Cu(II) and Co(II) ternary complexes of quinolone antimicrobial drug enoxacin and levofloxacin: Structure and biological evaluation

Wan-Yun Huang[a,b,d,1], Ji Li[c,1], Shi-Lin Kong[d], Zhong-Chang Wang[a], Hai-Liang Zhu[a,*]

[a]*State Key Laboratory of Pharmaceutical Biotechnology, Nanjing University, Nanjing 210093, China.*

[b]*Department of Pharmacology, Guilin Medical University, Guilin 541004, China.*

[c]*School of Life Sciences, Shandong University of Technology, Zibo 255049, China.*

[d]*Key Laboratory for the Chemistry and Molecular Engineering of Medicinal Resources (Guangxi Normal University), Ministry of Education of Chin, Guilin 541004, China.*

[1] These authors contributed equally to this work.

*Corresponding author. Tel. & fax: +86-25-83592672; e-mail: zhuhl@nju.edu.cn (H.L. Zhu).

Table S1 Selected bond lengths (Å) and angles (deg) for complex **1**–**4**.

1			
Cu(1)-O(1)	1.930(6)	N(6)-Cu(1)-N(5)	81.1(3)
Cu(1)-O(2)	1.939(6)	O(1)-Cu(1)-N(1)^{A}	97.3(3)
Cu(1)-N(6)	1.982(7)	O(2)-Cu(1)-N(1)^{A}	98.8(3)
Cu(1)-N(5)	2.015(7)	N(6)-Cu(1)-N(1)^{A}	94.5(3)
Cu(1)-N(1)^{A}	2.278(8)	N(5)-Cu(1)-N(1)^{A}	99.5(3)
Cu(2)-O(5)	1.917(6)	O(5)-Cu(2)-O(4)	93.4(3)
Cu(2)-O(4)	1.943(6)	O(5)-Cu(2)-N(11)	162.5(3)
Cu(2)-N(11)	2.000(7)	O(4)-Cu(2)-N(11)	90.9(3)
Cu(2)-N(12)	2.002(8)	O(5)-Cu(2)-N(12)	91.2(3)
Cu(2)-N(7)^{B}	2.291(7)	O(4)-Cu(2)-N(12)	165.0(3)
O(1)-Cu(1)-O(2)	93.9(3)	N(11)-Cu(2)-N(12)	80.7(3)
O(1)-Cu(1)-N(6)	166.1(3)	O(5)-Cu(2)-N(7)^{B}	95.9(3)
O(2)-Cu(1)-N(6)	91.6(3)	O(4)-Cu(2)-N(7)^{B}	96.4(3)
O(1)-Cu(1)-N(5)	89.6(3)	N(11)-Cu(2)-N(7)^{B}	100.5(3)
O(2)-Cu(1)-N(5)	160.7(3)	N(12)-Cu(2)-N(7)^{B}	97.4(3)
2			
Co(1)-O(5)	2.046(3)	O(4)-Co(1)-N(7)	97.24(12)
Co(1)-O(1)	2.073(2)	O(5)-Co(1)-O(2)	175.81(10)
Co(1)-O(4)	2.072(3)	O(1)-Co(1)-O(2)	84.48(10)
Co(1)-N(7)	2.096(3)	O(4)-Co(1)-O(2)	89.34(10)
Co(1)-O(2)	2.097(3)	N(7)-Co(1)-O(2)	89.72(11)
Co(1)-N(8)	2.106(3)	O(5)-Co(1)-N(8)	92.32(12)

O(5)-Co(1)-O(1)	93.43(11)	O(1)-Co(1)-N(8)	96.12(12)
O(5)-Co(1)-O(4)	87.00(11)	O(4)-Co(1)-N(8)	174.47(12)
O(1)-Co(1)-O(4)	89.40(10)	N(7)-Co(1)-N(8)	77.31(13)
O(5)-Co(1)-N(7)	92.77(12)	O(2)-Co(1)-N(8)	91.51(12)
O(1)-Co(1)-N(7)	171.14(12)		
3			
Cu(1)-O(2)	1.912(3)	O(2)-Cu(1)-N(5)	168.07(12)
Cu(1)-O(1)	1.937(2)	O(1)-Cu(1)-N(5)	92.79(11)
Cu(1)-N(4)	2.001(3)	N(4)-Cu(1)-N(5)	81.03(12)
Cu(1)-N(5)	2.005(3)	O(2)-Cu(1)-O(9)	97.88(12)
Cu(1)-O(9)	2.280(3)	O(1)-Cu(1)-O(9)	95.85(11)
O(2)-Cu(1)-O(1)	92.84(11)	N(4)-Cu(1)-O(9)	91.61(12)
O(2)-Cu(1)-N(4)	91.97(12)	N(5)-Cu(1)-O(9)	91.99(12)
O(1)-Cu(1)-N(4)	170.49(11)		
4			
Co(1)-O(2)	2.069(4)	O(3)^{A}-Co(1)-O(3)	96.5(2)
Co(1)-O(2)^{A}	2.069(4)	O(2)-Co(1)-N(4)	97.75(18)
Co(1)-O(3)^{A}	2.095(4)	O(2)^{A}-Co(1)-N(4)	89.68(18)
Co(1)-O(3)	2.095(4)	O(3)^{A}-Co(1)-N(4)	169.60(19)
Co(1)-N(4)	2.124(5)	O(3)-Co(1)-N(4)	92.63(19)
Co(1)-N(4)^{A}	2.124(5)	O(2)-Co(1)-N(4)^{A}	89.68(18)
O(2)-Co(1)-O(2)^{A}	170.4(2)	O(2)^{A}-Co(1)-N(4)^{A}	97.75(18)
O(2)-Co(1)-O(3)^{A}	87.85(17)	O(3)^{A}-Co(1)-N(4)^{A}	92.63(18)
O(2)^{A}-Co(1)-O(3)^{A}	85.77(17)	O(3)-Co(1)-N(4)^{A}	169.60(19)
O(2)-Co(1)-O(3)	85.77(17)	N(4)-Co(1)-N(4)^{A}	78.7(3)
O(2)^{A}-Co(1)-O(3)	87.85(17)		

Symmetry transformations used to generate equivalent atoms:

For **1**, A) $-x+1, -y+2, -z+1$; B) $-x+1, -y+1, -z$. For **4**, A) $-x+1, y, -z+3/2$.

Table S2 Hydrogen bonds (Å and °) for complexes **1**–**4**.

D–H···A	*d* (H···A)	*d* (D···A)	< DHA
1			
N(7)-H(7)...O(16)^{C}	2.26	3.157(11)	167.4
N(1)-H(1)...O(13)^{D}	2.33	3.129(12)	145.8
O(16)-H(16B)...O(9)^{A}	2.20(8)	2.924(19)	143(12)
O(16)-H(16A)...O(3)^{A}	2.17(9)	2.876(11)	140(11)
O(15)-H(15A)...O(3)^{E}	1.99(10)	2.83(2)	166(34)
Symmetry code: A) $-x+1, -y+2, -z+1$; C) $-x+1, -y+1, -z+1$; D) $x, y+1, z$; E) $-x+2, -y+2, -z+1$			
2			
O(11)-H(11A)...O(3)^{A}	2.01(2)	2.843(5)	165(7)
O(14)-H(14A)...O(13)^{B}	2.26(4)	2.965(17)	141(6)
O(11)-H(11B)...N(6)^{C}	2.14(4)	2.893(8)	149(7)

O(12)-H(12C)...O(5)	1.953(16)	2.802(5)	172(7)
O(12)-H(12D)...O(11)	2.12(5)	2.826(7)	140(6)
N(3)-H(3A)...O(9)^{D}	2.12	3.008(10)	166.7
N(3)-H(3B)...O(3)^{D}	1.82	2.712(4)	168.6
Symmetry code: A) $x + 1, y, z$; B) $–x + 1, –y + 1, –z + 1$; C) $–x + 1, –y + 1, –z$; D) $x, y + 1, z$			
3			
O(9)-H(9A)...O(10)^{A}	1.92(3)	2.708(5)	154(5)
O(9)-H(9B)...O(11)	1.905(13)	2.751(6)	172(5)
O(10)-H(10A)...O(5)^{A}	2.35(4)	3.021(7)	136(5)
O(11)-H(11F)...O(3)^{B}	2.17(4)	2.820(6)	133(5)
Symmetry code: A) $–x + 1, –y + 1, –z + 1$; B) $–x, –y + 1, –z + 1$.			

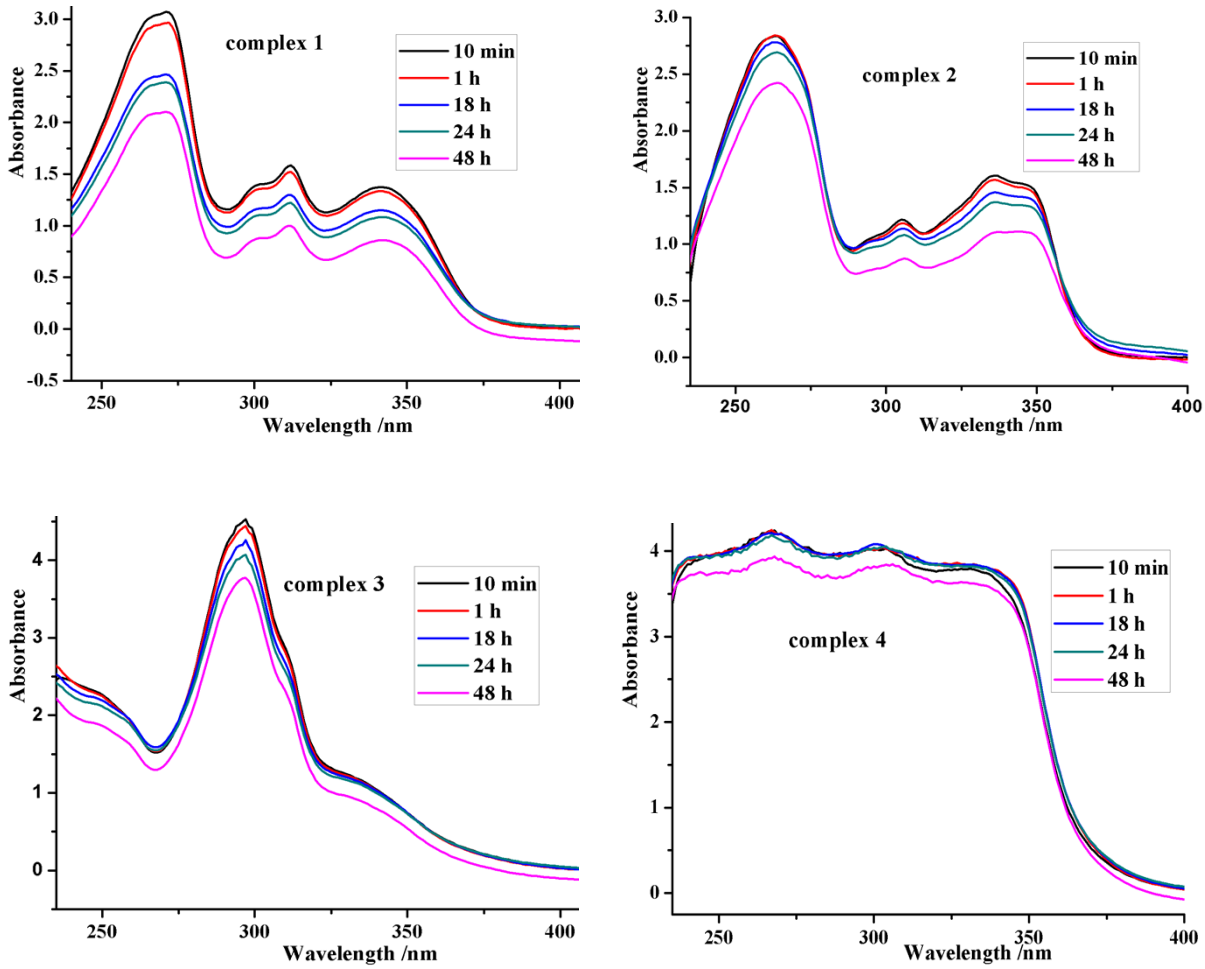


Fig. S1. Time-dependent stability studies on complexes **1-4** in TBS buffer solution monitored by UV-vis absorption spectra.

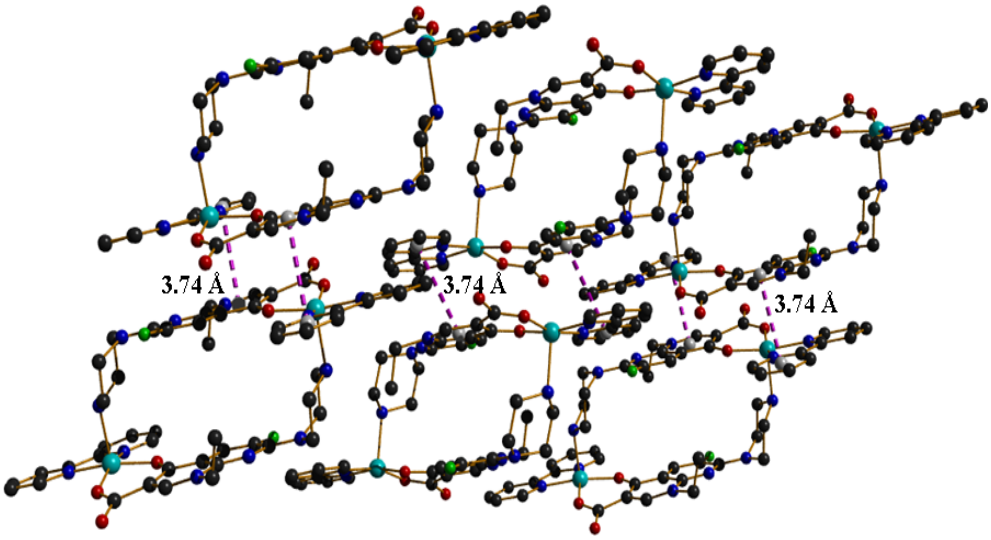


Fig. S2. π–π stacking (purple dashed lines) between the aromatic planes of the HEn ligand and bipyridine of complex **1**.

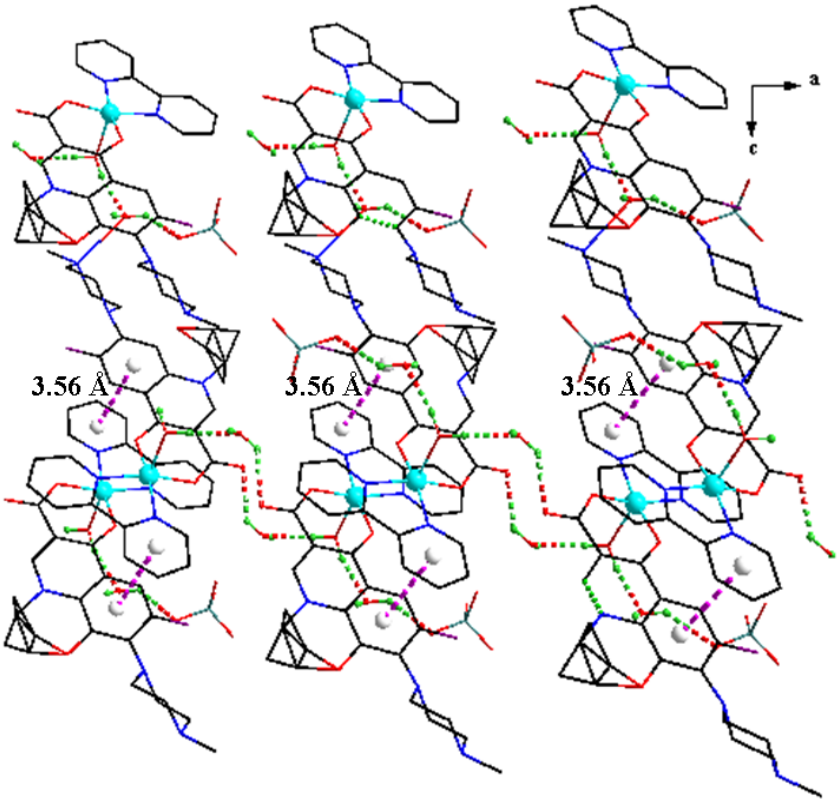
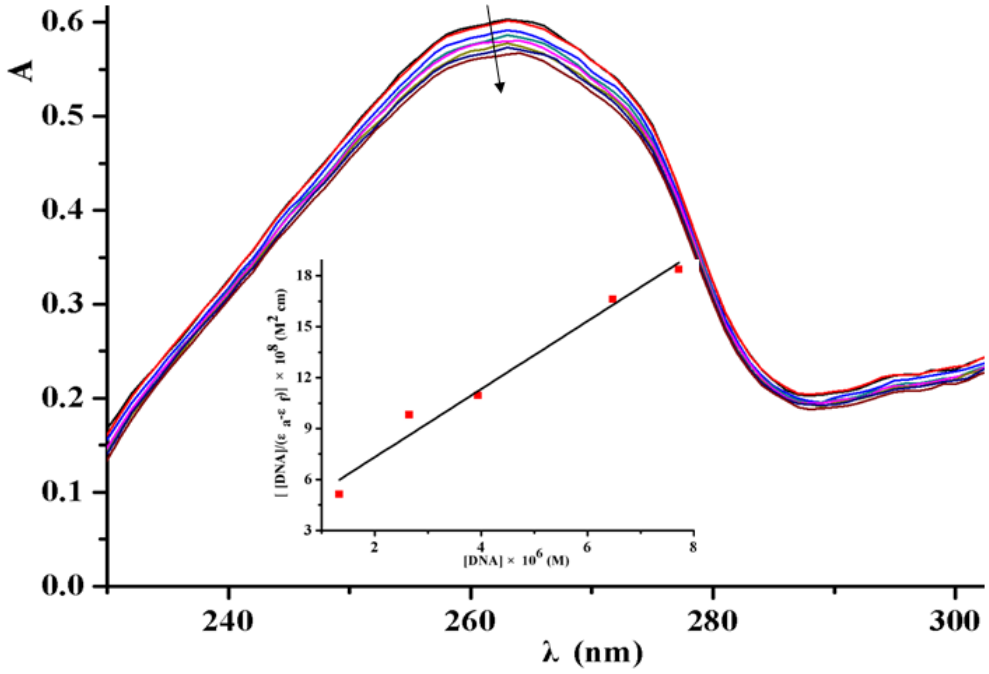
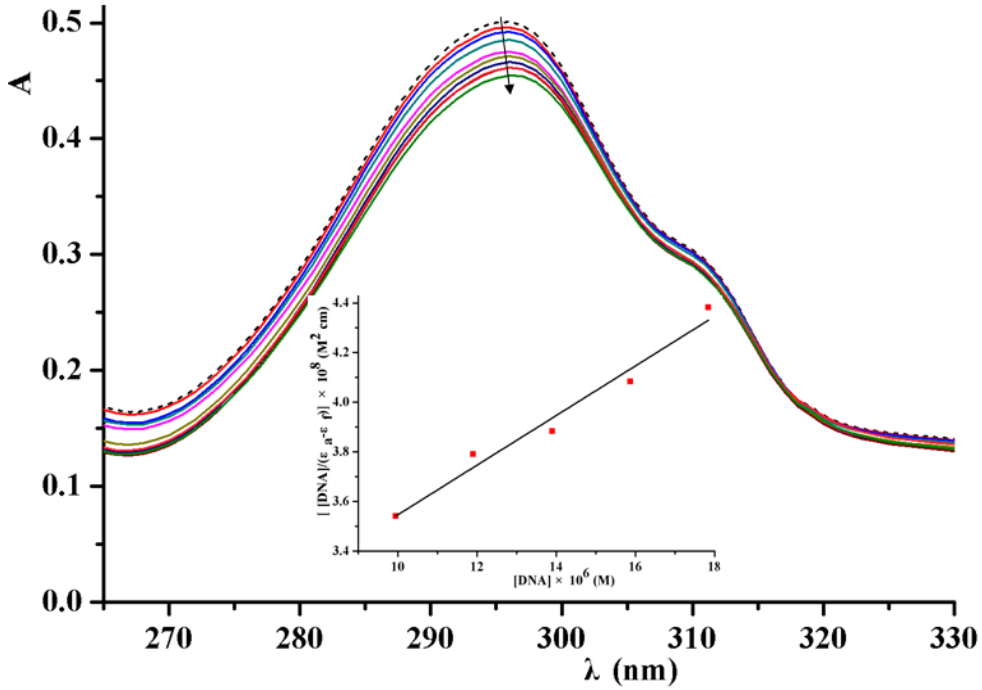


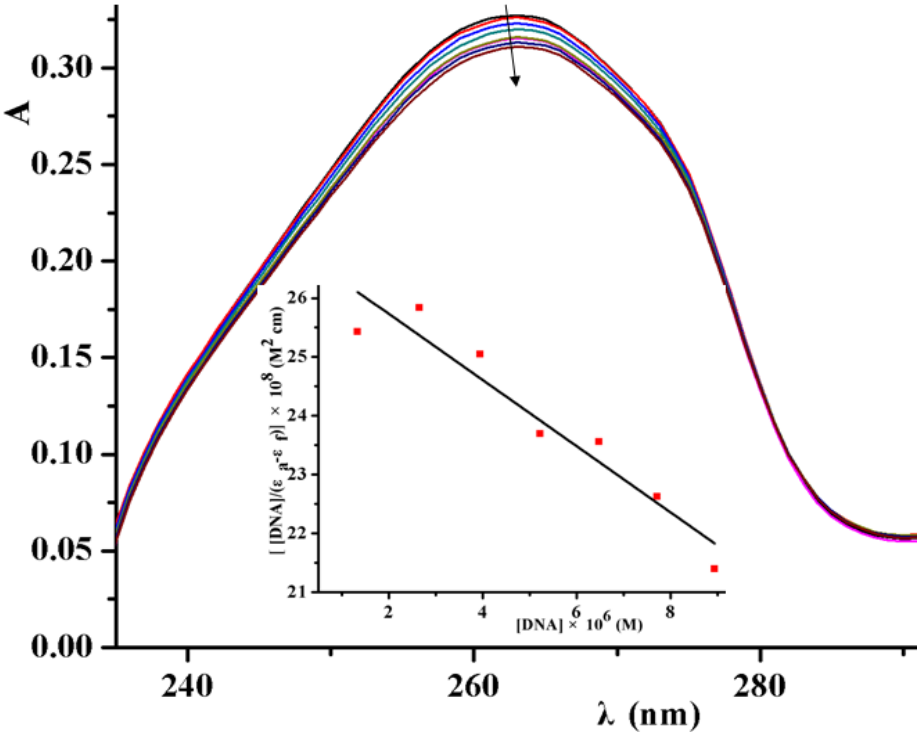
Fig. S3. 2D supramolecular network of **3** constructed by hydrogen bonding (green dashed lines) and π–π stacking interactions (purple dashed lines) in the *ac* plane.


A
λ (nm)
[DNA] × 10^6 (M)
[[DNA]/(ε_a-ε_f)] × 10^8 (M^2 cm)

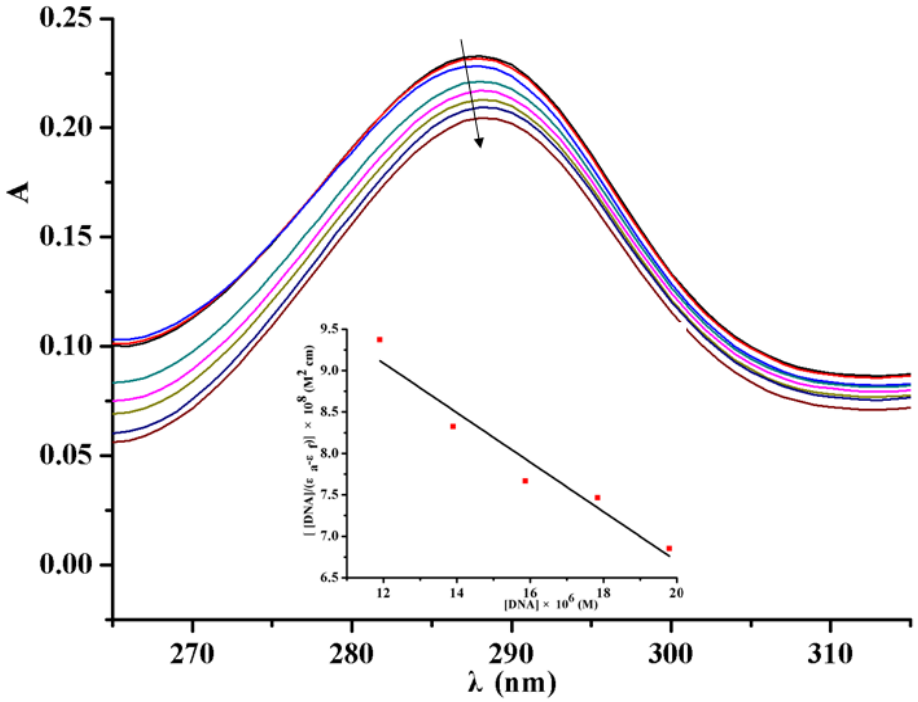
(A)


A
λ (nm)
[DNA] × 10^6 (M)
[[DNA]/(ε_a-ε_f)] × 10^8 (M^2 cm)

(B)

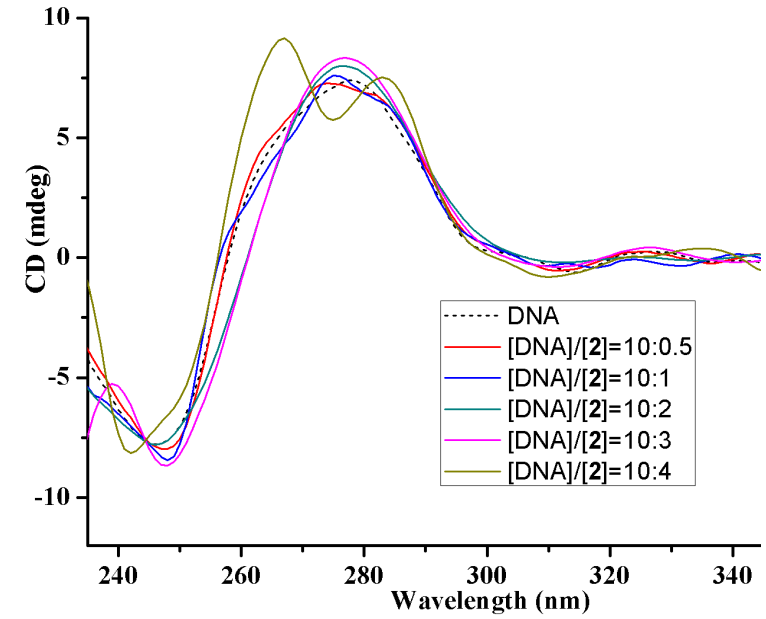


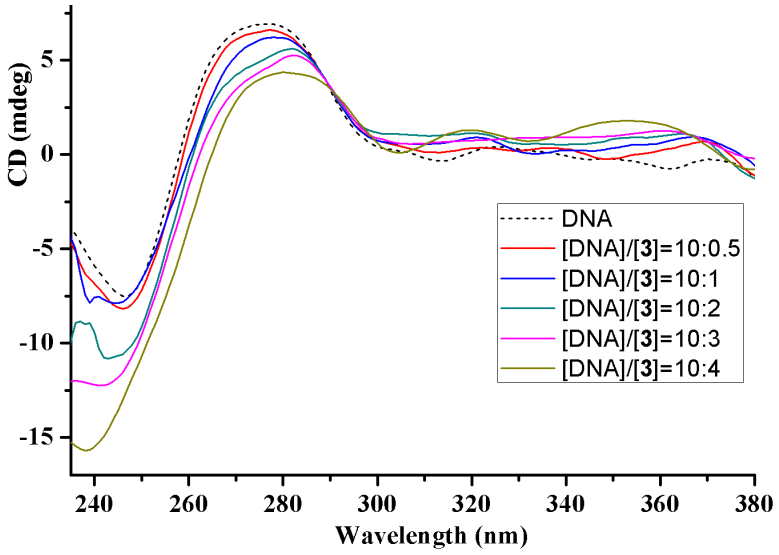
(C)

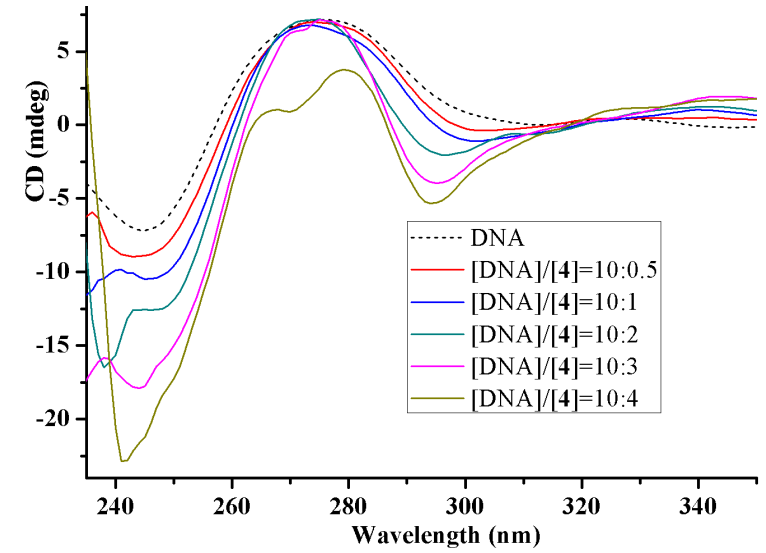


(D)

Fig. S4. UV absorption spectra of **2** (A), **3** (B), HEn (C) and HLevo (D) in the absence (---) and presence (—) of CT DNA with increasing [DNA]/[compound] ratios. The arrows show the changes upon increasing amounts of CT DNA. Inset: plot of [DNA]/(ε_a-ε_f) versus [DNA].


CD (mdeg)
Wavelength (nm)
DNA
[DNA]/[2]=10:0.5
[DNA]/[2]=10:1
[DNA]/[2]=10:2
[DNA]/[2]=10:3
[DNA]/[2]=10:4


CD (mdeg)
Wavelength (nm)
DNA
[DNA]/[3]=10:0.5
[DNA]/[3]=10:1
[DNA]/[3]=10:2
[DNA]/[3]=10:3
[DNA]/[3]=10:4


CD (mdeg)
Wavelength (nm)
DNA
[DNA]/[4]=10:0.5
[DNA]/[4]=10:1
[DNA]/[4]=10:2
[DNA]/[4]=10:3
[DNA]/[4]=10:4

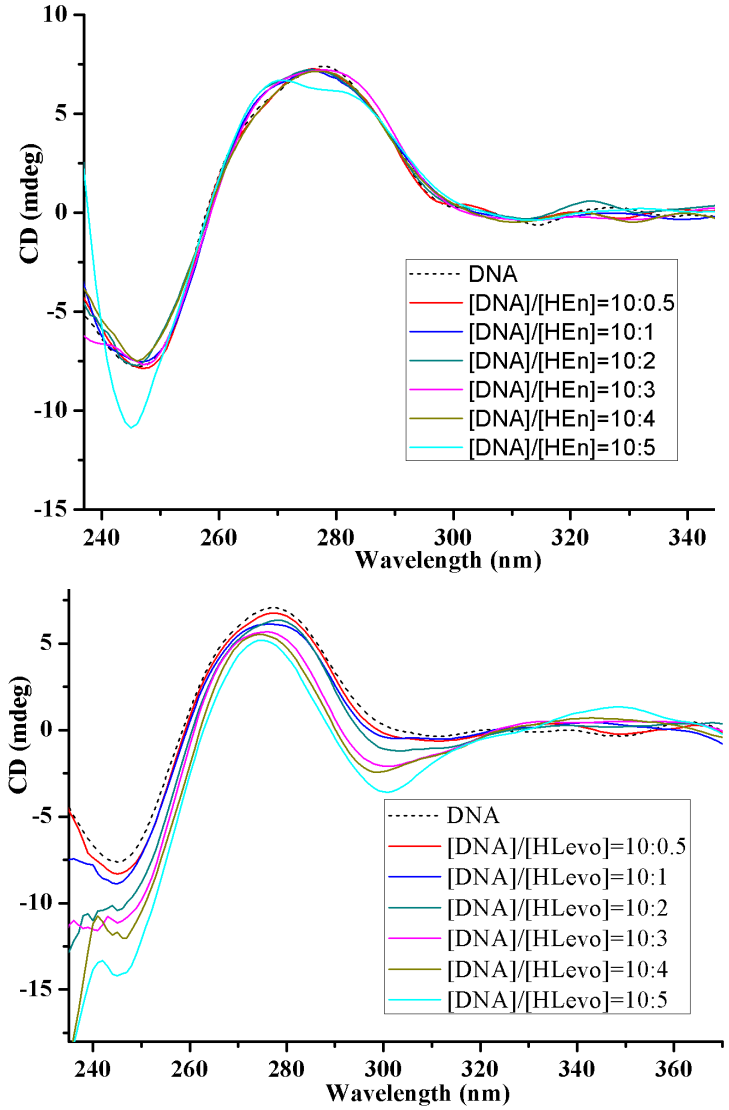
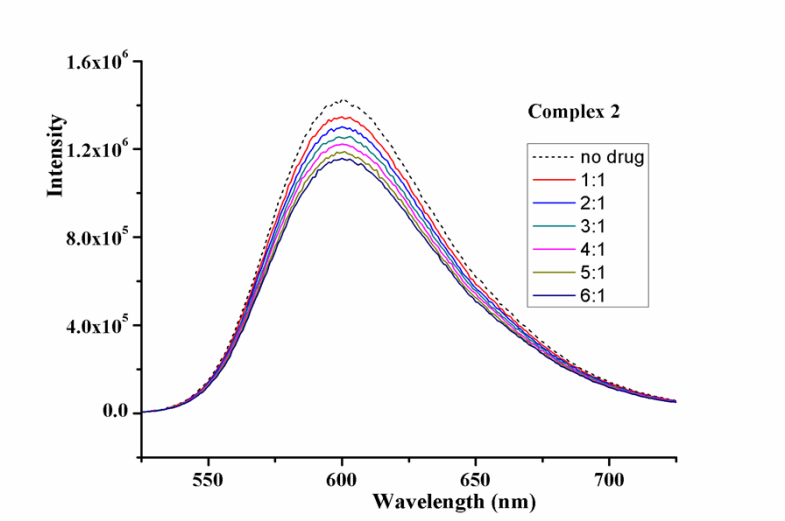
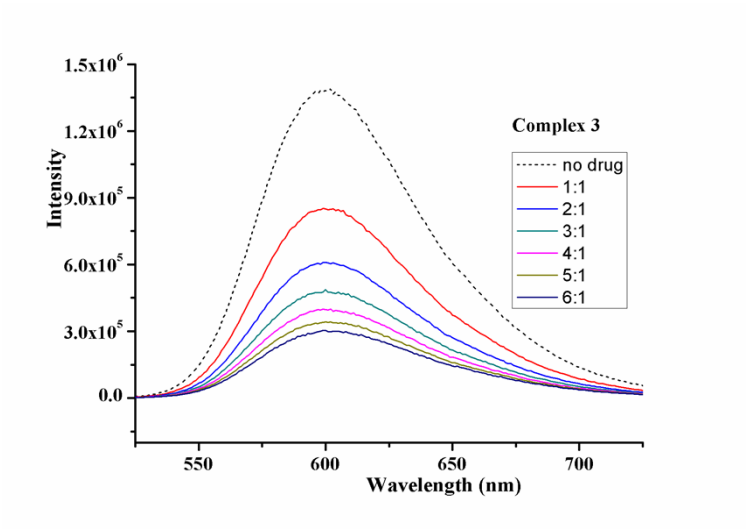
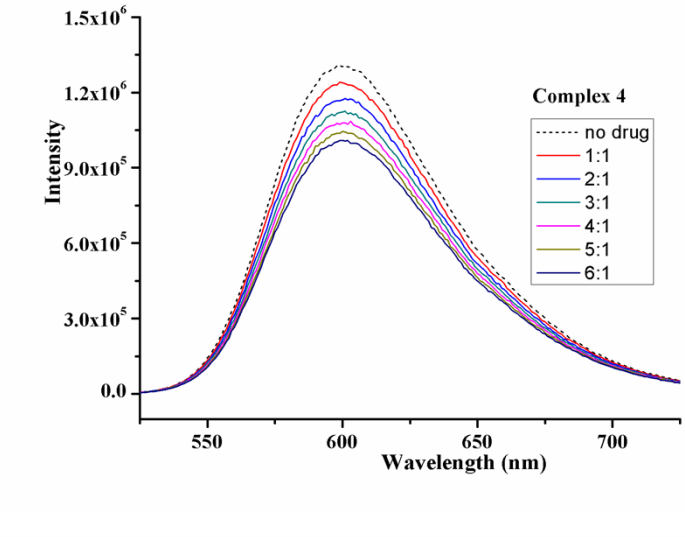
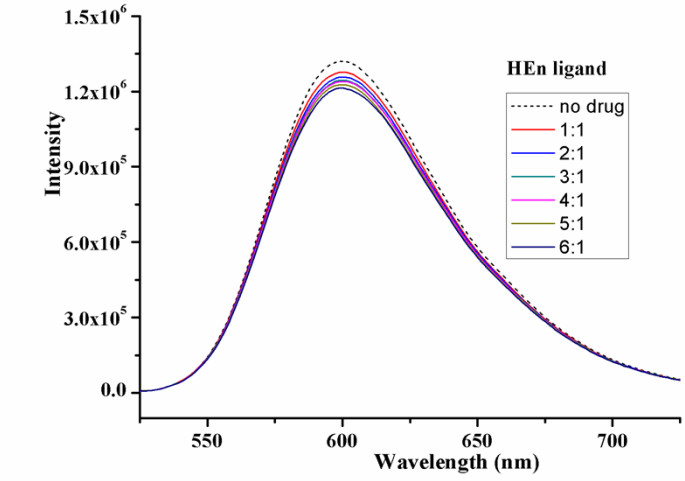


Fig. S5. Circular dichromism spectra of CT DNA bound with HEn, Hlevo and complexes **2**-**4** with [DNA] = 1×10^{-4} M.




Complex 3
no drug
1:1
2:1
3:1
4:1
5:1
6:1
Intensity
1.5x10^6
1.2x10^6
9.0x10^5
6.0x10^5
3.0x10^5
0.0
550
600
650
700
Wavelength (nm)


Complex 4
no drug
1:1
2:1
3:1
4:1
5:1
6:1
Intensity
1.5x10^6
1.2x10^6
9.0x10^5
6.0x10^5
3.0x10^5
0.0
550
600
650
700
Wavelength (nm)


HEn ligand
no drug
1:1
2:1
3:1
4:1
5:1
6:1
Intensity
1.5x10^6
1.2x10^6
9.0x10^5
6.0x10^5
3.0x10^5
0.0
550
600
650
700
Wavelength (nm)

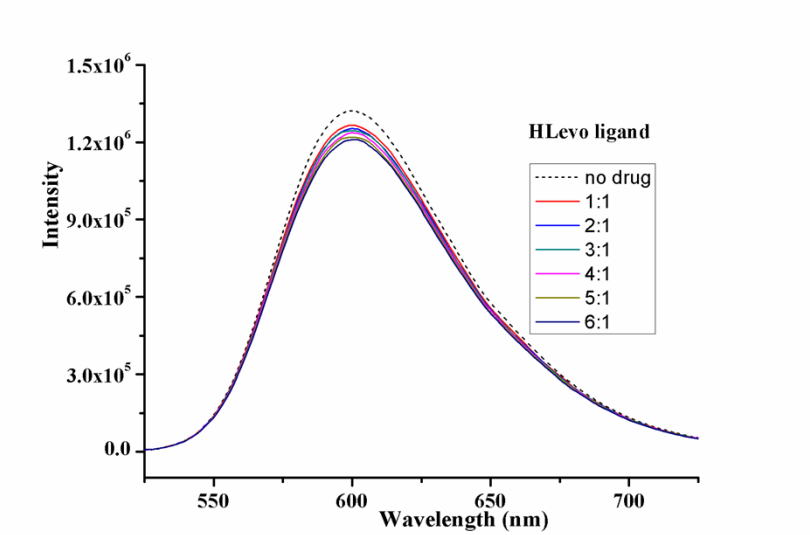


Fig. S6. Fluorescence emission spectra of EB-DNA in the absence (dashed line) and presence (colored solid lines) of HEn, HLevo and complexes **2-4** as competitive agents with increasing [compound]/[EB] ratios from 1:1 to 6:1.

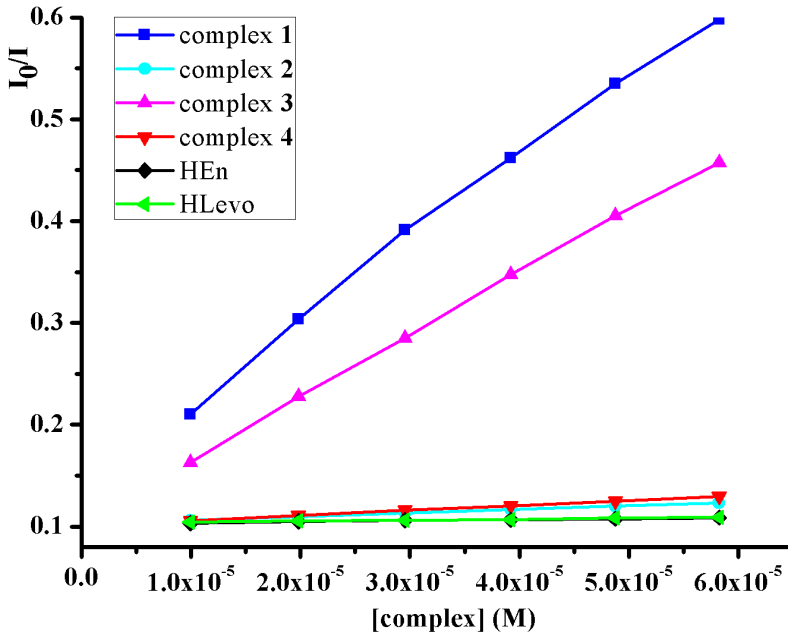
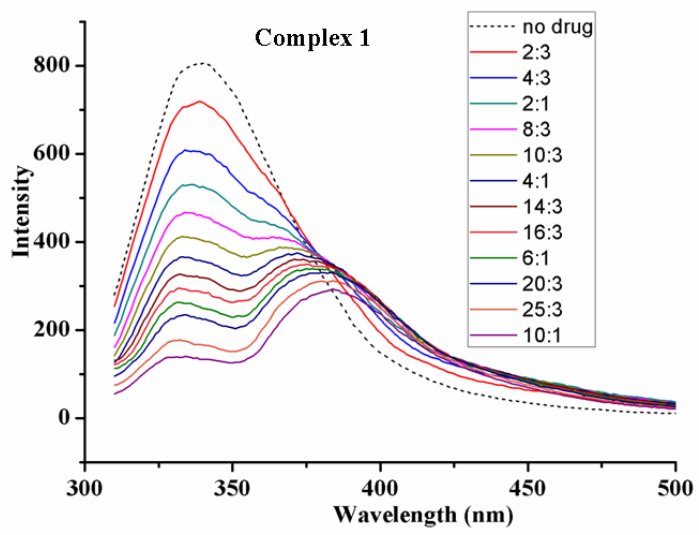
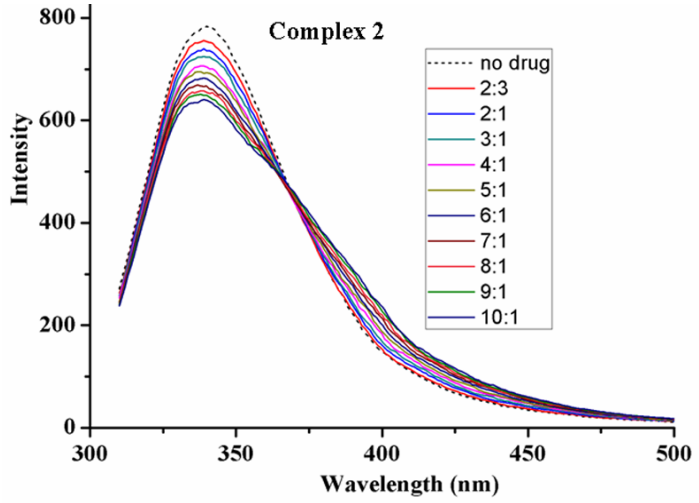
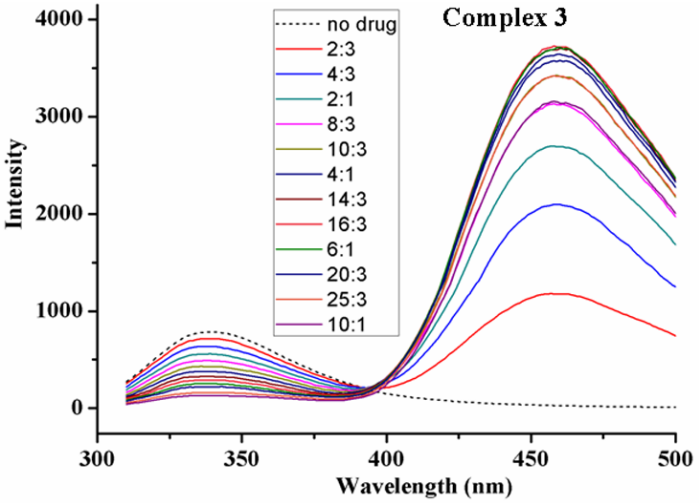


Fig. S7. Stern-Volmer quenching plot of EB bound to CT-DNA by complexes **1-4** and HEn, HLevo.


Complex 1
no drug
2:3
4:3
2:1
8:3
10:3
4:1
14:3
16:3
6:1
20:3
25:3
10:1
Intensity
Wavelength (nm)
0
200
400
600
800
300
350
400
450
500


Complex 2
no drug
2:3
2:1
3:1
4:1
5:1
6:1
7:1
8:1
9:1
10:1
Intensity
Wavelength (nm)
0
200
400
600
800
300
350
400
450
500


Complex 3
no drug
2:3
4:3
2:1
8:3
10:3
4:1
14:3
16:3
6:1
20:3
25:3
10:1
Intensity
Wavelength (nm)
0
1000
2000
3000
4000
300
350
400
450
500

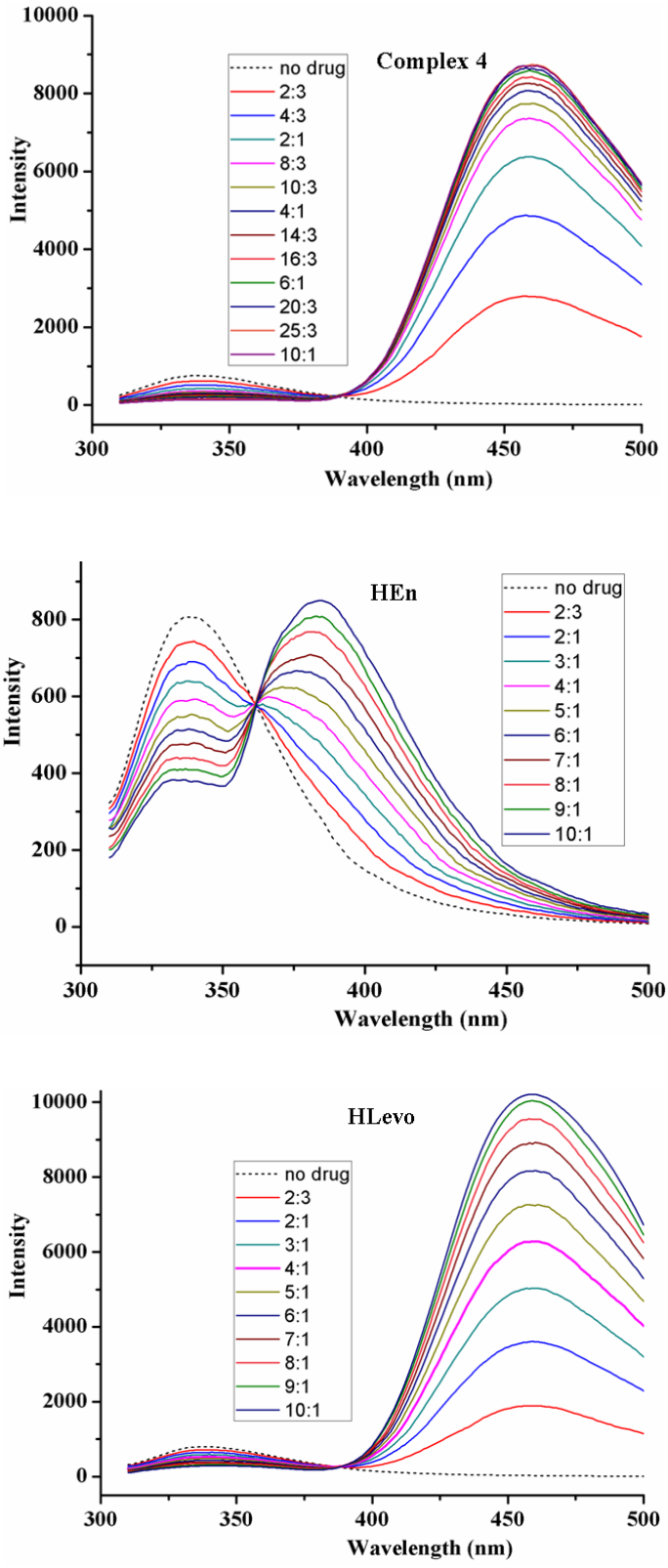
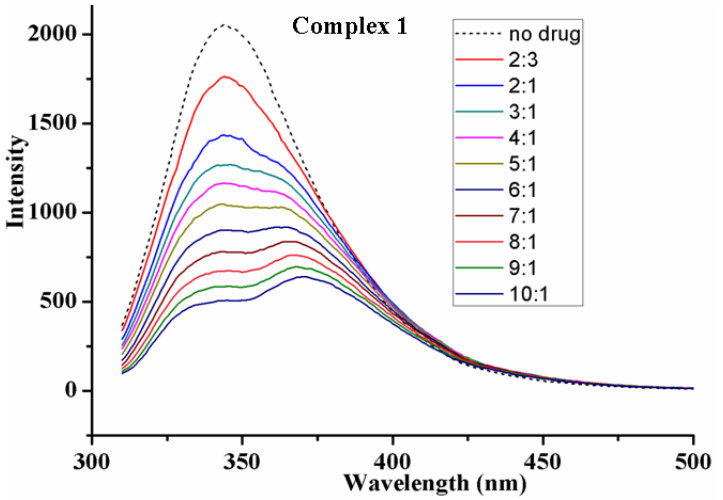
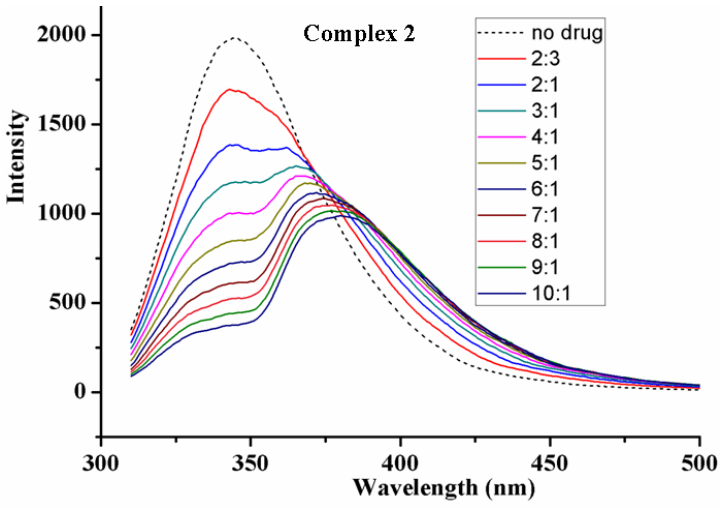
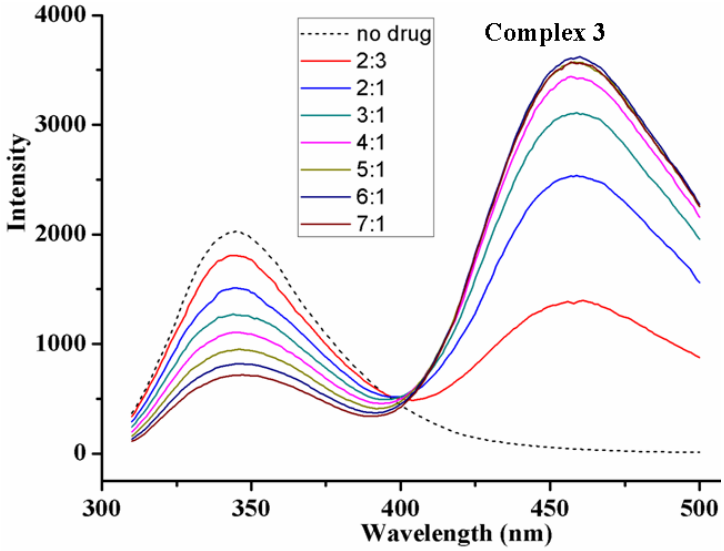


Fig. S8. Emission spectra (λ_{exit} = 295 nm) for HSA ([HSA] = 3 μM) in buffer solution in the absence (dashed line) and presence (colored solid lines) of increasing amounts of compound (r = [compound] / [HSA] = 0-10).


Complex 1
no drug
2:3
2:1
3:1
4:1
5:1
6:1
7:1
8:1
9:1
10:1
Intensity
Wavelength (nm)
0
500
1000
1500
2000
300
350
400
450
500


Complex 2
no drug
2:3
2:1
3:1
4:1
5:1
6:1
7:1
8:1
9:1
10:1
Intensity
Wavelength (nm)
0
500
1000
1500
2000
300
350
400
450
500


Complex 3
no drug
2:3
2:1
3:1
4:1
5:1
6:1
7:1
Intensity
Wavelength (nm)
0
1000
2000
3000
4000
300
350
400
450
500

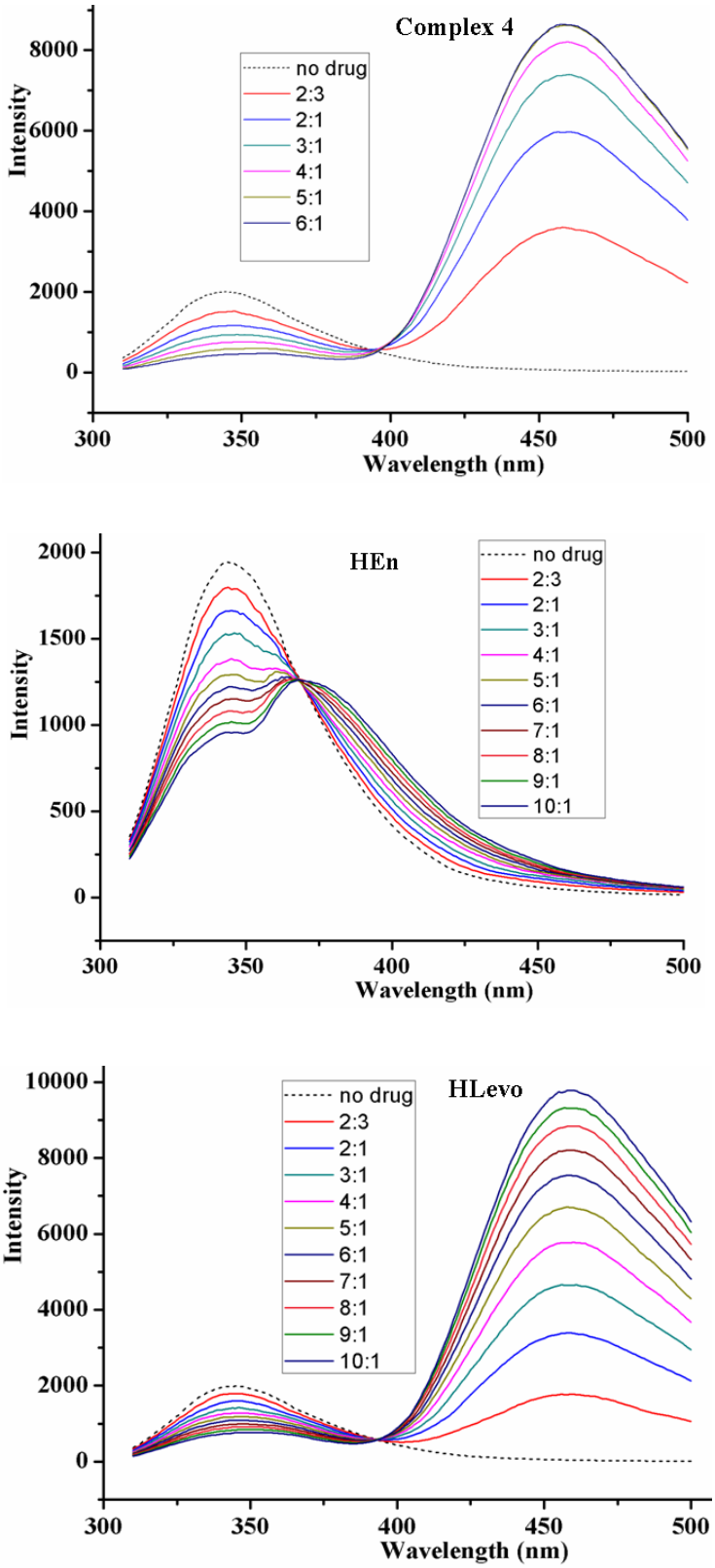


Fig. S9. Emission spectra (λ_{exit} = 295 nm) for BSA ([BSA] = 3 μM) in buffer solution in the absence (dashed line) and presence (colored solid lines) of increasing amounts of compound (r = [compound] / [BSA] = 0-10, except r = 0-7 for **3** and r = 0-6 for **4**).

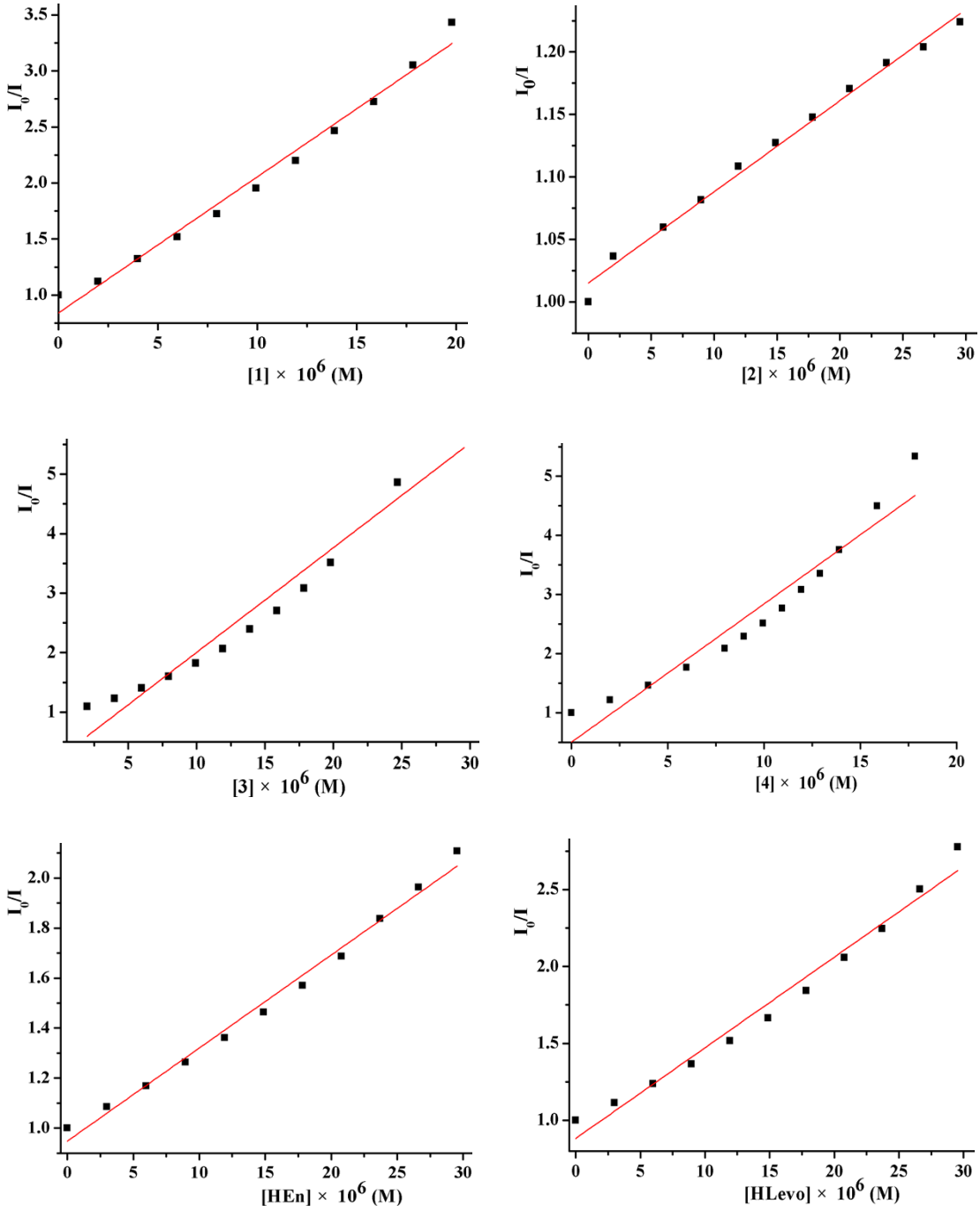


Fig. S10. Stern-Volmer quenching plot of HSA for complexes **1-4** and HEn, HLevo ligand, respectively.

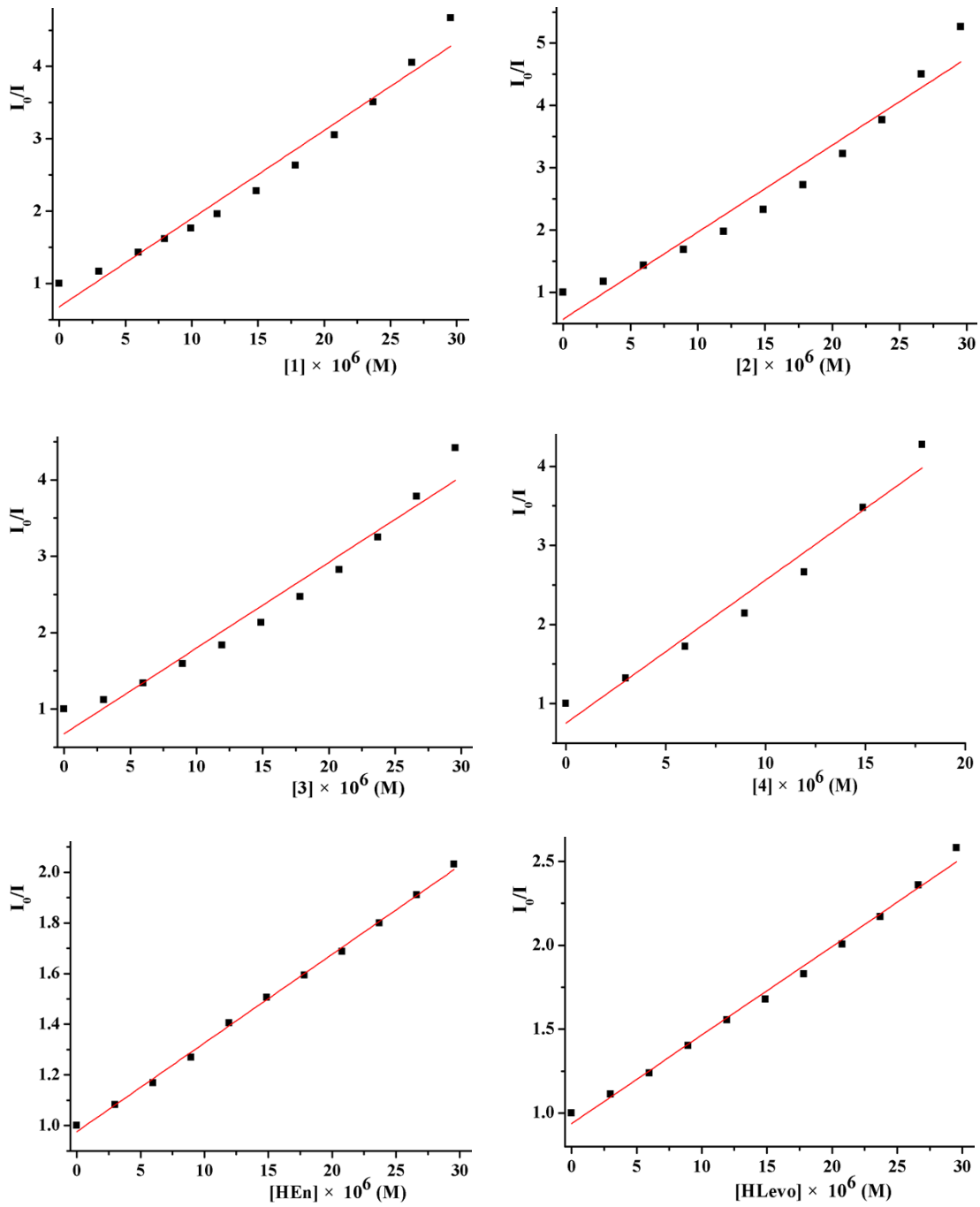


Fig. S11. Stern-Volmer quenching plot of BSA for complexes **1**-**4** and HEn, HLevo ligand, respectively.

Equations:

$$\frac{[DNA]}{(\varepsilon_a - \varepsilon_f)} = \frac{[DNA]}{(\varepsilon_b - \varepsilon_f)} + \frac{1}{K_b(\varepsilon_b - \varepsilon_f)} \qquad \text{Eq. (S1)}$$

where [DNA] is the concentration of DNA in base pairs, ε_f is the extinction coefficient for the free compound at the corresponding λ_{max}, $\varepsilon_a = A_{obsd}/[compound]$ and ε_b is the extinction

coefficient for the compound in the fully bound form.

$$\frac{I_0}{I} = 1 + K_{sv}[Q] \qquad \text{Eq. (S2)}$$

where I_0 and I are the emission intensities in the absence and the presence of the complex, respectively.

$$\frac{I_0}{I} = 1 + k_q \tau_0 [Q] = 1 + K_{sv}[Q] \qquad \text{Eq. (S3)}$$

where Io = the initial tryptophan fluorescence intensity of SA, I = the tryptophan fluorescence intensity of SA after the addition of the quencher, k_q = the quenching rate constants of SA, K_{sv} = the dynamic quenching constant, τ_o = the average lifetime of SA without the quencher, [Q] = the concentration of the quencher respectively, $K_{sv} = k_q \tau_o$ (taking as fluorescence lifetime (τo) of tryptophan in SA at around 10^{-8} s).

$$\frac{\Delta I / I_0}{[Q]} = nK - K\frac{\Delta I}{I_0} \qquad \text{Eq. (S4)}$$

Where n is the number of binding sites per albumin and K (M^{-1}) is the association binding constant, K (M^{-1}) may be calculated from the slope in plots $\Delta I/(I_0[Q])$ versus $\Delta I/I_0$ and n is given by the ratio of y intercept to the slope.