

Supplemental Materials

Molecular Biology of the Cell

Leśniewska et al.

Legends for Supplementary Figures

Figure S1: Peptide aptamers (A) Free or single peptide aptamers and double constrained aptamers. In a free (or double constrained) aptamer, a short peptide is fused to the end of the protein. In a double constrained aptamer, a short peptide is inserted into a small scaffold protein, such as thioredoxin. (B) An aptamer has a high affinity to a protein of interest (EB1), and competes with an endogenous interacting protein (SxIP-containing protein) for binding.

Figure S2: The composition of the unselected SxIP library. (A) The DNA sequences of 39 random unselected clones from the SxIP prey library. The sequences were a heterogeneous mixture of nucleotides. K = G/T, n = A/T/C/G. (B) The amino acid sequences encoded by unselected clones from the prey SxIP library. "-" represents a stop codon. (C) The frequencies of amino acids for each position encoded by these unselected prey clones. Each position was tested for statistical differences from the frequency expected from random DNA sequences. "-", $p \geq 0.05$. "±", $0.01 \leq p < 0.05$. No significant differences were observed.

Figure S3: Protein sequences of EB1 aptamers isolated from the SxIP library. The amino acid sequences encoded by 45 random EB1 aptamers isolated from the XXXX library based on the aptamer Perfect sequence in which SRIP was replaced with 4 random residues.

Figure S4: The compositions of the unselected XXXX library and double constrained library. (A) The amino acid sequences encoded by 17 random unselected prey clones from the XXXX library. "-" represents a stop codon. (B) The frequencies of amino acids for each position encoded by these unselected prey clones. Each position was tested for statistical differences from the frequency expected from random DNA sequences. "-", $p \geq 0.05$. "±", $0.01 \leq p < 0.05$. "+", $p < 0.01$. (C) The amino acid sequences encoded by 10 random unselected prey clones from the double constrained SxIP-containing library. "-" represents a stop codon. (D) The frequencies of amino acids for each position encoded by these

unselected clones from double constrained SxIP-containing library. Each position was tested for statistical differences from the frequency expected from random DNA sequences. “–”, $p \geq 0.05$. “±”, $0.01 \leq p < 0.05$.

Figure S5: Strength of interaction between all double constrained aptamers and *Drosophila* EB1. It is measured by a quantitative assay for β -galactosidase activity and normalised for the cell density (A_{420}/A_{600}). The five strongest ones were selected for further assays in triplicates together with aptamer 37 (Figure 2B).

Figure S6: Modelled structures of aptamer Perfect and an SxIP fragment of Sentin. *Drosophila* EB1 (green) complexed with aptamer Perfect (purple) or an SxIP fragment of the endogenous EB1 interactor Sentin (yellow) was modelled using a crystal structure of the complex between human EB1 Δ C8 and MACFp1 as a template. Some residues of *Drosophila* EB1 have changed their orientation after energy minimisation in comparison to human EB1 (pale blue). Aptamer Perfect is likely to interact with EB1 more strongly than the fragment of Sentin. Buried surface areas are estimated to be 3100.2 Å² with aptamer Perfect in comparison to 2972.7 Å² with the Sentin fragment. The main differences are W and V at the positions +2 and +3 on aptamer Perfect (highlighted in yellow), in contrast to P and S on Sentin.

Figure S7: (A) Residues at X, +1–+5 (underlined) in 18 known human EB1-interacting sequences are highlighted when they match to residues significantly overrepresented among aptamers to HsEB1 (green), HsEB3 (purple) or both (blue). (B) Residues at X, +1–+5 of SxIP sequences in three human proteins known to interact specifically with EB3, not EB1, are highlighted when they match to residues significantly overrepresented among aptamers to HsEB1 (green), HsEB3 (purple) or both (blue).

Figure S8: The sequences of oligonucleotides and DNA synthesised for this study. K = G/T,
n = A/T/C/G.

Legends for Supplementary Figures

Movie S1: Aptamer Perfect fused to GFP in haemocytes from third instar larvae carrying *Act5C-GAL4* and *UASp-GFP-aptamer Perfect*. This demonstrates that the aptamer is expressed in haemocytes and localised as expected. The width of the frame is 51 μm . The images were taken every two seconds.

Movie S2: Growing microtubule plus ends in haemocytes from third instar larvae carrying *Ub-EB1-GFP* and *Act5C-GAL4*. EB1-GFP marks microtubule plus ends. The width of the frame is 51 μm . The images were taken every two seconds.

Movie S3: Growing microtubule plus ends in haemocytes from third instar larvae carrying *Ub-EB1-GFP*, *Act5C-GAL4* and *UASp-GFP-aptamer Perfect*. The average growth rate is significantly slower in haemocytes expressing the aptamer and EB1-GFP than the control (Movie S2) only expressing EB1-GFP. The width of the frame is 51 μm . The images were taken every two seconds.

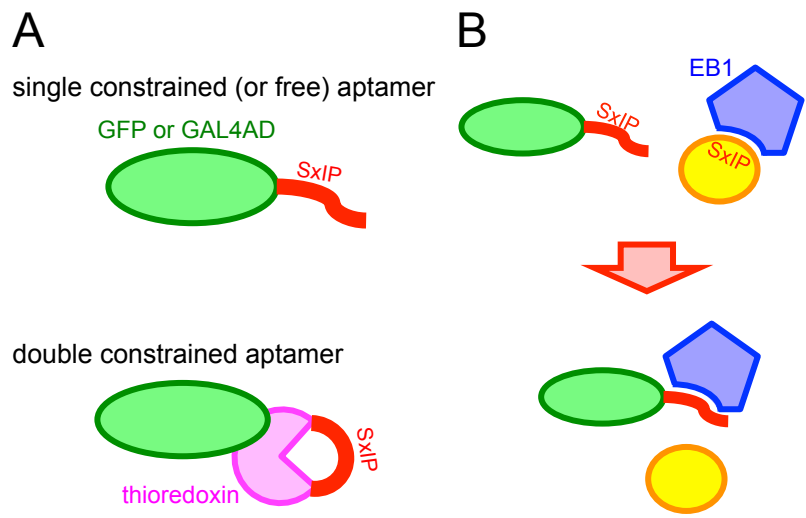


Figure S1

A unselected SxIP library

nnKnnKnnKnnKnnKTCCnnKATTCAnnKnnKnnKnnKnnKnnKnnKTGA
 1 GAGTTGTACTTGGTCCCTGGATTCCATCGGATGTTTGGGGGTGGCTTGA
 2 TTTATGCAGAGTAGGTCCGGTATTCCAGAGATGGTGAGGGGGTGGGGTGA
 3 GGTTCGTGGTCCGGTCCATTATTCCAATGGTTCGGTGGCTCAAGGGTGA
 4 GCTGGGCGTATTGGGTCCACTATTCCAAGGCAGTTCATGTGGATCATTGA
 5 ACTGAGACGCAGAAATCCAGTATTCCAGCTTTGTGGCGTGGCTTAGTTGA
 6 CAGTTGGCTTTGGTGTCCGCGATTCCAAGTGTAGTCTTTATAAGCTTTGA
 7 GTTGTGCGTGGGTTCCGTTATTCCAATGTGGGGTCAAGGGGGGGGGTGA
 8 AGGCGGATGCATCCTTCCAGTATTCCACAGCGGGTGTATTGGGCGAGGTGA
 9 TCGTAGTCGCTGTTGTCCCGTATTCCATAGATGCCGGAGCGGGTGTGTTGA
 10 GGTCTGCATGTGTTTTCCAGATTCCACTTCATGTTATGACGAGGGTGTGA
 11 CTGAAGCGAGTTTGTCCAATATTCCACCGTTGAGGGTTCAGACTAGGTGA
 12 AGGTTTTTGTCTCAGTCCAATATTCCAATGTAGTCTTCTTGTATTTGA
 13 GGGCTTGGGTTGGGGTCCGCGATTCCACTGGTGGATCGGGATGGTGCTTGA
 14 ATTGGGAGGACGCTTCCGGGATTCCACCCTGTCCGTATCGTTCGGGTTGA
 15 CGTGTGAGTATTGGTCCATGATTACAAATGTTTTACCTCGCATGATTGA
 16 TGGGAGTTGTTGGGGTCCCGATTCCATGGTATCCTTTTACTTATCGGTGA
 17 TCGCCTAGTGGTCCGGTCCAGGATCCAGGGCGGATATGGGGAGGTTTTGA
 18 GGGCGGCTGGGGCGTCCGCTATTCCATGTCGTTATAGGAGGAATGATGA
 19 CTGTGTTGAGTCTGCTCGGATTCCATAGTTTGGTGGGTGTATCAGTGA
 20 GGGTGGTTCGGGTAGTCCCTATTCCCGAGTTGGGGGGAATGTTCTTGA
 21 GGTATGTGTTGTTATCCGGGATTCCAGTGGGGTGGGGGGTCTTAAGTGA
 22 TCGCGGCTTTGTGCGTCCCTATTCCAGATCTTTAGGAGATGAGGAGTTGA
 23 CCTCGGTAGCGTACGTCCTGTATTCCAACGGCGTGTCTGGAGCGGTATTGA
 24 TGTGGTGGGGTTATTCCTAGATTCCAAGTCGGAGTGGTATGTTTCAGTGA
 25 GGTGTTTTGGTGTTCCTCCGATATTCCAAGGGTACGCCGCATCAGGGTTGA
 26 CAGTATGGTAGTATTCCCTGGATTCCAATGCGGGGGGGTATGATTGTTGA
 27 GGTGTGTTGAATCCGTCCTGTATTCCAGGGCGTTCGGTGGGTCGGCTTGA
 28 CAGGAAGTTGATGGTCCGGGATTCCAACGGCGGCGCATGGGTTATTTGA
 29 ATTAGTGGACGAGTCCATTATTCCAGCGGTGCGGTTGATTAAGTTGTTGA
 30 CCGCGGAGTGGCGTGTCCGAGATTCCATCGACTTGTGGGCTATTTAGTGA
 31 AGGCCGTGTCGGTGGTCCGGTACTCCATAGTAGAGTGTGAATGCCGAGTGA
 32 CGGTAGAGTTATCAGTCCATTATTCCAGAGTAGTAGCGTTGGCGGTCTTGA
 33 CTGAGGTATTCTGGTCCGGGATTCCATGGGTGAGGGGGCTGCTTGAGTGA
 34 AGTTTGGCGTGGCGGTCCAGGATTCCAAAGGGTAGTTCGATTCCGGTTGGA
 35 TATTCTGGTGGAGTCCTAGATTCCATTGAAGCTGGTGTGTACGATCGTG
 36 TGGGATGCGATGGGGTCCCTCGATTCCATGGAGGTAGCATCGGCCTTTTGA
 37 CAGTGTGGTGCATTCCCTGGATTCCAGGGGTGGTGGCGGGCCACTTGT
 38 CGGTTGTTGGTGGGTTCCCATATTCCATAGAAGTGGTGTGGACGGTGTGA
 39 CCGTGTGTGATGGGTTCCCGTATTCCACATTCTCGGGTGGATTCCGGTGA

B

1 **ELYTWSWIPSDVCGVA-**
 2 **FMQSRSGIPEMVRGLG-**
 3 **GSWCGSIIPMVRLAQG-**
 4 **AGRIGSTIPRQLHVDH-**
 5 **TE'QNSSIPALWRWLS-**
 6 **QLALVSAIPSVSLYKL-**
 7 **VVRAVSVIPLWGQGGG-**
 8 **RRMHPSSIPQRVDWAR-**
 9 **S-SLLSRIP-MPERGC-**
 10 **GLHVFSKIPLHVMTRV-**
 11 **LKASLSNIPRLRVQTR-**
 12 **RFLSQSNIPMLSLSCI-**
 13 **GLGLGSAIPLVDRDGA-**
 14 **IGRTSSGIPPCPYRSG-**
 15 **RVSI GSMITNCF'FSHD-**
 16 **WELLGSSIPWYPFYR-**
 17 **SPSGRSRIPGADMGRF-**
 18 **GRPGASAI PCRYRRE--**
 19 **LCCSLSWIP-FGEVYQ-**
 20 **GWLR-SSIPQLGGNCS-**
 21 **GMCWLSGIPVGLGMLK-**
 22 **SRLCASLIPDL-EMRS-**
 23 **PR-RTSCIPTACLERY-**
 24 **CGAGYS-IPSRSG-FQ-**
 25 **GVLVVDIPRVTPHQG-**
 26 **QYGSISWIPMRGGYDC-**
 27 **GVLNPSCIPRSLGSA-**
 28 **QEVDSGSIPTAAHWVI-**
 29 **IRWTSSII PAVRLIKL-**
 30 **PRSAVSEIPSTCWAI--**
 31 **RPCRWSGTP--SVNAQ-**
 32 **R-SYQSIPE--RWRS-**
 33 **LRYSWSGIPWVRGLLE-**
 34 **SLAWRSRIPKGRSIPVG**
 35 **YSGE-S-IPLKLVCTIV**
 36 **WDAMGSSIPWR-HAAF-**
 37 **QCWCSSWIPGVDRPTC**
 38 **RLLRGRSHIP-KWCQTV-**
 39 **PCV-GSRIPHSRVDSR-**

C

	position																Average amino acid occurrence		
	-5	-4	-3	-2	-1	S	x	I	P	+1	+2	+3	+4	+5	+6	+7	+8	observed	expected
Ala	1	0	5	2	2	0	3	0	0	2	3	1	0	3	3	3	0	2.15	2.44
Arg	6	7	3	4	3	0	4	0	0	3	6	6	6	3	5	4	0	4.62	3.66
Asn	0	0	0	1	1	0	2	0	0	1	0	0	0	2	0	0	0	0.54	1.22
Asp	0	1	0	1	0	0	1	0	0	1	1	2	1	2	2	1	0	1.00	1.22
Cys	1	3	3	3	0	0	2	0	0	1	2	2	2	1	2	2	1	1.92	1.22
Glu	1	3	0	1	0	0	1	0	0	2	0	0	3	1	1	1	0	1.08	1.22
Gln	4	0	1	1	2	0	0	0	0	2	1	0	1	1	2	3	0	1.38	1.22
Gly	8	3	3	3	9	0	6	0	0	3	2	4	5	8	3	5	1	4.85	2.44
His	0	0	1	1	0	0	1	0	0	1	1	0	3	1	1	1	0	0.85	1.22
Ile	2	0	0	2	1	0	3	38	0	0	0	0	0	2	1	3	0	1.08	1.22
Leu	3	6	7	4	4	0	1	0	0	4	5	3	5	1	4	2	0	3.77	3.66
Lys	0	1	0	0	0	0	1	0	0	1	2	0	0	0	2	1	0	0.62	1.22
Met	0	2	1	1	0	0	1	0	0	3	2	0	2	1	0	0	0	1.00	1.22
Phe	1	1	0	0	1	0	0	0	0	0	1	1	1	0	1	2	0	0.69	1.22
Pro	3	2	1	0	2	0	0	0	38	1	0	3	1	0	2	0	0	1.15	2.44
Ser	4	2	5	6	2	39	5	0	0	4	1	5	1	2	3	4	0	3.38	3.66
Thr	1	0	1	3	1	0	1	1	1	2	1	1	1	2	3	1	0	1.54	2.44
Trp	2	1	3	2	3	0	4	0	0	3	1	2	1	4	0	0	0	2.00	1.22
Val	1	4	2	2	4	0	1	0	0	1	7	5	5	2	2	3	1	3.08	2.44
Tyr	1	1	2	1	2	0	0	0	0	0	1	1	1	2	2	1	0	1.15	1.22
*	0	2	1	1	2	0	2	0	0	4	2	3	0	1	0	1	36	1.46	1.22
Different from expected	±	-	-	-	±	NA	-	NA	NA	-	-	-	-	-	-	±	NA		

Figure S2

* * *

1 RTRHVSLIPRWVDRT-
2 KYKWRSRFQSGRMLG
3 GAVKNSRIPRYIGWRG-
4 ITTRPSLIPRWVGRGG-
5 GARGRSKIPRWSAGG-
6 LTASTSRIPPLGVWEFV
7 GTPRRSRIPRWRNRGG-
8 KERHTSRIPRWQSGNS-
9 VARRQSLIPWRGRSR-
10 VARFRSRIPVFVGSGR-
11 RAVSRSRIPWRRAMG-
12 SPGGLSRIPRWTGL-GC
13 AGMFKSRIPIRRRVHKP-
14 REFGRSLIPWRGRGTG-
15 RNCKPSRIASDGEVEVL
16 SACAGSRIPVWRGRCG-
17 RFFQKSRIPRWTRGHD-
18 QICARSRIPKWVGRNS-
19 KWKVKSIPVKAARM-
20 PVRQSSSIPRWKGGRV-
21 KNCGKSRIPVLQRRVS-
22 ARVFGSRIPVRTAPSK-
23 FPPYRSRIPVVR-VL-
24 QRRAVSRIPRPLKVPS-
25 AISRRSRIPVMLRSQR-
26 RWMSVSRIPVLRGRR-
27 RCVSRSKIPKLCLSWYL
28 SNGWKSRIPRWCQR-W-
29 LTVAQSRIPRYLGRRS-
30 SPLRVSKIPRWLGWNR-
31 KMRGVSKIPIRKANSS-
32 RGKMPSLIPRWGGVGK-
33 ERRVRSVIPVWRGWRG-
34 HGSYRSVIPRWIGWKG-
35 NGGGRSRIPVYRLRRG-
36 VVVRASKIPRWVGRLP-
37 RWVGSRIPRWVGWES-
38 RRCRRSRIPRWFRGRL-
39 RPSGVSGIPRWRGRVG-
40 RTNFVSRIPRWRG-RLG
41 LQSRRSRIPRWVGCRQ-
42 RRRVVSkipRWCGAPT-
43 RGGWRSRIPRWGNVW-
44 KKNW-SRIPVMALESSV
45 AGGLRSRIPVVNACGL

Figure S3

A unselected XXXX library

```

*****      *****
1  RTRGRRWRFRWVGRRG-
2  RTRGRLFCSRWVGRRG-
3  RTRGRSGNFRWVGRRG-
4  RTRGRDLVFRWVGRRG-
5  RTRGR-HAARWVGRRG-
6  RTRGRRGRARWVGRRG-
7  RTRGRLRGCRWVGRRG-
8  RTRGRWPFLRWVGRRG-
9  RTRGRLELPRWVGRRG-
10 RTRGRVEYLRWVGRRG-
11 RTRGRL-LRRWVGRRG-
12 RTRGRR-YFRWVGRRG-
13 RTRGRL-LRRWVGRRG-
14 RTRGRMGRWRWVGRRG-
15 RTRGRLILFRWVGRRG-
16 RTRGR-KMLRWVGRRG-
17 RTRGREWSLRWVGRRG-

```

B

	Position				Average amino acid occurrence	
	X1	X2	X3	X4	observed	expected
Ala	0	0	1	2	0.75	1.13
Arg	3	1	3	2	2.25	1.69
Asn	0	0	1	0	0.25	0.56
Asp	1	0	1	0	0.25	0.56
Cys	0	0	1	1	0.50	0.56
Glu	1	2	0	0	0.75	0.56
Gln	0	0	0	0	0.00	0.56
Gly	0	3	1	0	1.00	1.13
His	0	1	0	0	0.25	0.56
Ile	0	1	0	0	0.25	0.56
Leu	6	1	4	4	3.75	1.69
Lys	0	1	0	0	0.25	0.56
Met	1	0	1	0	0.50	0.56
Phe	0	1	1	6	2.00	0.56
Pro	1	1	0	1	0.75	1.13
Ser	1	0	1	1	0.75	1.69
Thr	0	1	0	0	0.25	1.13
Trp	1	2	0	1	1.00	0.56
Val	1	0	1	0	0.50	1.13
Tyr	0	0	2	0	0.50	0.56
*	2	3	0	0	1.25	0.56
Different from expected	-	-	-	+		

C unselected double constrained library

```

* **
FKYLMSYIPYWCIRWR
RSIVSWSVIPINWLAGR
NACYYSWIPLWQRLWI
RTVSCSYIPLRDSWR
ERMGFSFIPLSRWGLR
TAMILSTIPMGGT-SW
VGRQKSRIPLSCATR
RPRVSSRIPWNAVDWE
GDYASLIPKDVTYVP
EWGLSQIPVSVYYVS

```

D

	position														Average amino acid occurrence			
	-5	-4	-3	-2	-1	S	x	I	P	+1	+2	+3	+4	+5	+6	+7	observed	expected
Ala	0	2	0	0	1	0	0	0	0	0	0	1	1	2	0	0	0.54	0.63
Arg	3	1	2	0	0	0	2	0	0	0	1	1	1	0	2	3	1.23	0.94
Asn	1	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0.23	0.31
Asp	0	0	1	0	0	0	0	0	0	0	1	1	0	1	0	0	0.31	0.31
Cys	0	0	1	0	1	0	0	0	0	0	0	2	0	0	0	0	0.31	0.31
Glu	1	1	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0.23	0.31
Gln	0	0	0	1	0	0	1	0	0	0	0	1	0	0	0	0	0.23	0.31
Gly	0	2	0	2	0	0	0	0	0	0	1	1	0	1	1	0	0.62	0.63
His	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.00	0.31
Ile	0	0	1	1	0	0	0	10	0	1	0	0	1	0	0	1	0.38	0.31
Leu	0	0	0	1	2	0	1	0	0	4	0	0	1	1	1	0	0.85	0.94
Lys	0	1	0	0	1	0	0	0	0	1	0	0	0	0	0	0	0.23	0.31
Met	0	0	2	0	1	0	0	0	0	1	0	0	0	0	0	0	0.31	0.31
Phe	1	0	0	0	1	0	1	0	0	0	0	0	0	0	0	0	0.23	0.31
Pro	0	1	0	0	0	0	0	10	0	0	0	0	0	0	1	0	0.15	0.63
Ser	0	1	0	1	1	10	0	0	0	0	3	0	1	0	1	1	0.69	0.94
Thr	1	1	0	0	0	0	1	0	0	0	0	0	2	0	1	0	0.46	0.63
Trp	0	0	1	0	1	0	1	0	0	1	2	1	1	2	2	2	1.08	0.31
Val	1	0	1	2	0	0	1	0	0	1	0	2	1	0	2	0	0.85	0.63
Tyr	0	0	1	2	1	0	2	0	0	1	0	0	1	2	0	0	0.77	0.31
*	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0.08	0.31
Different from expected	-	-	-	-	-	NA	-	NA	NA	-	±	-	-	±	-	-		

Figure S4

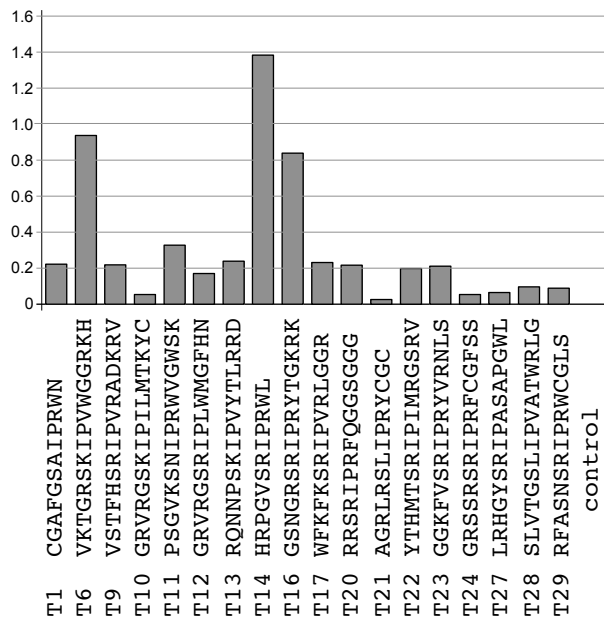


Figure S5

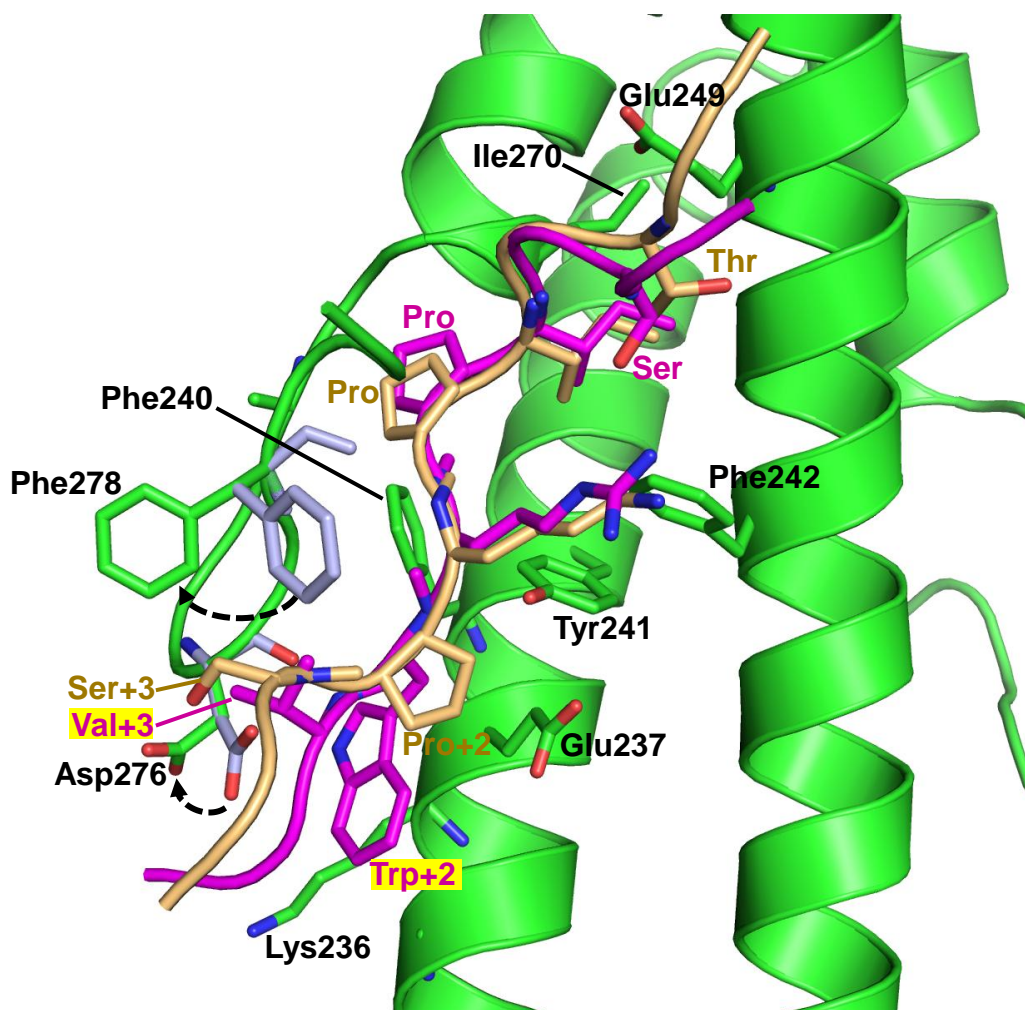


Figure S6

A

known human EB1-
interacting sequences

CLASP1 GLARS**SRIP**PSMSQG
CLASP2 SAQKR**SKIP**RSQ**G**CSR
CLASP2 SVARS**SRIP**PSVSQG
MACF1 GLNKP**SKIPT**MSKKTT
APC TSARP**SQIPT**PVNNNT
STIM1 QASRN**TRIP**HL**G**KKA
MCAK RRSVN**SKIP**AP**K**ESLR
KI18B KRQRQ**SFLP**CL**R**RGSL
KI18B GPKPT**SSLP**GT**S**ACKK
SLAIN2 FQVPNGG**IP**RMQ**P**QAS
SLAIN2 MQPQA**SAIP**SPG**K**FRS
SLAIN2 TTAMR**SGLP**PSAPSA
SLAIN2 AQPVRR**SLP**AP**K**TYGS
SLAIN2 SAPSAGG**IP**VPRSKLA
SALIN2 AQPVRR**SLP**AP**K**TYGS
CK5P2 REAKK**SRLP**IL**I**KPSR
PSRC1 AVKSS**SRLP**VPSAIPK
PSRC1 RLPVP**SAIP**KPAT**R**V
* - **

residues overrepresented
among aptamers to
HsEB1 only (11)
HsEB3 only (5)
Both (13)

B

known human proteins that
interact specifically with EB3

ANK2
EQKGR**SKIP**I**R**V**K**GKE
EVQPP**SPLP**SSMDSNS
FTESK**SKIP**V**R**TMPTS
EAKPK**SKLP**V**K**VPLQR
PKILT**SRLP**V**K**SRSTT
* - **

Drebrin
RRMAP**TPIPT**R**S**PSDS
* - **

Myosin 1B
no SxIP

residues overrepresented
among aptamers to
HsEB1 only (2)
HsEB3 only (5)
Both (7)

Figure S7

DmEB1 bait plasmid

oKMT17:cggaagagagtagtaacaaaggtcaaagacagttgactgtatcgccggaatggctgtaaactctactc
oKMT18:agaaattcgcccgaattagcttggtgcaggtcgacggatccccgggaattaatactctctcctctg

HsEB1 bait plasmid

oKMT82:cggaagagagtagtaacaaaggtcaaagacagttgactgtatcgccggaatggcagtgaaactataactcaac
oKMT83:agaaattcgcccgaattagcttggtgcaggtcgacggatccccgggaattaatactctctctctctctct

HsEB3 bait plasmid

oKMT84:cggaagagagtagtaacaaaggtcaaagacagttgactgtatcgccggaatggcgtcaatgtgtactccac
oKMT85:agaaattcgcccgaattagcttggtgcaggtcgacggatccccgggaatcagactcgtcctggtctctct

SxIP prey library

oKMT34:ACTTGCGGGGTTTTTTCAGTATCTACGATTCATAGATCTCTCGAGCTCGAA
oKMT46:TACGCTAGCTTGGGTGGTCATATGGCCATGGAGGCCCGGGGATCCGAnnknnknnknnknnkTCCnnkATTCCAn
nknnknnknnknnknnknnkTGATTCGAGCTCGAGAGATCTATG

XXXX prey library

oKMT34:ACTTGCGGGGTTTTTTCAGTATCTACGATTCATAGATCTCTCGAGCTCGAA
oKMT68:TACGCTAGCTTGGGTGGTCATATGGCCATGGAGGCCCGGGGATCCGACGAACCCGAGGCCGAnnknnknnknnk
GATGGGTGGGCCGACGAGGCTGATTCGAGCTCGAGAGATCTATG

Double constrained SxIP prey library

oKMT64:ATATTGCTCAGCGATTTTCATCCAGAATCGGGGCGATCATTTTGCACGG
oKMT63:AAAGCGGACGGGGCGATCCTCGTCGATTTCTGGGCAGAGTGGTGCGGTnnknnknnknnknnkTCCnnkATTCCAn
nknnknnknnknnknnknnkCCGTGCAAAATGATCGCCCCGATT

The prey plasmid for tetramer of aptamer 37

pKMT169:
ATCTGTATGGCTTACCCATACGATGTTCCAGATTACGCTAGCTTGGGTGGTCATATGGCCATGGAGGCCCGGGGATCCGA
cgt tgt gtt tcc cgt tcc aaa att cca aaa ttg tgt ttg tcc tgg tat ttg att cgt gct cgt gaa att tat gaa tcc tcc ggt tcc ggt
agg tgt gtg tgg agg tgg aaa ata cct aaa ctt tgt ctt tgg tgg tat ctt ata agg gca agg gaa ata tat gaa tgg tgg ggc agt ggc
aga tgc gtc agt aga agt aag atc ccc aag ctg tgc ctg agt tgg tac ctg atc aga gcc aga gag atc tac gag agt agt gga tgg gga
cgt tgc gta agc aga agc aag ata ccg aaa ttg tgc ttg agc tgg tac ctt att agg gcg aga gaa att tac gaa agc
TAGTTCGAGCTCGAGAGATCTATGAATCGTAGATACTGAAAAACCCCGCAAGTTCACCTCAACTGTGCATCGTG

The prey plasmid for septamer of aptamer Perfect

pKMT170:
ATCTGTATGGCTTACCCATACGATGTTCCAGATTACGCTAGCTTGGGTGGTCATATGGCCATGGAGGCCCGGGGATCCGA
cgt acg cgt ggt cgt agc cgt att ccg cgt tgg gtg ggt cgt cgt ggt tcc ggt agt ggt
cgt acc aga ggt cgt tcc cgt ata cca cgt tgg gta ggt aga agg ggt agc gcc tcc gcc
aga act aga gcc aga tgg aga atc ccc agg tgg gtc gcc cgt aga gcc agt gga agc gga
aga acg aga ggt aga agc aga atc ccg aga tgg gtg gcc aga aga gcc tgg gcc agc ggt
agg aca agg gga agg agt agg ata cct cgt tgg gtc ggt agg agg gga tcc gga tgg gcc
agg acg cgt ggt agg agc agg att ccg agg tgg gtg gga cgt cgt gga agt ggt tcc gga
cgt act agg ggt aga tgg cgt atc cct aga tgg gta gga aga cgt gcc
TAGTTCGAGCTCGAGAGATCTATGAATCGTAGATACTGAAAAACCCCGCAAGTTCACCTCAACTGTGCATCGTG

The prey plasmid for dimer of aptamer 37

oKMT31:GTATGGCTTACCCATACG
oKMT69:ATTCATAGATCTCTCGAGCTCGAACTACGATTCATATATTTCCCTTGCCCT

Figure S8