

Supplementary Figures

Figure S1: gBlock sequence (IDT DNA Technologies, Codon Optimized for E. coli K-12).

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CTCGAGGGAAGTGAAGCGGAAGTGTGACTGTTTCATAGCAGTGAGCCCGAGGTGCGCATTCCGG
AGAACATCCAGTGAAATTGAGTTGTGCGTACAGTGGTTTTAGTTCACCACGTGTAGAATGGAA
GTTTGACCAGGGGGATACTACACGCCCTGTGTGTTATAATAATAAAAATCACCGCTAGCTACGAG
GACCGTGTCACTTTTTTACCAACAGGTATTACGTTCAAGTCCGTGACCCGCGAGGACACAGGCA
CGTATACATGCATGGTATCGGAAGAGGGCGGGAACCTGACGGGGAGGTTAAGGTTAAATTGAT
TGTATTAGTCCCACCCTCTAAACCCACAGTGAACATCCCTAGTCCGCAACAATCGGCAATCGT
GCCGTTTTAACTTGCTCAGAACAAGATGGTTACCACCCTCAGAATACACATGGTTAAGGACG
GTATCGTTATGCCTACCAATCCAAAGTCCACCCGTGCATTAGCAACTCCTCTTATGTGCTTAA
TCCCCTACCGGTGAATTAGTTCGACCCCTGTCCGCTAGTGATACAGGAGAATACTCGTGC
GAAGCCCGTAACGGGTATGGTACACCGATGACTTCCAATGCTGTCGGTATGGAAGCCGTCGAGC
GCAACGTCGGAGTCATCGTAGCGGGGTGTTAGTACAGTAAATTCTGCTGGGCATTTTGGTGT
CGGAATTTGGTTGCTACTCGCGTGGCCATTCGATCGCACAAAAAGGGCACTAGTTCGAAG
AAAGTTATTATAGTCAGCCATCTGCTCGCTCGGAGGGAGAGTTAAGCAGACATCGTCTTTTC
TTGTTCTCGAG
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Figure S2: Primers for PCR amplification.

	Forward Primer	Reverse Primer
JAM-A	atataCATATGggaagtggaagcggaag	tatataCTCGAGcagcagcattggaagtcac

Figure S3: pET28-MBP (Kanamycin resistance) Vector.

Note:

- a. Number of amino acids: 374. Molecular weight: 40992.61
- Sites for NdeI (HM) amino acids and XhoI (LE amino acids) are present in the plasmid. pET28-MBP-JAM-A-6xHIS construct.
- Number of amino acids: 588. Molecular weight: 64250.35.

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10      20      30      40      50      60
MGKIEEGKLV IWINGDKGYN GLAEVGGKFE KDTGIKVTVE HPDKLEEKFP QVAATGDGPD
70      80      90      100     110     120
IIFWAHDFG GYAQSGLLAE ITPDKAFQDR LYPFTWDAVR YNGKLIAYPI AVEALSIIYN
130     140     150     160     170     180
KDLLPNPPKT WEEIPALDKE LKAKGKSALM FNLQEPYFTW PLIAADGGYA FKYENGYDI
190     200     210     220     230     240
KDVGVNAGÄ KAGLFLVDL IKNKHMNADT DYSIAEAAFN KGETAMTING PWAWSNIDTS
250     260     270     280     290     300
KVNYGVTVLF TFKGQPSKPF VGVLSAGINA ASPNKELAKE FLENYLLTDE GLEAVNKDKP
310     320     330     340     350     360
LGAVALKSYE EELAKDPRIA ATMENAQKGE IMPNIPQMSÄ FWYAVRTAVI NAASGRQTVD
370
EALKDAQTNÄ AAHM -INSERT-LEHHHHHH
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10      20      30      40      50      60
MGKIEEGKLV IWINGDKGYN GLAEVGKKFE KDTGIKVTVE HPDKLEEKFP QVAATGDGPD

70      80      90      100     110     120
IIFWAHDRFG GYAQSGLLAE ITPDKAFQDK LYPFTWDAVR YNGKLIAYPI AVEALS LIYN

130     140     150     160     170     180
KDLLPNPKT WEEIPALDKE LKAKGKSALM FNLQEPYFTW FLIAADGGYA FKYENGYDI

190     200     210     220     230     240
KDVGVNAGA KAGLTFVLVDL IKNKHMNADT DYSIAEAAFN KGETAMTING PWAWSNIDTS

250     260     270     280     290     300
KVNYGVTVLP TFKGQPSKPF VGVLSAGINA ASPNKELAKE FLENYLLTDE GLEAVNKDKP

310     320     330     340     350     360
LGAVALKSYE EELAKDPRIA ATMENAQKGE IMPNIPQMSA FWYAVRTAVI NAASGRQTVD

370     380     390     400     410     420
EALKDAQTNA AAHMGSSGSGS VTVHSSEPEV RIPENNPVKL SCAYSGFSSP RVEWKFDQGD

430     440     450     460     470     480
TTRLVCYNNK ITASYEDRVT FLPTGITFKS VTREDTGTYT CMVSEEGGNS YGEVKVKLIV

490     500     510     520     530     540
LVPPSKPTVN IPSSATIGNR AVLTCSEQDG SPPSEYTWFK DGIVMPTNPK STRAFSNSSY

550     560     570     580
VLNPTTGELV FDPLSASDTG EYSCEARNGY GTPMTSNAVR LEHHHHHH
    
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Human JAM-A (GFP) fusion. In bold is GFP sequence.

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10      20      30      40      50      60
MGGLYKQSWF      SEPSDQIFVD      PTIHSTHPPA      AAAKLMGTKA      QVERKLLCLF      ILAILLCSLA

70      80      90      100     110     120
LGSVTVHSSE      PEVRIPENNP      VKLSCAYSGF      SSPRVEWKFD      QGDTTRLVCY      NNKITASYED

130     140     150     160     170     180
RVTFLLPTGIT      FKSVTREDTG      TYTCMVSEEG      GNSYGEVKVK      LIVLVPPSKP      TVNIPSSATI

190     200     210     220     230     240
GNRAVLTCSE      QDGSPPSEYT      WFKDGIVMPT      NPKSTRAFSN      SSVYLNPTTG      ELVFDPLSAS

250     260     270     280     290     300
DTGEYSCEAR      NGYGTPMTSN      AVRMEAVERN      VGVIVA AVL V      TLILLGILVF      GIWFAYSRGH

310     320     330     340     350     360
FDRTKKGTSS      KKV IYSQPSA      RSEGEFKQTS      SFLVLEGAAA      GMADPVSKGE      ELFTGVVPIL

370     380     390     400     410     420
VELDGDVNGH      KFSVSGE GEG      DATYGKLT LK      FICTTGKLPV      PWPTLVTTLT      YGVQCFSRYP

430     440     450     460     470     480
DHMQHDFFK      SAMPEGYVQE      RTIFFKDDGN      YKTRAEVKFE      GDTLVNRIEL      KGIDFKEDGN

490     500     510     520     530     540
ILGHKLEyny      NSHNVYIMAD      KQKNGIKVNF      KIRHNIEDGS      VQLADHYQQN      TPIGDGPVLL

550     560     570     580
PDNHLSTQS ALSKDPNEKR DHMLLEFVT AAGITLGME LYK
    
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Human E-CADHERIN (GFP) fusion

	10	20	30	40	50	60
MGPWSRSLSA	LLLLLQVSSW	LCQEPEPCHP	GFDAESYFTT	VPRRHLEGR	VLGRVNFEDC	
	70	80	90	100	110	120
TGRQRTAYFS	LDTRFKVGTD	GVITVKRPLR	FHNPQIHFLV	YAWDSTYRKF	STKVTLNTVG	
	130	140	150	160	170	180
HHRPPPHQA	SVSGIQAEEL	TFPNSSPGLR	RQKRDWVIPP	ISCPENEKGP	FPKNLVQIKS	
	190	200	210	220	230	240
NKDKEGKVFY	SITGQGADTP	PVGVFIERE	TGWLVTEPL	DRERIATYTL	FSHAVSSNGN	
	250	260	270	280	290	300
AVEDPMEILI	TVTDQNDNKP	EFTQEVFKGS	VMEGALPGTS	VMEVTATDAD	DDVNTYNAAI	
	310	320	330	340	350	360
AYTILSQDPE	LPDKNMFTIN	RNTGVISVVT	TGLDRESFPT	YTLVVQAADL	QGEGLSSTAT	
	370	380	390	400	410	420
AVITVTDND	NPPIFNPTY	KGQVPENEAN	VVITTLKVTD	ADAPNTPAWE	AVYTILNDDG	
	430	440	450	460	470	480
GQFVVTTNPV	NNDGILKTAK	GLDFEAKQQY	ILHVAVTNVV	PFEVSLTST	ATVTVDVLDV	
	490	500	510	520	530	540
NEAPIFVPPE	KRVESEDFG	VGQEITSYTA	QEPDTFMEQK	ITYRIWRDTA	NWLEINPDGTG	
	550	560	570	580	590	600
AISTRAELDR	EDFEHVKNST	YALIIATDN	GSPVATGTGT	LLLILSDVND	NAPIPEPRTI	
	610	620	630	640	650	660
FFCERNPKPQ	VINIIDADLP	PNTSPFTAEL	THGASANWTI	QYNDPTQESI	ILKPKMALEV	
	670	680	690	700	710	720
GDYKINLKLM	DNQNKDQVTT	LEVSVCDCEG	AAGVCRKAQP	VEAGLQIPAI	LGILGGILAL	
	730	740	750	760	770	780
LILLLLLLF	LRRRAVVKEP	LLPPEDDTRD	NVYYYDEEGG	GEEDQDFDLS	QLHRGLDARP	
	790	800	810	820	830	840
EVTRNDVAPT	LMSVPRYLPR	PANPDEIGNF	IDENLKAADT	DPTAPPYDSL	LVFDYEGSGS	
	850	860	870	880	890	900
EAASLSSLNS	SESDKDQDYD	YLNEWGNRFK	KLADMYGGGE	DDLEGAAAGM	ADPVSKGEEL	
	910	920	930	940	950	960

Human JAM-A Copper Potential Docking Binding Sites		
No.	Binding Residues	Score
1	85D, 86R	1.103
2	65D, 66Q	0.931
3	167F, 188Y, 206D	0.903
4	217G, 218Y	0.877
5	102E, 103D	0.876
6	162S, 163E	0.839
7	112S, 113E	0.803
8	67G, 68D	0.779
9	116G, 117N	0.752
10	156Q, 157D	0.731
11	152T, 153C, 194T	0.704
12	168K, 169D	0.673

Human JAM-A Iron II Potential Docking Binding Sites		
No.	Binding Residues	Score
1	42E, 102E	0.65
2	42E, 43N, 102E	0.624
3	42E, 100T, 102E	0.599
4	42E, 100T, 102E	0.595
5	65D, 68D, 71R	0.555
6	42E, 100T, 102E	0.546
7	42E, 100T, 102E	0.528
8	65D, 66Q	0.521
9	65D, 66Q	0.515
10	42E, 43N, 98S	0.505
11	42E, 157D	0.464

Human JAM-A Iron III Potential Docking Binding Sites		
No.	Binding Residues	Score
1	153C, 212C	0.926
2	83Y, 107Y	0.684
3	42E, 102E	0.658
4	65D, 66Q	0.547

Figure S5: Predicted calcium- binding sites for human E-CAD PDBID 2072.

E-CAD Calcium Potential Docking Binding Sites		
No.	Binding Residues	Score
1	102N, 104N, 134D, 136D, 143N, 195D	4.721
2	102N, 104N, 134D, 136D, 143N, 195D	4.688
3	11E, 69E, 100D, 101Q, 103D, 136D	4.608
4	11E, 69E, 100D, 101Q, 103D, 136D	4.333
5	11E, 69E, 100D, 101Q, 103D, 136D	4.199

6	11E, 12N, 67D, 69E, 103D	3.779
7	102N, 104N, 134D, 136D, 143N, 195D	3.747
8	11E, 12N, 67D, 69E, 103D	3.737
9	11E, 69E, 100D, 101Q, 103D, 136D	2.904

Figure S6: Overexpression of E-CAD(GFP) in HEK 293 cells. Cells display a high concentration of E-CAD(GFP) in the interface between cells.

