Chemistry of Cyclopropyl-*p*-Benzoquinone: A Specific Organogenesis Inhibitor in Plants

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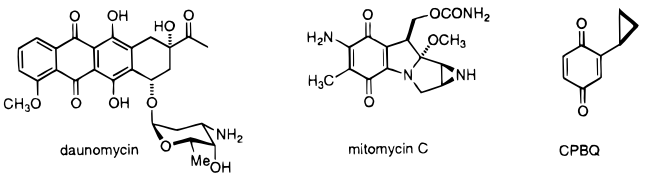
The activity of several classes of antitumor antibiotics (e.g., the mitomycins, anthracyclines, and enediynes) is based on unmasking a specific functionality after reduction of a quinone nucleus.[1,2] Daunomycin, for example, has an amino sugar positioned proximal to the quinone; this relatively poor leaving group appears to be jettisoned only after quinone reduction. The mitomycins undergo the more thermodynamically favorable aziridine ring-opening, again via the semiquinone. Here we explore the chemistry of a cyclopropane when it is attached to the quinone nucleus. This investigation has led to the generation of a new class of specific inhibitors of redox-controlled development, the simplest of which is cyclopropyl-*p*-benzoquinone (CPBQ).[3] We report on the unique reaction pathways open to various substituted cyclopropanes, each bearing a quinone as one substituent on the ring, and discuss their potential biological relevance to the mechanism of the *in vivo* inhibition.



Several mechanism-based inactivators are known in which a cyclopropane is activated for nucleophilic attack by substituent protonation.[4] CPBQ[5] adds solvent under acidic conditions (TsOH/MeOH), but to the quinone nucleus, as seen for the alkylquinones,[6] rather than to the cyclopropane (**1a**, Scheme 1). However, the phenyl substituent in *cis*-Ph-CPBQ[5] activates cyclopropane opening; clean addition of 2 equiv of solvent produces the hydroquinone as a mixture of all four stereoisomers, **1b**. The electron-deficient quinone uniquely activates the cyclopropane, via the quinone methide, for the addition of two nucleophile equivalents.

Scheme 1

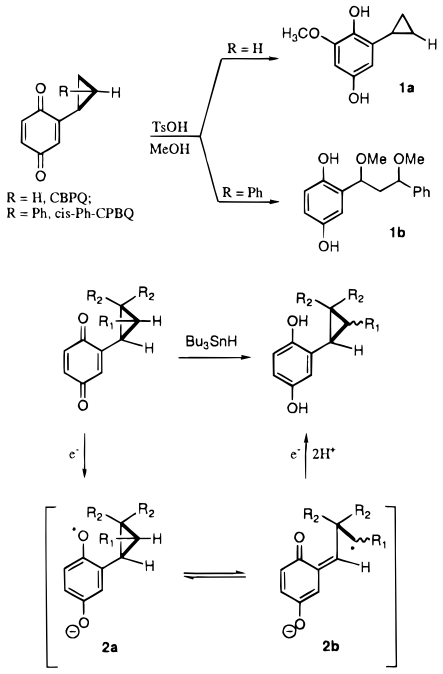


Figure 1. Reduction of cyclopropylbenzoquinones (Q) to -hydroquinones (HQ): CPBQ to CPHQ, $R_1 = R_2 = H$; CPBQ-d_3 to CPHQ-d_3, $R_1 = R_2 = D$ gives 3% *trans* isomer; *cis*-Ph-CPBQ to *cis*-Ph-CPHQ, $R_1 = Ph$, $R_2 = H$ gives 70% *trans* isomer.

One-electron reduction produces an electron-rich semiquinone. Under conditions that extend its lifetime,[7] cyclopropyl ring isomerization occurs more rapidly than further reduction to the hydroquinone (Figure 1). Neither CPBQ-d_3 nor *cis*-Ph-CPBQ isomerized in the absence of reductants, while the hydroquinones, CPHQ-d_3 and *cis*-Ph-CPHQ, did not isomerize in the presence or absence of reductants. Moreover, reduction in Bu_3SnD showed no incorporation of deuterium. Taken together, the isomerization does not originate with hydrogen atom abstraction; rather, isomerization must be the result of cyclopropyl ring-opening from the semiquinone.

The bioreductive antibiotics exploit the anionic character of the semiquinone.[1,2] With the cyclopropane, the reactions of the semiquinone of CPBQ are more related to the ketyl anion of the aryl cyclopropyl ketones.[8] Radical character is delocalized into the cyclopropyl ring, and the ring-opened form has a lifetime sufficient for bond isomerization.[9] **2b**, however, does not react under the conditions reported here, while the ring-opened aryl cyclopropyl ketyl anions are reduced under controlled-current electrolysis.[9] Similar conditions could be explored as models of the CPBQs' binding site.

Photochemical reduction of CPBQ was explored in an effort to extend the lifetime of the semiquinone. In contrast to the expected reduction, an unprecedented ring-expanded product, **3**, was produced in high yield.[10] In *t*-BuOH, the yield of **3** is 80%, and the quantum yield (0.033 ± 0.003 in MeOH and 0.044 ± 0.004 in *t*-BuOH) is unchanged.[11] The annulation reaction is, therefore, not dependent on hydrogen atom abstraction from the solvent, a reaction of the triplet state which is important in the reduction to the hydroquinone. Furthermore, the reaction does not appear to be limited by sterically hindered nucleophiles.

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(4) (a) Suckling, C. J. *Angew. Chem., Int. Ed. Engl.* **1988**, *27*, 537. (b) Liu, H.-w.; Walsh, C. In *The Chemistry of the Cyclopropyl Group*; Rappoport, Z., Ed.; Wiley-Interscience: New York, 1987; Part 2, p 969. (c) Haddow, J.; Suckling, C. J.; Wood, H. C. S. *J. Chem. Soc., Perkin Trans. 1* **1989**, 1297. (d) Kraus, G. A.; Kim, H.; Thomas, P. J.; Metzler, D. E.; Metzler, C. M.; Taylor, J. E. *Synth. Commun.* **1990**, *20*, 2667. (e) Croteau, R.; Alonso, W. R.; Koepp, A. E.; Shim, J. H.; Cane, D. E. *Arch. Biochem. Biophys.* **1993**, *307*, 397. (f) Silverman, R. B.; Ding, C. Z.; Borrillo, L.; Chang, J. T. *J. Am. Chem. Soc.* **1993**, *115*, 2982.

(5) Direct LAH reduction (McOmie, J. F. W.; Watts, M. L.; West, D. E. *Tetrahedron* **1968**, *24*, 2289−2292; *Org. Synth. Collect. Vol. 5*, **1973**, 412−414. Jorgenson, M. J.; Friend, A. W. *J. Am. Chem. Soc.* **1965**, *87*, 1815−1816; *Org. Synth. Collect. Vol. 5*, **1973**, 509−513) of ethyl *trans*-2,5-dimethoxycinnamate, demethylation ($(CH_3)_2S{\cdot}BBr_3$), and oxidation (MnO_2) give CPBQ in high yield. 2,5-Diacetoxystyrene is cyclopropanated with phenyldiazomethane to give a resolvable mixture of isomers, deprotected (LAH), and oxidized (MnO_2) to give *cis*-Ph-CPBQ.

(6) Bruce, J. M. In *The Chemistry of the Quinonoid Compounds*; Patai, S., Ed.; John Wiley & Sons: New York, 1974; Vol. 1, pp 465−538.

(7) Klingler, R. J.; Mochida, K.; Kochi, J. K. *J. Am. Chem. Soc.* **1979**, *101*, 6625−6637.

(8) For a review, see: Boche, G.; Walborsky, H. M. *Cyclopropane Derived Reactive Intermediates*; John Wiley & Sons: Chichester, 1990; p 256.

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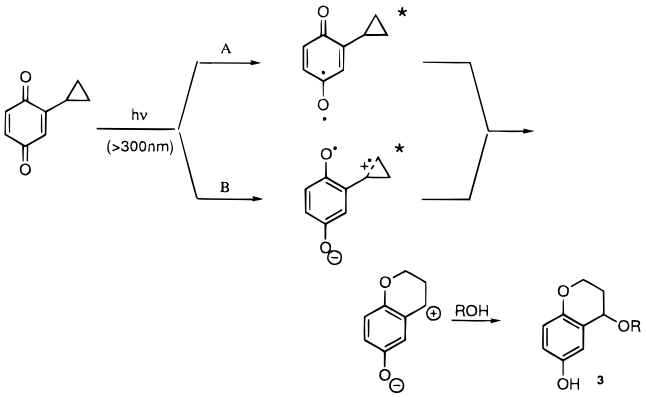
Scheme 2



3 may arise by rearrangement of the diradical and electron demotion, as suggested in Scheme 2, path A; however, these characteristics appear to be more consistent with previously described reactions of one-electron cyclopropyl σ bonds. When oxidized intermolecularly with photoexcited oxidants, arylcyclopropanes add nucleophiles with high regio- and stereoselectivity.[12] In the case of CPBQ, intramolecular oxidation of the cyclopropane could occur by direct low-energy excitation of the quinone.[13] The lowest energy CPBQ $\pi \rightarrow \pi^*$ transition is red-shifted from methylbenzoquinone by 30 nm to 345 nm (ϵ 1492 cm^{-1} M^{-1}). Irradiation at this frequency results in a broad, structureless fluorescence centered at 420 nm.[14] Alkylquinones, in general, have excited singlet and triplet states that are of comparable energy, ~50−60 kcal/mol, and fluorescence is not observed.[6] Here, the photochemical results are consistent with charge transfer character in the excited state[15] and with a dominant funneling through path B.

In conclusion, benzoquinone redox chemistry is critical both to the activation of several classes of antitumor antibiotics and to the function of several well-known redox circuits, most notably in the electron transport chains associated with oxidative phosphorylation and photosynthesis. Redox-responsive gene expression systems have been detected recently in both prokaryotes and eukaryotes.[16] For example, specific benzoquinones have been shown to induce organogenesis in plants,[17,18] and it was recently suggested that these compounds act to complete a critical redox circuit required for the initiating event in cellular development.[3] The chemistry described above appears to be particularly useful in the study of the mechanism of the initiation of plant development, as the simplest structure, CPBQ, functions as a specific irreversible inhibitor.

This inhibition could require trapping **2b**, an intermediate which contains radical and quinone methide character. Cyclopropylamines have been developed as mechanism-based inhibitors of monoamine oxidase; here, radical recombination with the ring-opened structure gives covalent adducts between the flavin cofactor and the inhibitor.[19] **2b** would have to be trapped by a reactive functional group unique to the binding site of the redox-active binding protein in order to explain the observed inhibition. The unique ability of CPBQs to accept two nucleophile equivalents, essentially cross-linking the binding site, may also explain the inhibition. The product distribution from this reaction should be readily distinguished from a redox-activated inhibition pathway. Both pathways appear to require nucleophilic residues within the binding site, and the unprecedented photoannulation reaction opens a separate and distinct opportunity to map the nucleophilic residues in the binding site through photochemical unmasking of the electrophilic center. The general synthetic utility of this reaction also deserves further scrutiny.

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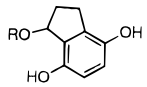
(10) In addition to **3**, **4** and the reduced hydroquinone **5** are isolated in a relative ratio of 8:1:1.



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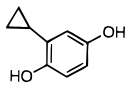

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(11) Quantum yields employed 1 M valerophenone in pentane as an actinometer (Murov, S. L. *Handbook of Photochemistry*; Marcel Dekker: New York, 1973); light from an oriel lamp was filtered to 320−380 nm. Wagner, P. J.; Kochevar, I. E.; Kemppainen, A. E. *J. Am. Chem. Soc.* **1972**, *94*, 7489−7494.

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(14) A solution of cyclopropylquinone in MeOH was deoxygenated by three freeze−thaw cycles and then irradiated in a MPF-66 fluorospectrometer at 350 nm.

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