Label-free fluorescence aptasensor for chloramphenicol based on hybridization chain reaction amplification and G-quadruplex/*N*methyl mesoporphyrin IX complexation

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Name	Sequence (5'→3')
Apt	5'- <u>ACT TCA GTG AGT TGT CCC AC</u> G GTC GGC GAG TCG GTG GTA G - 3'
Apt-C	5'- <u>GT GGG ACA ACT CAC TGA AGT</u> -3'
Initiator-MB	5'- <u>ACC TTC TTC T</u> ACT TCA GTG AGT TGT CCC AC <u>A GAA</u> <u>GAA GGT G</u> TT TAA GTA -3'
Hairpin DNA (H1)	5'- AGG GCG GGT G <u>GG TGT TTA AGT</u> TGG AGA ATT GT <u>A</u> <u>CTT AAA CAC C</u> TT CTT CTT GGG T -3'
Hairpin DNA (H2)	5'- TGG GTC AAT TCT <u>CCA ACT TAA AC</u> T AGA AGA AGG T <u>GT TTA AGT TGG</u> GTA GGG CGG G -3'

Table S1. Sequences of oligonucleotides

Spectra properties of samples were detected under the same experimental conditions using an F-4600 Hitachi fluorescence Spectrometer with a xenon lamp. At an excitation of λ = 399nm, the emission peak was recorded at 612 nm both for N-Methyl mesoporphyrin IX(NMM) and NMM/G4 (Figure S1). However, the fluorescence intensity of NMM/G4 was significantly higher than that of NMM, indicating that the NMM/G4 composite greatly enhanced the fluorescence of NMM.

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Figure S1. Spectra properties of NMM and NMM/G4

The fluorescence intensity of NMM was enhanced after the complexation reaction with G-quadruplexes. The systematic quantum yield was measured for G4/ NMM, and the quinine sulfate dihydrate which dissolved in 0.1 mol/L H_2SO_4 with a quantum yield of 51% was the reference. The quantum yield (y) is calculated by the following formula.

$$Y = Y_R \cdot \frac{F_s}{F_R} \cdot \frac{A_R}{A_s} \cdot \frac{\eta_s^2}{\eta_R^2}$$

F is the integral area of the fluorescence emission spectrum, A is the absorbance, and η is the refractive index of the solvent (R and S represent the reference quinine sulfate dihydrate solution and the sample respectively)

The systematic quantum yield measurements were shown in table S2.

	Test sample	λex/nm	А	F	Y
Reference	quinine sulfate dihydrate	399	0.024	13245	0.51
Sample	G/NMM	399	0.02	15319	0.64

Table S2. Systematic quantum yield measurements

Method	Detection limit	Detection range	Ref.
Electrochemiluminescence assay	1.7 pg·mL ⁻¹	0.01 - 4000 ng·mL ⁻¹	(Chen et al. 2020)
Structure switching signaling Assay	0.70 ng mL ⁻¹	1 - 100 ng·mL ⁻¹	(Ma et al. 2020)
MOF-based ratiometric fluorescent biosensor	0.08 pg mL ⁻¹	0.1pg ·mL ⁻¹ - 10ng·mL ⁻¹	(Liu et al. 2020)
Electrochemical sensor	$0.32 \ \mu mol \cdot L^{-1}$	$0.03 - 0.5 \text{ mmol} \cdot \text{L}^{-1}$	(Shad et al. 2019)
Photovoltammetric sensor	$0.14 \ \mu mol \cdot L^{-1}$	0.5 - $50 \ \mu mol \cdot L^{-1}$	(Zhu et al. 2019)
Chemiluminescence resonance energy transfer assay	2.0 pg·mL ⁻¹	0.01 - 100 ng·mL ⁻¹	(Jia et al. 2019)
Label free aptasensor based on HCR and G-quadruplex/NMM	0.8 pg·mL ⁻¹	$2.5 - 200 \text{ pg} \cdot \text{mL}^{-1}$	This work

Table S3. Comparison with other methods for CAP detection

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