



APTAMER INFORMATION

Peroxiredoxin aptamer #346

1a. Description:

- Identifiers: Peroxiredoxin aptamer (Oligo # 346)
- Number of DNA nucleotides: 35 bases (without 3'-12T), 47 bases (with 3'-12T)
- Molecular weight (including 3'-6T and biotin): 14,564.4 g/mol
- Target for selection: Peroxiredoxin protein, Novus Biologicals (Cat# NBPI-41141)

Aptamer was selected from a randomized 40-mer library against Peroxiredoxin 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 Peroxiredoxin protein by BLI (Bio-Layer Interferometry) method:

- Immobilized Ligand: Peroxiredoxin aptamer #346 with 3'-12T and biotin
- Analyte: Peroxiredoxin protein
- Buffer used for validation: 1X PBS, pH = 7.4
- Random aptamer as negative control: Random aptamer of same length, similar molecular weight and same 3'-modifications (i.e 12T and biotin) as Peroxiredoxin aptamer.

1c. Kinetics Screening Assay using Streptavidin Biosensors :

We validate the binding data by the double referencing method:

- Double reference data: For even more stringent validation of aptamer binding, we “double-reference” all data to both protein-free buffer (as above) as well as to a sensor with a control or “sham” aptamer (see Figures 1, 2 and Table 1). This random aptamer controls for any non-specific charge-only interactions.



1d. Double reference data with immobilized random aptamer:

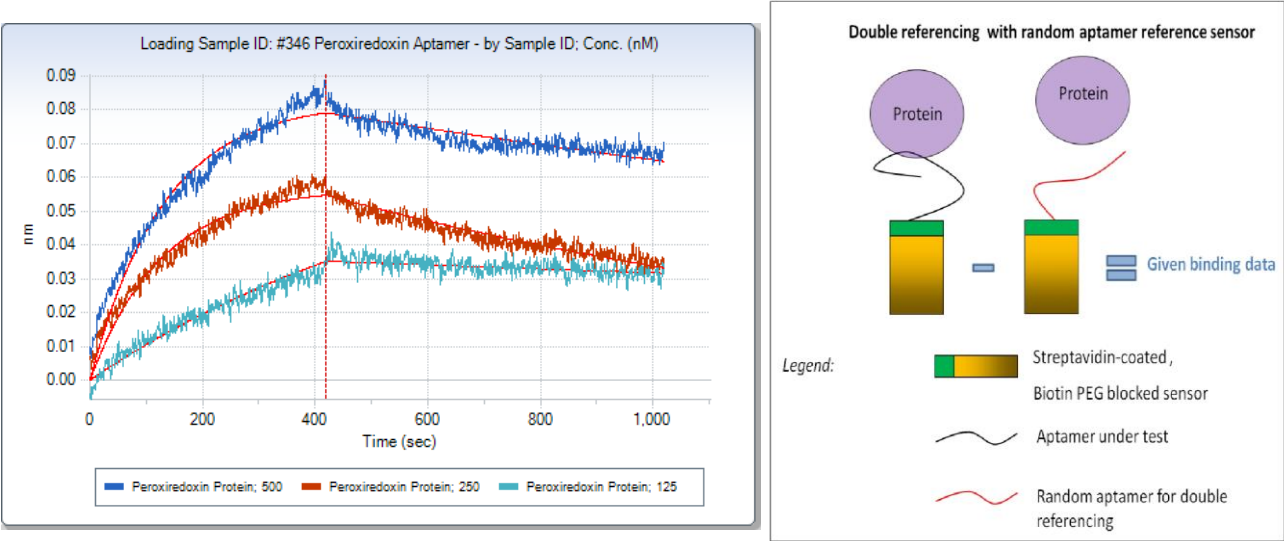


Figure 2. Association and dissociation graph of 1:1 fitting model of Peroxiredoxin aptamer #346 (biotinylated) to Peroxiredoxin protein concentrations 500, 250 and 125 nM, by double reference method with negative control aptamer.

Table 1. K_d , R^2 and χ^2 values by Local fitting for single reference method. Avg $K_d = 22.7$ nM

Immobilized Aptamer	Analyte	Conc. (nM)	Response	K_d (M)	Full χ^2	Full R^2
Peroxiredoxin aptamer #346	Peroxiredoxin	500	0.0842	2.27E-08	0.010179	0.952678
Peroxiredoxin aptamer #346	Peroxiredoxin	250	0.0575	2.92E-08	0.005786	0.944692
Peroxiredoxin aptamer #346	Peroxiredoxin	125	0.0318	1.63E-08	0.004226	0.955279