

Supplementary Materials: Aptamer-based Sandwich Assay Formats for Detection and Discrimination of High- and Low-Molecular-Weight uPA for Cancer Prognosis and Diagnosis

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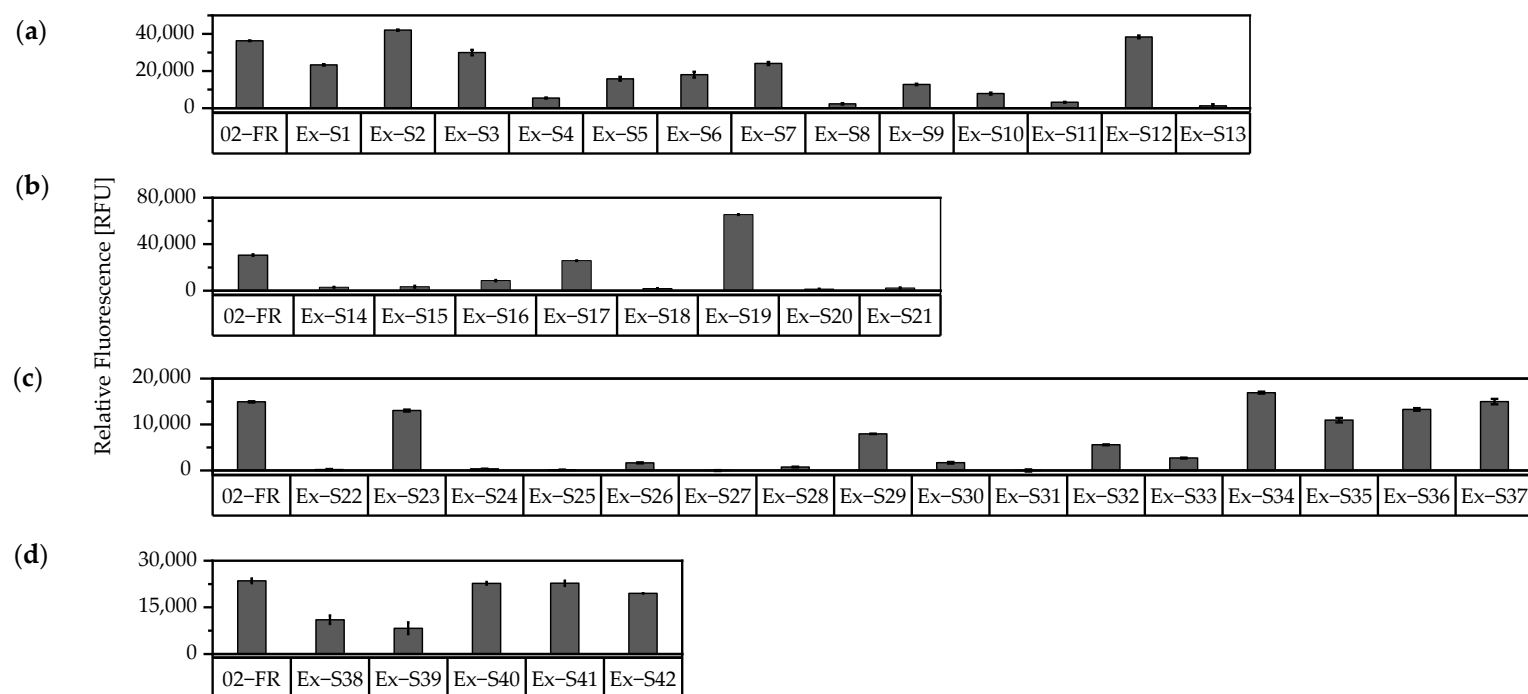


Figure S1. Binding of the exchange sequences (Ex-S1 – Ex-S42) to HMW-uPA by FLAA. Binding experiments were performed on different micro titer plates and are therefore shown as individual diagrams with the respective positive control uPAapt-02-FR (02-FR). (a) Ex-S1 – Ex-S13 (b) Ex-S14 – Ex-S21 (c) Ex-S22 – Ex-S37 (d) Ex-S38 – Ex-S42. The relative fluorescence unit [RFU] for each sample is given as the mean value of technical replicates and was measured using the multimode microplate reader Mithras² LB943. Negative control values were subtracted. Error bars represent the range of measured values of signals from the two target-coated wells plus the range of the measured values of the two negative control wells. Number of records $n=2$.

Table S1. Exchange sequences of uPAapt-02-FR. Each nucleotide of the sequence of uPAapt-02-FR was replaced individually (for G/C a T and for A/T a C). All 42 new sequence aptamers (Ex-S1 – Ex-S42) were tested for binding to HMW-uPA by FLAA. The reduction in fluorescence signal compared to the control uPAapt-02-FR is given as a percentage for each sequence. Exchanged nucleotides in each sequence are shown in red.

Name	Sequence	Signal reduction compared to control (uPAapt-02-FR)
Ex-S1	TAAGCGGGGGTGAGAGATCTGTCAGTACGAGCTGGGTTTGCG	36%
Ex-S2	CCAGCGGGGGTGAGAGATCTGTCAGTACGAGCTGGGTTTGCG	n.r.*
Ex-S3	CAACGCGGGGGTGAGAGATCTGTCAGTACGAGCTGGGTTTGCG	18%
Ex-S4	CAATCGGGGGTGAGAGATCTGTCAGTACGAGCTGGGTTTGCG	85%
Ex-S5	CAAGTCGGGGGTGAGAGATCTGTCAGTACGAGCTGGGTTTGCG	56%
Ex-S6	CAAGCTGGGGGTGAGAGATCTGTCAGTACGAGCTGGGTTTGCG	50%
Ex-S7	CAAGCGTGGGGTGAGAGATCTGTCAGTACGAGCTGGGTTTGCG	34%
Ex-S8	CAAGCGGTGGGTGAGAGATCTGTCAGTACGAGCTGGGTTTGCG	94%
Ex-S9	CAAGCGGGTGTGAGAGATCTGTCAGTACGAGCTGGGTTTGCG	65%
Ex-S10	CAAGCGGGGTGAGAGATCTGTCAGTACGAGCTGGGTTTGCG	78%
Ex-S11	CAAGCGGGGGCGAGAGATCTGTCAGTACGAGCTGGGTTTGCG	91%
Ex-S12	CAAGCGGGGGTTAGAGATCTGTCAGTACGAGCTGGGTTTGCG	n.r.*
Ex-S13	CAAGCGGGGGTGCGAGATCTGTCAGTACGAGCTGGGTTTGCG	97%
Ex-S14	CAAGCGGGGGTGATAGATCTGTCAGTACGAGCTGGGTTTGCG	91%
Ex-S15	CAAGCGGGGGTGAGCGATCTGTCAGTACGAGCTGGGTTTGCG	89%
Ex-S16	CAAGCGGGGGTGAGATATCTGTCAGTACGAGCTGGGTTTGCG	72%
Ex-S17	CAAGCGGGGGTGAGAGCTCTGTCAGTACGAGCTGGGTTTGCG	16%
Ex-S18	CAAGCGGGGGTGAGAGACCTGTCAGTACGAGCTGGGTTTGCG	94%
Ex-S19	CAAGCGGGGGTGAGAGATTGTCAGTACGAGCTGGGTTTGCG	0%*
Ex-S20	CAAGCGGGGGTGAGAGATCCGTCAGTACGAGCTGGGTTTGCG	95%
Ex-S21	CAAGCGGGGGTGAGAGATCTTTCAGTACGAGCTGGGTTTGCG	92%
Ex-S22	CAAGCGGGGGTGAGAGATCTGCCAGTACGAGCTGGGTTTGCG	99%
Ex-S23	CAAGCGGGGGTGAGAGATCTGTAGTACGAGCTGGGTTTGCG	13%
Ex-S24	CAAGCGGGGGTGAGAGATCTGTCGTACGAGCTGGGTTTGCG	98%
Ex-S25	CAAGCGGGGGTGAGAGATCTGTCA TTACGAGCTGGGTTTGCG	99%
Ex-S26	CAAGCGGGGGTGAGAGATCTGTCAGCACGAGCTGGGTTTGCG	89%
Ex-S27	CAAGCGGGGGTGAGAGATCTGTCAGTCCGAGCTGGGTTTGCG	100%
Ex-S28	CAAGCGGGGGTGAGAGATCTGTCAGTATGAGCTGGGTTTGCG	95%
Ex-S29	CAAGCGGGGGTGAGAGATCTGTCAGTACTAGCTGGGTTTGCG	47%
Ex-S30	CAAGCGGGGGTGAGAGATCTGTCAGTACCGCTGGGTTTGCG	89%
Ex-S31	CAAGCGGGGGTGAGAGATCTGTCAGTACGATCTGGGTTTGCG	100%
Ex-S32	CAAGCGGGGGTGAGAGATCTGTCAGTACGAGTTGGGTTTGCG	63%
Ex-S33	CAAGCGGGGGTGAGAGATCTGTCAGTACGAGCCGGGTTTGCG	82%
Ex-S34	CAAGCGGGGGTGAGAGATCTGTCAGTACGAGCTTGGTTTGCG	0%*
Ex-S35	CAAGCGGGGGTGAGAGATCTGTCAGTACGAGCTGTGTTTGCG	27%
Ex-S36	CAAGCGGGGGTGAGAGATCTGTCAGTACGAGCTGGTTTTGCG	11%
Ex-S37	CAAGCGGGGGTGAGAGATCTGTCAGTACGAGCTGGGCTTGCG	n.r.*
Ex-S38	CAAGCGGGGGTGAGAGATCTGTCAGTACGAGCTGGGTCTGCG	53%
Ex-S39	CAAGCGGGGGTGAGAGATCTGTCAGTACGAGCTGGGTTCCG	65%
Ex-S40	CAAGCGGGGGTGAGAGATCTGTCAGTACGAGCTGGGTTTTCG	3%
Ex-S41	CAAGCGGGGGTGAGAGATCTGTCAGTACGAGCTGGGTTTGTG	3%
Ex-S42	CAAGCGGGGGTGAGAGATCTGTCAGTACGAGCTGGGTTTGCT	17%

Note: n.r.*, no reduction (equal or higher signal compared to control uPAapt-02-FR).