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Published on Web 12/03/2008

Photochemical Generation and Reversible Cycloaromatization of a Nine-Membered Ring Cyclic Enediyne

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Abstract: Irradiation of the nine-membered ring enediyne precursor, which has one of its triple bonds masked as cyclopropenone, efficiently ($\Phi = 0.34$) generates the reactive 4,5-benzocyclonona-2,6-diynol. The latter rapidly equilibrates with the corresponding 1,4-didehydronaphthalene diradical and then undergoes rate-limiting hydrogen abstraction to produce the ultimate product of the Bergman cyclization, benz[f]indanol.

Introduction

Natural enediyne antibiotics possess an unusual (*Z*)-3-ene-1,5-diyne fragment (hence the name), which is responsible for their extreme cytotoxicity.[1] Upon binding to a double-stranded DNA molecule, this enediyne moiety is usually positioned within the minor grove of the host. Reductive or nucleophilic activation of the natural enediynes removes stereochemical blocking device and the (*Z*)-3-ene-1,5-diyne fragment undergoes Bergman[2] cyclization to produce a *p*-benzyne diradical,[1] which might exist in equilibrium with the enediyne precursor.[3] By it is size, *p*-benzyne (ca. 3.8 Å) is perfectly suitable to perform a double hydrogen abstraction from carbohydrate backbones of opposite DNA strands (1′H to 1′H distance is about 5.78 Å).Theoretical studies and experimental data suggest that this diradical has relatively low reactivity.[4,5] Initial hydrogen abstraction from one strand, however, converts *p*-benzyne into a 10−100 times more reactive phenyl radical,[4,6] which immediately abstracts the second hydrogen from the opposite DNA strand. This process leads to an oxidative double-strand dDNA scission.[1] While natural enediynes are very powerful dDNA-cleaving machines, the lack of antitumor selectivity results in a very high general toxicity, which hampers clinical applications of natural enediyne antibiotics.[7] Photochemical triggering of the cycloaromatization reaction might help to alleviate this problem by allowing for the spatial and temporal control of enediyne reactivity.[8] The direct irradiation of acyclic[9] and cyclic[10] enediynes, as well as the natural enediyne antibiotic Dynemicin A,[11] demonstrated that the Bergman cyclization can be induced photochemically, albeit with low efficiency. The quantum yield of the photochemical Bergman cyclization can be substantially improved by adjusting the electronic properties of substituents[12] and/or using different modes of excitation energy transfer, for example MLCT.[13] In addition, several caged enediynes have been prepared which undergo conventional chemical activation after the photochemical uncaging step.[14]

(1) (a) For recent reviews see: *Enediyne Antibiotics as Antitumor Agents*; Borders, D. B., Doyle, T. W., Eds.; Marcel Dekker: New York: 1995; (b) Jones, G. B.; Fouad, F. S. *Curr. Pharm. Design* **2002**, *8*, 2415. (c) Nicolaou, K. C.; Dai, W.-M. *Angew. Chem., Int. Ed. Engl.* **1991**, *30*, 1387. (d) Nicolaou, K. C.; Smith, A. L. *Acc. Chem. Res.* **1992**, *25*, 497. (e) Danishefsky, S. J.; Shair, M. D. *J. Org. Chem.* **1996**, *61*, 16. (f) Shao, R. G. *Curr. Mol. Pharm.* **2008**, *1*, 50–60. (g) Hamann, P. R.; Upeslacis, J.; Borders, D. B. *Anticancer Agents Nat. Prod.* **2005**, 451. (h) Smith, A. L.; Nicolaou, K. C. *J. Med. Chem.* **1996**, *39*, 2103. (i) Thorson, J. S.; Ahlert, J.; Shepard, E.; Whitwam, R. E.; Onwueme, K. C.; Sievers, E. L.; Ruppen, M. *Curr. Pharm. Des.* **2000**, *6*, 1841.

(2) Bergman, R. G. *Acc. Chem. Res.* **1973**, *6*, 25.

(3) (a) Iida, K.; Hirama, M. *J. Am. Chem. Soc.* **1995**, *117*, 8875. (b) Hirama, M. *Pure Appl. Chem.* **1997**, *69*, 525. (c) Hirama, M.; Akiyama, K.; Das, P.; Mita, T.; Lear, M. J.; Iida, K.-i.; Sato, I.; Yoshimura, F.; Usuki, T.; Tero-Kubota, S. *Heterocycles* **2006**, *69*, 83.

(4) (a) Logan, C. F.; Chen, P. *J. Am. Chem. Soc.* **1996**, *118*, 2113. (b) Schottelius, M. J.; Chen, P. *J. Am. Chem. Soc.* **1996**, *118*, 4896. (c) Hoffner, J. H.; Schottelius, M. J.; Feichtinger, D.; Chen, P. *J. Am. Chem. Soc.* **1998**, *120*, 376. (d) Sander, W. *Acc. Chem. Res.* **1999**, *32*, 669–676. (e) Chen, P. *Angew. Chem., Int. Ed. Engl.* **1996**, *35*, 1478.

(5) (a) Wenk, H. H.; Winkler, M.; Amegayibor, F. S.; Nash, J. J.; Lee, A. S.; Thoen, J.; Petzold, C. J.; Kenttämaa, H. I. *J. Am. Chem. Soc.* **2002**, *124*, 12066. (b) Sander, W. *Angew. Chem., Int. Ed.* **2003**, *42*, 502.

(6) Roth, W. R.; Hopf, H.; Wasser, T.; Zimmermann, H.; Werner, C. *Leib. Ann.* **1996**, 1691.

(7) (a) Galm, U.; Hager, M. H.; Lanen, S. G. V.; Ju, J.; Thorson, J. S.; Shen, B. *Chem. Rev.* **2005**, *105*, 739. (b) Gredicak, M.; Jeric, I. *Acta Pharm.* **2007**, *57*, 133. (c) Shen, B.; Nonaka, K. *Curr. Med. Chem.* **2003**, *10*, 2317.

(8) (a) Kar, M.; Basak, A *Chem. Rev.* **2007**, *107*, 2861. (b) Jones, G. B.; Fouad, F. S. *Curr. Pharm. Design* **2002**, *8*, 2415.

(9) (a) Kagan, J.; Wang, X.; Chen, X.; Lau, K. Y.; Batac, I. V.; Tuveson, R. W.; Hudson, J. B. *J. Photochem. Photobiol. B* **1993**, *21*, 135. (b) Turro, N. J.; Evenzahav, A.; Nicolaou, K. C. *Tetrahedron Lett.* **1994**, *35*, 8089. (c) Kaneko, T.; Takahashi, M.; Hirama, M. *Angew. Chem., Int. Ed.* **1999**, *38*, 1267. (d) Plourde II, G.; El-Shafey, A.; Fouad, F.; Purohit, A.; Jones, G. *Bioorg. Med. Chem. Lett.* **2002**, *12*, 2985. (e) Falcone, D.; Li, J.; Kale, A.; Jones, G. B. *Bioorg. Med. Chem. Lett.* **2008**, *18*, 934–7. (f) Spence, J. D.; Hargrove, A. E.; Crampton, H. L.; Thomas, D. W. *Tetrahedron Lett.* **2007**, *48*, 725. (g) Sud, D.; Wigglesworth, T. J.; Branda, N. R. *Angew. Chem., Int. Ed.* **2007**, *46*, 8071.

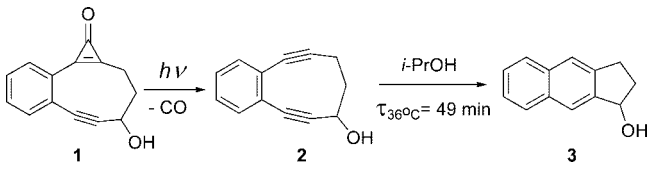
(10) (a) Choy, N.; Blanco, B.; Wen, J.; Krishan, A.; Russell, K. C. *Org. Lett.* **2000**, *2*, 3761. (b) Funk, R. L.; Young, E. R. R.; Williams, R. M.; Flanagan, M. F.; Cecil, T. L. *J. Am. Chem. Soc.* **1996**, *118*, 3291–2.

(11) Shiraki, T.; Sugiura, Y. *Biochemistry* **1990**, *29*, 9795.

(12) (a) Alabugin, I. V.; Manoharan, M. *J. Phys. Chem. A* **2003**, *107*, 3363. (b) Alabugin, I. V.; Manoharan, M.; Kovalenko, S. V. *Org. Lett.* **2002**, *4*, 1119. (c) Clark, A. E.; Davidson, E. R.; Zaleski, J. M. *J. Am. Chem. Soc.* **2001**, *123*, 2650.

(13) (a) Benites, P. J.; Holmberg, R. C.; Rawat, D. S.; Kraft, B. J.; Klein, L. J.; Peters, D. G.; Thorp, H. H.; Zaleski, J. M. *J. Am. Chem. Soc.* **2003**, *125*, 6434. (b) Kraft, B. J.; Coalter, N. L.; Nath, M.; Clark, A. E.; Siedle, A. R.; Huffman, J. C.; Zaleski, J. M. *Inorg. Chem.* **2003**, *42*, 1663. (c) Bhattacharyya, S.; Pink, M.; Baik, M. H.; Zaleski, J. M. *Angew. Chem., Int. Ed.* **2005**, *44*, 592.

Scheme 1



Our group explores an alternative strategy for the phototriggering of the cycloaromatization reaction: the in situ generation of an activated enediyne system.[15] Ideally, a photoswitchable analogue of natural enediyne antibiotics should be stable in the dark but undergo rapid and reversible cyclization to *p*-benzyne after irradiation. We have recently shown that masking one of the triple bonds in 10-membered cyclic enediynes with cyclopropenone moiety produces thermally stable enediyne precursors. While photolysis of cyclopropenone precursors under single-[16] or two-photon excitation[17] conditions results in efficient generation of corresponding enediynes, the cycloaromatization of the latter was not fast enough ($\tau_{40°C} > 16$ h) to achieve temporal and spatial resolution of *p*-benzyne generation in biological systems. To enhance the rate of Bergman cyclization of photoswitchable enediynes, we decided to design a nine-membered ring cyclopropenone-containing enediyne precursor (Scheme 1). Highly strained nine-membered enediynes are predicted to undergo very facile cycloaromatization under ambient conditions.[18] In fact, the only known monocyclic nine-membered enediyne with measurable lifetime, 3-chloro-3-cyclononene-1,5-diyne ($\tau_{40°C} \approx 8$ min), is stabilized by chlorine substituent in the vinylic position.[19] Natural antibiotic C-1027 and its analogues, which possess a bicyclic nine-membered ring enediyne core, are also extremely labile.[3,20] To achieve reversibility of the Bergman cyclization we decided to exploit the fact that hydrogen abstraction often becomes a partially rate-limiting step in the Bergman cyclization of benzannulated enediynes.[21]

Here we report the synthesis and photochemistry of cyclopropenone precursors 1, as well as theoretical and experimental studies of the reactivity of nine-membered ring enediyne 2 (Scheme 1).

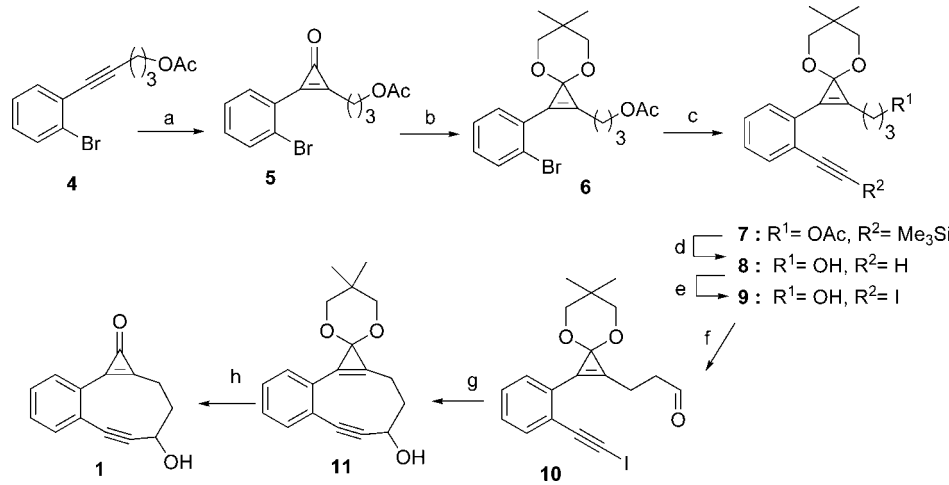
Results and Discussion

Synthesis of Cyclopropenone 1. A number of synthetic methods are available for the preparation of the cyclopropenone group, which is the key component of the photoactivatable enediyne precursor **1**.[22] Few of them, however, are compatible with a highly functionalized macrocycle **1**, which forced us to introduce cyclopropenone functionality on early stages of the synthesis.[23] The addition of difluorocarbene, which was generated by the thermolysis of trimethylsilyl fluorosulfonyldifluoroacetate ($FO_2SCF_2CO_2SiMe_3$, TFDA),[24] to the acetylene **4**, produced 1,1-difluorocyclopropene. The latter was hydrolyzed without isolation on wet silica gel to give cyclopropenone **5** (Scheme 2).[23] Introducing cyclopropenone moiety early in a multistep synthesis is difficult since this functionality is very susceptible to a nucleophilic attack, which usually results in ring opening.[25] Complexation with Lewis acids, on the other hand, produces relatively unreactive 2π-aromatic oxycyclopropenium cation. Masking cyclopropenone moiety as 2,2-dimethyl-1,3-propanediyl acetal **6** allows us to avoid these complications and broaden the range of reagents and reaction conditions that can be employed for the preparation of target compound (Scheme 2). It has to be noted, however, that cyclopropenone acetals are extremely susceptible to acid hydrolysis. They have life-times of only few milliseconds in an aqueous solution.[26]

The second acetylenic substituent was introduced using conventional Stille coupling condtions (**7**, Scheme 2). Simultaneous saponification of the acetate and removal of the TMS group followed by iodination[27] and Dess−Martin oxidation[28] gave rise to the iodoaldehyde **10**. Cyclization of the latter under

(14) (a) Nicolaou, K. C.; Dai, W.-M.; Wendeborn, S. V.; Smith, A. L.; Torisawa, Y.; Maligres, P.; Hwang, C.-K *Angew. Chem., Int. Ed. Engl.* **1991**, *30*, 1032. (b) Nicolaou, K. C.; Dai, W.-M. *J. Am. Chem. Soc.* **1992**, *114*, 8908. (c) Wender, P. A.; Zercher, C. K.; Beckham, S.; Haubold, E.-M. *J. Org. Chem.* **1993**, *58*, 5867. (d) Wender, P. A.; Beckham, S.; O'Leary, J. G. *Synthesis* **1994**, 1278. (e) Basak, A.; Bdour, H. M.; Shain, J. C.; Mandal, S.; Rudra, K. R.; Nag, S. *Bioorg. Med. Chem. Lett.* **2000**, *10*, 1321.
(15) Poloukhtine, A.; Karpov, G.; Popik, V. V. *Curr. Trends Med. Chem.* **2008**, *8*, 460.
(16) (a) Poloukhtine, A.; Popik, V. V. *J. Org. Chem.* **2005**, *70*, 1297. (b) Poloukhtine, A.; Popik, V. V. *Chem. Comm.* **2005**, 617.
(17) Poloukhtine, A.; Popik, V. V. *J. Org. Chem.* **2006**, *71*, 7417.
(18) (a) Nicolaou, K. C.; Ogawa, Y.; Zuccarello, G.; Schweiger, E. J.; Kumazawa, T. *J. Am. Chem. Soc.* **1988**, *110*, 4866. (b) Snyder, J. P. *J. Am. Chem. Soc.* **1990**, *112*, 5367. (c) Magnus, P.; Fortt, S.; Pitterna, T.; Snyder, J. P. *J. Am. Chem. Soc.* **1990**, *112*, 4986. (d) Jones, G. B.; Warner, P. M. *J. Am. Chem. Soc.* **2001**, *123*, 2134. (e) Gaffney, S. M.; Capitani, J. F.; Castaldo, L.; Mitra, A. *Int. J. Qant. Chem.* **2003**, *95*, 706. (f) Chen, W.-C.; Zou, J.-W.; Yu, C.-H. *J. Org. Chem.* **2003**, *68*, 3663. (g) Alabugin, I. V.; Manoharan, M. *J. Phys. Chem. A* **2003**, *107*, 3363.
(19) Jones, G. B.; Plourde, G. W. *Org. Lett.* **2000**, *2*, 1757.
(20) (a) Yoshida, K.; Minami, Y.; Azuma, R.; Saeki, M.; Otani, T. *Tetrahedron Lett.* **1993**, *34*, 2637. (b) Xu, Y. J.; Zhen, Y. S.; Goldberg, I. H. *Biochemistry* **1994**, *33*, 5947. (c) Iida, K.; Hirama, M. *J.Am. Chem. Soc.* **1995**, *117*, 8875. (d) Usuki, T.; Mita, T.; Lear, M. J.; Das, P.; Yoshimura, F.; Inoue, M.; Hirama, M.; Akiyama, K.; Tero-Kubota, S. *Angew. Chem., Int. Ed.* **2004**, *43*, 5249. (e) Inoue, M.; Ohashi, I.; Kawaguchi, T.; Hirama, M. *Angew. Chem., Int. Ed.* **2008**, *47*, 1777.
(21) (a) Zeidan, T. A.; Manoharan, M.; Alabugin, I. V. *J. Org. Chem.* **2006**, *71*, 954. (b) Stahl, F.; Moran, D.; Schleyer, P. R.; Prall, M.; Schreiner, P. R. *J. Org. Chem.* **2002**, *67*, 1453. (c) Koseki, S.; Fujimura, Y.; Hirama, M. *J. Phys. Chem. A.* **1999**, *103*, 7672. (d) Semmelhack, M. F.; Neu, T.; Foubelo, F. *J. Org. Chem.* **1994**, *59*, 5038. (e) Boger, D. L.; Zhou, J. *J. Org. Chem.* **1993**, *58*, 3018. (f) Semmelhack, M. F.; Neu, T.; Foubelo, F. *Tetrahedron Lett.* **1992**, *33*, 3277.
(22) (a) Breslow, R.; Posner, J.; Krebs, A. *J. Am. Chem. Soc.* **1963**, *85*, 234. (b) Köbrich, G.; Flory, K.; Fischer, R. H. *Chem. Ber.* **1966**, *99*, 1793. (c) Chikos, J. C.; Patton, E.; West, R. *J. Org. Chem.* **1974**, *39*, 1647. (d) Wadsworth, D. H.; Donatelli, B. A. *Synthesis* **1981**, 285. (e) Musigmann, K.; Mayr, H. *Tetrahedron Lett.* **1987**, *28*, 4517. (f) Weinder, C. H.; Wadsworth, D. H.; Knop, C. S.; Oyefesso, A. I.; Hafer, B. H.; Hartman, R. J.; Mehlenbacher, R. C.; Hogan, S. C. *J. Org. Chem.* **1994**, *59*, 4319. (g) Chiang, Y.; Grant, A. S.; Kresge, A. J.; Paine, S. W. *J. Am. Chem. Soc.* **1996**, *118*, 4366. (h) Gleiter, R.; Merger, M.; Altereuther, A.; Irngartinger, R. *J. Org. Chem.* **1996**, *61*, 1946. (i) Netland, K. A.; Gundersen, L.-L.; Rise, F. *Synth. Commun.* **2000**, *30*, 1767. (j) Poloukhtine, A.; Popik, V. V. *J. Org. Chem.* **2003**, *68*, 7833.
(23) See the Supporting Information file for details.
(24) (a) Cheng, Z.-L.; Xiao, J.-C.; Liu, C.; Chen, Q.-Y. *Eur. J. Org. Chem.* **2006**, 5581. (b) Cheng, Z.-L.; Chen, Q.-Y. *Chin. J. Chem.* **2006**, *24*, 1219.
(25) (a) Potts, K. T.; Baum, J. S *Chem. Rev.* **1974**, *74*, 189. (b) Halton, B.; Banwell, M. G. In *The Chemistry of Cyclopropyl Group*; Rappoport, Z., Ed.; Wiley: New York, 1987, 1300; (c) Ciabattoni, J.; Nathan, E. C. *J. Am. Chem. Soc.* **1969**, *91*, 4766. (d) Dehmlow, E. V.; Neuhaus, R.; Schell, H. G. *Chem. Ber.* **1988**, *121*, 569. (e) Dehmlow, S. S.; Dehmlow, E. V. *Z. Naturforsch. B* **1975**, *30b*, 404. (f) Dehmlow, E. V. *Leib. Ann.* **1969**, *729*, 64.
(26) McClelland, R. A.; Ahmad, M. *J. Am. Chem. Soc.* **1978**, *100*, 7027.
(27) Crevisy, C.; Beau, J.-M. *Tetrahedron Lett.* **1991**, *32*, 3171.
(28) Dess, D. B.; Martin, J. C. *J. Org. Chem.* **1983**, *48*, 4156.

Scheme 2[a]



[a] Reagents and conditions: (a) (i) NaF, TFDA, diglyme, 120 °C, 2 h; (ii) wet silica gel, hexanes; (b) Et_3OBF_4, CH_2Cl_2, r.t., 1 h; then neopentylglycol, Et_3N, CH_2Cl_2, r.t., 12 h, 51% (after two steps); (c) trimethyl[(tributylstannyl)ethynyl]silane, $Pd(PPh_3)_4$, toluene, 90 °C, 2 h, 76%; (d) K_2CO_3, MeOH, 2 h, 88%; (e) I_2, morpholine, benzene, 78%; (f) Dess−Martin periodinane, CH_2Cl_2, r.t., 3 h, 84%; (g) $CrCl_2$, $NiCl_2$, THF, 0 °C, 3 h, 36%; (h) Amberlyst, acetone, r.t., 1 h, 77%.

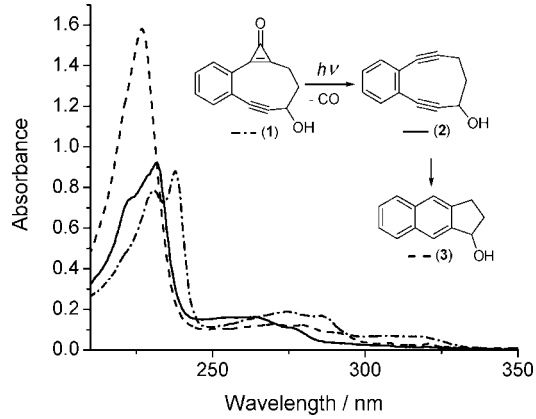


Figure 1. UV spectra of ca. 2×10^{-5} M solutions of the cyclopropenone **1** (dash-dotted line), enediyne **2** (solid line), and benz[f]indan-1-ol (**3**,dashed line) in 2-propanol.

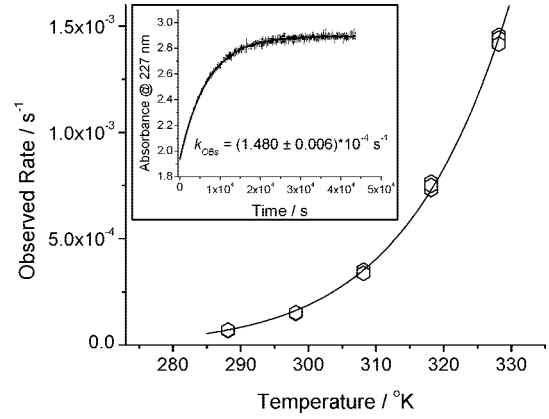


Figure 2. Temperature rate profile for the Bergman cyclization of the enediyne **2** generated in photolysis of **1** in 2-propanol. The line shown was drawn using parameters obtained by least-squares fitting of Eyring equation. The inset shows growth in absorbance at 227 nm after irradiation of **1** at 25 °C in neat 2-propanol. The line shown in the inset was drawn using parameters obtained by least-squares fitting of single exponential equation.

Nozaki−Hiyama−Kishi conditions,[29] followed by mild removal of an acetal protection, produced the target nine-membered ring cyclopropenone **1** (Scheme 2).[23]

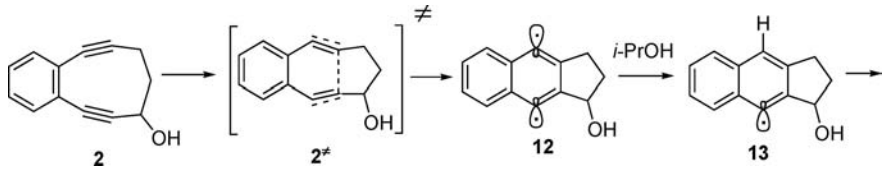
Photochemical Generation and Reaction of Nine-Membered Ring Enediyne 2. The UV spectrum of cyclopropenone **1** shows two close lying absorbance bands at 231 nm (log $\varepsilon = 4.5$) and 238 nm (log $\varepsilon = 4.6$, dash-dotted line in Figure 1). Photolysis of **1** in 2-propanol with 254 nm light resulted in efficient decarbonylation ($\Phi_{254nm} = 0.34 \pm 0.03$)[23] and the formation of the 4,5-benzocyclonona-2,6-diynol (**2**, Scheme 1). The latter undergoes spontaneous but relatively slow Bergman cyclization with a lifetime of ca. 2 h at 25 °C in 2-propanol. This process is surprisingly clean for a radical reaction producing quantitative yields of the ultimate product, benz[f]indan-1-ol (**3**, Scheme 1). The structural assignment of the product isolated from the photolysate of **1** was supported by the direct synthesis of benzindanol **3**.[23]

Kinetics of the Bergman Cyclization of 4,5-Benzocyclonona-2,6-diynol (2). The accurate rate measurements of cycloaromatization of the enediyne **2** were conducted by UV spectroscopy following the growth of the characteristic 227 nm band of benz[f]indan-1-ol (**3**, Figure 1) in the photolysate at various temperatures and solvent compositions. As shown on the inset in Figure 2, the reaction follows first-order equation well. The cyclization rates of the enediyne **2** were measured in 2-propanol solutions in the temperature range from 15 to 55 °C with 0.1 °C accuracy. The data so obtained are summarized in Table S1[23] and are displayed as the temperature rate profile in Figure 2.

The temperature dependence of the cyclization rate is surprisingly weak: every 10 K of temperature rise produces only about 2-fold rate increase (Figure 2). This observation suggests that the energy barrier ($\Delta G^{\ddagger}$) for the cyclization reaction grows with the temperature. In fact, least-squares fitting of the data to the Eyring equation[30] gives the following activation parameters: $\Delta H^{\ddagger} = 13.63 \pm 0.22$ kcal M^{-1} and $\Delta S^{\ddagger} = -30.66 \pm 0.61$ cal M^{-1} K^{-1}. The substantial negative entropy of activation indicates that in the transition state for the rate-determining step degrees

(29) (a) Takai, K.; Kuroda, T.; Nakatsukasa, S.; Oshima, K.; Nozaki, H. *Tetrahedron Lett.* **1985**, *26*, 5585. (b) Aicher, T. D.; Kishi, Y. *Tetrahedron Lett.* **1987**, *28*, 3463. (c) Brandstetter, T.; Maier, M. E. *Tetrahedron* **1994**, *50*, 1435. (d) Harwig, C. W.; Py, S.; Fallis, A. G. *J. Org. Chem.* **1997**, *62*, 7902. (e) Dai, W.-M.; Wu, A.; Hamaguchi, W. *Tetrahedron Lett.* **2001**, *42*, 4211.

Scheme 3



of freedom in the system are significantly reduced. The activation energy for the cyclization of the enediyne **2** to diradical **12** (Scheme 3) should have a low entropy component and is not consistent with the experimental value of $\Delta S^{\ddagger}$. The next step in the process, i.e., hydrogen abstraction, is a bimolecular reaction requiring proper orientation of the diradical **12** and a hydrogen donor. Such reactions are usually characterized by pronounced negative entropy of activation. In other words, the temperature dependence of the rate of Bergman cyclization of enediyne **2** suggests that hydrogen abstraction rather than formation of **12** is the rate limiting step of the reaction.

Further support for the rate-limiting hydrogen abstraction comes from the value of the solvent isotope effect on the rate of cyclization. When enediyne **2** was generated in 2-propanol-d_8 at 25 °C, the observed rate of its conversion to **3** was substantially lower than in 2-propanol ($k_D = (2.669 \pm 0.013) \times 10^{-5}$ s^{-1} vs $k_H = (1.497 \pm 0.019) \times 10^{-4}$ s^{-1}). The pronounced primary kinetic isotope effect, $k_H/k_D = 5.61$, clearly indicates that formation of a carbon−hydrogen bond happens on the rate-limiting step. This value, however, agrees well with both rate-limiting hydrogen transfer, as shown in Scheme 3, and rate-limiting proton transfer, which has been shown to induce cyclization of some enediyne compounds.[31] The use of 2-propanol-d_1 (*i*-PrOD) as a hydrogen donor provides a good method for a discrimination of the two pathways. Carbon-centered radicals are expected to abstract protium from 2-propanol-d_1 by cleaving the weakest C−H bond of a methine group, while proton (deuteron) transfer should involve a more acidic OH (OD) group. The rate of the cycloaromatization of **2** in 2-propanol-d_1, $k_{OBS} = (1.383 \pm 0.022) \times 10^{-4}$ s^{-1}, was essentially the same as in an all-protium solvent. This observation clearly shows that the hydrogen, but not proton, transfer is involved in the rate-limiting step of the Bergman cyclization of **2**. Proton transfer-initiated reactions are also expected to be catalyzed by acid, while conventional Bergman cyclization shows no acid catalysis.[32] Enediyne **2** generated in the presence of 0.025 M hydrochloric acid or absence of acid in 2-propanol−water mixture (4:1) at 25 °C undergoes conversion to **3** with the same rate ($k_{OBS} = (1.03 \pm 0.024) \times 10^{-4}$ vs $(9.87 \pm 0.82) \times 10^{-5}$ s^{-1}, respectively).

As was mentioned in the introduction, the rate of the Bergman cyclization of enediyne compounds often depends on the concentration and the nature of hydrogen donor.[3] This effect is especially prominent in case of benzannulated enediynes.[6,21] We have measured the rate of the cyclization of 4,5-benzocyclononona-2,6-diynol (**2**) in 2-propanol−water mixtures of various compositions. 2-Propanol is a good hydrogen donor with an α-C−H bond dissociation energy of 89 kcal mol^{-1}.[33] This value is similar to the C−H bond strength in tetrahydrofuran (92 kcal mol^{-1}),[34] a model compound for DNA deoxyribose. Water, on the other hand, is a poor hydrogen donor ($D_{0(H-OH)} = 118$ kcal mol^{-1}).[35] The rate of the cyclization of the enediyne 2 to benz[f]indan-1-ol (**3**) in 2-propanol−water mixtures was found to be linearly proportional to the concentration of alcohol up to neat 2-propanol solutions. (Figure 3, Table S2[23]). This observation indicates that equilibration between enediyne **2** and *p*-benzyne **12** is much faster than hydrogen abstraction even at the highest concentration of 2-propanol (Scheme 4).

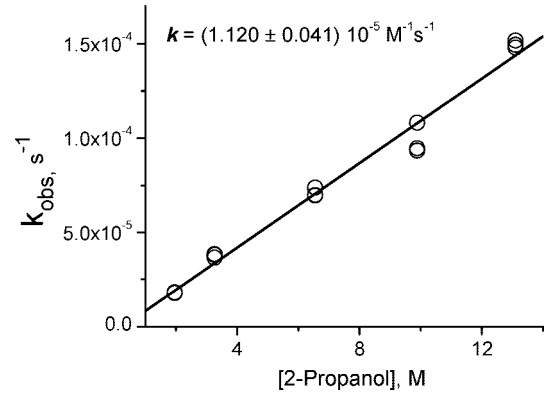
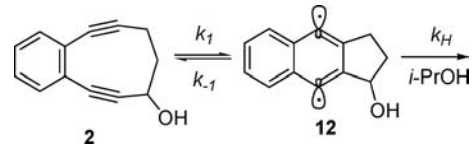


Figure 3. Observed rates of the Bergman cyclization of the enediyne **2** at various concentrations of 2-propanol in water at 25.0 ± 0.1 °C.

Scheme 4



The kinetics of the Bergman cyclization can be described by eq 1, where k_1 is the rate constant for the cycloaromatization of **2** to **12**, k_{-1} represents the rate of cycloreversion, k_H is a second order rate constant for hydrogen abstraction, and [H] is a concentration of a hydrogen donor.

$$k_{OBS} = \frac{k_1 \times k_H \times [H]}{k_{-1} + k_H \times [H]} \quad (1)$$

$$k_{OBS} = K_{eq} \times k_H \times [H] \quad (2)$$

When the rate of cycloreversion is much faster than the rate of hydrogen abstraction (i.e., $k_{-1} \gg k_H \times [H]$), the observed rate is linearly proportional to the hydrogen donor concentration,

(30) The transmission coefficient in the Eyring equation was set to unity. The nonlinear least square fitting calculation were conducted using Origin 7.5 by OriginLab. The correlation coefficient for this fit was $R^2 = 0.999$.

(31) (a) Karpov, G.; Kuzmin, A.; Popik, V. V. *J. Am. Chem. Soc.* **2008**, *130*, 11771. (b) Poloukhtine, A.; Popik, V. V. *J. Am. Chem. Soc.* **2007**, *129*, 12062.

(32) Perrin, C. L.; Rodgers, B. L.; O'Connor, J. M. *J. Am. Chem. Soc.* **2007**, *129*, 4795.

(33) Kanabus-Kaminska, J. M.; Gilbert, B. C.; Griller, D *J. Am. Chem. Soc.* **1989**, *111*, 3311.

(34) Golden, D. M.; Benson, S. W *Chem. Rev.* **1969**, *69*, 125.

(35) Ruscic, B.; Feller, D.; Dixon, D. A.; Peterson, K. A.; Harding, L. B.; Asher, R. L.; Wagner, A. F. *J. Phys. Chem. A* **2001**, *105*, 1.

as shown in eq 2, where $K_{eq} = k_1/k_{-1}$. The rate of hydrogen abstraction from methanol by 1,4-dehydronaphthaline is 4×10^5 M^{-1} s^{-1}.[6] Since 2-propanol is ~2.7 times more reactive than methanol as a hydrogen donor in radical reactions,[36] we can estimate the rate of hydrogen abstraction by the diradical **12** from 2-propanol as 1.1×10^6 M^{-1} s^{-1}. Using this value and eq 2 we can evaluate the enediyne **2** ↔ *p*-benzyne **12** equilibrium constant at $K_{eq} \approx 10^{-11}$. The free energy difference between **2** and **12** should be $\Delta G^o \approx 15$ kcal mol^{-1}. At high hydrogen donor concentration the hydrogen abstraction step often becomes faster than the cycloreversion (i.e., $k_H \times [H] \gg k_{-1}$) and observed rate levels off at $k_{OBS} = k_1$. Such rate saturation has been reported for the Bergman cyclization of acyclic[6,21a] and cyclic[21d] benzannulated enediynes in the presence of 1,4-cyclohexadiene or methanol. The rate of the cycloaromatization of **2** shows no signs of leveling off even in neat 2-propanol (Figure 3). This observation allows us to conclude that cycloreversion reaction (k_{-1}, Scheme 4) is at least an order of magnitude faster than the reaction of **12** in neat 2-propanol. The lower limit for the cycloreversion reaction can be therefore estimated at 1.4×10^8 s^{-1} and for the cycloaromatization reaction (k_1, Scheme 4) at 1.4×10^{-3} s^{-1}. The upper limit for the activation barrier for the Bergman cyclization of **2** at 25 °C is 21−22 kcal mol^{-1}.

It is also interesting to note that no additional products were observed in the reaction of enediyne **2** at low 2-propanol concentration. In wholly aqueous solutions UV spectrum of **2** remains virtually unchanged for four days at ambient temperatures. In other words, an enediyne **2**−diradical **12** equilibrated system has a relatively long lifetime in the absence of good hydrogen donors.

Theoretical Analysis. Theoretical analysis of the cycloaromatization reaction of enediyne **2** was conducted using a hybrid DFT B3LYP method with 6-31++G(d,p) orbital set. All geometries were optimized using either the restricted (**2** and **3**), unrestricted (**13a**,**b**), or broken-spin unrestricted (**12** and transition states **2**‡, **12**‡**a**,**b**) calculations (Figure 4). The BS-UB3LYP approach has been shown to provide good description of the energetics of Bergman reaction at reasonable computational costs.[37] The optimized geometries, as well as the details of theoretical procedures, are provided in the Supporting Information file. The relative energies of species involved in the Bergman cyclization of enediyne **2** are listed in the Table 1 and presented graphically in the Figure 4. Radical centers in 4,9-didehydrobenz[f]indanol (**12**) are different and there are two transition states for hydrogen transfer: **12a**‡ leading to 9-dehydrobenz[f]indanol radical (**13a**, this pathway is shown in Figure 4); and **12b**‡ leading to 4-dehydrobenz[f]indanol radical (13b). The energies of isomeric transition states **12a**‡ and **12b**‡, as well as radicals **13a** and **13b**, are very close (Table 1).[23]

According to BS-UB3LYP/6-31++G(d,p) calculations, the 4,9-didehydrobenz[f]indanol (**12**) is a ground-state open-shell singlet. The triplet state of this diradical is 4.9 kcal mol^{-1} higher in energy. Enhanced stability of singlet diradicals over triplet state is associated with their reduced reactivity in comparison to monoradicals.[4,38] Incorporation of the enediyne fragment into a nine-membered ring in 4,5-benzocyclonona-2,6-diynol (**2**)

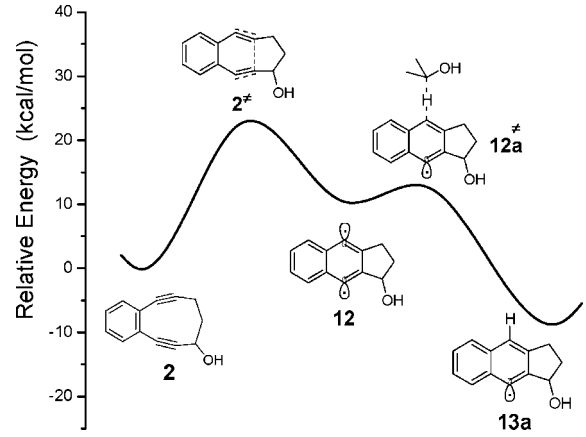


Figure 4. Schematic potential energy profile for the Bergman cyclization of 4,5-benzocyclonona-2,6-diynol (**2**) in the presence of 2-propanol.

Table 1. ZPVE-Corrected Relative Electronic Energies Calculated at the B3LYP/6-31++G(d,p) Level

	Δ*E* (kcal /mol)
2	0.00
2‡	22.30
12 singlet	10.74
12 triplet	15.64
12a‡	12.41
12b‡	12.65
13a	−7.97
13b	−7.68
3	−30.06

produces a moderate ring strain of ~11 kcal mol^{-1} according to B3LYP/6-31++G(d,p) calculations.[23] This value is similar to the ring strain of the isopropyl 2-oxo-5,6-benzocyclodeca-3,7-diyne-1-carboxylate, a benzannulated 10-membered ring enediyne which undergoes Bergman cyclization with a 5 h lifetime in 2-propanol at 36 °C.[39] The strain of the nine-membered ring is apparently reflected in a reduced endothermicity of enediyne **2** cycloaromatization. Thus, **12** lies only 10.7 kcal mol^{-1} above 4,5-benzocyclonona-2,6-diynol (2, Table 1), while the energy gap between the acyclic *o*-diethynylbenzene and 1,4-didehydronaphthaline is about 7 kcal mol^{-1} higher (ΔH^0_{exp}= 17.8 kcal mol^{-1},[6] $\Delta E_{(BCCD(T)/cc-pVDZ)}$ = 17.6 kcal mol^{-1} [40]). Frequency calculations indicate that entropy change accompanying the cyclization of **2** to **12** is quite small and free energy change at 25 °C ($\Delta G^0_{298°}$) is 11 kcal mol^{-1}. This value is 5 kcal mol^{-1} lower than an estimate based on experimental data (vide supra). We put more trust in experimental estimate because BS-DFT method is known for overestimation of the diradical stability. Thus, various BS-DFT calculations produce energy gap between *o*-diethynylbenzene and 1,4-didehydronaphthaline in the range of 11−14.4 kcal mol^{-1} [38b,40] vs experimental value of 17.8 kcal mol^{-1}.[6]

The activation energy predicted by BS-UB3LYP/6-31++G(d,p) calculations ($\Delta E^‡$ = 22.3 kcal mol^{-1}, Table 1) agrees with the upper estimate for the rate of the diradical **12** formation ($k_1 < 10^{-3}$ s^{-1}, vide supra). Surprisingly, ring strain

(36) Janzen, E. G.; Nutter, D. E., Jr.; Evans, C. A. *J. Phys. Chem.* **1975**, *79*, 1983.
(37) (a) Grafenstein, J.; Hjerpe, A. M.; Kraka, E.; Cremer, D. *J. Phys. Chem. A* **2000**, *104*, 1748. (b) Zeidan, T. A.; Kovalenko, S. V.; Manoharan, M.; Alabugin, I. V. *J. Org. Chem.* **2006**, *71*, 962. (c) Schreiner, P. R.; Navarro-Vasquez, A.; Prall, M. *Acc. Chem. Res.* **2005**, *38*, 29.
(38) (a) Clark, A. E.; Davidson, E. R. *J. Am. Chem. Soc.* **2001**, *123*, 10691. (b) Pickard, F. C.; Shepherd, R. L.; Gillis, A. E.; Dunn, M. E.; Feldgus, S.; Kirschner, K. N.; Shields, G. C.; Manoharan, M.; Alabugin, I. V. *J. Phys. Chem. A* **2006**, *110*, 2517.
(39) Karpov, G.; Popik, V. V. *J. Am. Chem. Soc.* **2007**, *129*, 3792.
(40) Prall, M.; Wittkopp, A.; Schreiner, P. R. *J. Phys. Chem. A* **2001**, *105*, 9265.

destabilization of the starting enediyne **2** has limited effect on the height of the activation barrier. Thus, for 10-membered ring analogue, 3,4-benzocyclodeca-1,5-diyne, experimental estimate puts activation energy at 25 kcal mol^{-1}.[21c] For the parent acyclic enediyne, *o*-diethynyl-benzene, experimental enthalpy of activation from the NO-trapping experiments is 25.2 kcal mol^{-1},[6] which agrees with BS-DFT value of 24.6 kcal mol^{-1} but high accuracy BCCD(T)/cc-pVDZ method put the barrier at 29.2 kcal mol^{-1}.[40] This phenomenon can be apparently explained by the fact that Bergman cyclization has an "electronically late" transition state. The geometry of the transition state for cycloaromatization of the parent (*Z*)-hex-3-ene-1,5-diyne is "80% product-like", while electronically it is "70% reactant-like".[41] This means that shortening of transannular distance results in a steady growth of a repulsive interaction between filled in-plain acetylenic π-orbitals, while stabilizing σ-bond formation and aromatization of the out-of-plain π-system lags behind.[12a] Benzannulation of the enediyne system results in further reduction of the aromatic stabilization energy for the transition state and the diradical.[21b] Subsequent abstraction of hydrogen by the diradical **12** from 2-propanol to give monoradicals **13a**,**b** is a highly exothermic process ($\Delta E^\circ = -18.7$ kcal mol^{-1}, Table 1), which makes this step practically irreversible. The barrier for the initial hydrogen abstraction is mostly of entropic nature ($\Delta E^\ddagger = 1.7$ kcal mol^{-1}, Table 1). Radicals **13a**,**b** should be even more reactive and the second hydrogen abstraction from 2-propanol has even higher exothermicity, $\Delta E^\circ = -22.1$ kcal mol^{-1}.

DNA Cleavage Experiments. An evaluation of the photogenerated nine-membered ring enediyne **2** nuclease activity was carried out using supercoiled plasmid DNA cleavage assays. Three forms of this DNA: native (RF I), circular relaxed (RF II, produced by single-strand cleavage), and linear (RF III, formed by scission of both strand in close proximity) are readily separated by the agarose gel electrophoresis.[23] To produce reactive enediyne **2**, 1 and 5 mM aqueous solutions of cyclopropenone **1** were irradiated with low-pressure mercury lamp until ca. 90% conversion. A solution of φX174 supercoiled circular DNA (10 ng/μL) in TE buffer was added to photolysate and incubated for 16 h at 25 °C.

Incubation of the DNA with cyclopropenone precursor **1** (lanes 2 and 4, Figure 5) does not induce any detectable DNA cleavage. The photogenerated enediyne **2**, on the other hand, induces substantial single strand cleavage of φX174 DNA (RF II), while linearized form (RF III) becomes prominent only at higher (5 mM) concentration of the cleaving agent (lanes 3 and 5, Figure 5). Integration of fluorescence of bands on the gel shown in Figure 5, allowed us to evaluate the relative abundance of the native, circular, and linearized forms of φX174 DNA. Thus, incubation of the latter with the 1 mM of enediyne **2** produces 45% of single strand cleavage (RF II) and less than 5% of the double strand cleavage (RF III, lane 3, Figure 5). At 5 mM concentration of **2** DNA-cleavage efficiency grows to 67% and 10%, respectively (lane 5, Figure 5). The relatively high concentrations of **2**, which are required to achieve double-strand DNA photoscission indicate that **2** has low affinity to a dDNA molecule. In order to improve the photonuclease activity of the enediyne **2**, we are working on the design and synthesis of enediyne **2** analogues containing a dDNA minor-groove binding or a dDNA-intercalating moiety. We also explore structural modification that would allow us to shift the absorbance of a cyclopropenone precursor to 350−400 nm.

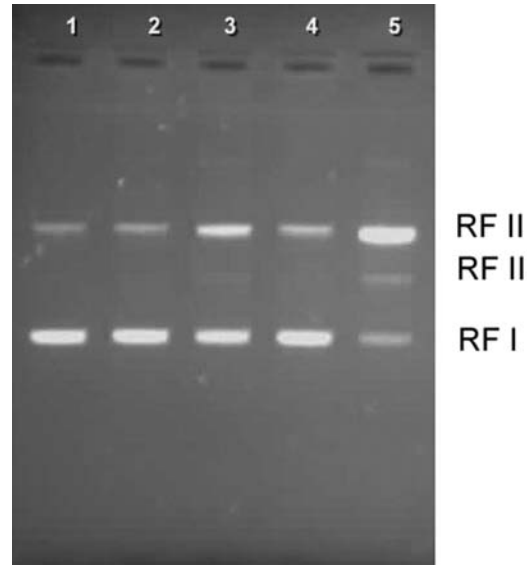


Figure 5. Cleavage of φX174 plasmid DNA by the photogenerated enediyne **2**. Lane 1: DNA alone incubated for 16 h at 25 °C; lane 2 and 4: DNA incubated in the dark with cyclopropenone precursors **1**, 1 and 5 mM; lane 3 and 5: DNA is incubated with the irradiated solution of **1**, 1 and 5 mM.

Conclusions

Irradiation of the thermally stable cyclopropenone precursor **1** produces the first known example of a benzannulated nine-membered ring enediyne, 4,5-benzocyclonona-2,6-diynol (**2**). The enediyne **2** exist in a rapid equilibrium with a corresponding *p*-benzyne analogue, 4,9-didehydrobenz[f]indanol (**12**), even at 25 °C ($K_{eq} \approx 10^{-11}$). In the presence of hydrogen donors, **12** undergoes double hydrogen abstraction quantitatively producing benz[f]indanol (**3**). Kinetic data indicates that the latter process is a rate-limiting step for the Bergman cyclization of the enediyne **2**. While **2** is rapidly consumed in neat 2-propanol ($\tau = 111$ min), the lifetime in aqueous solution is much longer, $\tau_{H2O} > 96$ h. In other words, if **2** is generated in a cellular environment, it will survive in active from until it encounters an appropriate hydrogen donor (DNA, thiol, protein, etc.). Enediyne **2** induces single and double strand cleavage of dDNA molecules, albeit with moderate efficiency. Cyclopropenone precursor **1**, on the other hand, does not produce DNA damage. Therefore, cyclopropenone **1** equipped with a DNA-binding moiety is a promising candidate for the development of an efficient in vivo photonuclease. Synthetic efforts in this direction are underway in our laboratory.

Acknowledgment. Authors thank the National Science Foundation (CHE-0449478), Georgia Cancer Coalition, and donors of the ACS Petroleum Research Fund (434444-AC4) for the support of this project.

Supporting Information Available: Experimental details; synthetic procedures for the preparation of compounds **1** and **3**; photochemical generation of **2**; details of kinetic measurements and DNA-cleaving experiments; Cartesian coordinates of DFT-optimized structures. This material is available free of charge via the Internet at http://pubs.acs.org.

JA8077076

(41) Galbraith, J. M.; Schreiner, P. R.; Harris, N.; Wei, W.; Wittkopp, A.; Shaik, S. *Chemistry* **2000**, *6*, 1446.