Specific Detection of the Cleavage Activity of Mycobacterial Enzymes using a Quantum Dots based DNA Nanosensor

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Supplementary Information  

1. Characterization of the QD-based DNA Nanosensor:  
The absorption and emission spectra of QDs, as shown in Supplementary Figure 1, were pre-evaluated prior the experiments. The Förster radius $R_0$, defined as where 50% of energy transfer occurs, was calculated to be 65.09 Å for the selected pair of QD (emission peak at 605 nm) and Cy5, according to Equation 1. Parameters used in the estimation included, the refractive index of the medium ($n$=1.4), the unperturbed donor photoluminescence quantum yield ($Q_D$=0.4), the spectral integral from the overlap of donor emission and acceptor absorption ($J(\lambda)$=1.24x10\textsuperscript{16} M\textsuperscript{-1}cm\textsuperscript{-1}nm\textsuperscript{4}, see also Supplementary Figure 1) and the relative orientation of the donor and acceptor dipoles ($\kappa^2 \sim 2/3$).  

$$R_0 = 0.211 \left[ \kappa^2 n^{-1} Q_D J(\lambda) \right]^{1/4} \quad (\text{in Å})$$  

Equation (1)
Supplementary Figure 1. Selection of QFRET Pair: The QFRET pair was selected for sufficient spectra overlap, rendering efficient energy transfer within the distance between donor and acceptor.

The molar ratio of Cy5-labeled MsTopoI DNA substrates to QD was kept at a ratio of 25 : 1 (1.25 pmol of MsTopol DNA substrate and 50 fmol of QD). The ratio was empirically determined at where the optimal quenching of QD fluorescence occurred, while the amount of MsTopol DNA substrate is in minimal excess.

Supplementary Figure 2. Optimal Quenching at the Initial Stage: (a) Emission spectra of the QD nanosensor conjugated with different ratio of MsTopol DNA substrate to QD. (b) Quenching efficiency measured at different ratio. The quenching was observed saturated at the ratio of 25-30. Further increase of the ratio did not observed significant quenching (Ratio 50).

References: