

Supporting information

High-throughput bioanalysis of bevacizumab in human plasma based on enzyme-linked aptamer assay using anti-idiotypic DNA aptamer

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Table S1. Effect of DNA aptamer concentration used for microplate immobilization on the developed absorbance.

Bevacizumab ($\mu\text{g/mL}$)	Aptamer (μM)		
	1	5	10
5	0.767	0.763	0.764
1	0.581	0.620	0.601
0.5	0.490	0.518	0.543
0.1	0.228	0.215	0.243
0.05	0.156	0.178	0.192
0.01	0.079	0.093	0.097

Table S2. LLOQ, ULOQ, linearity, precision and accuracy of the colorimetric method in standard solution analysis.

LLOQ (µg/mL)	ULOQ (µg/mL)	Linearity		Spiked concentration (µg/mL)				
				0.05	0.1	0.5	1.0	5.0
0.005	5.0	0.999	Precision	6.8	6.7	1.6	1.4	0.3
			Accuracy	-2.6	-1.6	12.4	-4.9	-2.3

Table S3. Effect of dilution ratio of bevacizumab-spiked plasma samples on the produced absorbance.

Dilution (fold)	Spiked concentration (µg/mL)							
	0.001	0.005	0.01	0.05	0.1	0.5	1	5
400	0.689	0.755	0.760	0.773	0.749	0.786	0.703	0.624
800	0.604	0.768	0.709	0.760	0.732	0.724	0.741	0.649
1200	0.520	0.749	0.678	0.752	0.714	0.745	0.823	0.680
1600	0.523	0.679	0.683	0.677	0.747	0.793	0.761	0.707
2000	0.596	0.612	0.556	0.568	0.543	0.608	0.666	0.727

Table S4. Changes in coefficient of variation (CV, %) of absorbance due to repeated use of assay plates,

Repeated use (times)	Bevacizumab (µg/mL)							
	standard solution				plasma-spiked sample			
	0.05	0.1	0.5	1	0.05	0.1	0.5	1
1	8.1	13.2	10.7	7.0	13.2	8.4	8.6	7.6
2	5.6	5.5	12.1	8.1	8.6	5.6	7.1	5.0
3	6.3	5.4	4.0	18.3	6.0	6.1	7.6	3.6
4	5.3	6.7	7.7	6.4	5.5	4.9	6.8	5.4
5	2.9	8.2	8.0	5.6	4.5	4.2	4.9	2.6

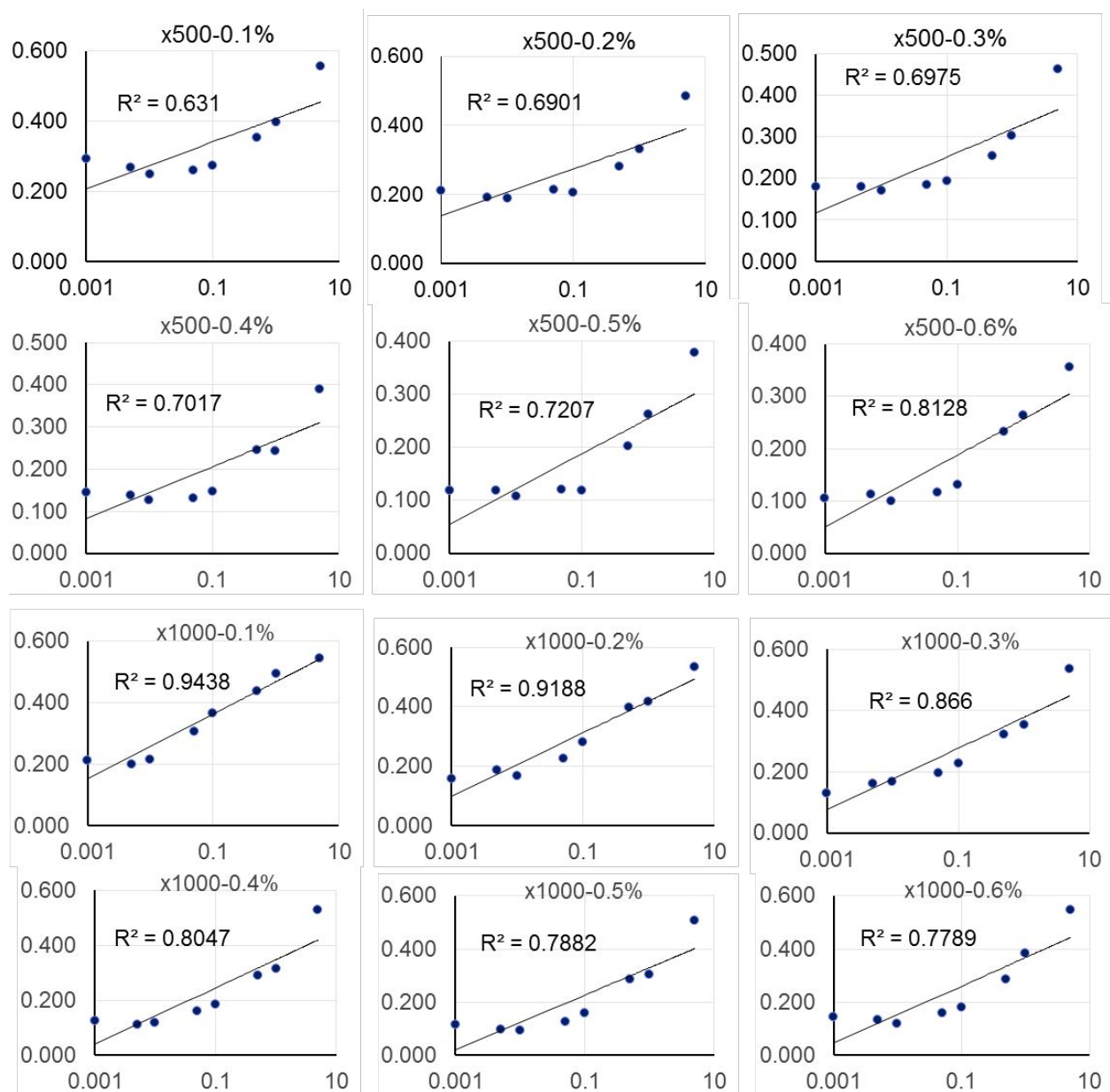


Figure S1. Optimization of sample dilution ratio of the plasma sample spiked at 0.001-10 $\mu\text{g/mL}$ and the concentration of skimmed milk.