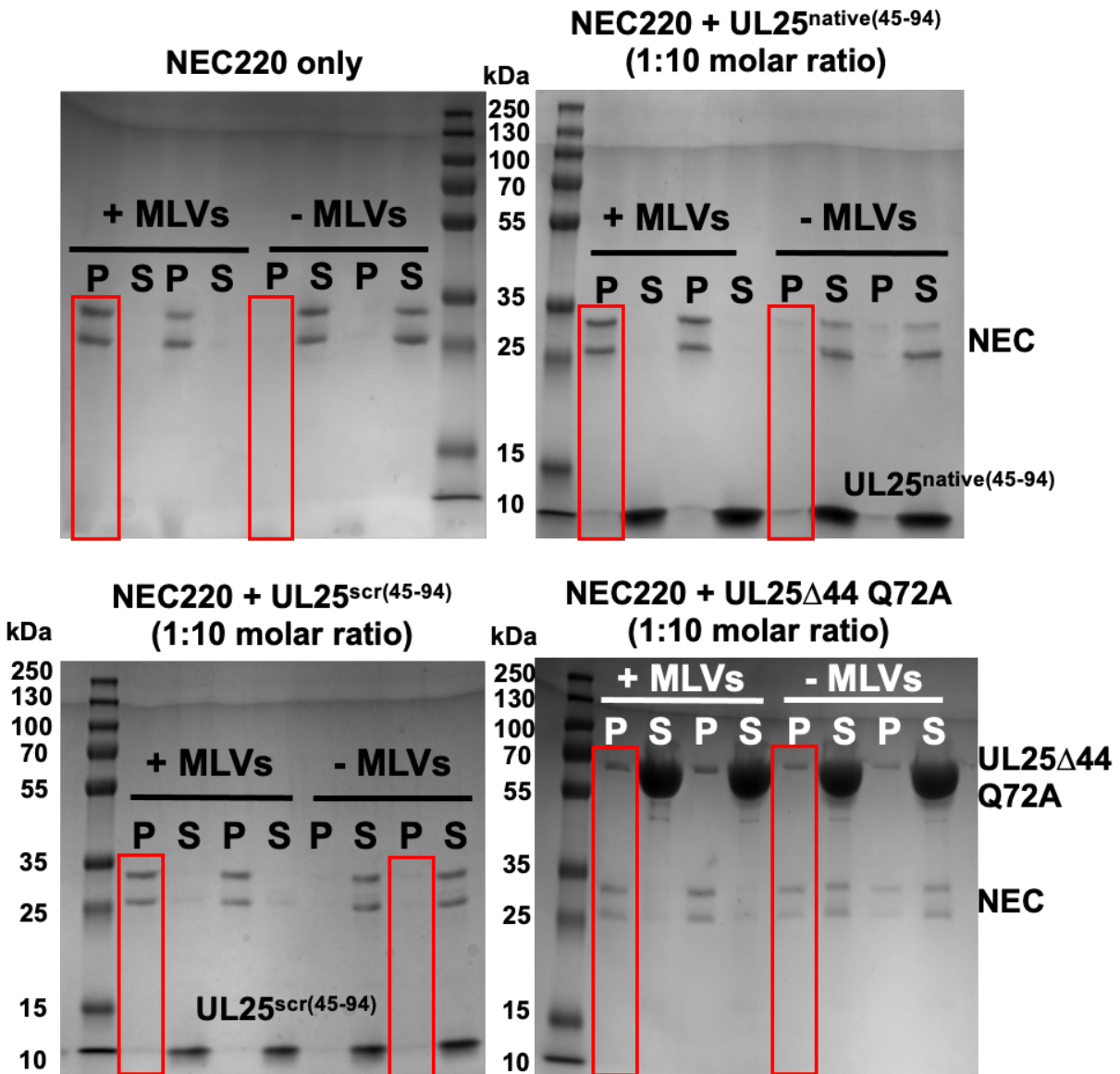


Supporting Information

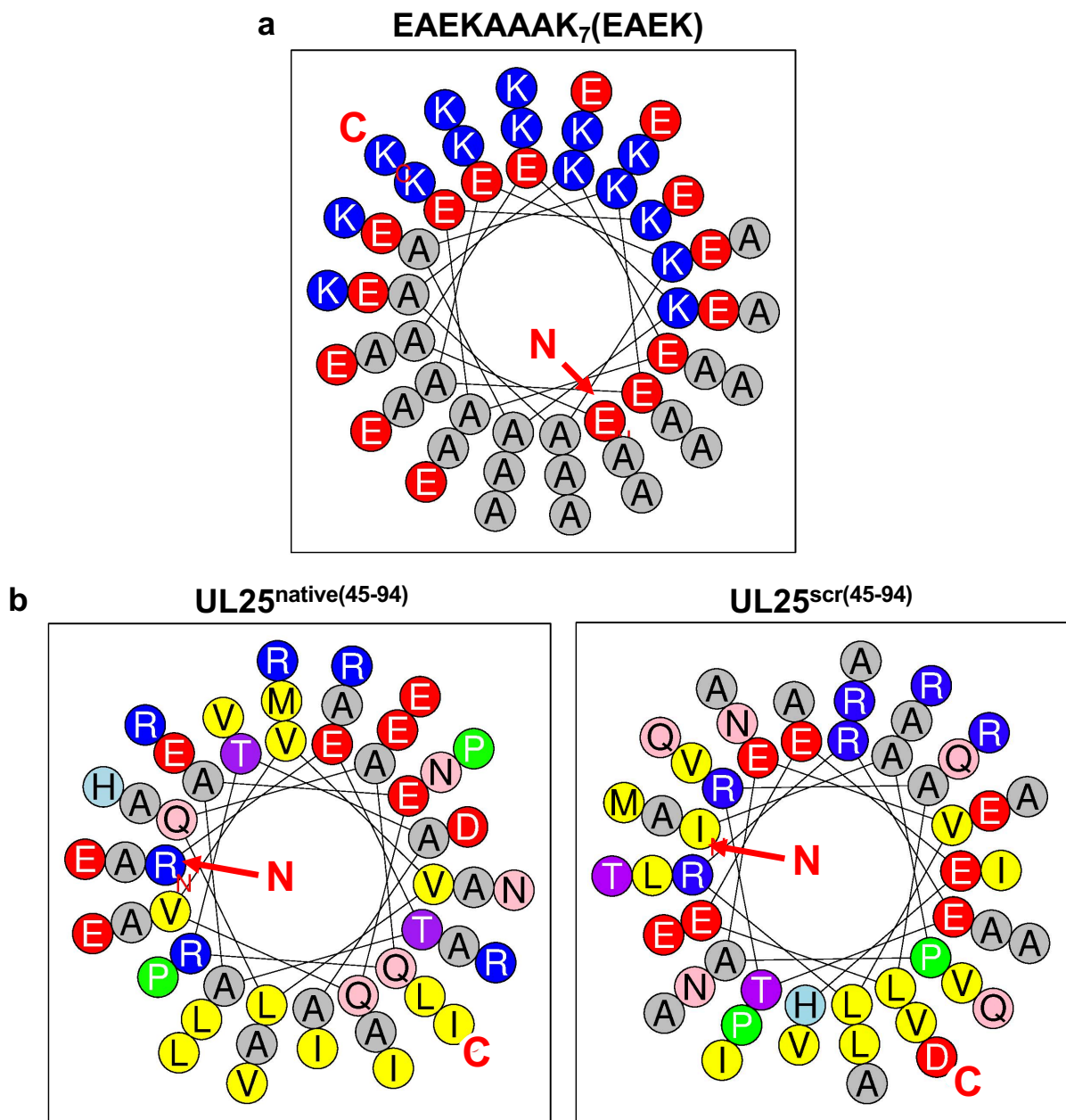
Virus-derived peptide inhibitors of the herpes simplex virus type 1 nuclear egress complex

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Supplementary Fig. S1. Original SDS-PAGE images of the co-sedimentation assay samples shown in Figure 5. Each gel represents one biological replicate used to generate the bar graph in Figure 5. All samples were run in parallel. Red boxes denote the gel lanes shown in the main figure. Gels were stained with Coomassie G-250. Gels were imaged together using a Syngene G:BOX. Both the pellet (P) and supernatant (S) samples, either in the presence (+MLVs) or absence (-MLVs) of MLVs are shown on the gel.



Supplementary Fig. S2. Helical wheel projections of the (EAEKAAAK)₇EAEK, UL25^{native(45-94)} and UL25^{scr(45-94)} peptides. **a)** The (EAEKAAAK)₇EAEK peptide is amphipathic as shown by the distribution of charged (blue and red) residues on one face of the helix versus the hydrophobic alanine residues (grey) on the other. **b)** The UL25^{native(45-94)} and UL25^{scr(45-94)} peptides are less amphipathic as indicated by the less even distribution of charged (blue and red) and polar residues (pink, light blue, and purple) versus hydrophobic (grey and yellow) residues. The N and C on each wheel indicate the N and the C termini. Wheel projections were made using Heliquest¹.

Supplementary Table S1. Secondary structure content (%) estimation of peptides. Spectral deconvolution using DichroWeb² was performed on samples in the absence or presence of 30% trifluoroethanol (TFE). Spectra were deconvoluted using the CONTINLL analysis program³. Data from reference datasets 4, 7 and SMP180 were averaged to obtain secondary structure estimates⁴.

Peptide	No TFE			30% TFE		
	α -Helix	β -sheet	Random coil	α -Helix	β -sheet	Random coil
	%					
UL25^{native(45-94)}	12	12	76	37	12	51
UL25^{scr(45-94)}	8	26	66	31	16	53
UL25⁷⁴⁻⁹⁴	7	35	58	7	35	58
gB¹⁰³⁻¹⁵²	8	26	66	13	24	63
(EAEKAAK)₇EAEK	63	3	29	75	2	23

Supplementary Table S2. List of primers and gBlock sequence used for cloning procedures described in Materials and Methods. All primers are listed in the 5'-3' direction. Restriction sites are underlined and mutations are bolded.

Primer Name	Primer Sequence (5'-3')	Restriction Site
UL25 rev (JB134)	aaaaa <u>actcgag</u> ttattacactgcgctcagatactgagg	XhoI
UL25Δ44 Q72A (ED06)	aatgcagcaatg gcg gcagcc	Site-directed
UL25Δ44 Q72A (ED07)	ggctg ccg cattgctgcatt	Site-directed
(EAEKAAK) ₇ EAEK peptide gBlock sequence	aaaaaa <u>ggatcc</u> gaa gcg gaa aaa gcg gcg aaa gaa gcg gaa aaa gcg gcg aaa gaa gcg gaa aaa gaa gcg gaa aaa gcg gcg aaa gaa gcg gaa aaa gcg gcg aaa gaa gcg gaa aaa gaa gcg gaa aaa gcg gcg aaa gaa gcg gaa aaa gcg gcg aaa gaa gcg gaa aaa taa taa <u>ctcgag</u> aaaaaa	BamHI/XhoI

Supplementary Table 3. Raw data and background values collected for each reported GUV budding assay. The reported biological replicate average values (%) are the points presented in Figure 2.

Biological Replicate	Technical Replicate	Background (BG)	ILVs/GUVs	Raw Value (ILV-BG)	Average (AVG)	ILVs/GUVs	Raw Value (ILV-BG)	Average (AVG)	% Normalized Budding (Raw values/NEC AVG) * 100	Biological Replicate Average (%)
			NEC220			NEC220 + UL25 ^{native(45-94)} (1:0.1 NEC:peptide)				
1	1	10/100 = 0.10	17/100 = 0.17	0.07	0.09	13/100 = 0.13	0.03	0.10	34	112
	2	6/100 = 0.06	15/100 = 0.15	0.09		19/95 = 0.20	0.14		161	
	3	8/100 = 0.08	18/100 = 0.18	0.10		19/95 = 0.20	0.12		138	
2	1	3/96 = 0.03	23/99 = 0.23	0.20	0.15	20/99 = 0.20	0.17	0.17	112	111
	2	7/95 = 0.07	22/100 = 0.22	0.15		26/100 = 0.26	0.19		122	
	3	9/100 = 0.09	20/100 = 0.20	0.11		24/100 = 0.24	0.15		98	
3	1	6/100 = 0.06	19/100 = 0.19	0.13	0.14	19/100 = 0.19	0.13	0.13	95	95
	2	7/100 = 0.07	19/100 = 0.19	0.12		19/90 = 0.21	0.14		103	
	3	2/100 = 0.02	18/100 = 0.18	0.16		14/100 = 0.14	0.12		88	
			NEC220			NEC220 + UL25 ^{native(45-94)} (1:0.3 NEC:peptide)				
1	1	3/96 = 0.03	23/99 = 0.23	0.20	0.15	15/100 = 0.15	0.12	0.11	78	71
	2	7/95 = 0.07	22/100 = 0.22	0.15		18/100 = 0.18	0.11		70	
	3	9/100 = 0.09	20/100 = 0.20	0.11		19/100 = 0.19	0.10		67	
2	1	6/100 = 0.06	19/100 = 0.19	0.13	0.14	14/100 = 0.14	0.08	0.9	59	68
	2	7/100 = 0.07	19/100 = 0.19	0.12		13/100 = 0.13	0.06		44	
	3	2/100 = 0.02	18/100 = 0.18	0.16		16/100 = 0.16	0.14		102	
3	1	1/100 = 0.01	17/90 = 0.19	0.18	0.14	14/100 = 0.14	0.13	0.11	91	75
	2	6/100 = 0.06	18/100 = 0.18	0.14		14/100 = 0.14	0.08		56	
	3	5/100 = 0.05	18/100 = 0.18	0.13		16/100 = 0.16	0.11		77	
			NEC220			NEC220 + UL25 ^{native(45-94)} (1:0.6 NEC:peptide)				
1	1	8/100 = 0.08	19/100 = 0.19	0.11	0.11	16/100 = 0.16	0.08	0.06	71	56
	2	9/100 = 0.09	18/100 = 0.18	0.09		17/100 = 0.17	0.08		71	
	3	8/100 = 0.08	22/100 = 0.22	0.14		11/100 = 0.11	0.03		26	
2	1	6/100 = 0.06	19/100 = 0.19	0.13	0.14	13/95 = 0.14	0.08	0.08	56	58
	2	7/100 = 0.07	19/100 = 0.19	0.12		13/100 = 0.13	0.06		44	
	3	2/100 = 0.02	18/100 = 0.18	0.16		12/100 = 0.12	0.10		73	
3	1	1/100 = 0.01	17/90 = 0.19	0.18	0.14	10/100 = 0.10	0.09	0.07	63	49
	2	6/100 = 0.06	18/100 = 0.18	0.14		11/100 = 0.11	0.05		35	
	3	5/100 = 0.05	18/100 = 0.18	0.13		12/100 = 0.12	0.07		49	
			NEC220			NEC220 + UL25 ^{native(45-94)} (1:1 NEC:peptide)				
1	1	8/100 = 0.08	19/100 = 0.19	0.11	0.11	10/100 = 0.10	0.02	0.04	18	36
	2	9/100 = 0.09	18/100 = 0.18	0.09		13/97 = 0.13	0.04		39	

Biological Replicate	Technical Replicate	Background (BG)	ILVs/GUVs	Raw Value (ILV-BG)	Average (AVG)	ILVs/GUVs	Raw Value (ILV-BG)	Average (AVG)	% Normalized Budding (Raw values/NEC AVG) * 100	Biological Replicate Average (%)
	3	8/100 = 0.08	22/100 = 0.22	0.14		14/100 = 0.14	0.06		53	
2	1	3/90 = 0.03	10/81 = 0.12	0.09	0.09	2/80 = 0.03	0.00	0.02	9	19
	2	3/100 = 0.03	12/90 = 0.13	0.10		2/75 = 0.03	0.00		3	
	3	3/85 = 0.04	9/80 = 0.11	0.07		7/80 = 0.09	0.07		58	
3	1	5/100 = 0.05	12/100 = 0.12	0.07	0.10	7/100 = 0.07	0.02	0.03	21	27
	2	6/100 = 0.06	16/105 = 0.15	0.09		8/100 = 0.08	0.02		21	
	3	5/100 = 0.05	18/100 = 0.18	0.13		9/100 = 0.09	0.04		41	
			NEC220			NEC220 + UL25 ^{native(45-94)} (1:10 NEC:peptide)				
1	1	3/96 = 0.03	23/99 = 0.23	0.20	0.15	11/100 = 0.11	0.08	0.05	52	30
	2	7/95 = 0.07	22/100 = 0.22	0.15		7/100 = 0.07	0.00		0	
	3	9/100 = 0.09	20/100 = 0.20	0.11		15/100 = 0.15	0.06		39	
2	1	3/100 = 0.03	13/94 = 0.14	0.11	0.07	3/100 = 0.03	0.00	0.02	0	33
	2	4/100 = 0.04	8/90 = 0.09	0.05		5/91 = 0.05	0.01		23	
	3	5/100 = 0.05	8/91 = 0.09	0.04		7/70 = 0.10	0.05		77	
3	1	5/100 = 0.05	12/100 = 0.12	0.07	0.10	8/100 = 0.08	0.03	0.03	31	34
	2	6/100 = 0.06	16/105 = 0.15	0.09		9/100 = 0.09	0.03		31	
	3	5/100 = 0.05	18/100 = 0.18	0.13		9/100 = 0.09	0.04		41	
			NEC220			NEC220 + UL25 ^{scr(45-94)} (1:0.1 NEC:peptide)				
1	1	10/100 = 0.10	17/100 = 0.17	0.07	0.09	21/100 = 0.21	0.11	0.10	127	115
	2	6/100 = 0.06	15/100 = 0.15	0.09		13/100 = 0.13	0.07		81	
	3	8/100 = 0.08	18/100 = 0.18	0.10		20/100 = 0.20	0.12		138	
2	1	3/96 = 0.03	23/99 = 0.23	0.20	0.15	17/100 = 0.17	0.14	0.13	91	84
	2	7/95 = 0.07	22/100 = 0.22	0.15		18/100 = 0.18	0.11		70	
	3	9/100 = 0.09	20/100 = 0.20	0.11		23/100 = 0.23	0.14		92	
3	1	6/100 = 0.06	19/100 = 0.19	0.13	0.14	19/96 = 0.20	0.14	0.14	101	104
	2	7/100 = 0.07	19/100 = 0.19	0.12		18/100 = 0.18	0.11		80	
	3	2/100 = 0.02	18/100 = 0.18	0.16		20/100 = 0.20	0.18		132	
			NEC220			NEC220 + UL25 ^{scr(45-94)} (1:0.3 NEC:peptide)				
1	1	3/96 = 0.03	23/99 = 0.23	0.20	0.15	20/100 = 0.20	0.17	0.10	111	65
	2	7/95 = 0.07	22/100 = 0.22	0.15		13/100 = 0.13	0.08		37	
	3	9/100 = 0.09	20/100 = 0.20	0.11		16/98 = 0.16	0.07		48	
2	1	6/100 = 0.06	19/100 = 0.19	0.13	0.14	14/90 = 0.16	0.10	0.11	70	82
	2	7/100 = 0.07	19/100 = 0.19	0.12		18/100 = 0.18	0.11		80	
	3	2/100 = 0.02	18/100 = 0.18	0.16		15/100 = 0.15	0.13		95	
3	1	1/100 = 0.01	17/90 = 0.19	0.18	0.14	15/100 = 0.15	0.14	0.11	98	77
	2	6/100 = 0.06	18/100 = 0.18	0.14		15/100 = 0.15	0.09		63	
	3	5/100 = 0.05	18/100 = 0.18	0.13		15/100 = 0.15	0.10		70	

Biological Replicate	Technical Replicate	Background (BG)	ILVs/GUVs	Raw Value (ILV-BG)	Average (AVG)	ILVs/GUVs	Raw Value (ILV-BG)	Average (AVG)	% Normalized Budding (Raw values/NEC AVG) * 100	Biological Replicate Average (%)
			NEC220			NEC220 + UL25 ^{scr(45-94)} (1:0.6 NEC:peptide)				
1	1	8/100 = 0.08	19/100 = 0.19	0.11	0.11	15/90 = 0.17		0.07	76	61
	2	9/100 = 0.09	18/100 = 0.18	0.09		15/100 = 0.15	0.06		52	
	3	8/100 = 0.08	22/100 = 0.22	0.14		14/100 = 0.14	0.06		53	
2	1	6/100 = 0.06	19/100 = 0.19	0.13	0.14	13/100 = 0.13	0.07	0.08	51	62
	2	7/100 = 0.07	19/100 = 0.19	0.12		12/90 = 0.13	0.06		46	
	3	2/100 = 0.02	18/100 = 0.18	0.16		14/100 = 0.14	0.12		88	
3	1	1/100 = 0.01	17/90 = 0.19	0.18	0.14	12/100 = 0.12	0.11	0.08	78	56
	2	6/100 = 0.06	18/100 = 0.18	0.14		11/100 = 0.11	0.05		35	
	3	5/100 = 0.05	18/100 = 0.18	0.13		13/100 = 0.13	0.08		56	
			NEC220			NEC220 + UL25 ^{scr(45-94)} (1:1 NEC:peptide)				
1	1	8/100 = 0.08	19/100 = 0.19	0.11	0.11	13/100 = 0.13	0.05	0.04	44	38
	2	9/100 = 0.09	18/100 = 0.18	0.09		13/100 = 0.13	0.04		35	
	3	8/100 = 0.08	22/100 = 0.22	0.14		12/100 = 0.12	0.04		35	
2	1	3/90 = 0.03	10/81 = 0.12	0.09	0.09	7/76 = 0.09	0.06	0.02	65	27
	2	3/100 = 0.03	12/90 = 0.13	0.10		4/100 = 0.04	0.01		11	
	3	3/85 = 0.04	9/80 = 0.11	0.07		4/100 = 0.04	0.00		5	
3	1	5/100 = 0.05	12/100 = 0.12	0.07	0.10	11/100 = 0.11	0.06	0.02	62	25
	2	6/100 = 0.06	16/105 = 0.15	0.09		6/97 = 0.06	0.00		2	
	3	5/100 = 0.05	18/100 = 0.18	0.13		6/100 = 0.06	0.01		10	
			NEC220			NEC220 + UL25 ^{scr(45-94)} (1:10 NEC:peptide)				
1	1	3/96 = 0.03	23/99 = 0.23	0.20	0.15	9/95 = 0.09	0.06	0.05	42	31
	2	7/95 = 0.07	22/100 = 0.22	0.15		13/100 = 0.13	0.08		37	
	3	9/100 = 0.09	20/100 = 0.20	0.11		11/100 = 0.11	0.02		13	
2	1	3/100 = 0.03	13/94 = 0.14	0.11	0.07	6/100 = 0.06	0.03	0.02	46	29
	2	4/100 = 0.04	8/90 = 0.09	0.05		5/81 = 0.06	0.02		33	
	3	5/100 = 0.05	8/91 = 0.09	0.04		4/72 = 0.06	0.01		9	
3	1	5/100 = 0.05	12/100 = 0.12	0.07	0.10	9/100 = 0.09	0.04	0.03	41	32
	2	6/100 = 0.06	16/105 = 0.15	0.09		7/100 = 0.07	0.01		10	
	3	5/100 = 0.05	18/100 = 0.18	0.13		9/95 = 0.09	0.04		46	
			NEC220			NEC220 + UL25 ⁷⁴⁻⁹⁴ (1:0.1 NEC:peptide)				
1	1	10/100 = 0.10	17/100 = 0.17	0.07	0.09	15/100 = 0.15	0.05	0.08	58	96
	2	6/100 = 0.06	15/100 = 0.15	0.09		17/100 = 0.17	0.11		127	
	3	8/100 = 0.08	18/100 = 0.18	0.10		17/100 = 0.17	0.09		104	
2	1	3/96 = 0.03	23/99 = 0.23	0.20	0.15	21/100 = 0.21	0.18	0.15	117	97

Biological Replicate	Technical Replicate	Background (BG)	ILVs/GUVs	Raw Value (ILV-BG)	Average (AVG)	ILVs/GUVs	Raw Value (ILV-BG)	Average (AVG)	% Normalized Budding (Raw values/NEC AVG) * 100	Biological Replicate Average (%)
	2	7/95 = 0.07	22/100 = 0.22	0.15		23/100 = 0.23	0.16		102	
	3	9/100 = 0.09	20/100 = 0.20	0.11		20/100 = 0.20	0.11		72	
3	1	6/100 = 0.06	19/100 = 0.19	0.13	0.14	17/90 = 0.19	0.13	0.13	94	97
	2	7/100 = 0.07	19/100 = 0.19	0.12		21/100 = 0.21	0.14		102	
	3	2/100 = 0.02	18/100 = 0.18	0.16		15/100 = 0.15	0.13		95	
			NEC220			NEC220 + UL25 ⁷⁴⁻⁹⁴ (1:0.3 NEC:peptide)				
1	1	3/96 = 0.03	23/99 = 0.23	0.20	0.15	25/100 = 0.25	0.22	0.16	143	102
	2	7/95 = 0.07	22/100 = 0.22	0.15		23/100 = 0.23	0.16		103	
	3	9/100 = 0.09	20/100 = 0.20	0.11		18/100 = 0.18	0.09		59	
2	1	6/100 = 0.06	19/100 = 0.19	0.13	0.14	17/100 = 0.17	0.11	0.13	80	98
	2	7/100 = 0.07	19/100 = 0.19	0.12		17/100 = 0.17	0.11		73	
	3	2/100 = 0.02	18/100 = 0.18	0.16		21/100 = 0.21	0.19		139	
3	1	1/100 = 0.01	17/90 = 0.19	0.18	0.14	17/90 = 0.19	0.18	0.14	125	98
	2	6/100 = 0.06	18/100 = 0.18	0.14		17/100 = 0.17	0.11		77	
	3	5/100 = 0.05	18/100 = 0.18	0.13		18/100 = 0.18	0.13		91	
			NEC220			NEC220 + UL25 ⁷⁴⁻⁹⁴ (1:0.6 NEC:peptide)				
1	1	6/100 = 0.06	19/100 = 0.19	0.13	0.14	18/100 = 0.18	0.12	0.12	88	87
	2	7/100 = 0.07	19/100 = 0.19	0.12		16/95 = 0.17	0.10		72	
	3	2/100 = 0.02	18/100 = 0.18	0.16		16/100 = 0.16	0.14		102	
2	1	1/100 = 0.01	17/90 = 0.19	0.18	0.14	18/93 = 0.19	0.18	0.15	128	103
	2	6/100 = 0.06	18/100 = 0.18	0.14		20/100 = 0.20	0.14		98	
	3	5/100 = 0.05	18/100 = 0.18	0.13		17/100 = 0.17	0.12		84	
3	1	8/100 = 0.08	19/100 = 0.19	0.11	0.11	17/100 = 0.17	0.09	0.10	80	90
	2	9/100 = 0.09	18/100 = 0.18	0.09		20/100 = 0.20	0.11		97	
	3	8/100 = 0.08	22/100 = 0.22	0.14		18/96 = 0.19	0.11		95	
			NEC220			NEC220 + UL25 ⁷⁴⁻⁹⁴ (1:1 NEC:peptide)				
1	1	8/100 = 0.08	19/100 = 0.19	0.11	0.11	15/96 = 0.16	0.08	0.09	67	75
	2	9/100 = 0.09	18/100 = 0.18	0.09		18/100 = 0.18	0.09		79	
	3	8/100 = 0.08	22/100 = 0.22	0.14		17/100 = 0.17	0.09		79	
2	1	1/100 = 0.01	17/90 = 0.19	0.18	0.14	16/100 = 0.16	0.15	0.13	105	93
	2	6/100 = 0.06	18/100 = 0.18	0.14		18/100 = 0.18	0.12		84	
	3	5/100 = 0.05	18/100 = 0.18	0.13		18/100 = 0.18	0.13		91	
3	1	5/100 = 0.05	12/100 = 0.12	0.07	0.10	10/100 = 0.10	0.05	0.05	51	55
	2	6/100 = 0.06	16/105 = 0.15	0.09		10/100 = 0.10	0.04		41	
	3	5/100 = 0.05	18/100 = 0.18	0.13		12/100 = 0.12	0.07		72	

Biological Replicate	Technical Replicate	Background (BG)	ILVs/GUVs	Raw Value (ILV-BG)	Average (AVG)	ILVs/GUVs	Raw Value (ILV-BG)	Average (AVG)	% Normalized Budding (Raw values/NEC AVG) * 100	Biological Replicate Average (%)
			NEC220			NEC220 + UL25 ⁷⁴⁻⁹⁴ (1:10 NEC:peptide)				
1	1	3/96 = 0.03	23/99 = 0.23	0.20	0.15	17/100 = 0.17	0.14	0.10	91	64
	2	7/95 = 0.07	22/100 = 0.22	0.15		14/96 = 0.15	0.08		47	
	3	9/100 = 0.09	20/100 = 0.20	0.11		17/100 = 0.17	0.08		52	
2	1	3/100 = 0.03	13/94 = 0.14	0.11	0.07	6/97 = 0.06	0.03	0.04	49	66
	2	4/100 = 0.04	8/90 = 0.09	0.05		6/100 = 0.06	0.02		31	
	3	5/100 = 0.05	8/91 = 0.09	0.04		12/95 = 0.13	0.08		117	
3	1	5/100 = 0.05	12/100 = 0.12	0.07	0.10	15/100 = 0.15	0.10	0.07	103	72
	2	6/100 = 0.06	16/105 = 0.15	0.09		11/100 = 0.11	0.05		51	
	3	5/100 = 0.05	18/100 = 0.18	0.13		11/100 = 0.11	0.06		62	
			NEC220			NEC220 + gB RC ¹⁰³⁻¹⁵² (1:0.1 NEC:peptide)				
1	1	10/100 = 0.10	17/100 = 0.17	0.07	0.09	20/100 = 0.20	0.10	0.10	115	115
	2	6/100 = 0.06	15/100 = 0.15	0.09		17/100 = 0.17	0.11		127	
	3	8/100 = 0.08	18/100 = 0.18	0.10		17/100 = 0.17	0.09		104	
2	1	3/96 = 0.03	23/99 = 0.23	0.20	0.15	22/100 = 0.22	0.19	0.18	124	120
	2	7/95 = 0.07	22/100 = 0.22	0.15		25/98 = 0.26	0.19		119	
	3	9/100 = 0.09	20/100 = 0.20	0.11		24/90 = 0.27	0.18		116	
3	1	6/100 = 0.06	19/100 = 0.19	0.13	0.14	19/90 = 0.21	0.15	0.15	111	107
	2	7/100 = 0.07	19/100 = 0.19	0.12		16/100 = 0.16	0.09		66	
	3	2/100 = 0.02	18/100 = 0.18	0.16		21/97 = 0.22	0.20		144	
			NEC220			NEC220 + gB RC ¹⁰³⁻¹⁵² (1:0.3 NEC:peptide)				
1	1	3/96 = 0.03	23/99 = 0.23	0.20	0.15	21/100 = 0.21	0.18	0.16	117	102
	2	7/95 = 0.07	22/100 = 0.22	0.15		23/100 = 0.23	0.16		103	
	3	9/100 = 0.09	20/100 = 0.20	0.11		22/100 = 0.22	0.13		85	
2	1	6/100 = 0.06	19/100 = 0.19	0.13	0.14	18/100 = 0.18	0.12	0.14	88	105
	2	7/100 = 0.07	19/100 = 0.19	0.12		21/100 = 0.21	0.14		102	
	3	2/100 = 0.02	18/100 = 0.18	0.16		19/100 = 0.19	0.17		124	
3	1	1/100 = 0.01	17/90 = 0.19	0.18	0.14	17/100 = 0.17	0.16	0.14	112	96
	2	6/100 = 0.06	18/100 = 0.18	0.14		18/100 = 0.18	0.12		84	
	3	5/100 = 0.05	18/100 = 0.18	0.13		17/100 = 0.17	0.12		91	
			NEC220			NEC220 + gB RC ¹⁰³⁻¹⁵² (1:0.6 NEC:peptide)				
1	1	6/100 = 0.06	19/100 = 0.19	0.13	0.14	16/100 = 0.16	0.10	0.12	73	85
	2	7/100 = 0.07	19/100 = 0.19	0.12		14/100 = 0.14	0.07		51	
	3	2/100 = 0.02	18/100 = 0.18	0.16		20/100 = 0.20	0.18		132	
2	1	1/100 = 0.01	17/90 = 0.19	0.18	0.14	16/100 = 0.16	0.15	0.13	105	93
	2	6/100 = 0.06	18/100 = 0.18	0.14		19/100 = 0.19	0.13		91	

Biological Replicate	Technical Replicate	Background (BG)	ILVs/GUVs	Raw Value (ILV-BG)	Average (AVG)	ILVs/GUVs	Raw Value (ILV-BG)	Average (AVG)	% Normalized Budding (Raw values/NEC AVG) * 100	Biological Replicate Average (%)
	3	5/100 = 0.05	18/100 = 0.18	0.13		16/94 = 0.17	0.12		84	
3	1	8/100 = 0.08	19/100 = 0.19	0.11	0.11	16/100 = 0.16	0.08	0.10	71	91
	2	9/100 = 0.09	18/100 = 0.18	0.09		18/100 = 0.18	0.09		80	
	3	8/100 = 0.08	22/100 = 0.22	0.14		22/100 = 0.22	0.14		124	
			NEC220			NEC220 + gB RC ¹⁰³⁻¹⁵² (1:1 NEC:peptide)				
1	1	8/100 = 0.08	19/100 = 0.19	0.11	0.11	18/95 = 0.19	0.11	0.12	97	103
	2	9/100 = 0.09	18/100 = 0.18	0.09		19/100 = 0.19	0.10		88	
	3	8/100 = 0.08	22/100 = 0.22	0.14		22/100 = 0.22	0.14		124	
2	1	1/100 = 0.01	17/90 = 0.19	0.18	0.14	17/100 = 0.17	0.16	0.14	112	98
	2	6/100 = 0.06	18/100 = 0.18	0.14		16/100 = 0.16	0.10		70	
	3	5/100 = 0.05	18/100 = 0.18	0.13		19/90 = 0.21	0.16		113	
3	1	5/100 = 0.05	12/100 = 0.12	0.07	0.10	13/100 = 0.13	0.08	0.08	82	82
	2	6/100 = 0.06	16/105 = 0.15	0.09		15/100 = 0.15	0.09		92	
	3	5/100 = 0.05	18/100 = 0.18	0.13		12/100 = 0.12	0.07		72	
			NEC220			NEC220 + gB RC ¹⁰³⁻¹⁵² (1:10 NEC:peptide)				
1	1	3/96 = 0.03	23/99 = 0.23	0.20	0.15	20/91 = 0.22	0.19	0.12	124	82
	2	7/95 = 0.07	22/100 = 0.22	0.15		19/100 = 0.19	0.12		76	
	3	9/100 = 0.09	20/100 = 0.20	0.11		16/100 = 0.16	0.07		46	
2	1	3/100 = 0.03	13/94 = 0.14	0.11	0.07	9/90 = 0.10	0.07	0.05	108	73
	2	4/100 = 0.04	8/90 = 0.09	0.05		8/90 = 0.09	0.05		75	
	3	5/100 = 0.05	8/91 = 0.09	0.04		5/67 = 0.07	0.02		38	
3	1	5/100 = 0.05	12/100 = 0.12	0.07	0.10	9/97 = 0.09	0.04	0.06	44	63
	2	6/100 = 0.06	16/105 = 0.15	0.09		12/100 = 0.12	0.06		62	
	3	5/100 = 0.05	18/100 = 0.18	0.13		13/100 = 0.13	0.08		82	
			NEC220			NEC220 + (EAEKAAK) ₇ EAEK (1:0.1 NEC:peptide)				
1	1	3/96 = 0.03	23/99 = 0.23	0.20	0.15	22/100 = 0.22	0.19	0.15	124	97
	2	7/95 = 0.07	22/100 = 0.22	0.15		24/100 = 0.24	0.17		109	
	3	9/100 = 0.09	20/100 = 0.20	0.11		18/100 = 0.18	0.09		59	
2	1	6/100 = 0.06	19/100 = 0.19	0.13	0.14	21/93 = 0.23	0.17	0.15	121	109
	2	7/100 = 0.07	19/100 = 0.19	0.12		19/100 = 0.19	0.12		88	
	3	2/100 = 0.02	18/100 = 0.18	0.16		18/100 = 0.18	0.16		117	
3	1	8/100 = 0.08	19/100 = 0.19	0.11	0.11	17/100 = 0.17	0.09	0.09	80	82
	2	9/100 = 0.09	18/100 = 0.18	0.09		16/100 = 0.16	0.07		62	
	3	8/100 = 0.08	22/100 = 0.22	0.14		20/100 = 0.20	0.12		106	
			NEC220			NEC220 + (EAEKAAK) ₇ EAEK (1:0.3 NEC:peptide)				
1	1	3/96 = 0.03	23/99 = 0.23	0.20	0.15	23/100 = 0.23	0.20	0.15	130	99

Biological Replicate	Technical Replicate	Background (BG)	ILVs/GUVs	Raw Value (ILV-BG)	Average (AVG)	ILVs/GUVs	Raw Value (ILV-BG)	Average (AVG)	% Normalized Budding (Raw values/NEC AVG) * 100	Biological Replicate Average (%)
	2	7/95 = 0.07	22/100 = 0.22	0.15		20/100 = 0.20	0.13		83	
	3	9/100 = 0.09	20/100 = 0.20	0.11		21/96 = 0.22	0.13		84	
	1	6/100 = 0.06	19/100 = 0.19	0.13		20/100 = 0.20	0.14		102	
2	2	7/100 = 0.07	19/100 = 0.19	0.12	0.14	18/100 = 0.18	0.11	0.13	80	95
	3	2/100 = 0.02	18/100 = 0.18	0.16		16/100 = 0.16	0.14		102	
	1	1/100 = 0.01	17/90 = 0.19	0.18		17/100 = 0.17	0.16		112	
3	2	6/100 = 0.06	18/100 = 0.18	0.14	0.14	16/100 = 0.16	0.10	0.14	70	96
	3	5/100 = 0.05	18/100 = 0.18	0.13		20/100 = 0.20	0.15		105	
				NEC220			NEC220 + (EAEKAAK)₇EAEK (1:0.6 NEC:peptide)			
1	1	6/100 = 0.06	19/100 = 0.19	0.13	0.14	18/100 = 0.18	0.12	0.12	88	88
	2	7/100 = 0.07	19/100 = 0.19	0.12		15/100 = 0.15	0.08		58	
	3	2/100 = 0.02	18/100 = 0.18	0.16		18/100 = 0.18	0.16		117	
2	1	1/100 = 0.01	17/90 = 0.19	0.18	0.14	19/100 = 0.19	0.18	0.16	126	112
	2	6/100 = 0.06	18/100 = 0.18	0.14		21/100 = 0.21	0.15		105	
	3	5/100 = 0.05	18/100 = 0.18	0.13		20/100 = 0.20	0.15		105	
3	1	8/100 = 0.08	19/100 = 0.19	0.11	0.11	16/95 = 0.17	0.09	0.10	78	85
	2	9/100 = 0.09	18/100 = 0.18	0.09		15/100 = 0.15	0.06		53	
	3	8/100 = 0.08	22/100 = 0.22	0.14		22/100 = 0.22	0.14		124	
			NEC220			NEC220 + (EAEKAAK)₇EAEK (1:1 NEC:peptide)				
1	1	8/100 = 0.08	19/100 = 0.19	0.11	0.11	19/90 = 0.21	0.13	0.12	116	106
	2	9/100 = 0.09	18/100 = 0.18	0.09		21/100 = 0.21	0.12		106	
	3	8/100 = 0.08	22/100 = 0.22	0.14		19/100 = 0.19	0.08		97	
2	1	6/100 = 0.06	19/100 = 0.19	0.13	0.14	18/100 = 0.18	0.12	0.12	88	90
	2	7/100 = 0.07	19/100 = 0.19	0.12		17/100 = 0.17	0.10		73	
	3	2/100 = 0.02	18/100 = 0.18	0.16		17/100 = 0.17	0.15		110	
3	1	1/100 = 0.01	17/90 = 0.19	0.18	0.14	15/90 = 0.17	0.16	0.16	110	102
	2	6/100 = 0.06	18/100 = 0.18	0.14		18/100 = 0.18	0.12		84	
	3	5/100 = 0.05	18/100 = 0.18	0.13		21/100 = 0.21	0.16		112	
			NEC220			NEC220 + (EAEKAAK)₇EAEK (1:10 NEC:peptide)				
1	1	9/100 = 0.09	0.04	0.05	0.06	19/99 = 0.19	0.15	0.15	34	40
	2	11/100 = 0.11	0.04	0.07		16/100 = 0.16	0.12		45	
	3	10/100 = 0.10	0.04	0.06		21/100 = 0.21	0.17		41	
2	1	15/101 = 0.15	0.06	0.09	0.11	26/100 = 0.26	0.20	0.21	41	53
	2	19/98 = 0.19	0.06	0.13		22/100 = 0.22	0.16		63	
	3	14/100 = 0.14	0.02	0.12		30/100 = 0.30	0.28		56	
3	1	9/100 = 0.09	0.04	0.05	0.08	17/97 = 0.18	0.14	0.14	35	54
	2	12/100 = 0.12	0.04	0.08		16/96 = 0.16	0.12		57	

Biological Replicate	Technical Replicate	Background (BG)	ILVs/GUVs	Raw Value (ILV-BG)	Average (AVG)	ILVs/GUVs	Raw Value (ILV-BG)	Average (AVG)	% Normalized Budding (Raw values/NEC AVG) * 100	Biological Replicate Average (%)
	3	13/100 = 0.13	0.03	0.10		19/100 = 0.19	0.13		71	
			NEC220			NEC220 + UL25Δ44 Q72A (1:0.1 NEC:UL25)				
1	1	10/100 = 0.10	17/100 = 0.17	0.07	0.09	14/100 = 0.14	0.04	0.09	46	107
	2	6/100 = 0.06	15/100 = 0.15	0.09		17/90 = 0.19	0.13		149	
	3	8/100 = 0.08	18/100 = 0.18	0.10		19/100 = 0.19	0.11		127	
2	1	3/96 = 0.03	23/99 = 0.23	0.20	0.15	21/100 = 0.21	0.18	0.17	117	110
	2	7/95 = 0.07	22/100 = 0.22	0.15		28/100 = 0.28	0.21		135	
	3	9/100 = 0.09	20/100 = 0.20	0.11		21/100 = 0.21	0.12		79	
3	1	6/100 = 0.06	19/100 = 0.19	0.13	0.14	17/100 = 0.17	0.11	0.13	80	97
	2	7/100 = 0.07	19/100 = 0.19	0.12		14/96 = 0.15	0.08		55	
	3	2/100 = 0.02	18/100 = 0.18	0.16		23/100 = 0.23	0.21		154	
			NEC220			NEC220 + UL25Δ44 Q72A (1:0.3 NEC:UL25)				
1	1	3/96 = 0.03	23/99 = 0.23	0.20	0.15	27/97 = 0.28	0.25	0.15	162	101
	2	7/95 = 0.07	22/100 = 0.22	0.15		17/100 = 0.17	0.10		63	
	3	9/100 = 0.09	20/100 = 0.20	0.11		21/100 = 0.21	0.11		79	
2	1	6/100 = 0.06	19/100 = 0.19	0.13	0.14	16/100 = 0.16	0.10	0.12	73	88
	2	7/100 = 0.07	19/100 = 0.19	0.12		16/100 = 0.16	0.09		66	
	3	2/100 = 0.02	18/100 = 0.18	0.16		19/100 = 0.19	0.17		124	
3	1	1/100 = 0.01	17/90 = 0.19	0.18	0.14	21/100 = 0.21	0.20	0.14	140	98
	2	6/100 = 0.06	18/100 = 0.18	0.14		15/100 = 0.15	0.09		63	
	3	5/100 = 0.05	18/100 = 0.18	0.13		18/100 = 0.18	0.13		91	
			NEC220			NEC220 + UL25Δ44 Q72A (1:0.6 NEC:UL25)				
1	1	8/100 = 0.08	19/100 = 0.19	0.11	0.11	16/100 = 0.16	0.08	0.11	71	97
	2	9/100 = 0.09	18/100 = 0.18	0.09		21/100 = 0.21	0.12		106	
	3	8/100 = 0.08	22/100 = 0.22	0.14		21/100 = 0.21	0.13		115	
2	1	6/100 = 0.06	19/100 = 0.19	0.13	0.14	18/100 = 0.18	0.08	0.13	88	93
	2	7/100 = 0.07	19/100 = 0.19	0.12		16/94 = 0.17	0.10		73	
	3	2/100 = 0.02	18/100 = 0.18	0.16		18/100 = 0.18	0.16		117	
3	1	1/100 = 0.01	17/90 = 0.19	0.18	0.14	20/100 = 0.20	0.19	0.15	133	105
	2	6/100 = 0.06	18/100 = 0.18	0.14		15/90 = 0.17	0.11		75	
	3	5/100 = 0.05	18/100 = 0.18	0.13		19/93 = 0.20	0.15		108	
			NEC220			NEC220 + UL25Δ44 Q72A (1:1 NEC:UL25)				
1	1	8/100 = 0.08	19/100 = 0.19	0.11	0.11	19/91 = 0.21	0.13	0.13	113	112
	2	9/100 = 0.09	18/100 = 0.18	0.09		21/80 = 0.27	0.18		152	
	3	8/100 = 0.08	22/100 = 0.22	0.14		16/100 = 0.16	0.08		71	
2	1	1/100 = 0.01	17/90 = 0.19	0.18	0.14	16/100 = 0.16	0.15	0.15	105	105

Biological Replicate	Technical Replicate	Background (BG)	ILVs/GUVs	Raw Value (ILV-BG)	Average (AVG)	ILVs/GUVs	Raw Value (ILV-BG)	Average (AVG)	% Normalized Budding (Raw values/NEC AVG) * 100	Biological Replicate Average (%)
	2	6/100 = 0.06	18/100 = 0.18	0.14		22/100 = 0.22	0.16		112	
	3	5/100 = 0.05	18/100 = 0.18	0.13		19/100 = 0.19	0.14		98	
	1	3/90 = 0.03	10/81 = 0.12	0.09		11/97 = 0.11	0.08		89	
3	2	3/100 = 0.03	12/90 = 0.13	0.10	0.09	11/90 = 0.12	0.09	0.09	102	100
	3	3/85 = 0.04	9/80 = 0.11	0.07		13/97 = 0.13	0.09		109	
			NEC220			NEC220 + UL25Δ44 Q72A (1:10 NEC:UL25)				
1	1	3/100 = 0.03	13/94 = 0.14	0.11	0.07	4/100 = 0.04	0.01	0.00	15	12
	2	4/100 = 0.04	8/90 = 0.09	0.05		4/100 = 0.04	0.00		0	
	3	5/100 = 0.05	8/91 = 0.09	0.04		5/80 = 0.06	0.01		19	
2	1	5/100 = 0.05	12/100 = 0.12	0.07	0.10	5/100 = 0.05	0.00	0.00	0	7
	2	6/100 = 0.06	16/105 = 0.15	0.09		6/97 = 0.06	0.00		2	
	3	5/100 = 0.05	18/100 = 0.18	0.13		7/100 = 0.07	0.02		21	
3	1	3/87 = 0.03	17/85 = 0.20	0.17	0.14	3/85 = 0.03	0.00	0.01	0	7
	2	2/96 = 0.02	15/94 = 0.16	0.14		5/101 = 0.05	0.03		20	
	3	3/103 = 0.03	15/97 = 0.15	0.12		3/95 = 0.03	0.00		2	

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