# Electronic supplementary materials

*Mendeleev Commun.*, 2007, **17**, 37−39

## Porphyrin dimers and their interaction with DNA

**Kai Wang, Shi-tao Fu, Lei Wu and Zao-ying Li***

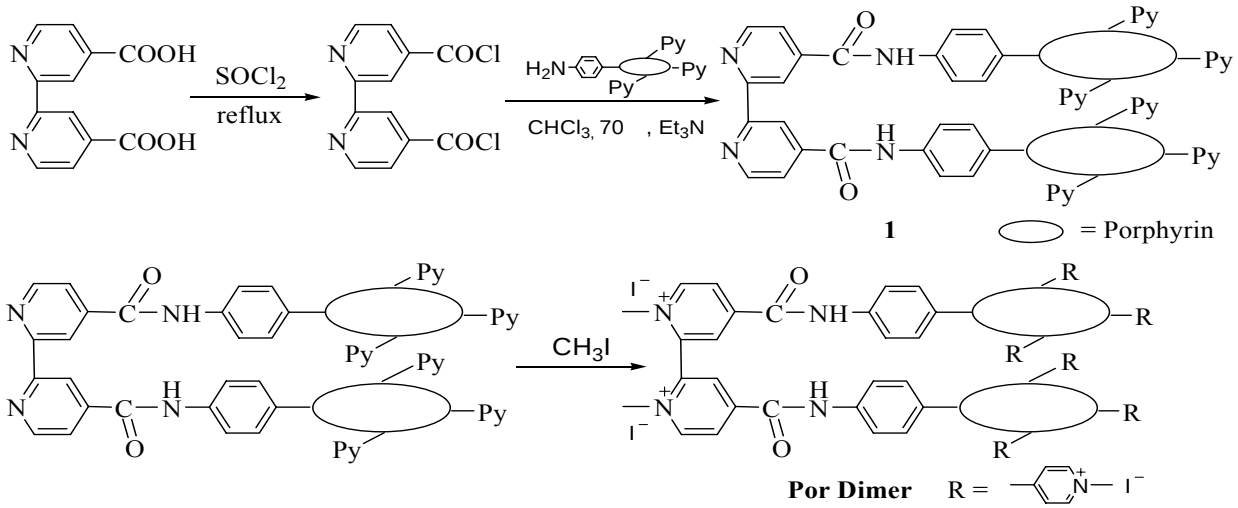
## Experimental

### General

All reagents and solvents were purchased from commercial sources and used with standard purification. Chromatographic separations were performed using silica gel G (200-300 mesh). All UV-visible spectra were obtained on a Shimadzu 1601 spectrophotometer. Fluorescence spectra were recorded on Perkin Elmer LS-55 spectrometer. Circular dichroism was measured on a JASCO J-810 spectrometer. IR spectra were obtained on a Shimadzu FT-IR 3000 spectrometer. Proton NMR spectra were measured using a Varian Mercury-VX 300 spectrometer. Mass spectra were obtained on a TSQ 7000 instrument.

The spectral measurements were performed at room temperature in buffer (pH=7.4, 0.05 M Tris-HCl, 0.1 M NaCl). An extinction coefficient of $1.31\times10^4$ $M^{-1}$ $cm^{-1}$ at 260 nm was used to determine the CT DNA concentration in base pairs. UV-vis absorption spectra of porphyrins were recorded within a range of 380-510 nm. Fluorescence spectra of porphyrins were recorded within a range of 550-750 nm. CD spectra were recorded within a range of 380-520 nm.

5-(4-aminophenyl)-10,15,20-tripyridyl porphyrin was prepared according to literature method.[1]




Synthetic routes

**Procedure for preparing porphyrin dimer 1.**

4,4'-dicarboxy-2,2'-bipyridine (1 eq) and large excess of thionyl chloride were mixed and refluxed for three hours. The solvent was evaporated to dryness, and 5-(4-aminophenyl)-10,15,20-tripyridyl porphyrin (2.5 eq), triethylamine (5 eq) and dried chloroform were added and refluxed gently for 18 hours. The solution was concentrated and purified by silica gel column using mixture of chloroform and methanol as eluent. The main band was collected and recrystallized from a mixture of chloroform and petroleum ether.

For **1**: Yield: 40%. $^1$H NMR ($CDCl_3$, 300 MHz), δ: 9.06 (s, 2H, $Py^3$), 9.02 (m, 16H, $H_\beta$), 8.94 (d, 4H, CONH-$Ph_m$), 8.90 (d, 2H, $Py^6$), 8.83 (d, 12H, Por-$Py_o$), 8.67 (d, 2H, $Py^5$), 8.24 (d, 4H, CONH-$Ph_o$), 8.13 (d, 12H, Por-$Py_m$), -2.85(s, 4H, pyrrole-H); UV-vis ($\lambda_{max}$/nm, in $CHCl_3$): 420, 517, 553, 594, 652; IR (KBr, $cm^{-1}$): 1654 (-CONH-); MS (FAB), m/z: 1472 $[M-1]^+$.

**Procedure for preparing Por Dimer.**

To a solution of porphyrin dimer **1** in 5 mL DMF was added methyl iodide (>400-fold), and the mixture was stirred at room temperature for three hours. After concentrating, 50 mL ether was poured into the mixture, and the precipitate was obtained with a centrifuge and dried under vacuum.

**Por Dimer**: Yield: 94%. $^1$H NMR (DMSO-$d_6$, 300 MHz), δ: 11.24 (s, 2H, $Py^3$), 9.47 (m, 16H, $H_\beta$), 9.19 (br s, 6H, CONH-$Ph_m$, $Py^6$), 9.11(br s, 12H, Por-$Py_o$), 9.00(m, 6H, $Py^5$, CONH-$Ph_o$), 8.18(br s, 12H, Por-$Py_m$), 4.70(s, 24H, -$CH_3$), -2.99(s, 4H, pyrrole-H); MS (FAB), m/z: 1594 $[M-8I]^+$.

**The ability to producing singlet oxygen:**

Measurement of singlet oxygen production was carried out by DPBF (1, 3-diphenyl isobenzofuran) decomposition reaction. Porphyrin ($1.0\times10^{-6}$ M) and DPBF ($1.0\times10^{-4}$ M) were dissolved in the buffer (pH=7.4, 0.05 M Tris-HCl, 0.1 M NaCl), and irradiated under a high-pressure lamp (50W). A decrease of DPBF concentration was measured by an absorbance at 415 nm. The result was shown as below. The slopes of the plots of bleached absorption of DPBF versus illumination time were 0.02511 for **$H_2$TMPyP**, 0.02500 for **Por Dimer**. And the slope is proportional to the rate of production of singlet oxygen.

**Apparent binding constant measurements:**

The apparent affinity binding constants were determined by using a competition method with EB. This assay consists of the measurement of the fluorescence intensity of EB bound to CT DNA in the presence of porphyrin. Fluorescence spectra of EB binding to DNA were recorded at 540 nm for excitation and 610 nm for emission. The decrease of fluorescence in the cuvettes containing porphyrins determined the apparent binding constants ($K_{app}$) of the porphyrin for CT DNA according to literature 2. The measurement was performed at room temperature in buffer (pH=7.4, 0.05 M Tris-HCl, 0.1 M NaCl). The binding constant of EB for DNA in the experimental conditions was $5.93\times10^5$ $M^{-1}$.

**DNA photocleavage assay:**

The photocleavages of pBR322 plasmid DNA by the porphyrins were investigated using agarose gel electrophoresis. All experiments were performed in buffer . The photo-inducing experiments were performed by illumination with 50 W high-pressure mercury at 37 °C and the distance from the sample to the filament of the mercury lamp was 15 cm. DNA was analyzed by 0.9% agarose gel electrophoresis. The gel was incubated in a solution of ethidium bromide for 30 min and the DNA bands were filmed by gel imaging instrument of Vilber Lourmat Bio Print. 10 μL reaction mixtures contained 1.0 μg of plasmid DNA, and the concentration of porphyrins was 2.0 or 0.5 μM. Time of illumination was 12 min.

## References


1 D. Li, C. Casas, G. Etemad-Moghadam and B. Meunier, *New J. Chem.,* 1990, **14**, 421.

2 J.B. LePecq and C. Paoletti, *J. Mol. Biol.,* 1967, **27**, 87.