Chinese Chemical Letters Vol. 11, No. 6, pp. 479–482, 2000

Photoinduced One-Electron Reduction of 1,4-Dihydroxyanthraquinone

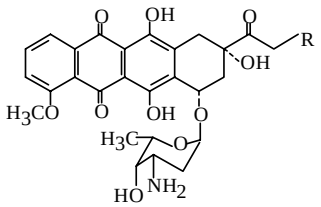
Zhi Cheng XU, Jing Yi AN*, Yi Zhen HU, Fei TIAN

Institute of photographic chemistry, Academi Sinica, Beijing 100101

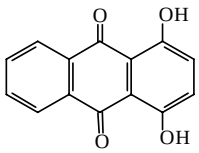
Abstract: The photoinduced one-electron reduction of 1,4-dihydroxyanthraquinone(1,4-DHAQ) was studied by fluorescence, absorption and ESR spectroscopics.

Keywords: 1,4-Dihydroxyanthraquinone, photochemistry , reduction .

Anthracyclines, such as adriamycin **a** and daunomycin **b**, containing the 1,4-dihydro-xyanthraquinone(1,4-DHAQ) moiety, are widely used as anti-cancer drugs[1-3]. It showed that the photosensitizing ability of anthracyclines may allow their uses in the treatment of tumors. One of the possible modes of action that has received attention in recent years is the ability to undergo photochemical reduction to generate semiquinone radical anions and a number of reactive oxygen species[4]. Therefore, it is importance to characterize the corresponding semiquinone radical anion by studying simple model compounds, *e.g.* 1,4-DHAQ, rather than the daunomycin ,adriamycin themselves. 1,4-DHAQ forms the semiquinoid moiety of the anthracycline can be studied as a model compound of anthracycline drugs. In this study we reported the photoinduced one-electron reduction of 1,4-DHAQ upon visible light illumination in the presence of 1-benzyl-1,4-dihydronicotinmide (BNAH), a typical electron donor. The semiquinone radical anion was characterized photochemically and spectrophotometrically.



a R=OH, adriamycin; **b** R=H, daunomycin



1,4-DHAQ

Materials and Methods

1,4-DHAQ and 5,5-dimethyl-1-pyrroline-N-oxide(DMPO) were purchased from Aldrich Chemical Company. BNAH was a generous gift from Prof. Huijun Xu. The fluorescence spectra were run on a Hitachi MPF-4 fluorometer. Perylene (Φ_f=0.99 in benzene) was used

as the standard for the determination of fluorescence quantum yield of 1,4-DHAQ at the excitation wavelength of 438 nm. The concentration of 1,4-DHAQ was 1.1×10^{-6} mol/L.

For the spectrophotometric study, the solution of 1,4-DHAQ and BNAH was deoxygenated by passing argon through for 3 minutes and then illuminated using a 300 W medium pressure sodium lamp in company with a filter cutting off the light of wavelength 470 nm. The absorption spectra were measured employing a HP diode-array spectrophotometer Model 8451 A. The spectra lines were recorded after each exposure. In the ESR experiments, samples were illuminated directly inside the microwave cavity of the spectrometer. A Shoeffel 1 KW Xe Arc lamp was used as light source and a monochromator was used to isolate the 470 nm emission. The ESR measurements were carried out on a Varian E-109 spectrometer. For the measurement of semiquinone radical anion of 1,4-DHAQ the concentration of the sample in dimethylsulfoxide were 1×10^{-3} mol/L for 1,4-DHAQ and 5×10^{-4} for BNAH respectively. The deaerated sample in 1mm quartz capillary was illuminated for 1 minute.

The superoxide radical anion produced was identified by means of spin-trapping method for ESR detection. In the experiments, 20 μL of DMPO (0.3 mol/L) and oxygen were involved into 250 μL of deaerated solution of 1,4-DHAQ in DMSO. The concentration of 1,4-DHAQ was 1×10^{-4} mol/L.

Results and Discussion

Photophysics

The first excited singlet state of 1,4-DHAQ fluorescents medium(ϕ_f=0.11,λem 535 and 565 nm), and possesses a short lifetime(τ_f=2.44 ns in CH_3CN).The mechanism was believed to be as **(1),(2)**.

$$1,4\text{-DHAQ} \xrightarrow{h\nu > 470\ nm} {}^1(1,4\text{-DHAQ}) \qquad \textbf{(1)}$$

$${}^1(1,4\text{-DHAQ}) \longrightarrow {}^0(1,4\text{-DHAQ}) + h\nu_f(\phi_f) \qquad \textbf{(2)}$$

The singlet energy of 1,4-DHAQ was measured to be 2.38 V. Cyclic voltammetry studies carried out with 1,4-DHAQ in CH_3CN showed that the redox potential for one-electron reduction was –0.31V *vs.* NHE. Using the above singlet energy, the redox potential for one-electron reduction of the singlet excited state can be calculated to be 2.07 V *vs.* NHE. Thus, on thermodynamic grounds, the singlet of 1,4-DHAQ should be fairly easy to reduce.

BNAH, a model compound of nicotinamide adenine dinucleotide, was used as electron donor in this study. The fluorescence of 1,4-DHAQ could be efficiently quenched by BNAH. The Stern-Volmer behavior was observed by adding various amounts of BNAH to the solution of 1,4-DHAQ. The fluorescence quenching rate constant of 1,4-DHAQ by BNAH was calculated to be 6.3×10^{10}(mol/L)$^{-1}$S^{-1}. The result suggested that the electron transfer between 1,4-DHAQ and BNAH proceeds *via* the excited singlet state of 1,4-DHAQ.

Spectrophotometric measurements

Typical absorption spectral change upon illumination of 1,4-DHAQ in the presence of BNAH is shown in **Figure 1**.The mechanism of photoreduction of 1,4-DHAQ is believed to occur as **(3)** .

$$1,4\text{-DHAQ}^{*} + \text{BNAH} \longrightarrow 1,4\text{-DHAQ}^{\cdot -} + \text{BNAH}^{\cdot +} \quad \textbf{(3)}$$

1,4-DHAQ shows absorption maximum at 472nm in CH_3CN. The absorption band at 348 nm is attributed to BNAH in CH_3CN since 1,4-DHAQ has very low absorption at this wavelength. This ensure a select excitation of 1,4-DHAQ with visible light. Deaerated sample containing 1,4-DHAQ (1,1 x10 $^{-4}$mol/L) and BNAH (3 x10 $^{-4}$mol/L) was illuminated with visible light of wavelength longer than 470 nm, absorbed only by the sensitizer, at 0.5, 1.0, 1.5, 2.0 and 2.5 min. intervals. The absorption spectra were recorded after each exposure. It may be seen that illumination induces decay of the maxima at 348 nm and 472 nm, characteristic of BNAH and 1,4-DHAQ respectively, and is concomitant with formation of the absorption maxima at 492 and 540 nm (sh). There are only one group of isobestic points within the spectra region examined (300-800 nm), at 320, 375, 410 and 475 nm, indicating that the presence of two light-absorbing forms : the starting 1,4-DHAQ and its semiquinone radical anion,1,4-DHAQ$^{\cdot -}$. The absorption spectrum with maximum at 492 nm is attributed to the semiquinone radical anion, 1,4-DHAQ$^{\cdot -}$, since the maximum wavelength at 492 nm and the ε_{492nm} of the reduced form are in good agreement with those of 1,4-DHAQ$^{\cdot -}$ obtained previously by controlled-potential electrolysis[5].

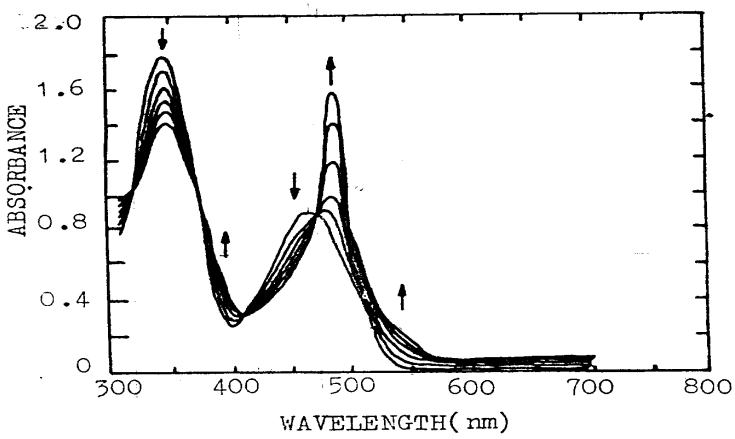
To support the suggestion an ESR study was performed. When the reaction is followed by ESR, the buildup of a singlet radical species is observed concurrently with the changes in the ESR spectrum, as shown in **Figure 2**. The ESR spectrum is indefinitely stable and approximately the same spectrum and total intensity are observed after the illumination stopped. The 17-line ESR spectrum recorded is readily attributed to the semiquinone radical anion 1,4-DHAQ$^{\cdot -}$, since the ESR hyperfine structure observed is exactly the same as that generated thermochemically[6]. The intensity of the ESR signal increased as the illumination continued, the parallel relationship between the intensities of the ESR signal and the absorption band at 492nm was also observed, which demonstrates strongly that the species with absorption maximum at 492nm could be assigned to 1,4-DHAQ$^{\cdot -}$.

The ESR signal and absorption at 492 nm were indefinitely stable in the absence of oxygen, but disappear rapidly on exposure to air to generate $O_2^{\cdot -}$. The mechanism was believed to occur as (4).

$$1,4\text{-DHAQ}^{\cdot -} + O_2 \longrightarrow 1,4\text{-DHAQ} + O_2^{\cdot -} \quad \textbf{(4)}$$

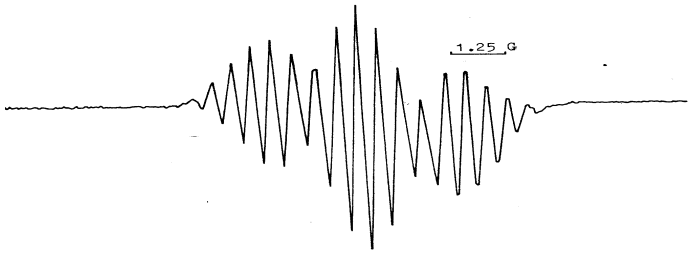
When DMPO (an efficient scavenger of $O_2^{\cdot -}$) and oxygen were involved in the solution of the photoinduced 1,4-DHAQ radical, a multiple ESR signal (12-line) with hyperfine coupling constants of a^N=12.75G, a_β^H=10.25G and a_γ^H=1.25G, which are characteristics of the DMPO-superoxide radical adduct[7], was immediately observed. These results further support our suggestion that the species with absorption maximum at 492nm is attributed to 1,4-DHAQ$^{\cdot -}$. In summmary, the identification of the photochemically generated semiquinone radical anion of 1,4-DHAQ will facilitate future investigation on the photochemistry and photobiology of 1,4-DHAQ, and is important for understanding the biological functions of anthracycline antibitics *in vivo*.

Figure 1 The changes of absorption spectra during the photoinduced one-electron reduction of 1,4-DHAQ.



Deaerated sample containing 1,4-DHAQ ($1.1x10^{-4}$mol/L) and BNAH ($3x10^{-4}$mol/L) in CH_3CN was illuminated for 0.5, 1.0, 1.5, 2.0 and 2.5 min. Arrows indicate the direction of changes.

Figure 2 ESR spectrum of the semiquinone radical anion of 1,4-DHAQ produced by illumination of 1,4-DHAQ and BNAH in dimethylsulfoxide (DMSO).



THE ESR spectrum obtained in CH_3CN is similar to that in DMSO. Instrumental settings: microwave power 5 mW, modulation amplitude $3.2x10^{-5}$T, scan width $2.5x10^{-3}$T, gain $6.3x10^3$.

References

1. T. Mukherjee, P. M. Guyan, J. M. Bruce, *J. Chem. Soc. Faraday Trans*, **1990**, *86* (9), 1483.
2. I. Gutierrez, S. G. Bertolotti, M. A. Biasutti, A. T. Soltermann, N. A. Garcia. *Can. J. Chem.*, **1997**, *75*, 422.
3. D. K. Palit, H. Pal, T mukherje, J. P. Mittal. *J. Photochem. Photobiol. A:chemistry*, **1990,** *52*, 375.
4. K. Gollnik, S. Held, D. O. Martire, S. E. Braslarsky, *J. ptotochem. phtotbiol. A: Chem.*, **1992,** *69*, 155.
5. A. Ann, J. Moiroux, *Nouveau Journal De Chimie*, **1984**, *8*(4), 259.
6. M. Tachibana, S. Tero-Kubota, M. Iwaizumi, *J. Cord. Chem.*, **1988**, *18*, 77.
7. J. R. Harbour, M. L. Hair, *J. Phys. Chem.*, **1978**, *82*(12), 1397.

Received 21 January 2000