Supporting Information

Materials and Methods: Tris-hydroxymethylaminomethane (Tris) was purchased from USB. All the other reagents used in the experiments were purchased from Sigma. All the DNA oligonucleotides were purchased from Sangon Biotech (ShangHai, China) and purified by HPLC. The concentration of each DNA sequence was estimated by absorption at 260 nm. The molar extinction coefficients were estimated by the nearest neighbor method. All DNA oligonucleotides used here are listed in Table S1.

Electrophoresis: The native PAGE experiments were carried out on 15% polyacrylamide (19:1 acrylamide/bisacrylamide ratio) gel and run for 2.5 h with a field of 80 V at 4 ^{o}C. The tris-acetic (TB) running buffer consisted of Tris-HCl (50 m M), pH 7.0 and pH 5.0, boric acid (20 mM). Gels were stained with stains-all.

Fluorescence Spectra: Fluorescence measurements were carried out on Jasco-FP-6500 spectrofluorometer (Jasco International Co. LTD. Tokyo, Japan) using a quartz cell of 1 cm path length, at an excitation wavelength of 480 nm (doxorubicin) or 346 nm (hoechst 33258). Fluorescence emission spectra were monitored from 500 to 700 nm and from 400 to 650 nm, respectively. The slits for the excitation and emission monochromator were both set to 5 nm.

Absorbance and UV melting: Absorbance measurements and melting experiments

were carried out on a Cary 300 UV/vis spectrophotometer equipped with a Peltier temperature control accessory. All UV/vis spectra were measured in 1.0-cm-path-length cell .Absorbance changes at 260 nm versus temperature were collected[1] eating rate of 1 $^{o}C \cdot min^{-1}$.

CD spectra : CD spectra were measured on a JASCO J-810 spectropolarimeter equipped with a temperature controlled water bath[1] optical chamber of CD spectrometer was deoxygenated with dry purified nitrogen (99.99%) for 45 min before use and kept the nitrogen atmosphere during experiments. Three scans were accumulated and automatically averaged.

Determination of Binding Constants:[2] Binding constants were measured by fluorescence titration methods, in which fixed concentrations of either doxorubicin or hoechst 33258 titrated with increasing DNA concentrations.

According to the 1 : n binding mode, the binding reaction can be described as:

$$P + nM = PM_n \quad (1)$$

Where P denotes small drug molecules, M denotes DNA, and PM denotes drug molecules /DNA conjugates.

So, the binding constant can be described as:

$$K_b=[PM_n]/[P][M]^n \quad (2)$$

With

$$[P] =[P_0] -[PM_n] \quad (3)$$

$$[M]= [M_0]-n[PM_n] \quad (4)$$

Where $[P_0]$ is the concentration of small drug molecules and $[M_0]$ is the concentration of DNA.

So,

$$K_b=[PM_n]/\{ [P_0] -[PM_n] \}\{[M_0]-n[PM_n]\}^n \quad (5)$$

The formation of drug molecules/DNA conjugates PM_n can be quantitated by the SPR signal that satisfies the following equation:

$$F=F_0+(F_\infty-F_0)[PM_n]/[P_0] \quad (6)$$

So

$$K_b=\{\Delta F\times[P_0]/ \Delta F_{max}\} /\{ [P_0] -\Delta F\times[P_0]/ \Delta F_{max} \}\{[M_0]-n\Delta F\times[P_0]/ \Delta F_{max} \}^n \quad (7)$$

Where $\Delta F= F - F_0$, $\Delta F_{max}= F_{max}- F_0$

The free energy change ($\triangle G$) was calculated from the equation:

$\triangle G_b = - RTlnK_b$

Name	Sequence
gcDNA	CGC CCC TAA CCC TAA CCC TAA CCC TGC G
atDNA	AAA CCC TAA CCC TAA CCC TAA CCC TTT T
dT_{12}	TTT TTT TTT TTT
dT_{22}	TTT TTT TTT TTT TTT TTT TTT T

Table S1. The nucleic acid name and sequences.

DNA	Drug	K_b	ΔG_{4oC} (kJ/mol)
gcDNA	Doxorubicin	3.73×10^5	-29.54
atDNA	Hoechst 33258	2.32×10^4	-23.15

Table S2. Summary of Thermodynamic Parameters for drugs binding to DNA in 100 mM NaCl, 1 mM cacodylic buffer (pH 7.0) at 4 ^oC.

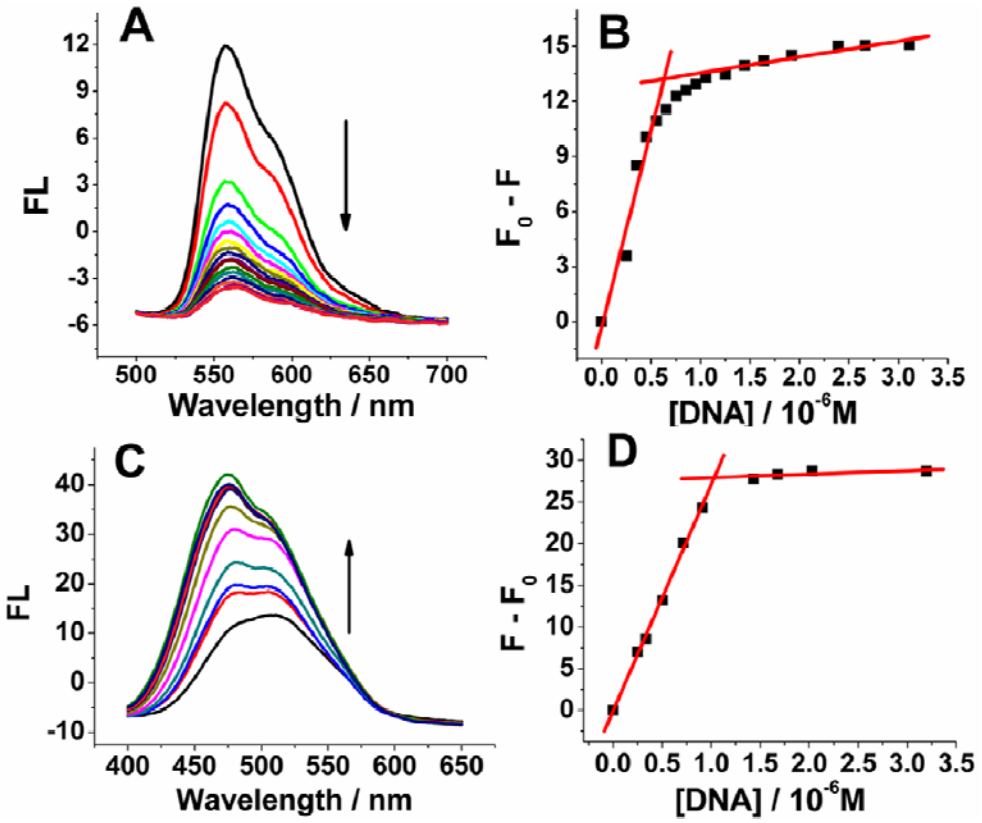

Fig. S1 (A) Fluorescence titration of doxorubicin with gcDNA in 100 mM NaCl, 1 mM cacodylic buffer (pH 7.0) at 4 ^{o}C. The concentration of gcDNA was varied from 0 μM to 3 μM. λ_{ex} = 480 nm and λ_{em}=560 nm. (B) The change in Fluorescence intensity at 560 nm with increased concentration of gcDNA derived from the fluorescence titration. Doxorubicin concentration was 2 μM. A breakpoint was observed at 0.67 μM gcDNA (3:1 ratio of [doxorubicin] / [gcDNA]). C) Fluorescence titration of hoechst 33258 with atDNA in 100 mM NaCl, 1 mM cacodylic buffer (pH 7.0) at 4 ^{o}C. The concentration of atDNA was varied from 0 μM to 3 μM. λ_{ex} = 346 and λ_{em} = 475 nm (with atDNA), λ_{em} = 525 nm (without atDNA). (D) The change in Fluorescence intensity at 475 nm with increased concentration of atDNA derived from the Fluorescence titration. A breakpoint was observed at 1 μM atDNA (2:1 ratio of [hoechst 33258] / [atDNA] . Hoechst 33258 concentration was 2 μM.

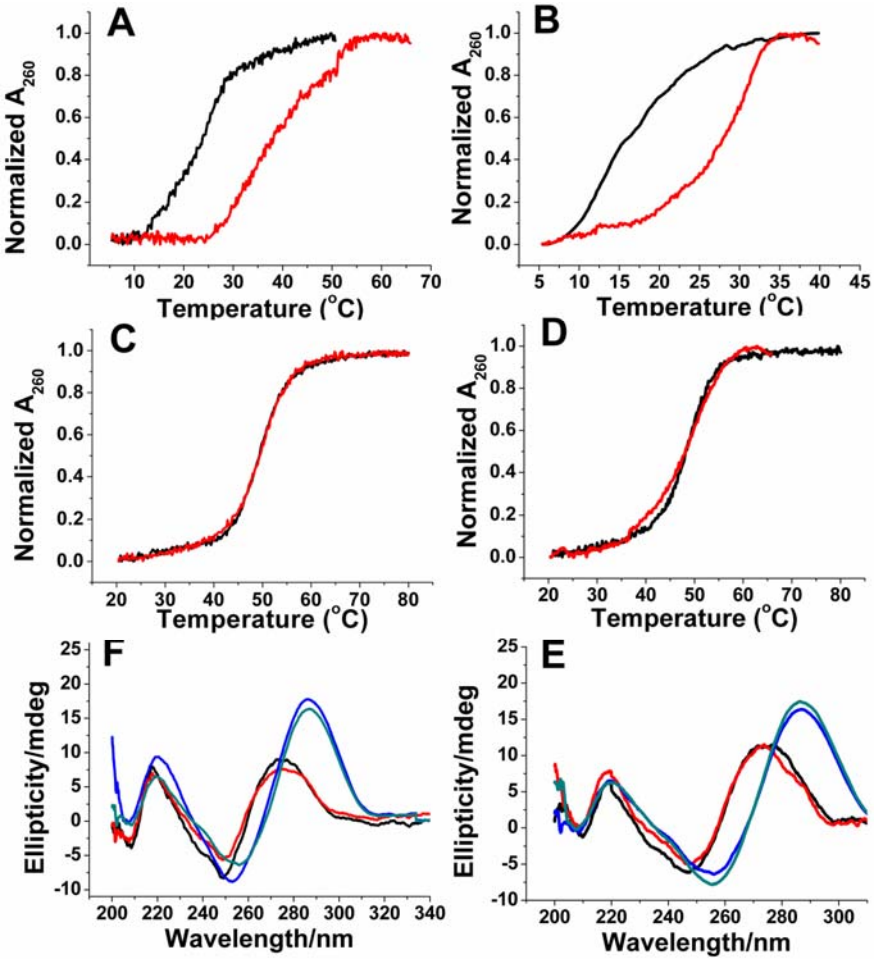

Fig. S2 DNA UV meltings and CD spectral changes at different pH (1 mM cacodylic acid/sodium cacodylate/100 mM NaCl). (A) UV melting profiles of gcDNA at pH 7.0 in the absence (black line) or presence (red line) of doxorubicin. (B) UV melting profiles of atDNA at pH 7.0 in the absence (black line) or presence (red line) of hoechst 33258. (C) UV melting profiles of gcDNA at pH 5.0 in the absence (black line) or presence (red line) of doxorubicin. (D) UV melting profiles of atDNA at pH 5.0 in the absence (black line) or presence (red line) of hoechst 33258. (E) CD spectral of gcDNA in the absence or presence of doxorubicin at different solution environment. (F) CD spectral of atDNA in the absence or presence of hoechst 33258 at different solution environment. DNA concentration was 1 μM (UV meltings) or 2 μM (CD spectral) in strand.

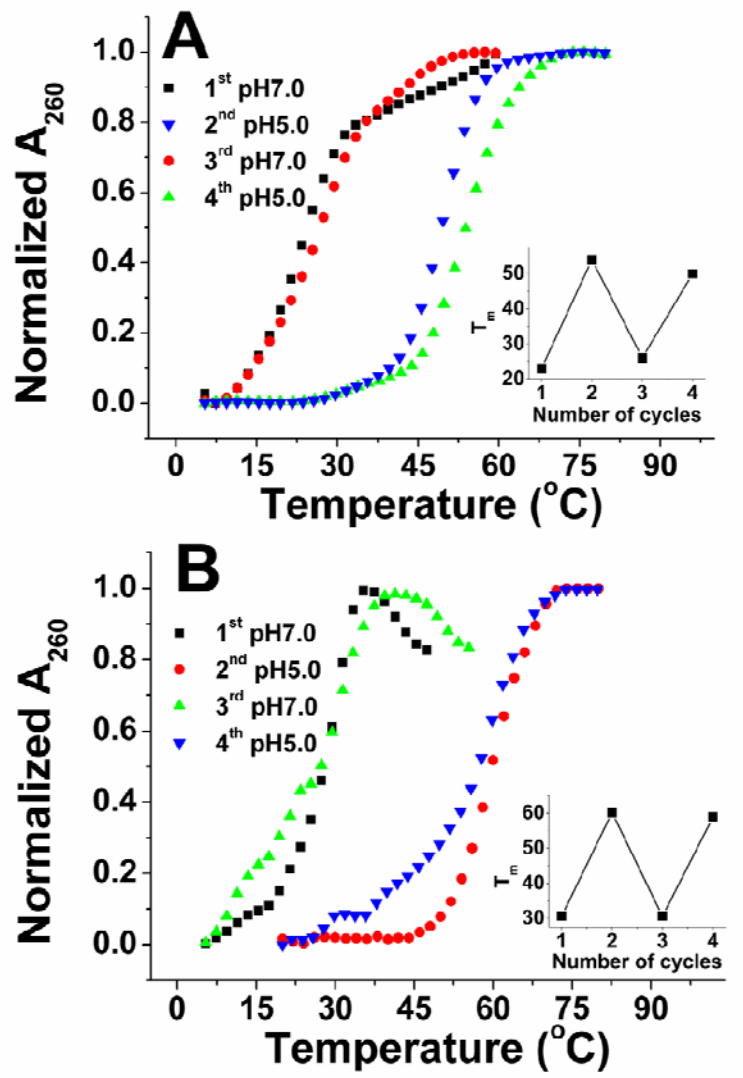

Fig. S3 Switches between drug binding and release monitored by UV melting study while the solution pH value oscillated between 5.0 and 7.0 (1 mM cacodylic acid/sodium cacodylate/100 mM NaCl). A) UV melting curves of gcDNA-doxorubicin while the solution pH value oscillated between 5.0 and 7.0. (inset) Changes of Tm of gcDNA-doxorubicin system while the solution pH value oscillated between 5.0 and 7.0. B) UV melting curves of atDNA-hoechst 33258 while the solution pH value oscillated between 5.0 and 7.0. (inset) Changes of T_m of atDNA-hoechst 33258 system while the solution pH value oscillated between 5.0 and 7.0.

References

1. (a) C. Chen, F. Pu, Z. Huang, Z. Liu, J. Ren and X. Qu, *Nucleic Acids Res.*, 2011, **39**, 1638; (b) X. Qu, J. O. Trent, I. Fokt, W. Priebe and J. B. Chaires, *Proc. Natl. Acad. Sci. USA*, 2000, **97**, 12032

2. (a) X. Qu and J. B. Chaires, *Method Enzymol.*, 2000, **321**, 353; (b) H. J. Yu J. Ren and X. Qu, *Chembiochem*, 2008, **9**, 879; (c) W. Garzon-Rodriguez, A. K. Yatsimirsky and C. G. Glabe, *Bioorg. Med. Chem. Lett.*, 1999, **9**, 2243.