Negative SPR Signals During Low Molecular Weight Analyte Recognition

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SPR response monitored during the aptasensor build-up

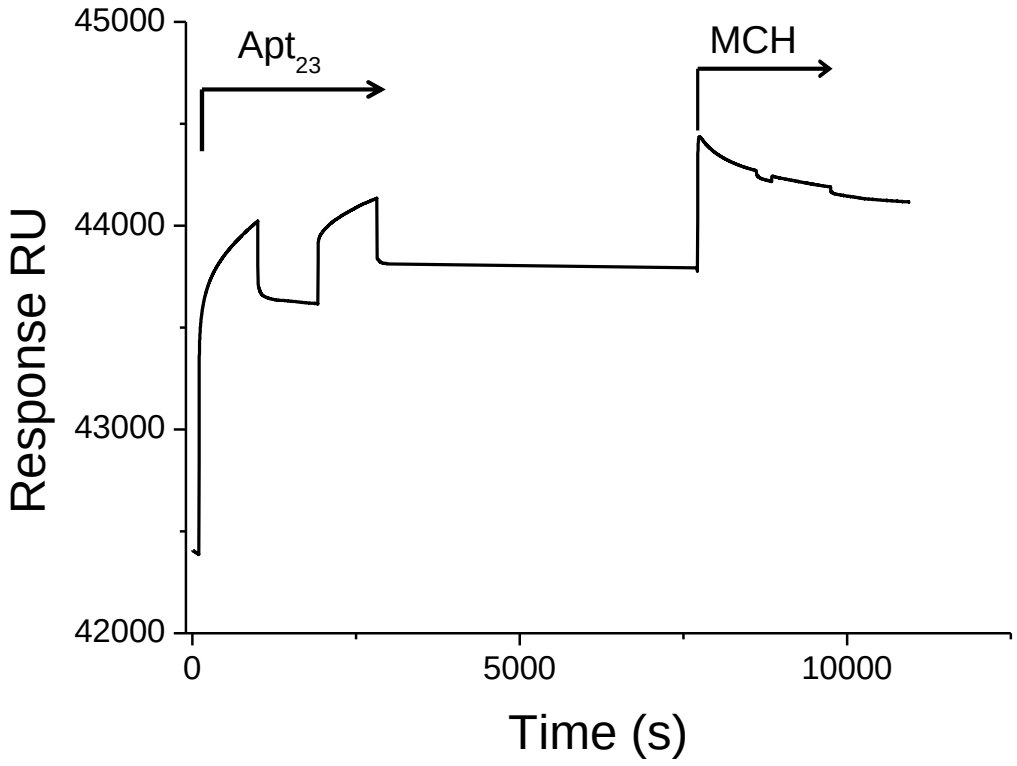

Figure SI-1: Sensorgram recorded during the build-up of the aptasensor on the gold surface. Thiolated 23-mer aptamer (1 µM) was injected followed by MCH (1mM) injection. T= 25°C. Flow rate: 2µL/min

Circular dichroism of T_6-Apt_{23} with and without L-Tym or D-Tym

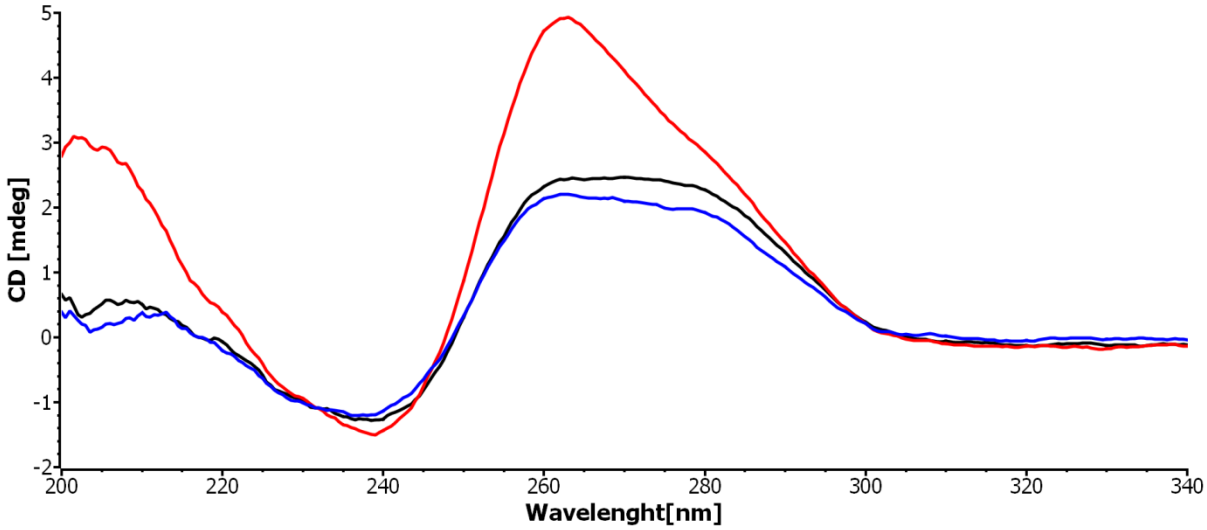

Figure SI-2: CD spectra of T_6-Apt_{23} with and without L-Tym or D-Tym (15µM in 5 mM Tris, 10 mM $MgCl_2$, 50mM NaCl). (black line) T6-Apt23; (red line) T_6-Apt_{23} +L-Tym (1eq), (blue line) T_6-Apt_{23} +D-Tym (1eq)

QCM-D responses monitored during the aptasensor build-up

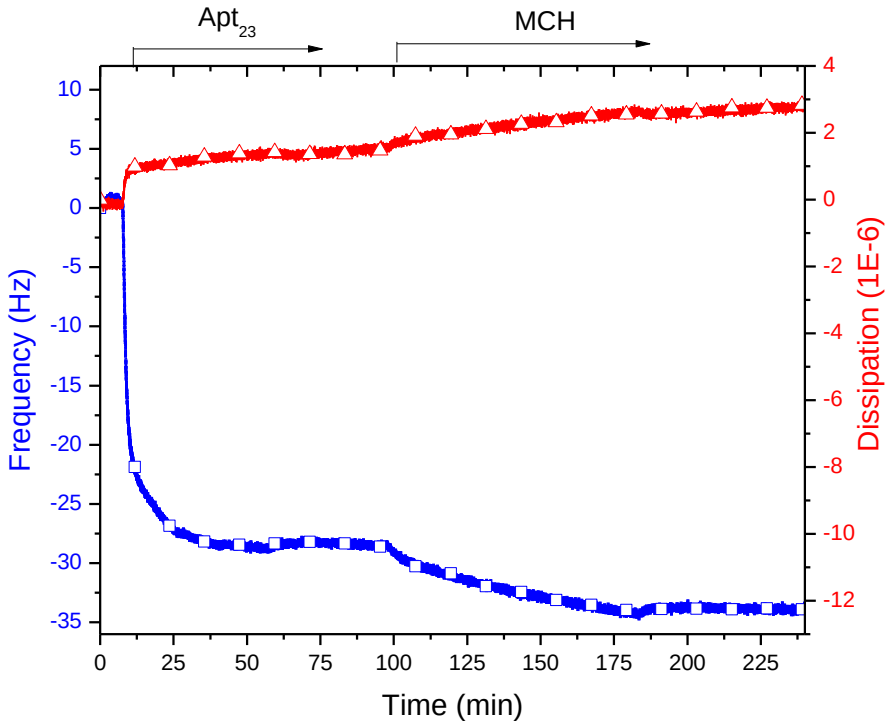

Figure SI-3: QCM-D profile characterizing the build-up of an aptasensor on the quartz gold sensor. Thiol aptamer (1 µM) was injected during fixed duration followed by MCH (1mM). The frequency shift and the dissipation shift are represented for overtones: $i = 7$ (□ blue line for frequency and Δ red line for dissipation). Start and duration of aptamer and MCH injections are indicated by arrows on top of the profile. Before sample injection and after saturated binding, the surfaces were exposed to pure running buffer. Flow rate: 50µL/mL.

Sensorgram recorded during the D-Tym injection on T_6-Apt_{23} monolayer

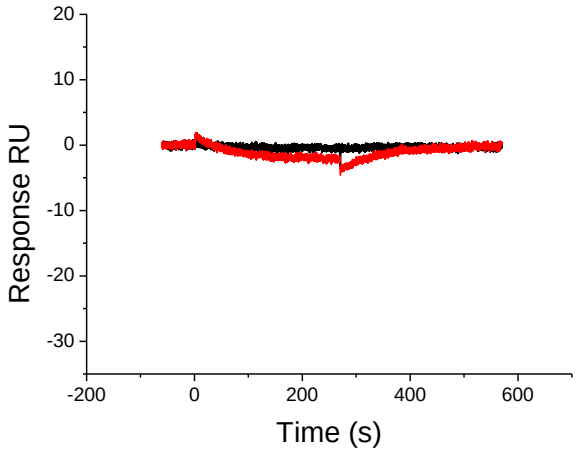

Figure SI-4: Sensorgram recorded upon injection of D-Tym at 100µM (black curve) and 1mM (red curve) on the active flowcell after double subtraction procedure.

Model developed by Dejeu et al for the maximal SPR response

See reference 4 below for further details.

- **Refractive Index Increment correction:**

Classically, the molar refractive index increment of the complex analyte/ligand can be defined from the RII of both the analyte and the ligand ones by equation (SI-1):

$$\left(\frac{dn}{dc}\right)_{LA} = \left(\frac{dn}{dc}\right)_{L} + V.\left(\frac{dn}{dc}\right)_{A} \qquad \text{(SI-1)}$$

The L-Tym/aptamer interaction having a 1:1 stoichiometry,[3] V=1 in our study

Considering a deviation factor for the RII of the analyte/ligand complex, eq (SI-1) is not valid anymore and a correction term has to be included. As a consequence, the RII of the L-Tym/aptamer complex is expressed as a function of the RII of the aptamer by introducing a correction factor, x:

$$\left(\frac{dn}{dc}\right)_{correction} = x\left(\frac{dn}{dc}\right)_{L} \qquad \text{(SI-2)}$$

where $(dn/dc)_{correction}$ is the correction of the L-Tym/aptamer complex RII defined as a function of the RII of the aptamer. By adding this correction factor to the sum of the RII of the two entities separately considered eq ((SI-1)), we obtain eq (SI-3):

$$\left(\frac{dn}{dc}\right)_{LA} = \left(\frac{dn}{dc}\right)_{L} + V.\left(\frac{dn}{dc}\right)_{A} + \left(\frac{dn}{dc}\right)_{correction} = (1+x)\left(\frac{dn}{dc}\right)_{L} + V.\left(\frac{dn}{dc}\right)_{A} \qquad \text{(SI-3)}$$

- **Dejeu's Formula[4-5]**

$$RU_{A_{\max}} = RU_L \left(\frac{1}{\rho} \frac{1 - e^{-\frac{\rho d_L}{d_p}}}{1 - e^{-\frac{d_L}{d_p}}} (\beta \times V + 1 + x) - 1 \right) \quad \text{(SI-4)}$$

Where RU_{Amax} is the expected maximum response at a single site, RU_L is the aptamer immobilization level, dp is the effective penetration depth of the SPR wave (175 nm), d_L and d_{LA} are the thicknesses before and during the interaction respectively, ρ the folding ratio of the aptamer ($\rho = d_{LA}/d_L$), $\beta = MW_A.(dn/dc)'_A / MW_L.(dn/dc)'_L$ is the ratio of the mass-weighted RII of the analyte versus the ligand and the V, the valency i.e. the ratio number of analytes per number of ligands involved in the recognition (V=1 in the present system). MW_A and MW_L are the molecular weights of the injected analyte (L-Tym, 180.2 $g.mol^{-1}$) and of the immobilized ligand (aptamer, 9182 $g.mol^{-1}$), respectively. $(dn/dc)'_A$ and $(dn/dc)'_L$ are the refractive index increments (RII) for the analyte (L-Tym, 0.215 $cm^3.g^{-1}$)[1] and for the ligand (aptamer, 0.238 $cm^3.g^{-1}$) respectively.

Reference

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