Molecular Beacon Based Bioassay for Highly Sensitive and Selective Detection of Nicotinamide Adenine Dinucleotide and the Activity of Alanine Aminotransferase

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We investigated the effect of LDH concentration on ALT activity assay. As shown in Figure S1, the signal/background ratio of different LDH concentration effect, from left to right, respectively was 1.97, 4.9, 8.3, 3.8. The assay got the highest signal/background ratio with 0.5 kU/L LDH so we chose 0.5 kU/L as the optimum LDH concentration in ALT activity assay.

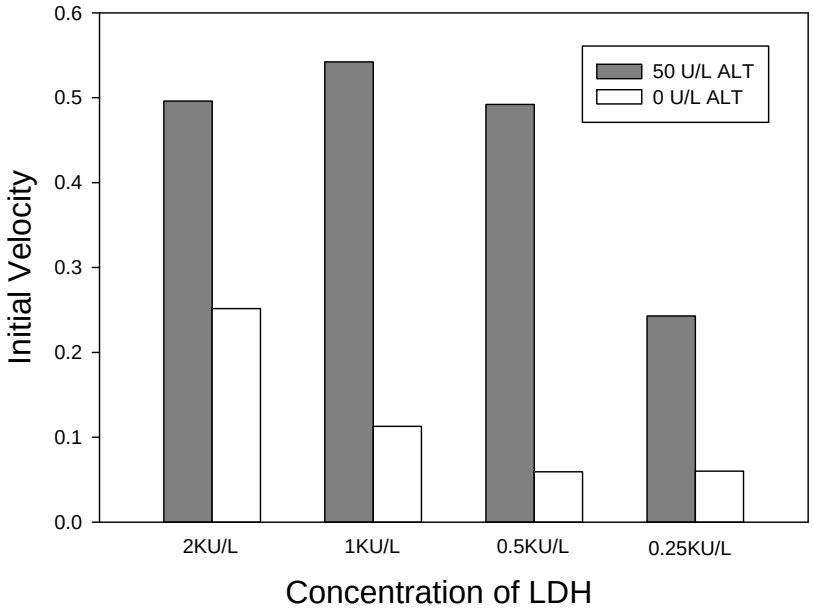

Figure S1. Optimization of LDH concentration in ALT activity assay. The highest signal to background ratio was obtained when the final LDH concentration in solution 1 was 0.5 kU/L. Measurements were performed in a Tris-HCl buffer (50 mM, pH = 8.0, 10 mM $MgCl_2$, 5 mM DTT, 2.5 mM $CaCl_2$, and 0.05% BSA).The concentration of LDH in the Solution 1 ranged from 0.25 KU/L to 2.0 KU/L. All samples were prepared and detected in a 80 μL aliquot of solution containing 250 nM MB2, 250 nM Oligo1, 250 nM Oligo2, and 6.0 units of *E.coli* DNA ligase. The initial velocity was determined by the average of fluorescence enhancement rate in 200 s after the addition of *E. coli* DNA ligase.