

# Nickel(II) tetrakis(dimethylpyrazolium-4-yl)-porphyrin: Experimental and DFT studies on its interaction with poly(dG-dC)<sub>2</sub> and poly(dA-dT)<sub>2</sub>

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**Abstract:** The interaction of nickel(II) complex of cationic porphyrins bearing five-membered rings, *meso*-tetrakis(1,2-dimethylpyrazolium-4-yl)porphyrinatonicel(II) (NiPzP), with synthetic polynucleotides poly(dG-dC)<sub>2</sub> and poly(dA-dT)<sub>2</sub> has been characterized by viscometric, visible absorption, CD and MCD spectroscopic, and melting temperature measurements. The nickel(II) complex NiPzP is intercalated into poly(dG-dC)<sub>2</sub> but outside bound to the major groove of poly(dA-dT)<sub>2</sub>. The binding constants of NiPzP to poly(dG-dC)<sub>2</sub> and poly(dA-dT)<sub>2</sub> are in the order of 10<sup>6</sup> M<sup>-1</sup> and comparable to those of other reported cationic metalloporphyrins. The binding process of NiPzP to poly(dG-dC)<sub>2</sub> and poly(dA-dT)<sub>2</sub> is endothermic and entropically driven.

**Keywords:** metallopyrazolium-4-ylporphyrin, poly(dG-dC)<sub>2</sub>, poly(dA-dT)<sub>2</sub>, porphyrin-DNA interaction, thermodynamic parameters.

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## Introduction

A great deal of interest has been aroused in cationic porphyrins and their non-covalent interactions with DNA since the first synthesis [1] and the prior discovery by Fiel et al. [2]. Substantial interests in cationic porphyrins are stemmed from the viewpoint of their role in biological systems. It has been far reported that cationic porphyrins act as a DNA cleaver [3-6], a specific probe of DNA structure [7], and a receptor for peptides [8]. Recently, cationic porphyrin has been also reported to act as an inhibitor of human telomerases through G-quadruplex stabilization, which involve in stopping of cancer cell proliferation [9-12]. In the studies of porphyrin-DNA interaction, .....

In this paper, we report the binding characteristics of NiPzP with polynucleotides of poly(dG-dC)<sub>2</sub> and poly(dA-dT)<sub>2</sub> on the basis of the binding mode, the binding affinity and thermodynamic parameters of the interaction as well as the influence of the axial ligands on the interaction between NiPzP and polynucleotides. Meanwhile, the theoretical calculations applying DFT were also carried out to explain its polynucleotide-binding behaviors.

## Experimental

### Materials and instruments

Nickel(II) pyrazoliumylporphyrin (NiPzP) was synthesized and characterized according to the previous method [13]. Duplex synthetic DNA (poly(dG-dC)<sub>2</sub> and poly(dA-dT)<sub>2</sub>) were purchased from Sigma Co. Ltd. and used as received. Other chemicals were used as received without further purification and all solvents were of reagent grade. The viscosity of buffer solution of DNA was measured by a Ubbelohde viscometer. The UV-visible absorption spectra were recorded in solution at 25 °C (unless otherwise stated) on a JASCO V-

570 spectrophotometer equipped with a JASCO ETC-505T temperature controller using 10 mm quartz cells. The CD and MCD spectra were recorded on a JASCO J-720 WI spectropolarimeter using 10 mm quartz cells.

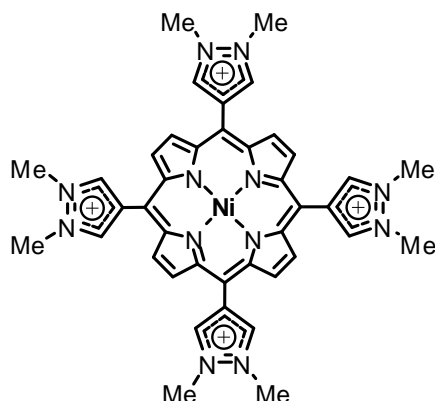


Fig. 1. Structure of nickel(II) tetrakis(dimethylpyrazolium-4-yl)porphyrin

### Molecular modeling

The models were constructed using the HyperChem program-package [14]. The full geometry optimization and frequency analysis of NiPzP were computed by the DFT-B3LYP method with 6-31G\* basis set level provided by HyperChem. A single point calculation was also further performed based on the basis set level of B3LYP/6-31G\*. The bonding energy of water molecule to nickel ion of NiPzP and NiTMPyP as well as the hydration of NiPzP and NiTMPyP in gas phase were calculated according to the published method [15, 16] using the Gaussian 03 package [17]. As a model for poly(dG-dC)<sub>2</sub> and poly(dA-dT)<sub>2</sub>, the double stranded B-DNA of [d(5'-GCGCGCGCGC-3')]<sub>2</sub> and [d(5'-ATATATATAT-3')]<sub>2</sub> were generated using the nucleic acid feature in the database of HyperChem. The models were geometrically minimized using Fletcher-Reeves conjugate-gradient minimization. The convergence was defined when the gradient of the average root mean square (RMS) shift reached 0.001 kcal Å<sup>-1</sup> mol<sup>-1</sup>. Models for intercalation and face-on were constructed by hand-docking of the porphyrins to DNA. The final structures were obtained by geometrical optimization using Amber force field.

## Results and discussion

### Spectroscopic evidence for porphyrin-DNA interaction

The visible spectrum of NiPzP is very different from that of NiTMPyP. The latter NiTMPyP persistently exists in both four- and six-coordinate form as shown in its Soret band spectrum, and it appeared as a peak at 422 nm and a shoulder at 445 nm. The observed NiTMPyP spectrum is in accordance with the previous finding. In contrast, the Soret band of NiPzP shows a single peak at 411 nm, indicating that NiPzP exists in one form as four-coordinate complex. It is also supported by a theoretical study that NiPzP has lower preference to bind to the H<sub>2</sub>O ligand compared to NiTMPyP. The formation of NiPzP.H<sub>2</sub>O and NiPzP.2H<sub>2</sub>O has more positive enthalpy than that of NiTMPyP.H<sub>2</sub>O and NiTMPyP.2H<sub>2</sub>O (Table 1).

**Table 1.** Energetics (kcal.mol<sup>-1</sup>) for hydration process studied in gas phase for NiPzP and NiTMPyP at 298 K

Porphyrin	$\Delta E$	$\Delta H$	$-T\Delta S$	$\Delta G$
NiPzP-H <sub>2</sub> O	-20.7	-17.2	6.8	-10.4
NiPzP-2H <sub>2</sub> O	-22.2	-15.4	7.3	-8.1
NiTMPyP-H <sub>2</sub> O	-23.9	-19.2	5.1	-12.1
NiTMPyP-2H <sub>2</sub> O	-25.3	-22.5	8.7	-13.8

Upon interaction of NiPzP with poly(dG-dC)<sub>2</sub>, the substantial hypochromicity (*ca* 27%) and large red shift (12 nm) of the Soret band were observed for the binding of NiPzP to poly(dG-dC)<sub>2</sub>, indicating that the

perturbation to the localized  $\pi, \pi^*$  MOs ( $1a_{1u}$   $3a_{2u}$   $4e_g$ ) of the porphyrin is large. For this system, a conservative CD pattern with a negative and a positive signal appeared at 414 nm and 430 nm, respectively. .... It is easy to find that the energies of the LUMO and LUMO+x ( $x=1-4$ ) of NiPzP (Fig. 2) are from  $-11.21$  eV to  $-8.46$  eV, which are much lower than those of the HOMO and HOMO-x ( $x=1-6$ ) of the DNA base-pairs model.

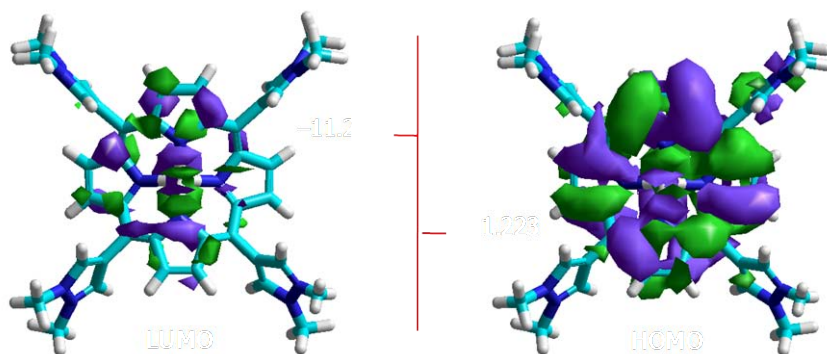


Fig. 2. Energy levels (eV) of the molecular orbitals of NiPzP (LUMO and HOMO) and its frontier orbital contour calculated at the level of DFT-B3LYP/6-31G\*

#### **Binding affinity and selectivity**

The melting temperature ( $T_m$ ) of DNA is sensitive to its double helix stability, and the binding of porphyrins to DNA alters the  $T_m$  depending on the strength of interactions [2]. .....

#### **Conclusions**

The binding properties of nickel(II) pyrazoliumylporphyrins to poly(dG-dC)<sub>2</sub> and poly(dA-dT)<sub>2</sub> has been studied by viscometric measurements, spectroscopic methods as well as in silico approach. NiPzP is intercalated into poly(dG-dC)<sub>2</sub>, while it is bound face-on at the 5'TA3' step of the major groove of poly(dA-dT)<sub>2</sub>. The binding constants of NiPzP to poly(dG-dC)<sub>2</sub> and poly(dA-dT)<sub>2</sub> were  $4.02 \times 10^6$  and  $3.21 \times 10^6$  M<sup>-1</sup> at 298 K, respectively, and are comparable to those of other reported cationic porphyrins. The DNA-binding process of NiPzP is endothermic and entropically driven. These experimental results have demonstrated that the insertion of nickel(II) ions into H<sub>2</sub>PzP changes the DNA-binding properties. This finding is important to get some insights into porphyrin-DNA interactions at the molecular level.

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