 1.5 h, 58%; (h) i. 1.0 equiv of mCPBA, CH$_2$Cl$_2$, −30 °C, 0.5 h, 90%; ii. 1.1 equiv of SO$_2$Cl$_2$, 3.6 equiv of pyridine, CH$_2$Cl$_2$, −78 °C, 10 min, 79%; iii. 7.7 equiv of mCPBA, CH$_2$Cl$_2$, 18 h, 99%; (i) 1.2 equiv of MeLi, Et$_2$O, −78 °C, 15 min, 20%; (j) 1.0 equiv of n-Bu$_4$NF, THF, 1 h, 84%.

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Registry No. **1**, 116531-31-8; **3**, 10074-98-3; **4**, 20141-17-7; **5**, 116503-81-2; **6**, 116503-82-3; **7**, 116503-83-4; **8**, 116503-84-5; **9**, 116503-85-6; **10**, 116503-86-7; **11**, 116503-87-8; **12**, 116503-88-9; **13**, 116503-89-0; CBr$_4$, 558-13-4; ClCO$_2$Me, 79-22-1; ^{t}BuPh$_2$SiCl, 58479-61-1; calcicheamicin, 113440-58-7; esperamicin, 114797-28-3.

Supplementary Material Available: R_f values and ^{1}H NMR data for compounds **11**, **12**, **13**, **1**, and **3** and kinetic data for **1** → **2** (2 pages). Ordering information is given on any current masthead page.

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(10) The estimated theoretical value for E_a (for **1** → **2**) was derived from a linear plot of distance cd versus experimental E_a for a series of conjugated enediynes (see ref 8 and 9 and unpublished results, these laboratories). The estimated experimental value for E_a (for **1** → **2**) was determined by using the experimental rate constant ($k = 9.76 \times 10^{-4}$ min^{-1}) and the Arrhenius constant ($\ln A = 31.337$), obtained for the parent cyclodecaenediyne (see ref 8 and Supplementary Material).

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(14) The possibility that traces of transition metal and/or other impurities present under the reaction conditions used are involved in the cleavage pathway is not ruled out at the present time. The effect of oxygen and other cofactors on the DNA-cleaving properties of **1** and its precise mechanism of action, including the possible involvement of hydroxyl radicals, is under further investigation.

(15) Although a number of model systems of the calicheamicin–esperamicin class of antibiotics have been described [(a) ref 8; (b) Schreiber, S. L.; Kiessling, L. L. *J. Am. Chem. Soc.* **1988**, *110*, 631; (c) Magnus, P.; Carter, P. A. *J. Am. Chem. Soc.* **1988**, *110*, 1626], no DNA-cleaving properties have been reported as yet for such systems.

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