Supporting Information

Label-free G-quadruplex aptamer fluorescent aptasensor for visual and real-time kanamycin detection in the lake and human samples

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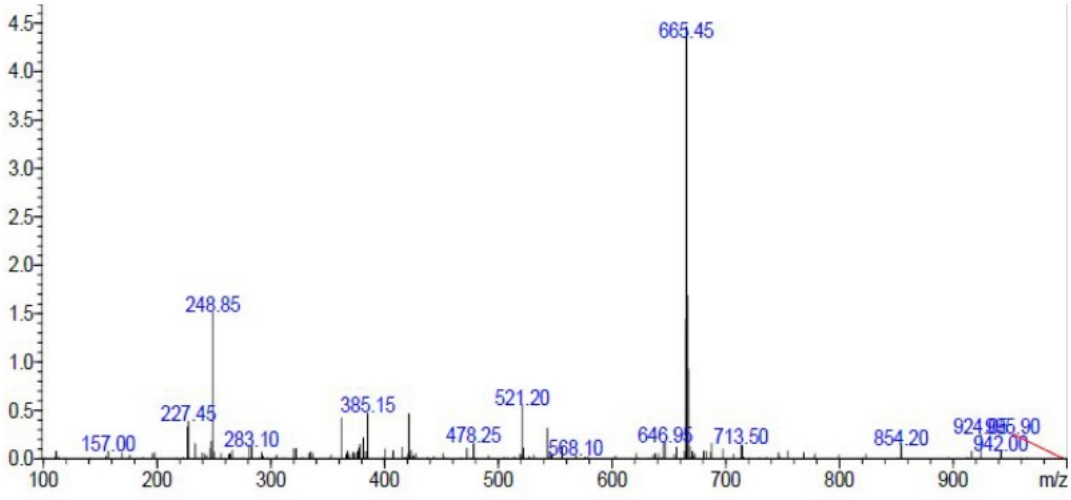

Fig. S1. MS-ESI spectrum of CyT.

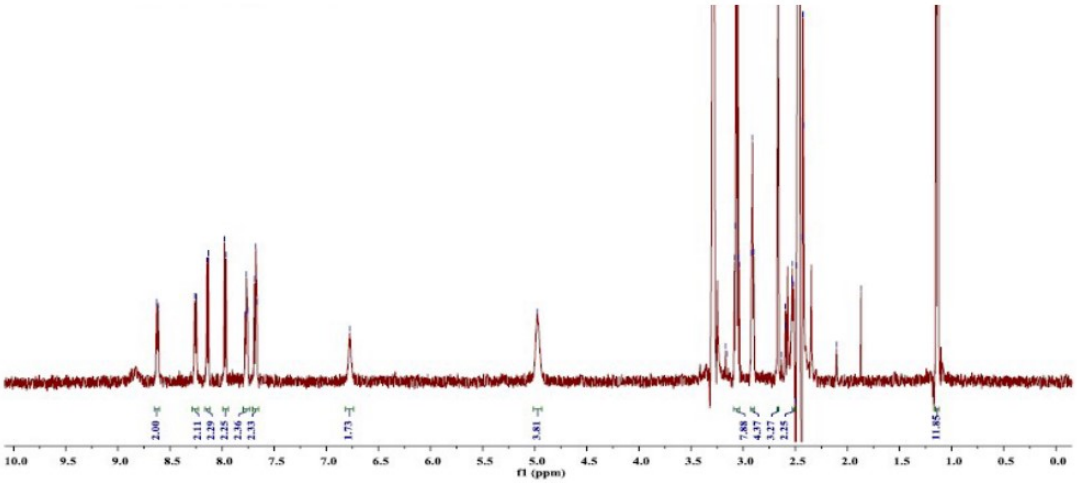

Fig. S2. ^{1}H-NMR spectrum of cyanine dye CyT in DMSO-d_6. δ^{1}H: 8.63−8.61 (d, 2H),8.26−8.25 (d, 2H), 8.15−8.13 (d, 2H), 7.98−7.96 (d, 2H), 7.77 (t, 2H), 7.68 (t, 2H), 6.78 (s, 2H),4.97 (s, 2H), 3.06 (m, 8H), 2.91 (t, 4H), 2.67 (s, 3H, CH_3), 2.51 (s, 2H), 1.14 (s, 12H) ppm.

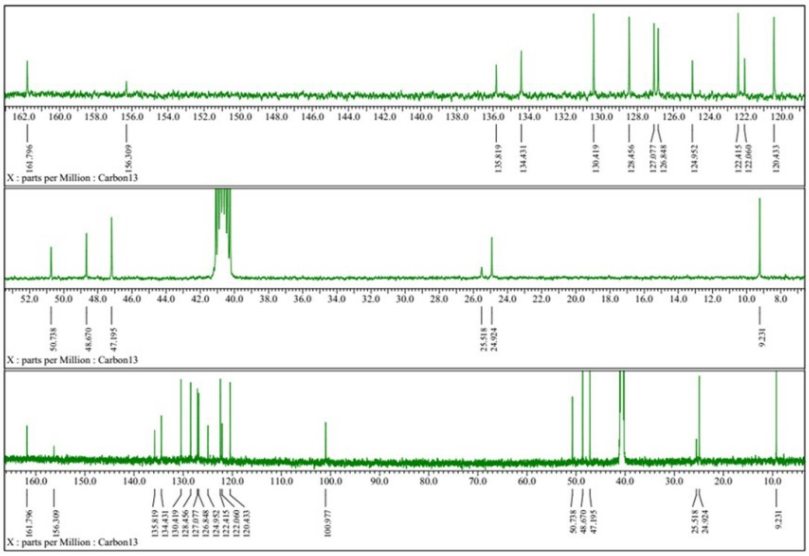

Fig. S3. ^{13}C-NMR spectrum of CyT in DMSO-d6. δ 161.80, 156.31, 135. 82, 134.43, 130.42, 128.46, 127.08, 126.85, 124.95, 122.41, 122.06, 120.43, 100.98, 50.74, 48.67, 47.20, 25.52, 24.92, 9.23.

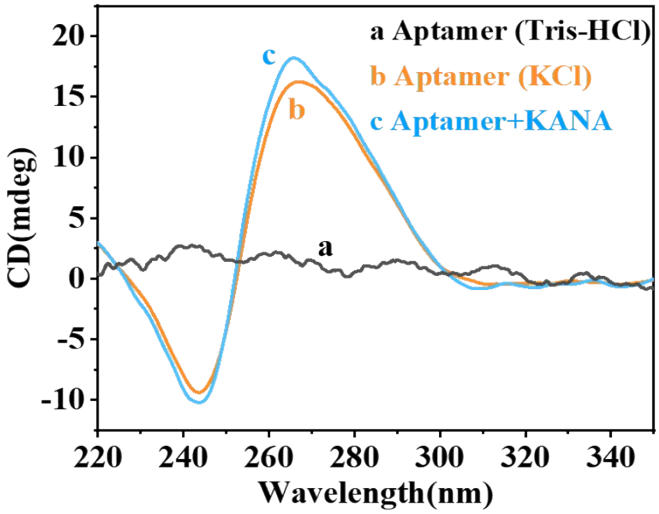

Fig. S4. The CD spectra of (a) KANA-aptamer in 20 mM Tris-HCl, (b) KANA-aptamer in 5 mM KCl, (c) KANA aptamer+KANA in 5 mM KCl. The concentrations of KANA-aptamer and KANA were 3.0 μM, 0.6 μM , respectively.

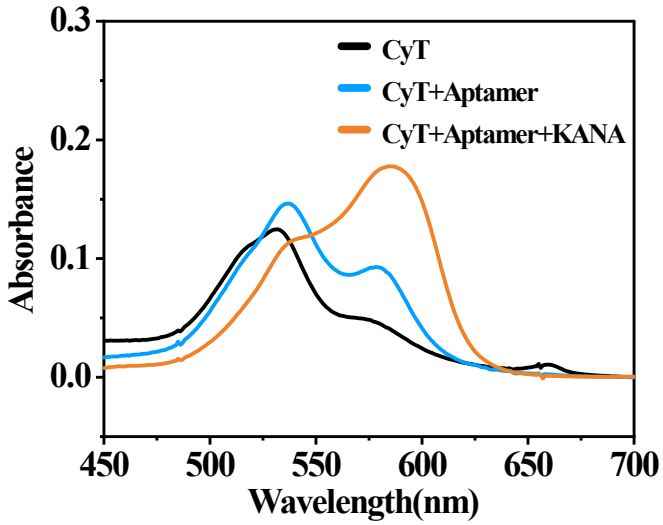

Fig. S5. The absorption spectra of CyT upon addition of KANA-aptamer and KANA.The concentrations of CyT, KANA-aptamer and KANA were 2.0 μM, 2.0 μM, 5.0 μM , respectively.

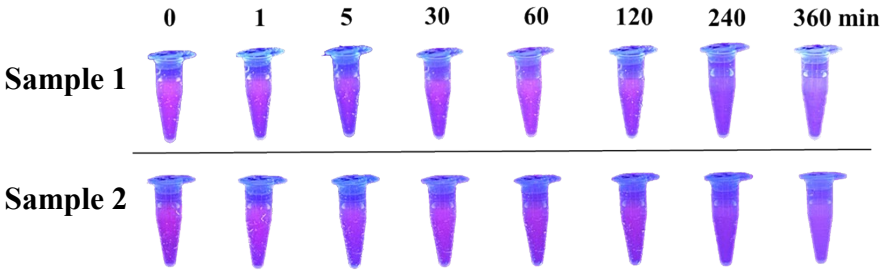

Fig. S6. Photographs of samples under UV lamp (302 nm) at different times.The concentrations of KANA were 0.6μM.

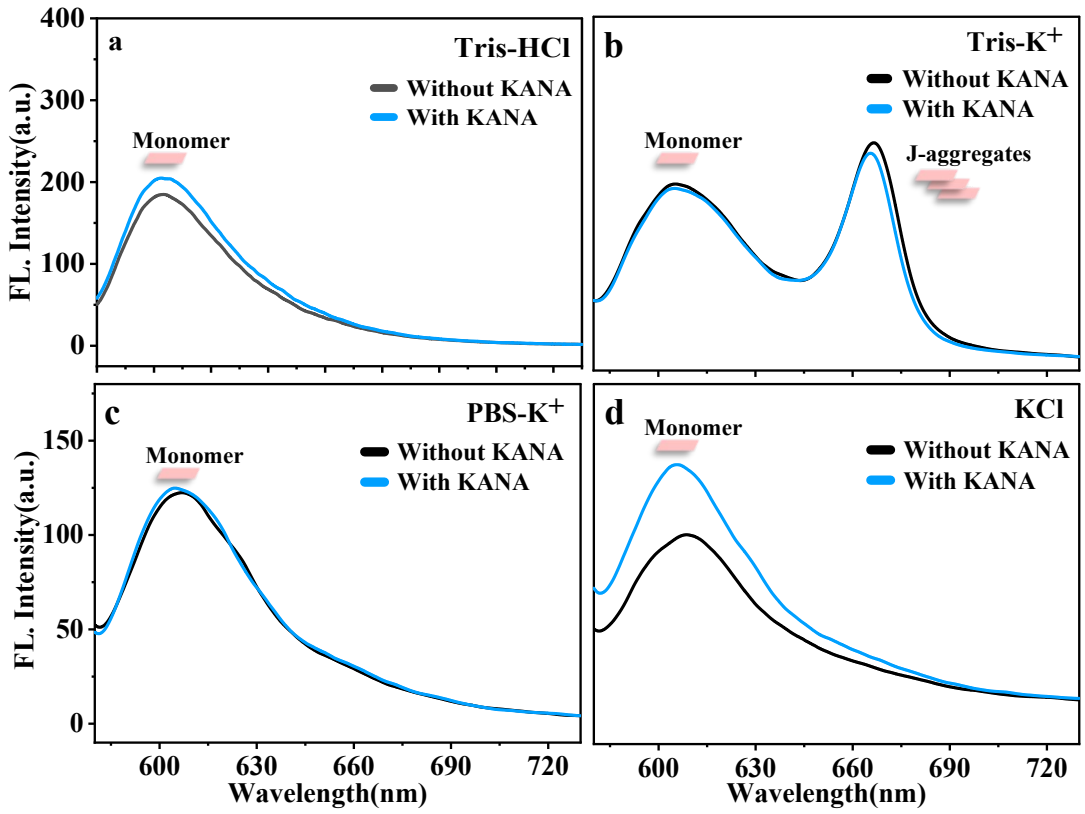

Fig. S7. Fluorescence spectra of CyT in different buffer solutions with (blue) or without (black) KANA (0.2 μM). (a) 20 mM Tris-HCl, (b) Tris-K^+ (20 mM Tris-HCl and 5 mM KCl), (c) PBS-K^+ (5 mM K^+), (d) 5 mM KCl.

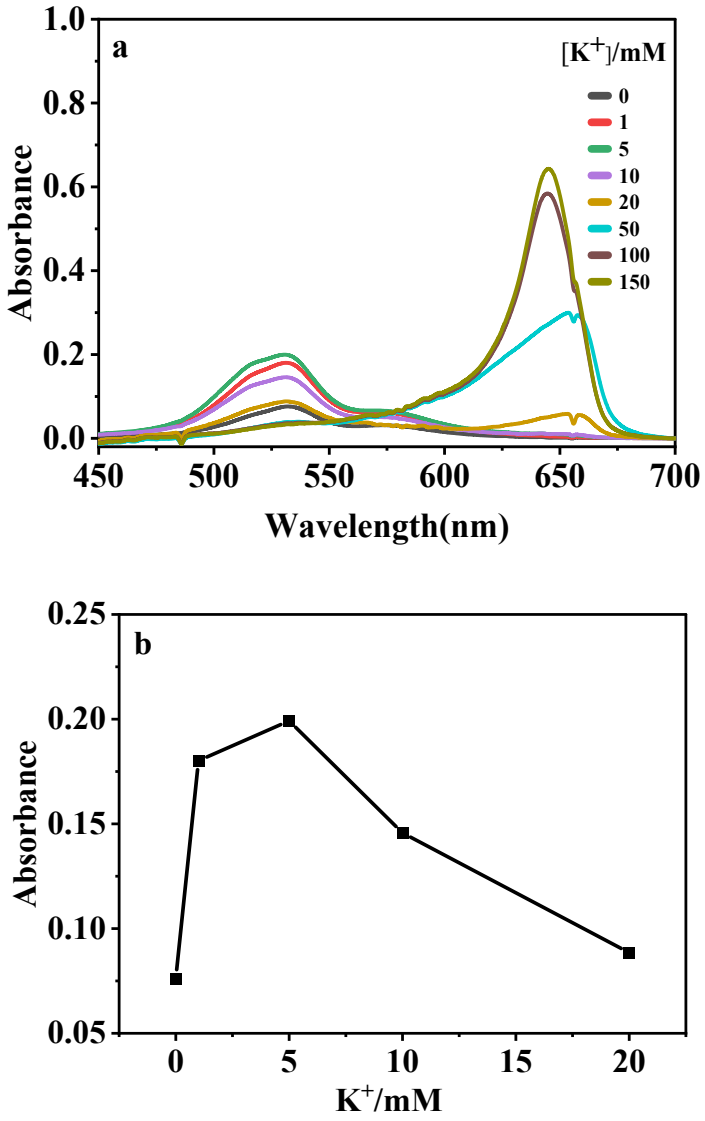

Fig. S8. a) The absorption spectra of 4 μM CyT with increasing concentrations of K^+. b) The plot of the absorbance at 532 nm of 4 μM CyT versus [K^+].

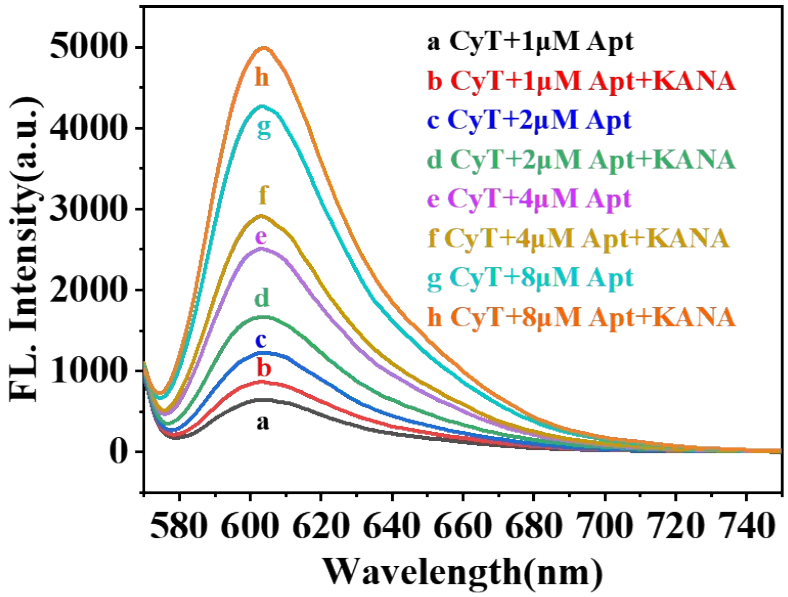

Fig. S9. The fluorescence spectra of 2 μM CyT with various concentrations of KANA-aptamer in the absence and presence of KANA.

Table S1 The reported G-rich sequence of antibiotic aptamers

Antibiotic Classes	Antibiotics	Aptamer sequence 5'→3'	Confor -mation	Ref.
Tetracyclines	Tetracycline	CGT ACG GAA TTC GCT AGC CCC CCG GCA GGC CAC GGC TTG GGT TGG TCC CAC TGC GCG TGG ATC CGA GCT CCA CGT G	Antipar allel G4	[1]
		TGGGTAGGGCGGGTTGGGA	-	[2]
		CGGTGGTG		[3]
	Oxytetracycline	CGTACGGAATTCGCTAGCCGAGTTGAGCCGGGCGC GGTACGGGTACTGGTATGTGTGGGGATCCGAGCTC CACGTG	-	[4]
Quinolones	Ofloxacin Ciprofloxacin	ATA CCA GCT TAT TCA ATT AGT TGT GTA TTG AGG TTT GAT CTA GGC ATA GTC AAC AGA GCA CGATCG ATC TGG CTTGTT CTA CAA TCG TAA TCA GTT AG	Parallel G4	[5][6]
	Enrofloxacin	CCC ATC AGG GGG CTA GGC TAA CAC GGT TCG GCT CTC TGA GCC CGG GTT ATT TCA GGG GGA	G-rich	[7]
Aminoglycosides		TGGGGGTTGAGGCTAAGAAGA	Parallel G4	[3]
	Kanamycin	TGGGGGTTGAGGCTAAGCCGA	Parallel G4	[8]
		TGG GGG TTG AGG CTA AGC CGA CCA TGT ACT TTT	G-rich	[9]
	Tobramycin	GGG ACT TGG TTT AGG TAA TGA GTC CC GA CTA GGC ACT AGT C	G-rich	[10] [11]
	Gentamicin	GGG ACT TGG TTT AGG TAA TGA GTC CC	G-rich	[12]
	Streptomycin	GGGGTCTGGTGTTCTGCTTTGTTCTGTCGGGTCGT	G-rich	[13]
Sulfonamides	Sulfame/thazine	TTA GCT TA T GCG TTG GCC GGG A TA AGG A TC CAG CCG TTG TAG A TT TGC GTT CTA ACT CTC	-	[14]
β-Lactams	Ampicillin	GCGGGCGGTTGTA TAGCGG	G-rich	[15]
	Penicillin	TTA GTT GGG GTT CAG TTG G	G-rich	[16]
		GGGTCTGAGGAGTGCGCGGTGCCAGTGAGT	G-rich	[17]
Anthracyclines	Chloramphenicol	ACT TCA GTG AGT TGT CCC ACG GTC GGC GAG TCG GTG GTA G	G-rich	[18]

Table S2 Determination of KANA concentration in lake water and urine by proposed method and standard HPLC-ELSD[a]

	Sample	Proposed method *	HPLC-ELSD *
Lake water	1	Not detected	Not detected
	2	Not detected	Not detected
Urine	1	Not detected	Not detected
	2	Not detected	Not detected

* Average value of three determinations ±standard deviation

[a]The standard HPLC-ELSD method as described in the Chinese pharmacopoeia

Table S3 Comparison with the reported methods for KANA determination

Detection method	LOD	Linear range	Application	Ref.
HPLC	1.23 nM	10-31 μM	-	[19]
ELISA	0.4 nM	0.4-6.0 nM	Fc fusion protein	[20]
CE	0.83 μM	4.0-65 μM	-	[21]
FRET	1.1 μM	4-25 μM	milk	[22]
Fluorescent	1.05 nM	50-2000 nM	pork, chicken, beef	[23]
Electrochemical	0.06 nM	0.1-60 nM	honey	[24]
Electrochemical	0.23 nM	2.5-155 nM	milk	[25]
Luminescence	143 nM	0.2-150 μM	fish	[26]
Colorimetric	0.06 μM	0.1-0.5μM	honey, milk, pork	[27]
Colorimetric	0.68 μM	1-40 μM	milk	[28]
Fluorescent	43 nM	0.1-1μM	lake water, urine	This work

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