Supporting Information

A Dual-Colored Ratiometric-Fluorescent Oligonucleotide Probe for Detection of Human Telomerase RNA in Cell Extracts

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UV and fluorescence spectra of TAMRA, FAM and Dabcyl



Fig. S1 (A) UV absorption spectra and (B) fluorescence spectra for oligonucleotides modified with TAMRA, FAM and Dabcyl, respectively. Excitation wavelengths of FAM and TAMRA were 485 nm and 545 nm, respectively. The involved oligonucleotide sequences are listed (from 5' to 3'): TTTdT(TAMRA)GTCTAACCCTAATTTTTGGTTAGACAAAA, TTTTGTCTAACCCTAATTTTTGGTTAGACAAAA-FAM,

CTTCTCAGTTAGGGTTAGACAAAA-Dabcyl. All spectra have been normalized.

FRET efficiencies

A ratiometric method¹⁻³ was used to calculate FRET efficiencies (E) according to the equation: $E=F_A/(F_D+F_A)$

	Sn (%)	Sn+D1+Target (%)
n = 1	66.8 %	65.0 %
n = 4	72.5 %	72.1 %
n = 6	58.6 %	58.0 %

Table S1 FRET efficiencies of Sn and Sn+D1+Target (n = 1, 4, 6).

Signal stability of the DRO probe

To study the signal stability, enzyme digestion reaction in cellular environment and some external factor, such as incubation time and pH value, had been taken into consideration. For incubation time effects, 250 nM DRO probe was incubated at 37 °C, and the fluorescence was recorded every 20 minutes. For pH value effects, a series of 250 nM DRO probes in PB buffer with different pH value from 6.0 to 10.0 were incubated at 37 °C for 2 h. For enzyme digestion reaction, two groups of 250 nM DRO probes were separately treated with 1 μ L 70 U/ μ L DNase I and 1 μ L 200 U/ μ L Exo III at 37 °C, then the fluorescence was collected every 10 min.



Fig. S2 Signal stability of DRO probe with (A) different incubation time, (B) pH value, and in the presence of (C) DNase I and (D) Exo III.

Selectivity and specificity of DRO probe



Fig. S3 The fluorescent spectra of DRO probes with target and different number of mismatched bases.

Study of color variation

Several 1.0 μ M DRO probes were reacted with different concentrations of targets. As other ratiometric fluorescent sensors, this dual-labeled fluorescent oligonucleotide also exhibits color variations from green to olivine to orange with increased concentration of target by the excitation of UV lamp at 365 nm.



Fig. S4 The evolution of colors when DRO probes responded to various concentrations of targets.



Fig. S5 The fluorescent responses to different concentrations of cell extracts of (A) KB and (B) QSG, respectively.

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Fig. S6 The theoretical description of fluorescent color variations of DRO probe with the addition of HeLa cell extracts in CIE chromaticity chart.



Fig. S7 The left is the reversibility of the FRET probe with increasing cycle times. The right is the color variation of 1 and 2 cycle times, respectively.

References

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