

APTAMER INFORMATION Human Serum Albumin (HSA) aptamer_23G03 #372

1a. Description:

- Identifiers: 23 G03 (Oligo #372)
- <u>Number of DNA nucleotides</u>: 39 bases (without 3'-6T), 45 bases (with 3'-6T)
- Molecular weight (includes 3'-biotin and 6T): 14,237.4 g/mol
- <u>Target for selection</u>: HSA protein, Sigma (Cat# A3782)

Aptamer was selected from a randomized 40-mer library against HSA protein. Proprietary methods were then used to select the aptamer.

Aptamer folding instruction before use:

Once the aptamer is in its working concentration, it needs to be heated to 85-90 °C for 2 minutes, and then cooled to room temperature before use.

1b. Validation data with HSA protein by BLI (Bio-Layer Interference) method:

- *Immobilized Ligand:* HSA aptamer_23G03 #372 with 3'-6T and biotin
- Analyte: HSA protein
- Buffer used for validation: 20 mM Tris, 100 mM NaCl, 0.005% Tween20 in nuclease free water, pH 7.4

1c. Kinetics Screening Assay using Streptavidin Biosensors :

We validate the binding data, by single reference method.

• <u>Single reference data:</u> All curves are referenced to a sensor dipped in buffer alone (no protein) (see Figures 1, 2 and Table 1).





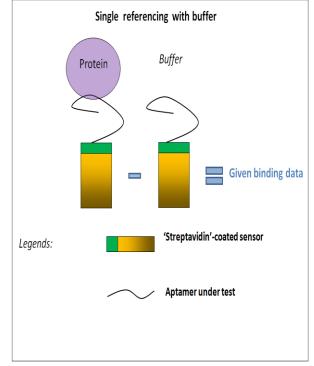


Figure 1. Diagram showing aptamer: protein binding validation scheme.



1d. Single reference data:

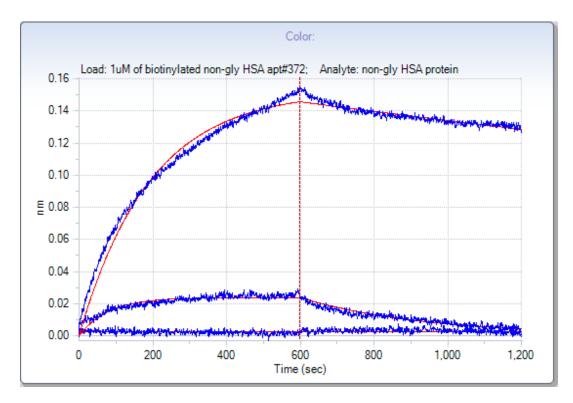


Figure 2. Association and dissociation graph of 1:1 fitting model of HSA aptamer (biotinylated) oligo #372 to HSA protein concentrations 500, 125 and 31.25 nM, by single reference method.

Table 1. K_d , R^2 and Chi^2 values by Local fitting for single reference method. $K_d = 29.1 \text{ nM}$						
Immobilized Aptamer	Analyte	Conc. (nM)	Response	К _d (М)	Full R ²	Full X ²
HSA apt#372 biotin	HSA protein	500	0.1512	2.09E-08	0.985181	0.016463
HSA apt#372	hox protein	500	0.1312	2.092-00	0.805161	0.010403
biotin	HSA protein	125	0.0268	3.72E-08	0.933033	0.004003







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