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Study on synthesis, structure, and DNA-binding of Ni, Zn complexes with 2-phenylquinoline-4-carboylhydrazide

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ABSTRACT

2-Phenylquinoline-4-carboylhydrazide (HL), and its novel nickel(II), zinc(II) complexes $[M(HL)_2(L)]\cdot 2H_2O\cdot NO_3$ (M = Ni (**1**), M = Zn (**2**)), have been synthesized and characterized by elemental analysis, molar conductivity, and IR spectra. The crystal structure of $[Ni(HL)_2(L)]\cdot 2H_2O\cdot NO_3$ obtained from ethanol solution was determined by X-ray diffraction analysis, crystallized in the rhombohedral system, space group $R\bar{3}$, Z = 18, a = 31.913(3) Å, b = 31.913(3) Å, c = 27.709(2) Å, $\alpha = 90°$, $\beta = 90°$, $\gamma = 120°$, R_1 = 0.0647. The interactions of the complexes and the ligand with calf thymus DNA had been investigated using UV–Vis spectra, fluorescent spectra, CD (circular dichroism) spectra, CV (cyclic voltammetry) and viscosity measurements. These compounds were tested against MFC (mouse forestomach carcinoma) cell lines. The complex **1** showed significant cytotoxic activity against MFC cell lines. The cleavage reaction on plasmid DNA has been monitored by agarose gel electrophoresis. Results suggest that the two complexes bound to DNA via a groove binding mode and the complexes can cleave pBR322 DNA.

1. Introduction

Binding studies of small molecules to DNA are very important in the development of DNA molecular probes and new therapeutic reagents. Over the past decades, the DNA-binding metal complexes have been extensively studied as DNA structural probes, DNA-dependent electron transfer probes, DNA footprinting and sequence-specific cleaving agents and potential anticancer drugs [1]. Transition-metal complexes are being used at the forefront of many of these efforts. Stable, inert, and water-soluble complexes containing spectroscopically active metal centers are extremely valuable as probes for biological systems. As both spectroscopic tags and functional models for the active centers of proteins, metal complexes have helped elucidate the mechanisms by which many metalloproteins function [2].

In order to develop new antitumor drugs which specifically target DNA, it is necessary to understand the different binding modes a metal complex is capable of undergoing. Basically, the complex bound to DNA through three non-covalent modes: intercalation, groove binding and external static electronic effects. Among these modes, the groove binding mode plays an important role in the efforts of the drugs targeted to DNA [3]. Some of the transition metal complexes activate endonucleolytic cleavage reaction in DNA in the presence of external agents or photolytically [4–7]. These endonucleolytic cleavage of DNA by metal complexes are used as models for the reactivity of some antitumor antibiotics. Previously some work demonstrated that 4-quinolinecarboxylic acid amides and hydrazides, exhibit pronounced antiinflammatory and analgesic activity at a quite low toxicity [8–12], but no metal complexes of such drug have been reported in the past which may have better pharmaceutical effect. Therefore, studies of the metal complexes are important to explore the possible new drug compare with the present drug.

As a part of our continuing research [13], 2-phenylquinoline-4-carboylhydrazide as a ligand and its complexes of transition metal (Ni, Zn) had been synthesized. We got the crystal of the Ni(II) complex, and described a comparative study of the interactions of complexes **1**, **2** and ligand with Calf thymus DNA (CT-DNA) using UV–Vis, CD, fluorescence, and CV measurements in order to gain a better understanding of the antitumor mechanism of these compounds. In order to provide more details about the relationship of the better antitumor properties of complexes and the mode of DNA binding, we have investigated the interaction of the complexes with DNA in this work. The results of agarose gel electrophoresis patterns show that the complex **1** is capable of cleaving Plasmid DNA at physiological pH and temperature. Information obtained from this study will be helpful to the understanding of the mechanism of interactions of aroylhydrazines and their complexes with nucleic acids, and should be useful in the development of potential probes of DNA structure and conformation and new therapeutic regents for some diseases.

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2. Experimental

2.1. Materials and instrumentation

All reagents and solvents were purchased commercially and used without further purification unless otherwise noted. CT-DNA and pBR322 DNA were obtained from Sigma Chemicals Co. (USA). Agarose was purchased from Promega Co. (German), ethidium bromide (EB) were obtained from Huamei Chemical Co.(Beijing, China). The concentration of DNA was determined spectrophotometrically using a molar absorptivity of 6600 M^{-1} cm^{-1} (260 nm) [14].

The melting points of the compounds were determined on a Beijing XT4-100 × microscopic melting point apparatus (the thermometer was not corrected). Carbon, hydrogen, and nitrogen were analyzed on an Elemental Vario EL analyzer. Infrared spectra (4000–400 cm^{-1}) were determined with KBr disks on a Therrno Mattson FTIR spectrometer. The UV–Vis spectra were recorded on a Varian Cary 100 UV–Vis spectrophotometer. The fluorescence spectra were recorded on a Hitachi RF-4500 spectrofluorophotometer. ^{1}H NMR spectra were measured on a Varian VR 300-MHz spectrometer, using TMS as a reference. Mass spectra were performed on a VG ZAB-HS Fast-atom bombardment (FAB) instrument and electrospray mass spectra (ESI-MS) were recorded on a LQC system (Finngan MAT, USA) using CH_3OH as mobile phase.

2.2. Preparation of the ligand (H_2L)

The ligand (Fig. 1) was prepared according to the literature [13]. 2-Phenylquinoline-4-carboxylic acid (12.49 g; 50 mmol) was esterified to 2-phenylquinoline-4-carboxylate, treatments of the esters with $N_2H_4{\cdot}H_2O$ gave the corresponding hydrazine (10.98 g; (90%)). ester: yellowish solid. ^{1}H NMR (300 MHz, $CDCl_3$, s, singlet; d, doublet; t, triplet; q, quartet; m, multiplet) δ (ppm): 1.49–1.53 (t, 3H, $-CH_3$), 4.52–4.59 (q, 4H, $-CH_2-$), 7.49–7.81 (m, 5H, –Ph), 8.20–8.27 (m, 3H, -quinoline), 8.40 (s, 1H, -quinoline), 8.73–8.76 (d, 1H, -quinoline). Hydrazine: white solid, m.p. 220–222 °C. ^{1}H NMR (300 MHz, DMSO-d_6) δ (ppm): 4.73 (s, 2H, $-NH_2$), 7.58–8.31 (m, 10H, –Ar), 10.04 (s, 1H, –NH–). FAB-MS: m/z = 264.1 $[M+H]^+$. Anal. Calcd for $C_{16}H_{13}N_3O$: C, 72.99; H, 4.98; N, 15.96. Found: C, 73.01; H, 4.94; N, 16.01. IR ν_{max} (cm^{-1}): ν(C=O): 1631 cm^{-1}, $\gamma(NH_2)$: 1125 cm^{-1}, ν(N–H): 3258 cm^{-1} U_{max} (nm): 210, 255, 330.

2.3. Preparation of the complexes

The ligand (1.0 mmol, 0.263 g) and the nickel nitrate (1.0 mmol, 0.120 g) were added to ethanol (10 mL). The mixtures were stirred at 60 °C. After 5 min, the mixtures were filtered to remove any insoluble residues and then stirring was continued for 24 h at room temperature. A green precipitate, the Ni(II) complex (**1**), was separated from the solution by suction filtration, purified by washing several times with ethanol, and dried for 24 h under vacuum, yield 85%. The Zn(II) complex (**2**) was prepared by the same method, yield 78%. Anal. Calcd (Found) for complex **1** $C_{48}H_{43}N_{11}O_{11}Ni$: C, 57.11 (57.216); H, 4.23 (4.30); N, 15.37 (15.28). Λ_m (s cm^2 mol^{-1}): 87.6. ESI-MS [CH_3OH, m/z]: 846.5, $[Ni(HL)_2(L)]^+$. IR ν_{max} (cm^{-1}): ν(C=O): 1608 cm^{-1}, $\gamma(NH_2)$: 1068 cm^{-1}, ν(N–H): 3236 cm^{-1} U_{max} (nm): 210, 255, 330. Anal. Calcd for complex **2** $C_{48}H_{43}N_{11}O_{11}Zn$: C, 56.72 (56.80); H, 4.25 (4.31); N, 15.25 (15.18). Λ_m (s cm^2 mol^{-1}): 93.1. ESI-MS [CH_3OH, m/z]: 852.2, $[Zn(HL)_2(L)]^+$. IR ν_{max} (cm^{-1}): ν(C=O): 1610 cm^{-1}, $\gamma(NH_2)$: 1073 cm^{-1}, ν(N–H): 3245 cm^{-1} U_{max} (nm): 210, 255, 330.

2.4. X-ray crystallography

Details of the crystal parameters, data collection and refinements are listed in Table 1. The crystal of the complex **1** was determined on a BRUKER SMART 1000 CCD diffractometer equipped with a graphite crystal monochromatized Mo K_α radiation (λ = 0.71073 Å) at 298(2) K. Cell constants and an orientation matrix for data collection were obtained by least-squares refinement of diffraction data from 34 reflections in the range $1.84 < h < 24.97$. The intensity data were collected by the ω scan mode within $1.28° < \theta < 25.01°$ for hkl ($-20 \leqslant h \leqslant 37$, $-37 \leqslant k \leqslant 31$, $-17 \leqslant l \leqslant 32$) in the rhombohedral system. The structure was solved by Patterson methods and completed by iterative cycles of least-squares refinement and DF-syntheses. H-atoms were located in their calculated positions and treated as riding on the atoms to which they are attached. All non-hydrogen atoms were refined anisotropically. All calculations were carried out using the SHELXS-97 program [15].

2.5. Cytotoxicity assay

The in vitro cytotoxicity of the complexes and the ligand was investigated as follows. The MFC cell lines, which was established

Table 1
Crystal data and structure refinement for $[Ni(HL)_2(L){\cdot}2H_2O{\cdot}NO_3]$.

Empirical formula	$C_{48}H_{42}N_{10}NiO_8$
Formula weight	945.63
Temperature (K)	298(2)
Wavelength	0.71073
Crystal system/space group	Rhombohedral, $R\bar{3}$
Unit cell dimensions	
a (Å)	31.913(3)
b (Å)	31.913(3)
c (Å)	27.709(2)
α (°)	90
β (°)	90
γ (°)	120
Volume (Å^3)	24439(4)
Z	18
Density (calculated) (Mg/m^3)	1.157
Absorption coefficient (mm^{-1})	0.413(11)
F(000)	8856
Crystal size (mm)	0.38 × 0.35 × 0.34
θ Range for data collection (°)	1.28–25.01
Index ranges	$-20 \leqslant h \leqslant 37$, $-37 \leqslant k \leqslant 31$, $-17 \leqslant l \leqslant 32$
Reflections collected/unique	33,369/9566
Absorption correction	Semi-empirical from equivalents
Refinement method	Full-matrix least-squares on F^2.
Data/restraints/parameters	9566/0/640
Goodness-of-fit on F^2	1.000
Final R indices [$I > 2\sigma(I)$]	R_1 = 0.0647, wR = 0.1351
R indices (all data)	R_1 = 0.1272, wR = 0.1518
Largest diff. peak and hole (e Å^{-3})	0.458 and −0.262

Crystallographic Data Center as supplementary publication number CCDC 690629

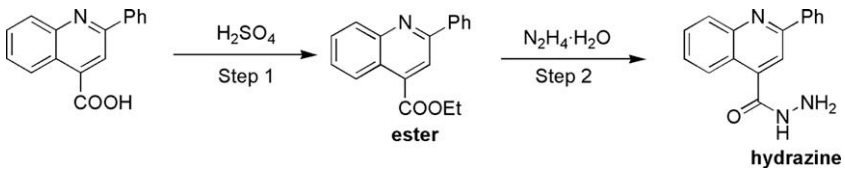

Fig. 1. Scheme of the synthesis of the ligand.