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Interaction of three new tetradentates Schiff bases containing N_2O_2 donor atoms with calf thymus DNA

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ABSTRACT

Interaction of 1,3-bis(2-hydroxy-benzylidene)-urea (H2L1), 1,3-bis(2-hydroxy-3-methoxy-benzylidene)-urea (H2L2) and 1,3-bis(2-hydroxy-3-methoxy-benzylidene)-urea nickel(II) (NiL2) with calf-thymus DNA were investigated by UV–vis absorption, fluorescence emission and circular dichroism (CD) spectroscopy as well as cyclic voltammetry, viscosity measurements, molecular docking and molecular dynamics simulation. Binding constants were determined using UV–vis absorption and fluorescence spectra. The results indicated that studied Schiff-bases bind to DNA in the intercalative mode in which the metal derivative is more effective than non metals. Their interaction trend is further determined by molecular dynamics (MD) simulation. MD results showed that Ni derivative reduces oligonucleotide intermolecular hydrogen bond and increases solvent accessible surface area more than other compounds.

1. Introduction

Schiff-bases are the important class of compounds in both medicinal and pharmaceutical fields [1,2]. In recent years, Schiff-base metal complexes have also found important applications in the biological field [3–7]. Schiff-bases play an important role in bioinorganic chemistry as they exhibit remarkable biological activity.

On the other hand, nucleic acids have important influence in biological systems and carry out a broad range of biological functions. DNA is the primary intracellular target of anticancer drugs and so the interactions between small molecules and DNA causes DNA damage in cancer cells, blocking their division and resulting in cell death [8]. Therefore, the interaction of metal complexes with DNA has attracted much attention [9,10]. Among the transition metal ions, zinc and nickel are biologically active [11,12].

The knowledge of intracellular speciation of nickel is essential for deeper understanding of the toxicity mechanism of this carcinogenic metal [13,14]. However, the interaction of drug molecules with DNA has become an active research area [15]. Since DNA is the intracellular target for a wide range of anticancer and antibiotic drugs [16–18], generally, there are three interaction modes between small molecules and DNA: (i) intercalative binding that small molecules intercalate into the base pairs of nucleic acids; (ii) groove binding in which the small molecules bound on nucleic acids in major or minor groove; (iii) long-range assembly on the molecular surfaces of nucleic acids [19]. The intercalative binding is stronger than other two binding modes because the surface of intercalative molecule is sandwiched between the aromatic, heterocyclic base pairs of DNA [20,21]. Tremendous interest has been attracted to interactions between transition metal complexes of morin and nucleic acids due to potential applications of the metal complexes as anticancer drugs or as complexes with other biological functions [8,22]. These studies are also important to understand the toxicity of drugs containing metal ions [23–25].

CT-DNA is a polymer of alternate sugar phosphate sequence with high polymerized skeleton. The investigation of drug–DNA interaction is important for understanding the molecular mechanism of drug action and for the design of specific DNA targeted drug. DNA binding is the critical step for DNA activity. To design effective chemotherapeutic agents and better anticancer drugs, it is essential to explore the interactions of drug with DNA. Interaction of a few ligands with DNA were investigated by different experimental methods such as UV–vis, fluorescence, CD, calorimetry, CV and computational methods such as MD, docking and QSAR techniques were investigated [26–31].

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In the present study, the interaction of new tetradentate Schiff-bases containing N_2O_2 donor atoms and its nickel complex with calf thymus DNA were investigated using various spectroscopy methods, molecular dynamics simulation and docking calculations.

2. Experimental

2.1. Materials

All materials were purchased from Sigma–Aldrich Company. All experiments involving interaction of the complexes with DNA were carried out in buffer (10 mM Tris–HCl, pH = 7.4). The DNA concentration per nucleotide was determined by absorption spectroscopy using the molar absorption coefficient (6600 M^{-1} cm^{-1}) at 260 nm. The structure of studied Schiff base (SB)s which synthesized by inorganic chemistry laboratory of Damghan University were shown in Fig. 1.

2.2. UV–vis spectroscopy measurement

UV–vis spectra were recorded on a Perkin-Elmer UV-Vis spectrophotometer model Lambda 25. Absorption titration experiments were performed in the wavelength range, 200–600 nm in a constant concentration of studied SBs and titrated by varying concentration of DNA ([DNA]/[compounds] = 0.00, 0.02, 0.04, 0.06, 0.08, 0.10, 0.12, 0.14, 0.16, 0.18 and 0.20) in (10 mM Tris–HCl, pH = 7.4). These solutions were incubated for 5 min.

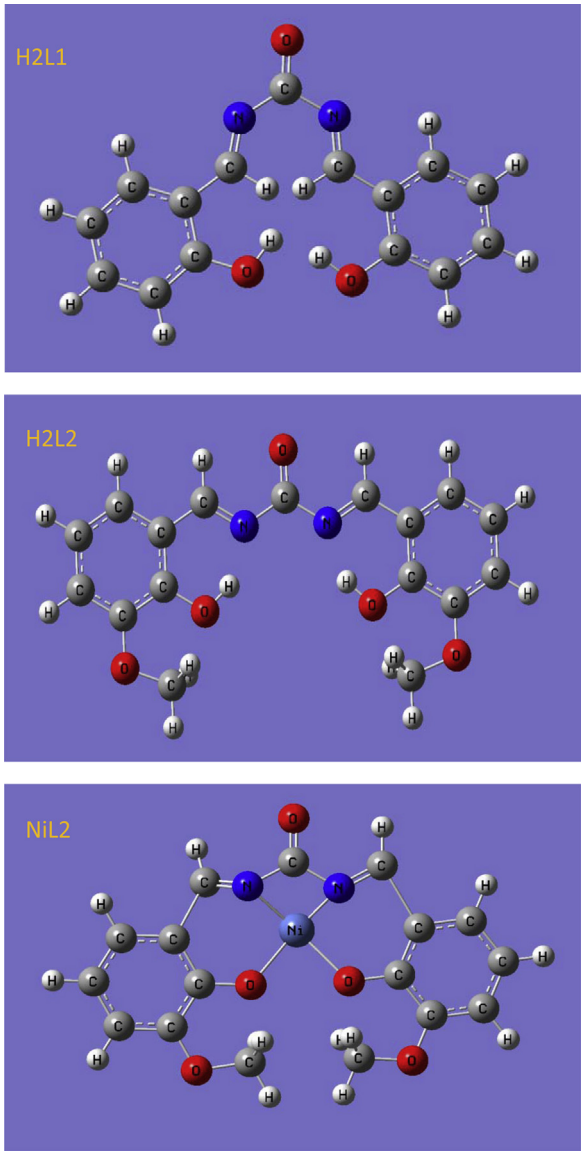

Fig. 1. Chemical structure of three studied SBs.

2.3. Thermal stability

DNA thermal stability was investigated by indicated absorbance value versus temperature. The absorbance at 260 nm was scanned from 35 to 85 °C at 5 °C per min scan rate and fixed concentration ratio ([SB]/[DNA] = 0.5). The melting temperature T_m, was defined as the mid-point of transition temperature.

2.4. Fluorescence measurements

Emission spectra were recorded on a Jasco spectrofluorometer model FP6200 luminescence spectrometer at 298 K. To compare quantitatively the affinity of the cited compounds to DNA, the average binding constants, K_b, were obtained by fluorescence spectroscopy [9]. Fluorescence measurements were performed on fixed amounts of SBs in the presence of different amounts of DNA.

Ethidium bromide (EB) is a fluorescent probe for DNA structure which has been employed in examination of the ligand binding mode to DNA. The excitation wavelengths for 5×10^{-4} M H2L1, H2L2 and NiL2 were 276, 484 and 514 nm, respectively and average of fluorescence emission intensity was monitored for H2L1, H2L2 and NiL2 as 250–600 nm, 470–550 nm and 500–550 nm, respectively.

2.5. Viscosity measurement

Viscosity was measured by an Ostwald viscometer immersed in a thermostatic water-bath maintained to 25.0 °C. In all samples, the DNA concentration was kept constant (1×10^{-8} M). Calculation was carried out using Eq. (1);

$$\eta_0 = \frac{t_{DNA} - t_0}{t_0} \quad (1a)$$

$$\eta = \frac{t - t_0}{t_0} \quad (1b)$$

where η_0 and η are the viscosity of DNA in the absence and presence of SB, t_0, t_{DNA} and t are the observed flow time of buffer, DNA and DNA containing solution upon the addition of SBs, respectively [32]. The values of $(\eta/\eta_0)^{1/3}$ versus the concentration ratio of SB to DNA were plotted.

2.6. Circular dichroism

Cyclic voltammetry measurements were done using Potentiostat/Galvanostat Autolab. The CD spectra are quite sensitive to the changes in the secondary structure of nucleic acids, which any conformational modification of DNA provoked by its interaction with SBs. CD spectrum of each sample was scanned in the ranges of 220–320 nm. The concentration of DNA was 1.0×10^{-3} M. A CD spectrum was generated by subtracting the CD spectrum of the native DNA and mixture of DNA-SB from the CD spectrum of the buffer and buffer-SB solutions.

2.7. Cyclic voltammetry

Circular dichroism spectra were recorded by AVIV circular dichroism spectrophotometer model 215. Cyclic voltammetry measurements were carried out by glassy carbon as working electrode, a platinum wire as an auxiliary electrode, and