Available online at www.sciencedirect.com
ScienceDirect
Bioorganic & Medicinal Chemistry Letters 18 (2008) 298–303
Bioorganic & Medicinal Chemistry Letters

Synthesis, characterization and DNA-binding properties of four Zn(II) complexes with bis(pyrrol-2-yl-methyleneamine) ligands

Yuan Wang,[a] Zheng-Yin Yang[a,*] and Zhong-Ning Chen[b]

[a]*College of Chemistry and Chemical Engineering and State Key Laboratory of Applied Organic Chemistry, Lanzhou University, Lanzhou 730000, PR China*
[b]*Fujian Institute of Research on the Structure of Matter, Chinese Academy of Sciences, Fuzhou 350002, PR China*

Received 20 May 2007; revised 27 September 2007; accepted 25 October 2007
Available online 30 October 2007

Abstract—A novel series of bis(pyrrol-2-yl-methyleneamine) ligands H_2L^n (n = 1–4) were synthesized via condensation of diamines with two equivalents of 2-formyl-pyrrole (**2**). Their Zn(II) complexes were characterized by elemental analyses, mass spectra and IR spectra. The crystal structures of $[ZnL^1]_2$ and $[ZnL^4]_2$ obtained from ethanol solution was determined by X-ray diffraction analysis, each of them possesses a double-stranded helical geometry. In addition, the DNA-binding properties of the compounds have been fully investigated by absorption, fluorescence and viscosity measurements.

It is well known that DNA is an important cellular receptor. The interaction of metal complexes with DNA has recently gained much attention because it is in close relationship with their potential biological and pharmaceutical activity.[1–5] Furthermore, the factors that determine the affinity and selectivity in binding of complexes to DNA would be valuable in the rational design of new diagnostic and therapeutic agents.[6–8]

The metal complexes of Schiff base bearing pyrrole units have been extensively investigated for a long time.[9,10] Most interesting research has been focused on macrocycles, such as texaphyrins and expanded porphyrins,[11–18] because they could stabilize mono- or binuclear metal complexes which have various structures and special properties. Due to the excellent fluorescent properties and good solubilities of their complexes, linear spaced bis(pyrrol-2-yl-methyleneamine) ligands have attracted much recent attention through the work of Ma and his co-workers.[19–23] By varying the spacers between two pyrrol-2-yl-methyleneamine units, dinuclear dimeric helicates,[19–22] trinuclear trimeric triangle[19] and tetranuclear tetrameric square[19] complexes have been generated. Yet it is noticed that the DNA-binding investigations of such complexes have been relatively few. These facts encouraged us to synthesize a novel series of bis(pyrrol-2-yl-methyleneamine) ligands and their Zn(II) complexes. In addition, the DNA-binding properties of all compounds were discussed in detail.

The ligands (H_2L^n) were prepared as shown in Figure 1, and were fully characterized by ^{1}H NMR, MS and elemental analysis (see Supplementary S1.1). Their Zn(II) complexes were obtained from EtOH/H_2O (1:1) solution in the yields of 50–60% (see Supplementary S1.2). All compounds, except for the complexes of H_2L^2 and H_2L^3, are soluble in DMF, THF, and DMSO, slightly soluble in methanol, ethanol, ethyl acetate, and acetone, insoluble in water and ether. The elemental analyses (Table 1) show that the formulas of the Zn(II) complexes are $[ZnL^n]_2$ (n = 1–4).

Some main IR spectra data are listed in Table 1. The free ligands exhibit bands of the ν_{NH} vibration at 3279–3317 cm^{-1}, but they disappeared in the corresponding complexes, showing that the nitrogen atoms of the pyrrole rings take part in coordination and the active H atoms are substituted by Zn(II) ions. The $\nu_{C=N}$ bands of the ligands appear at 1617–1641 cm^{-1}, they become 1588–1597 cm^{-1} in the complexes, indicating that the azomethine nitrogen atoms of the ligands should coordinate to Zn(II) ions. It is also interesting that the $\nu_{C=O}$ of the ester groups in the complexes (1693–1700 cm^{-1}) are higher than those in the ligands (1669–1673 cm^{-1}). This is probably due to the coordination be-

Keywords: Pyrrole; Complex; Schiff base; DNA-binding.
* Corresponding author. Tel.: +86 931 8913515; fax: +86 931 8912582; e-mail: yangzy@lzu.edu.cn

0960-894X/$ - see front matter © 2007 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmcl.2007.10.085

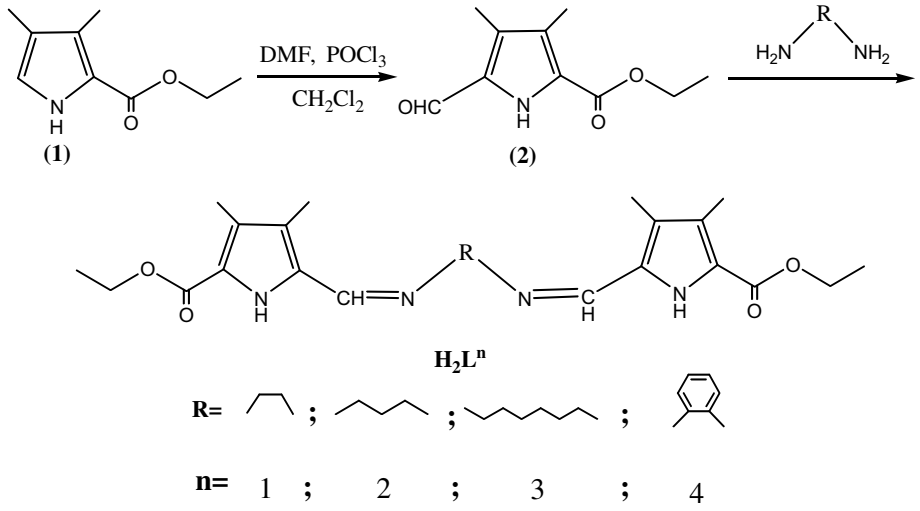

Figure 1. The preparation of the ligands.

Table 1. Elemental analyses and some main IR spectra data

Compound	C% (Cal)%	N% (Cal)%	H% (Cal)%	M% (Cal)%	IR data (cm^{-1})		
					ν_{NH}	$\nu_{C=O}$	$\nu_{C=N}$
H_2L^1	63.43 (63.75)	12.96 (13.52)	6.91 (7.30)		3317	1672	1639
$[ZnL^1]_2$	54.68 (55.29)	11.13 (11.72)	5.41 (5.91)	13.05 (13.68)		1696	1588
H_2L^2	64.29 (64.46)	12.73 (13.07)	7.20 (7.53)		3296	1667	1638
$[ZnL^2]_2$	55.76 (56.04)	11.51 (11.37)	6.34 (6.53)	12.87 (13.27)		1693	1595
H_2L^3	66.00 (66.36)	12.34 (11.91)	7.93 (8.14)		3291	1669	1641
$[ZnL^3]_2$	58.64 (58.37)	10.31 (10.47)	7.31 (6.97)	12.63 (12.22)		1694	1597
H_2L^4	67.30 (67.51)	12.25 (12.11)	6.71 (6.54)		3279	1673	1617
$[ZnL^4]_2$	58.79 (59.38)	10.15 (10.65)	4.92 (5.37)	12.85 (12.43)		1700	1592

tween the nitrogen atoms of the pyrrole rings and Zn(II) ions.

From the X-ray diffraction data given in Table 2, it can be confirmed that the formulas of the Zn(II) complexes with H_2L^n are $[ZnL^n]_2$ (n = 1,4). The IR spectra indicate that all complexes have similar structures. This means that all Zn(II) complexes of H_2L^n should be $[ZnL^n]_2$ (n = 1–4). As shown in Figure 2, the crystal structures of $[ZnL^1]_2$ and $[ZnL^4]_2$ are similar. Each of them consists of two unsymmetrical Zn(II) ions and each Zn(II) ion binds to four nitrogen atoms from two ligands with two covalent bonds and two coordination bonds, and displays a distorted tetrahedral geometry. Their molecular structures are similar to that of $[ZnL^a]_2\{H_2L^a$ = bis[3-ethyl-4-methyl-5-ethyloxy-carbonyl-pyrrol-2-yl-methyleneamino]ethane$\}$[22] and $[ZnL^b]_2\{H_2L^b$ = 1,2-bis[3-ethyl-4-methyl-5-ethyloxy-carbonyl-pyrrol-2-yl-methyleneamino]benzene$\}$.[19]

X-ray analysis of $[ZnL^n]_2$ (n = 1, 4) also indicates that each of them possesses a double-stranded helical geometry (Fig. 3). The twist around the ethyl or phenyl bridge divides the ligand into two pyrrol-2-yl-methyleneamine subunits, each of which is bound to a different Zn(II) ion. Selected bond lengths and angles are summarized in Tables 3 and 4. For $[ZnL^1]_2$, the N–Zn–N angles range from 82.42(9)° to 145.19(10), and the bond distances of Zn–N span from 1.944(3) to 2.112(3) Å. The distance between two zinc centers is 4.389 Å, which is longer than that of $[ZnL^a]_2$ (4.2 Å).[22] For $[ZnL^4]_2$, the N–Zn–N angles range from 82.00(10)° to 147.31(11)°, and the bond distances of Zn–N span from 1.964(2) to 2.101(2) Å. The distance between two zinc centers is 3.588 Å, which is somewhat shorter than that of $[ZnL^b]_2$ (3.7 Å).[19]

Because of their slight solubility in organic solvents, the DNA-binding properties of $[ZnL^2]_2$ and $[ZnL^3]_2$ were not measured. It is a general observation that varieties in both the absorption spectra and the emission spectra accompany the binding of molecules to DNA. The extent of spectral change is related to the strength of binding.[1–7] Due to the obscure spectral changes (see Supplementary data S2-4), the DNA-binding properties of H_2L^n (n = 1–3) were not discussed in this work. The absorption and emission spectra of the other compounds in the absence and presence of CT-DNA (at a constant concentration of the compounds) are given in Figures 4 and 5.

For $[ZnL^1]_2$ and H_2L^4, addition of increasing amounts of CT-DNA (calf thymus DNA) results in observation of weak hypochromicities in the absorption spectra, indicating that $[ZnL^1]_2$ and H_2L^4 can bind to DNA. It is generally accepted that the magnitude of both the

Table 2. Crystal data and experimental data

	$[ZnL^1]_2$	$[ZnL^4]_2$
Formula	$C_{44}H_{56}N_8O_8Zn_2$	$C_{52}H_{56}N_8O_8Zn_2$
Formula weight	955.71	1051.79
Crystal color	colorless	orange
Crystal size (mm)	$0.32 \times 0.18 \times 0.18$	$0.46 \times 0.15 \times 0.12$
Crystal system		
Space group	Monoclinic	Monoclinic
Z	$P2_1/c$	$P2_1/n$
T (K)	4	4
a (Å)	294 (2)	294 (2)
b (Å)	20.899 (3)	14.4772 (7)
c (Å)	14.918 (2)	21.4823 (11)
α (°)	14.787 (2)	16.3439 (8)
β (°)	90.00	90.00
γ (°)	90.00	92.655 (2)
V (Å^3)	90.00	90.00
D_{calc} (g cm^{-3})	4610.3 (12)	5077.6 (4)
Radiation (Å) (Mo K_a)	1.377	
Reflections collected	0.71073	1.376
Independent reflections (R_{int})	24,291	0.71073
F(000)	8554 (0.0345)	29,107
Number of parameters refined	2000	10,370 (0.0530)
Final R indices [$I > 2a$ (I)]	572	2192
R indices (all data)	$R_1 = 0.0383$, $wR_2 = 0.1086$	653
	$R_1 = 0.0649$, $wR_2 = 0.1311$	$R_1 = 0.0464$, $wR_2 = 0.0956$
		$R_1 = 0.0908$, $wR_2 = 0.1145$
Measurement	CCD area detector	
Monochromator	Graphite	
Structure determination	SHELXS-97 and SHELXL-97	
Refinement	Full-matrix least-squares on F^2	

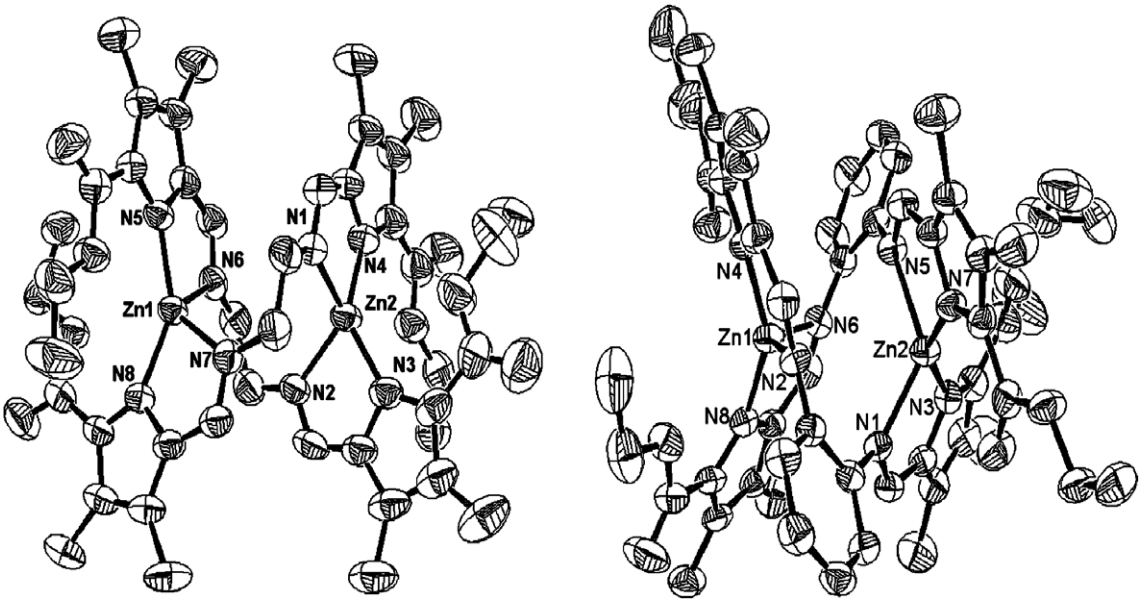

Figure 2. X-ray crystal structures of $[ZnL^1]_2$ (left) and $[ZnL^4]_2$ (right); H atoms are omitted for clarity.

red shift and hypochromism in the absorption spectra is found to correlate with the strength of the intercalative interaction.[7] The hypochromicity is mostly attributed to the interaction between the electronic states of the compound chromophore and those of the DNA bases. On the other hand, the red shift is associated with a decrease in the energy gap between the highest and the lowest occupied molecular orbitals (HOMO and LUMO) when the drug compound binds to DNA.[24] The lack of the red shift suggests that the major binding modes of both $[ZnL^1]_2$ and H_2L^4 with DNA are not classical intercalative interaction. However, for $[ZnL^4]_2$, the prime three additions of increasing amounts of CT-DNA result in hypochromicities, but the subsequent

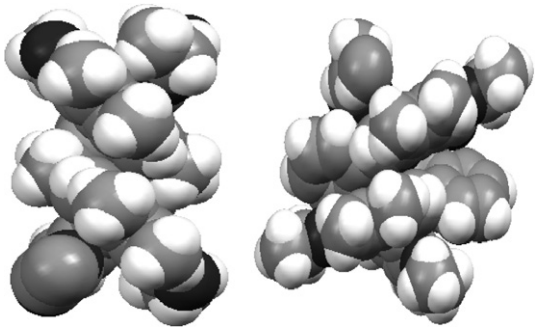

Figure 3. Spacefill structures of $[ZnL^1]_2$ (left) and $[ZnL^4]_2$ (right).

Table 3. Selected bond lengths [Å] and angles [°] of $[ZnL^1]_2$

Bond lengths [Å]			
Zn(1)–N(5)	1.955(2)	Zn(2)–N(1)	2.112(3)
Zn(1)–N(6)	2.099(2)	Zn(2)–N(2)	2.102(2)
Zn(1)–N(7)	2.110(2)	Zn(2)–N(3)	1.964(3)
Zn(1)–N(8)	1.954(2)	Zn(2)–N(4)	1.944(3)
Bond angles [°]			
N(8)–Zn(1)–N(5)	145.19(10)	N(4)–Zn(2)–N(3)	136.23(11)
N(8)–Zn(1)–N(6)	116.65(10)	N(4)–Zn(2)–N(2)	125.98(11)
N(5)–Zn(1)–N(6)	82.80(10)	N(3)–Zn(2)–N(2)	82.50(11)
N(8)–Zn(1)–N(7)	82.42(9)	N(4)–Zn(2)–N(1)	82.64(11)
N(5)–Zn(1)–N(7)	119.38(9)	N(3)–Zn(2)–N(1)	117.15(10)
N(6)–Zn(1)–N(7)	110.42(9)	N(2)–Zn(2)–N(1)	115.56(9)

Table 4. Selected bond lengths [Å] and angles [°] of $[ZnL^4]_2$

Bond lengths [Å]			
Zn(1)–N(4)	1.964(3)	Zn(2)–N(7)	1.964(2)
Zn(1)–N(8)	1.967(3)	Zn(2)–N(3)	1.965(3)
Zn(1)–N(6)	2.082(3)	Zn(2)–N(1)	2.083(3)
Zn(1)–N(2)	2.101(2)	Zn(2)–N(5)	2.092(3)
Bond angles [°]			
N(4)–Zn(1)–N(8)	146.78(11)	N(7)–Zn(2)–N(3)	147.31(11)
N(4)–Zn(1)–N(6)	110.55(11)	N(7)–Zn(2)–N(1)	111.85(10)
N(8)–Zn(1)–N(6)	83.00(11)	N(3)–Zn(2)–N(1)	83.01(10)
N(4)–Zn(1)–N(2)	82.00(10)	N(7)–Zn(2)–N(5)	82.79(10)
N(8)–Zn(1)–N(2)	113.43(10)	N(3)–Zn(2)–N(5)	111.42(10)
N(6)–Zn(1)–N(2)	129.42(10)	N(1)–Zn(2)–N(5)	128.35(10)

two result in hyperchromicity. It shows that there may be two phases of binding between the $[ZnL^4]_2$ and DNA.[6,8] Since $[ZnL^4]_2$ contains phenyl ring in its ligand structure, the spectral changes observed in the presence of DNA may be rationalized in terms of partial intercalation and groove binding.[25]

In the emission spectra, for $[ZnL^1]_2$, with increasing CT-DNA concentration the emission intensity is decreased seriously, due to self-stacking of some free bases in the compound along the DNA surface.[26] According to the classical Stern–Volumer equation, a plot of F_0/F versus [Q] gave the binding constant $1.866 \times 10^6\ M^{-1}$. However, for H_2L^4 and $[ZnL^4]_2$, upon addition of CT-DNA, the notable emission intensity enhancement is observed, indicating that a certain extent of partial intercalation should occur in the DNA-binding progress.[7,27] According to the Scatchard equation, a plot of r/C_f versus r from the fluorescence data gave the binding constant $8.567 \times 10^6\ M^{-1}$ and $1.527 \times 10^6\ M^{-1}$, respectively[5] (see Supplementary S5). It is noticed that our experimental K_b values are much higher than the tested zinc complex ($[Zn(Me\text{-}Hct_C)_2(OH_2)_2]\cdot 4H_2O$ 3.9×10^4; $[Zn(Ph\text{-}Hct_C)_2(OH_2)_2]\cdot 4H_2O$, 8.1×10^5 in 5 mM Tris–HCl/50 mM NaCl buffer, pH = 7.2)[27] and comparable with the classical intercalators (EB–DNA, 3.0×10^6 in 5 mM Tris–HCl/50 mM NaCl buffer, pH = 7.2).[28] These also indicate that such three compounds can bind to DNA effectively.

The emission spectra of DNA–EB system upon the addition of increasing amounts of the compounds were measured (see Supplementary S6). It is well known that EB can intercalate nonspecifically into DNA, which causes it to fluoresce strongly. Competitive binding of other drugs to DNA and EB will result in displacement of bound EB and a decrease in the fluorescence intensity. However, there are even no changes with increasing compound concentrations in the spectra.[7,8] This probably means that such three compounds bind to DNA not via classical intercalative interaction.[5–8] It is in accordance with the results obtained from the absorption and emission titration experiments.

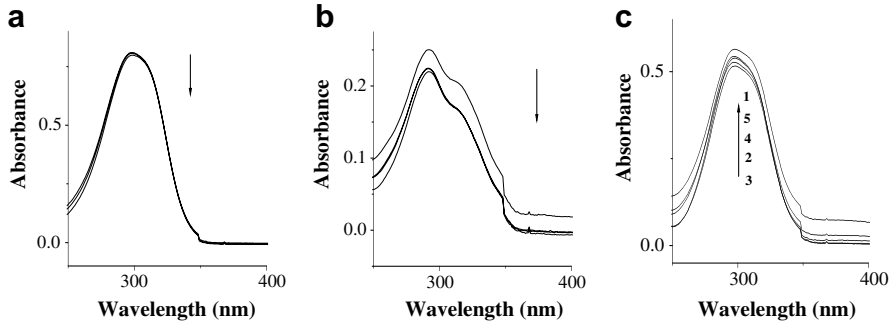

Figure 4. (a) Electronic spectra of $[ZnL^1]_2$ (10 μM) in the presence of increasing amounts of CT-DNA. [CT-DNA] = 0–10 μM. Arrow shows the absorbance changes upon increasing CT-DNA concentration. (b) Electronic spectra of H_2L^4 (10 μM) in the presence of increasing amounts of CT-DNA. [CT-DNA] = 0.7.5 μM. Arrow shows the absorbance changes upon increasing CT-DNA concentration. (c) Electronic spectra of $[ZnL^4]_2$ (10 μM) in the presence of increasing amounts of CT-DNA. [CT-DNA] = 0–10 μM. Lines 1–5 show the absorbance changes upon increasing CT-DNA concentration.

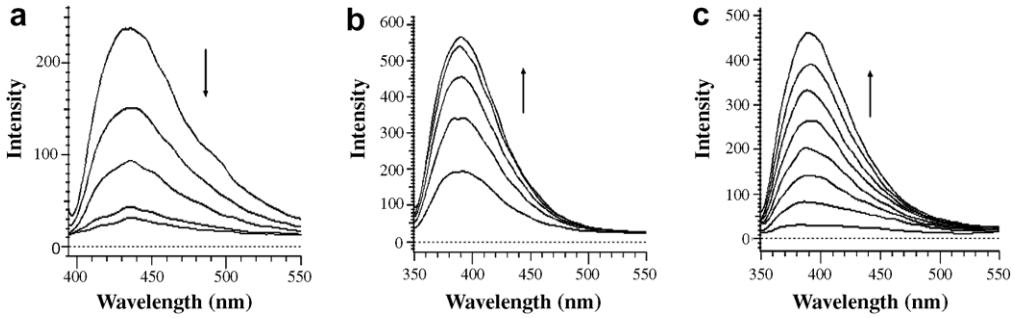

Figure 5. (a) The emission decrease spectra of $[ZnL^1]_2$ (10 μM) in the presence of increasing amounts of CT-DNA. [CT-DNA] = 0–4 μM. Arrow shows the emission intensity changes upon increasing CT-DNA concentration. (b) The emission enhancement spectra of H_2L^4 (10 μM) in the presence of increasing amounts of CT-DNA. [CT-DNA] = 0.10 μM. Arrow shows the emission intensity changes upon increasing CT-DNA concentration. (c) The emission enhancement spectra of $[ZnL^4]_2$ (10 μM) in the presence of increasing amounts of CT-DNA. [CT-DNA] = 0–20 μM. Arrow shows the emission intensity changes upon increasing CT-DNA concentration.

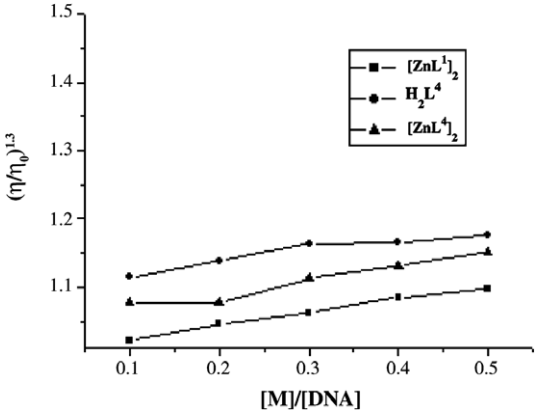

Figure 6. Effect of increasing amounts of $[ZnL^1]_2$, H_2L^4, and $[ZnL^4]_2$ on the relative viscosity of CT-DNA at 25 °C.

The interaction of the small molecular could bend the DNA helix, and reduce its effective length, concomitantly, its viscosity.[5,7,8] The effects of the three compounds on the viscosity of CT-DNA at 25 °C are shown in Figure 6. With an increasing amount of compounds, the relative viscosity of CT-DNA increased slightly, which also suggests that the three compounds can bind to DNA.[7]

From the comparison of tested cyclic free ligands with those in the corresponding linear form, it is obvious that cyclic compounds bind nucleic acid more strongly than the linear ones. In addition, it is noticed that the DNA-binding ability of $[ZnL^1]_2$ is better than that of H_2L^1, while it is reverse to H_2L^4 and $[ZnL^4]_2$. The possible reasons are as following: first, the presence of the metal ion interferes with the interaction processes of the parent free ligands; second, the double-stranded helical structure of $[ZnL^4]_2$ influences the effective partial intercalation of the phenyl ring in its ligand with DNA base pairs. These conclusions may be helpful to the understanding of the DNA interaction mechanism.

X-ray data are available from the CCDC, 12 Union Road, Cambridge CB2 1EZ, UK, www.ccdc.cam.ac.uk/data_request/cif, on request quoting the deposition number CCDC634595 (for $[ZnL^4]_2$) and 634596 (for $[ZnL^1]_2$).

Acknowledgment

This project was supported by the National Natural science Foundation in China (20475023).

Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.bmcl.2007.10.085.

References and notes

1. Jiao, K.; Wang, Q. X.; Sun, W.; Jian, F. F. *J. Inorg. Biochem.* **2005**, *99*, 1369.
2. Brodie, C. R.; Collins, J. G.; Aldrich-Wright, J. R. *J. Chem. Soc., Dalton Trans.* **2004**, 1145.
3. Pellegrini, P. P.; Aldrich-Wright, J. R. *J. Chem. Soc., Dalton Trans.* **2003**, 176.
4. Wu, J. Z.; Yuan, L.; Wu, J. F. *J. Inorg. Biochem.* **2005**, *99*, 2211.
5. Wang, Y.; Yang, Z. Y. *Trans. Met. Chem.* **2005**, *30*, 902.
6. Vijayalakshmi, R.; Kanthimathi, M.; Subramanian, V.; Nair, B. U. *Biochim. Biophys. Acta* **2000**, *1475*, 157.
7. Zeng, Y. B.; Yang, N.; Liu, W. S.; Tang, N. *J. Inorg. Biochem.* **2003**, *97*, 258.
8. Yang, G.; Wu, J. Z.; Wang, L.; Ji, L. N.; Tian, X. *J. Inorg. Biochem.* **1997**, *91*, 141.
9. Chakravorty, A.; Holm, R. H. *Inorg. Chem.* **1964**, *3*, 1521.
10. Weber, J. H. *Inorg. Chem.* **1967**, *6*, 258.
11. Acholla, F. V.; Takusagawa, F.; Mertes, K. B. *J. Am. Chem. Soc.* **1985**, *107*, 6908.
12. Arnold, P. L.; Blake, A. J.; Wilson, C.; Love, J. B. *Inorg. Chem.* **2004**, *43*, 8206.
13. Sessler, J. L.; Tomat, E.; Mody, T. D.; Lynch, V. M.; Veauthier, J. M.; Mirsaidov, U.; Markert, J. T. *Inorg. Chem.* **2005**, *44*, 2125–2127.
14. Veauthier, J. M.; Tomat, E.; Lynch, V. M.; Sessler, J. L.; Mirsaidov, U.; Markert, J. T. *Inorg. Chem.* **2005**, *44*, 6736.
15. Veauthier, J. M.; Cho, W.; Lynch, V. M.; Sessler, J. L. *Inorg. Chem.* **2004**, *43*, 1220.

16. Sessler, J. L.; Mody, T. D.; Dulay, M. T.; Espinoza, R.; Lynch, V. *Inorg. Chim. Acta* **1996**, *246*, 23.
17. Chen, X. B.; Gui, M. D.; Zhu, S. J.; Yang, Y.; Guo, C. C. *Chem. J. Chin. Universities* **1999**, *8*, 1238.
18. Chen, X. B.; Yang, Y. *Chin. J. Org. Chem.* **2000**, *20*, 547.
19. Wu, Z. K.; Chen, Q. Q.; Xiong, S. X.; Xin, B.; Zhao, Z. W.; Jiang, L. J.; Ma, J. S. *Angew. Chem. Int. Ed.* **2003**, *42*, 3271.
20. Yang, L. Y.; Chen, Q. Q.; Yang, G. Q.; Ma, J. S. *Tetrahedron* **2003**, *59*, 10037.
21. Yang, L. Y.; Chen, Q. Q.; Li, Y.; Xiong, S. X.; Li, G. P.; Ma, J. S. *Eur. J. Inorg. Chem.* **2004**, 1478.
22. Wu, Z. K.; Yang, G. Q.; Chen, Q. Q.; Liu, J. G.; Yang, S. Y.; Ma, J. S. *Inorg. Chem. Commun.* **2004**, *7*, 249.
23. Wu, Z. K.; Chen, Q. Q.; Yang, G. Q.; Xiao, C. B.; Liu, J. G.; Yang, S. Y.; Ma, J. S. *Sensors Actuators B* **2004**, *99*, 511.
24. Tan, J. H.; Lu, Y.; Huang, Z. S.; Gu, L. Q.; Wu, J. Y. *Eur. J. Inorg. Chem.* **2007**, *42*, 1169.
25. Uma, V.; Castineiras, A.; Nair, B. U. *Polyhedron* **2007**, *26*, 3008.
26. Nyarko, E.; Hanada, N.; Habib, A.; Tabata, M. *Inorg. Chim. Acta* **2004**, *357*, 739.
27. Baldini, M.; Belicchi-Ferrari, M.; Bisceglie, F.; Capacchi, S.; Pelosi, G.; Tarasconi, P. *J. Inorg. Biochem.* **2005**, *99*, 1504.
28. Baldini, M.; Belicchi-Ferrari, M.; Bisceglie, F.; Pelosi, G.; Pinelli, S.; Tarasconi, P. *Inorg. Chem.* **2003**, *42*, 2049.