

Aptamer based electrochemical biosensor for tumor necrosis factor-alpha detection in whole blood

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Supporting Information – Table of Contents

Part 1: SPR Analysis of Aptamer Assembly and TNF- α Binding (Figure S1)

Part 2: Effects of Aptamer Surface Density on Biosensor Performance in HEPES buffer (Figure S2)

Part 3: Effects of Aptamer Surface Density on Biosensor Performance in whole blood (Figure S3)

Part 4: Verification of Sensor Performance in Cellular Environment (Figure S4)

Part 1: SPR Analysis of Aptamer Assembly and TNF- α Binding

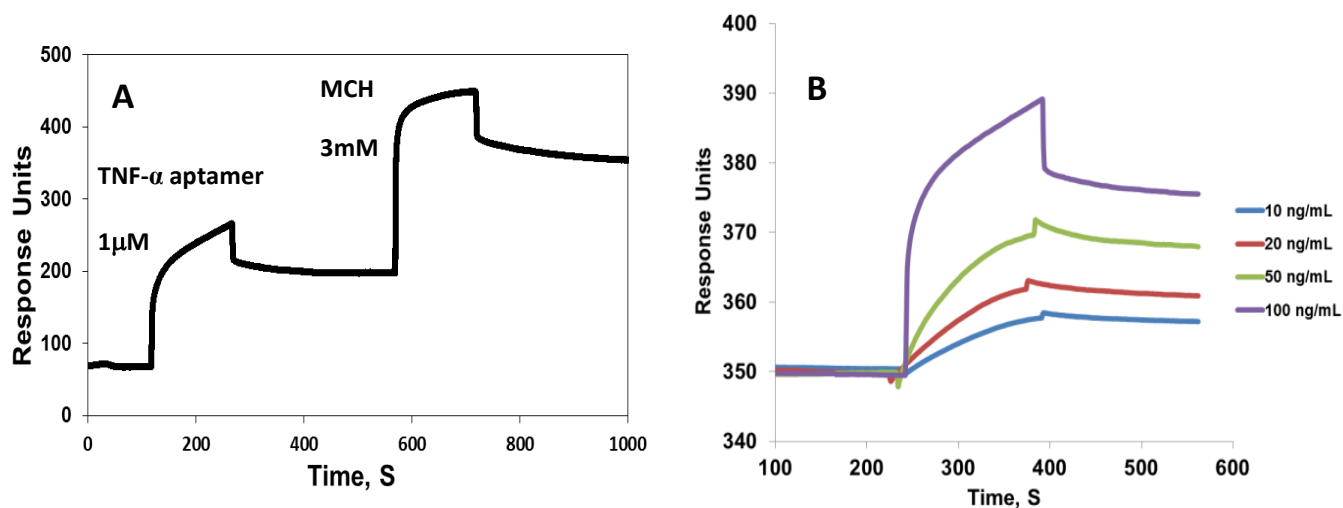


Figure S1: SPR analysis of MB labeled aptamer interaction with TNF- α . (A) SPR sensorgram showing assembly of 1 μ M aptamer in solution (3' thiolated and 5' MB labeled), followed by blocking with 3 mM MCH. Both solutions were prepared in HEPES (pH=7.4) buffer. (B) SPR sensorgrams indicate the binding of serially diluted TNF- α to RNA aptamers.

Part 2: Effects of Aptamer Surface Density on Biosensor Performance in HEPES buffer

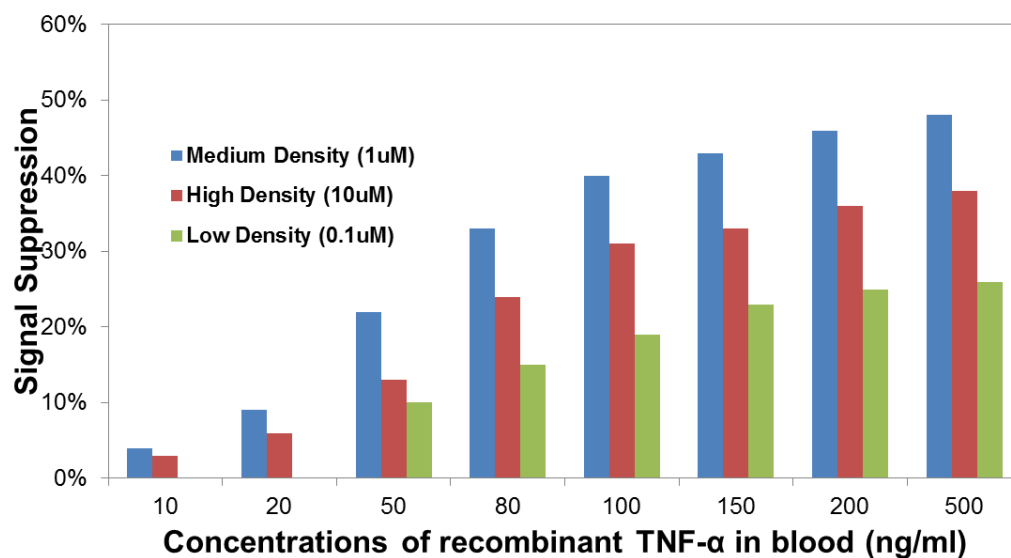


Figure S2: Sensitivity of electrodes with different aptamer packing densities was expressed as the loss or suppression of signal (current) upon binding of different concentrations of TNF- α in HEPES. The larger the signal suppression (or current loss), the higher is the sensitivity of the biosensor. Electrodes with low, medium, and high packing density were prepared using 0.1, 1, and 10 μ M aptamer concentrations. Aptasensors with medium packing density were found to be most sensitive with a detection limit of 0.6 nM or 10 ng/mL TNF- α .

Part 3: Effects of Aptamer Surface Density on Biosensor Performance in Whole Blood

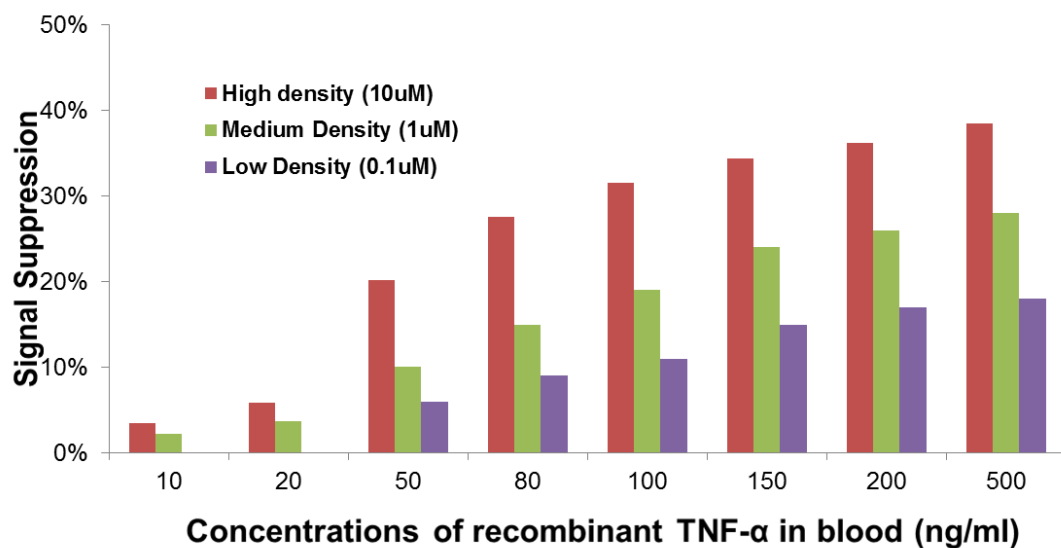


Figure S3: Sensitivity of electrodes with different aptamer packing densities was expressed as the loss or suppression of signal (current) upon binding of different concentrations of TNF- α in whole blood. The larger the signal suppression (or current loss), the higher is the sensitivity of the biosensor. Electrodes with low, medium, and high packing density were prepared using 0.1, 1, and 10 μ M aptamer concentrations. Aptasensors with high packing density were found to be most sensitive for this case with a detection limit of 0.6 nM or 10 ng/mL TNF- α .

Part 4: Verification of Sensor Performance in Cellular Environment

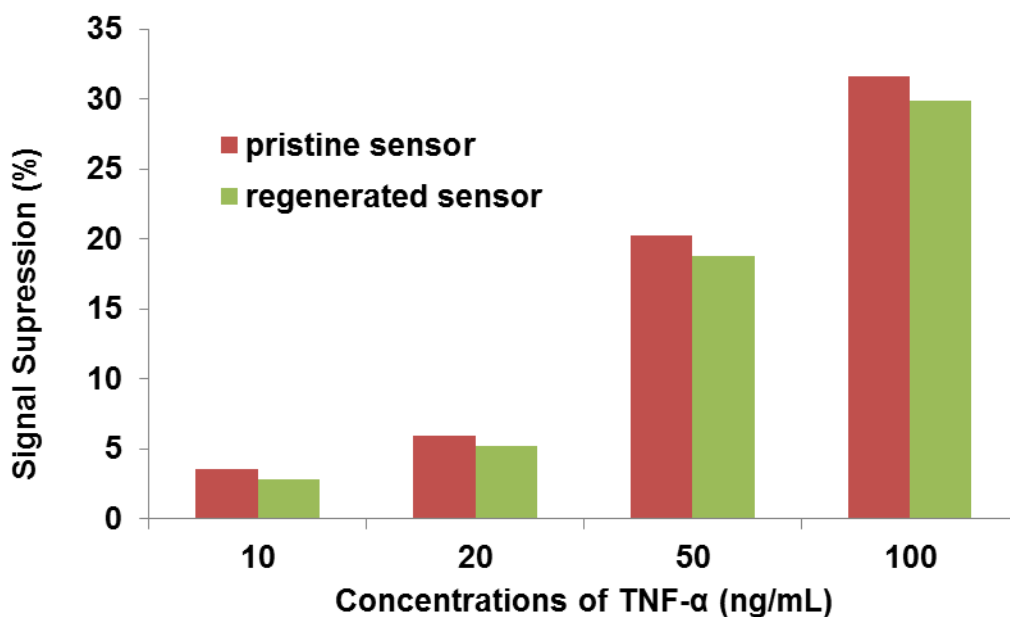


Figure S4: After detecting TNF- α secretion from blood, aptamer modified electrodes were regenerated using 7M Urea buffer for 30s. These “used” aptasensors were then challenged with varying concentrations of recombinant TNF- α in RPMI media solution. Similarity in responses of pristine and “used” sensors confirms stability of aptamer haripins against digestion with cell nucleases.