

Figure 1 (cont'd)

LCMV WE-DUE S-SEGMENT			HUMAN CELL LINES							MOUSE CELL LINES							LCMV MUTATION FAST EVOLUTION PLATFORM				
AMINOACID NUMBER	AMINOACID WE-DUE	AMINOACID MUTANT	Ma-Mel-86a MELANOMA	UKE-Mel-118b MELANOMA	UKE-Mel-118c MELANOMA	C643 ANAPLASTIC THYROID CA	A549 LUNG CA	H1975 LUNG ADENO CA	Sw480 COLORECTAL ADENO CA	B16F10 MELANOMA	511950 PANCAREAS CA	511950TR PANCAREAS CA	PANCAREAS CA	60590 PANCAREAS CA	60590TR PANCAREAS CA	TRAMP2 CA	PROSTATE ADENO CA	MC38 COLON ADENO CA	MC57 FIBROSARCOMA	LCMV MUTATION FAST EVOLUTION PLATFORM	
POS	WT	MUT	-	5FU	-	5FU	-	5FU	-	5FU	-	5FU	-	5FU	-	5FU	-	5FU	-	5FU	Y60H
60	Y	H																			G61D
61	G	D			2/4	1/4												1/4			(L62P)
62	L	P																			V71F
71	V	F				1/4		1/4													(F74L)
74	F	L																			M88T
88	M	T				1/4															A94S
94	A	S	1/4												1/4	2/4					(S102N)
102	S	N		1/4			4/4	4/4													M103L
103	M	L												1/4							(S105N)
105	S	N								1/5											S106F
106	S	F																			F112L
112	F	L																			H120Y
120	H	Y																			(N121K)
121	N	K		1/4														1/4			(F122L)
122	F	L																			A128T
128	A	T															1/4				F129S
129	F	S	1/4						1/4						1/4						F129V
132	K	Q																			K132Q
132	K	Q																			T133I
133	T	I																			H136Q
136	H	Q																			I141V
141	I	V												2/4							S144N
144	S	N																			(I149V)
149	I	V																			G151E
151	G	E																			N152Y
152	N	Y	1/4																		(S153P)
153	S	P																			(N154D)
154	N	D																			H155Y
154	N	D		1/4	4/4	2/4	1/4	1/4	1/4	4/4	1/4							3/4	1/4		(H155Y)
155	H	N																			H155N
155	H	N																			(H155N)
163	N	D																1/4			(N163D)
188	R	S	1/4																		R188S
198	A	T												1/4							A198T
203	Y	H							1/4												(Y203H)
207	G	S												1/4							G207S
211	A	T																			(A211T)
211	A	T																			A211V
217	T	I	1/4																		T217I
218	T	I				1/4								1/4							T218I
236	E	G																			E236G
252	F	S																			(F255S)
255	E	G																			E255G
256	K	R	1/4																1/4		K256R

B

Figure 1 (cont'd)

[illegible]

Figure 2

LCMV-WE S-Segm GPC AA No	LCMV-p52 [[181M, R185W]]	A375 (66 x)	A649 (66 x)	A549 (66 x)	RPMI-7951 (66 x)	RPMI-7951 (66 x)	SW620 (32 x)	SW620 (66 x)	HCC1954 (31 x)	HCC1954 (23 x)	LCMV MUTATION NOVEL
		-	-	5FU	-	5FU	-	5FU	-	5FU	
63	N					K					N63K
66	D			N							D66N
119	N						D	D			N119D
121	N	S									N121S
122	F							L			F122L
132	K						R				K132R
153	S			P			P	P	P	P	S153P
155	H	Y					Y				H155Y
163	N		D								N163D
181	M			V							M181V
185	W										
217	T				I	I / S		I			T217I T217S
260	L			I							L260I
327	A	V									A327V
358	R								K	K	R358K
451	I	V									I451V
477	N							S			N477S
492	K							E			K492E

Figure 3

LCMV-WE S-Segm GPC AA No	LCMV-WE S-Segm GPC [AA]	LCMV-YF13 [A211T]	C643 (10 x)	C643 (10x)	LCMV MUTATION NOVEL
			-	5FU	
48	S		G	G	G48S
211	A	T	T	T	
284	Y			H	Y284H
328	A		S	S	A328S
332	F		L	L	F332L
342	V		A		V342A

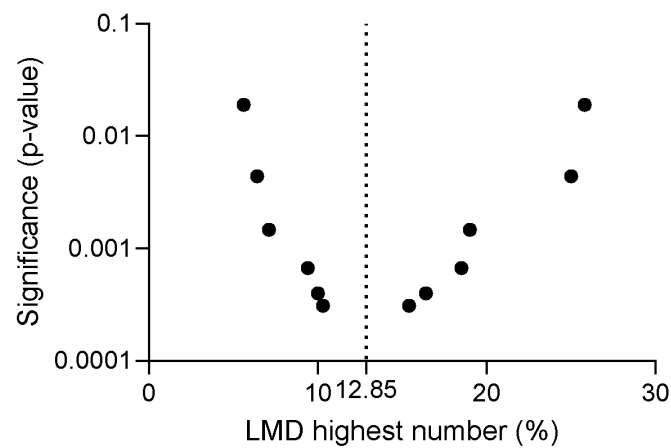
Figure 4

A

GPC	Domains	Position	Mutated AA positions	Total AA (N)	Mutated AA (N)	Mutation frequency (%)
SSP	H1, H2	1-58	18, 28, 36, 39, 48, 51	58	6	10,3
GP1	Strand β 1 N-terminal	59-89	60, 61, 62, 63, 66, 71, 74, 88	31	8	<u>25,8</u>
	Strand β 2, β 3, β 4,	90-113	94, 102, 103, 105, 106, 112	24	6	<u>25,0</u>
	Helix α 1, α 2	114-147	119, 120, 121, 122, 128, 129, 132, 133, 136, 141, 144	34	11	<u>32,4</u>
	Loop 1	148-157	149, 151, 152, 153, 154, 155	10	6	<u>60,0</u>
	Strand β 5, β 6	158-171	163	14	1	7,1
	Helix α 3	187-199	188, 198	13	2	<u>15,4</u>
	Loop 3	200-226	203, 207, 211, 217, 218	27	5	<u>18,5</u>
	Strand β 7	227-244	236	18	1	5,6
GP2	α 4, α 5	245-265	252, 255, 256, 260	21	4	<u>19,0</u>
	Fusion Peptide Fusion Loop Fusion Helix	266-312	280, 284, 308	47	3	6,4
	N-Helix	313-373	327, 328, 332, 335, 339, 342, 344, 357, 358, 369	61	10	<u>16,4</u>
	T loop and Extended Peptide	374-405	374, 382, 388	32	3	9,4
	C-Helix	406-438	426	33	1	3,0
	TM Cytoplasmic	439-498	451, 456, 471, 477, 491, 492	60	6	10,0

Figure 4 (cont'd)

B



C

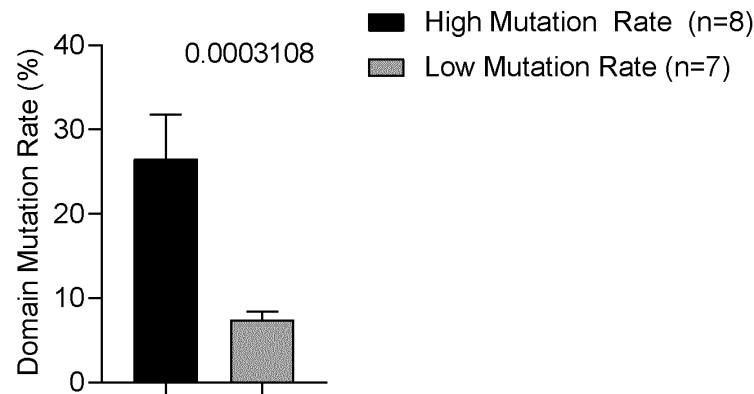


Figure 5

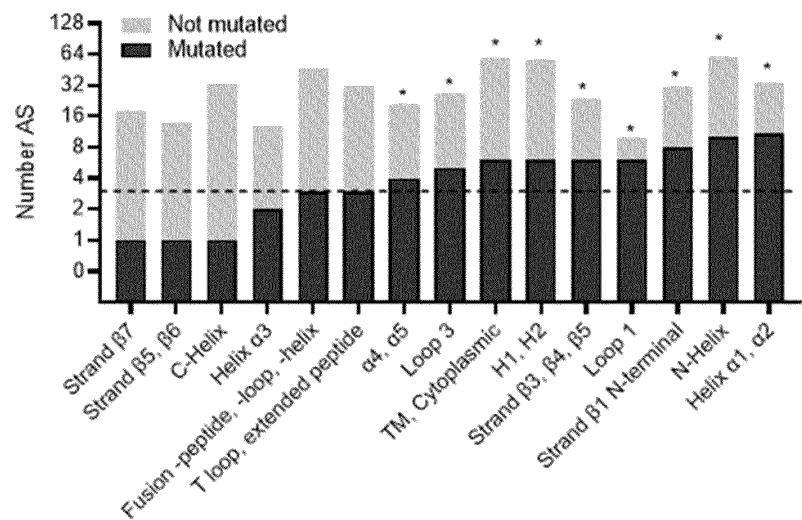


Figure 6

	Functional region	Aminoacid position	Total AA (N)	Mutated AA (N)	Total AA (N)	Mutated AA (N)	Mutation frequency (%)
GPC subunits	SSP	1-58	58	6	58	6	10.3
	GP1	59-265	192	44	192	44	22.9
	GP2	266-498	233	23	233	23	9.9
Cleavage	SPase	58-59	2	0	2	0	0.0
	SKI-1/S1P	265-266	2	0	2	0	0.0
Topological	Extracellular	[SSP] 33-40	8	2	373	63	16.9
		[GP1] 59-265	192	44			
		[GP2] 266-438	173	17			
	Transmembrane	[SSP] 18-32, 41-54	29	4	50	6	12.0
		[GP2] 439-459	21	2			
	Cytoplasmic	[SSP] 1-17, 55-58	21	0	60	4	6.7
		[GP2] 460-498	39	4			
Protein-Protein Interaction structure	Dimerization	[GP1] 122-127, 136, 143, 146-155	18	8	18	8	44.4
Receptor binding	Affinity to Alpha DG	[GP1] 136, 153, 155, 190, 260	5	4	5	4	80.0
Posttranslational modification	Lipidation / Myristoylation	[SSP] 2	1	0	1	0	0.0
	Disulfidbonds	[GP1] 92-239, 123-160, 184-220, [GP2] 285-298, 307-316, 370-391	11	0	11	0	0.0
	Glycosylation	[GP1] 85, 95, 114, 124, 171, 232 [GP2] 371, 396, 401	9	0	9	0	0.0

Figure 7

A

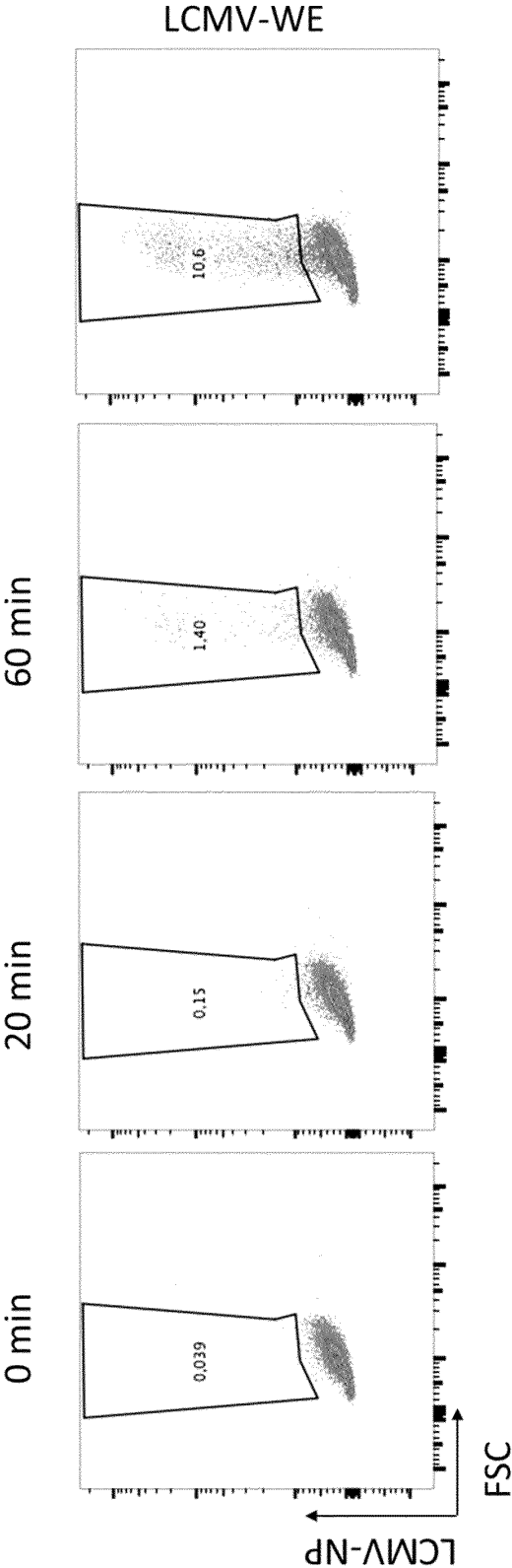


Figure 7 (cont'd)

B

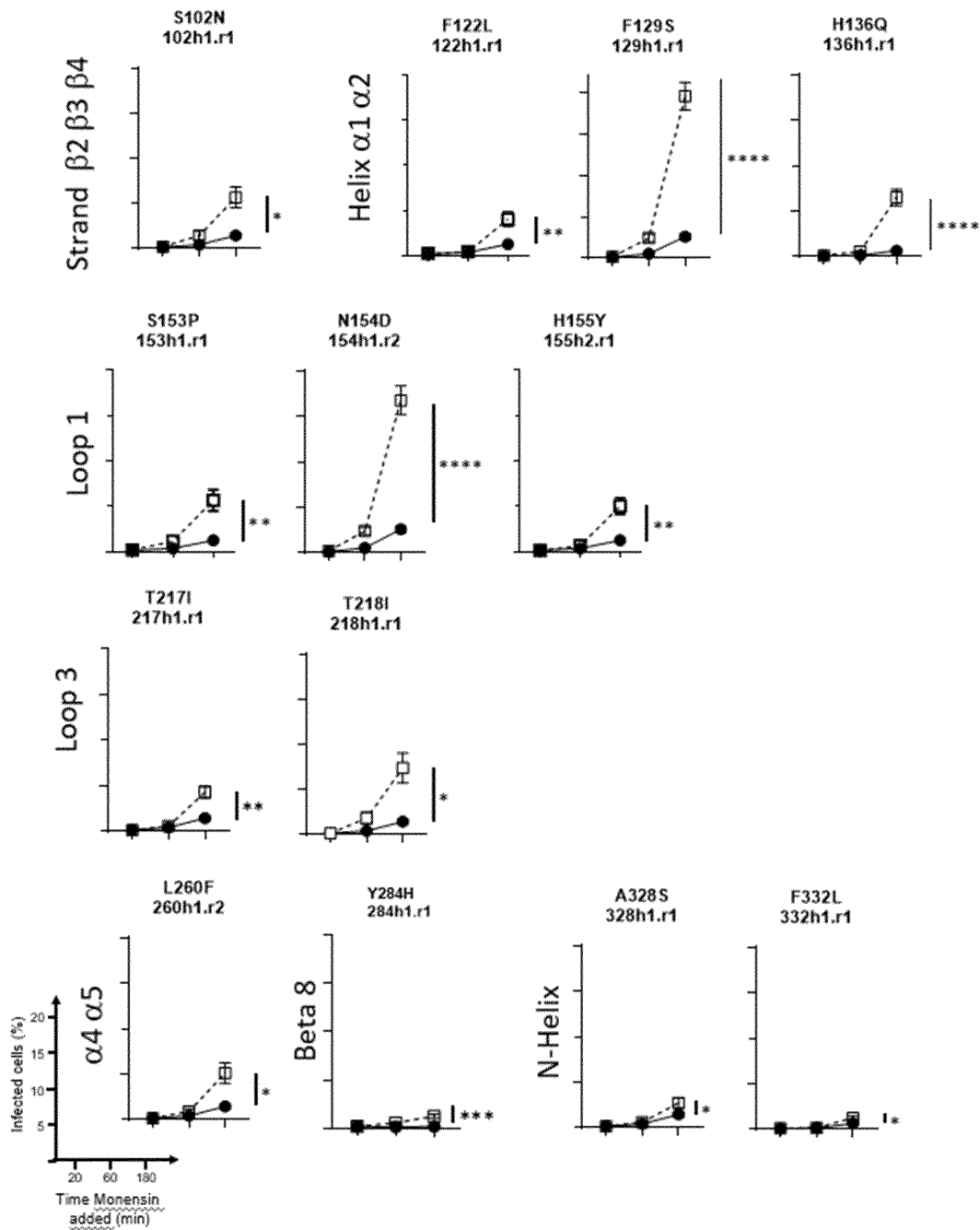
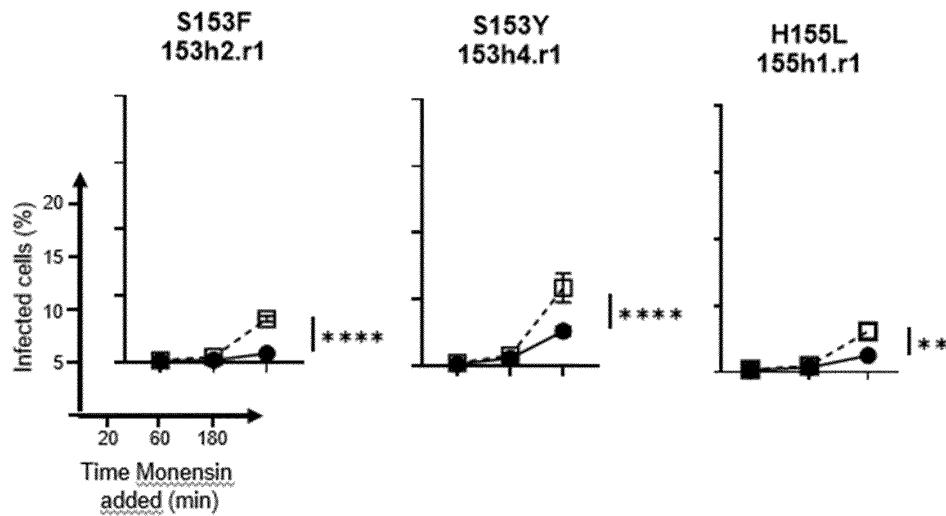


Figure 7 (cont'd)

C



D

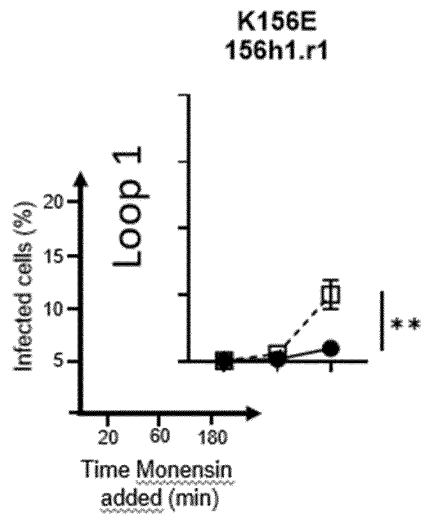
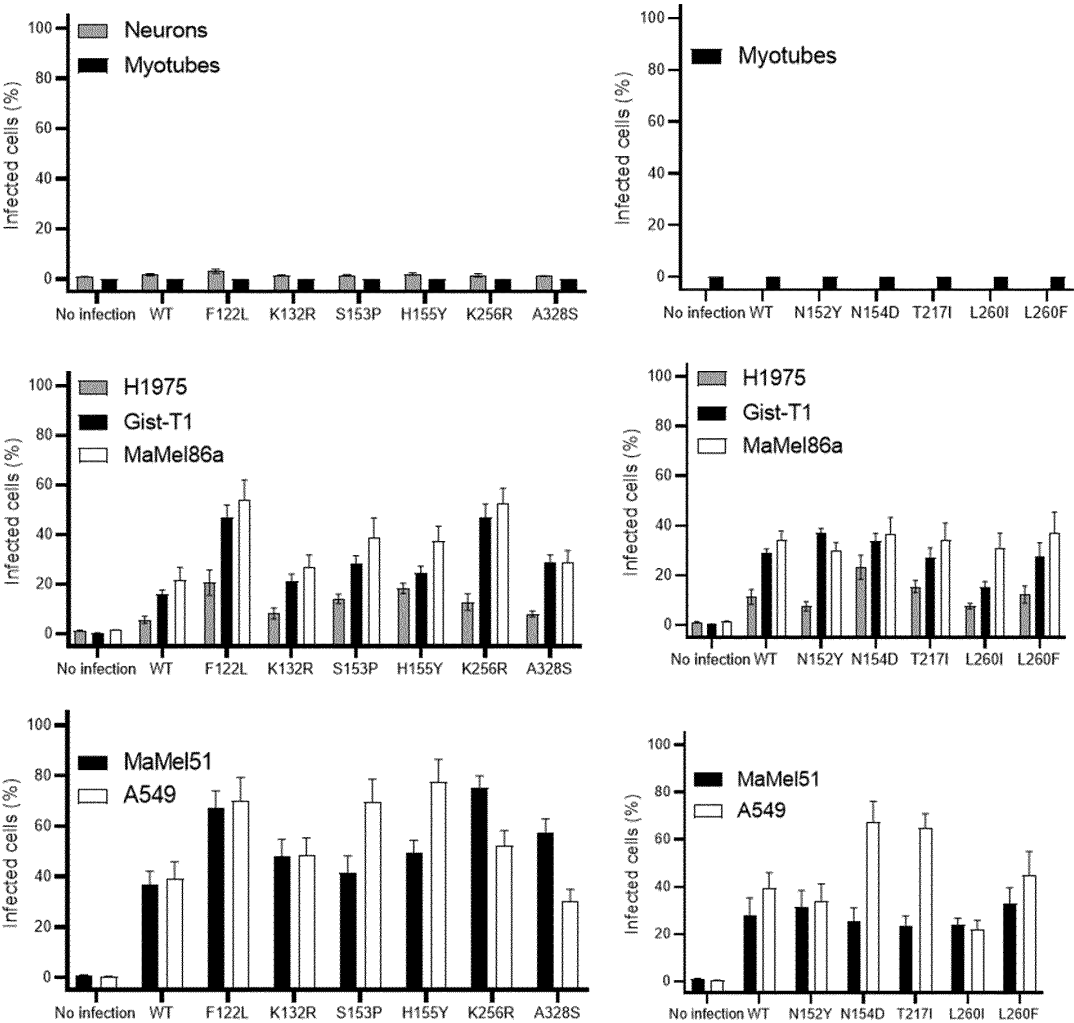


Figure 8

A



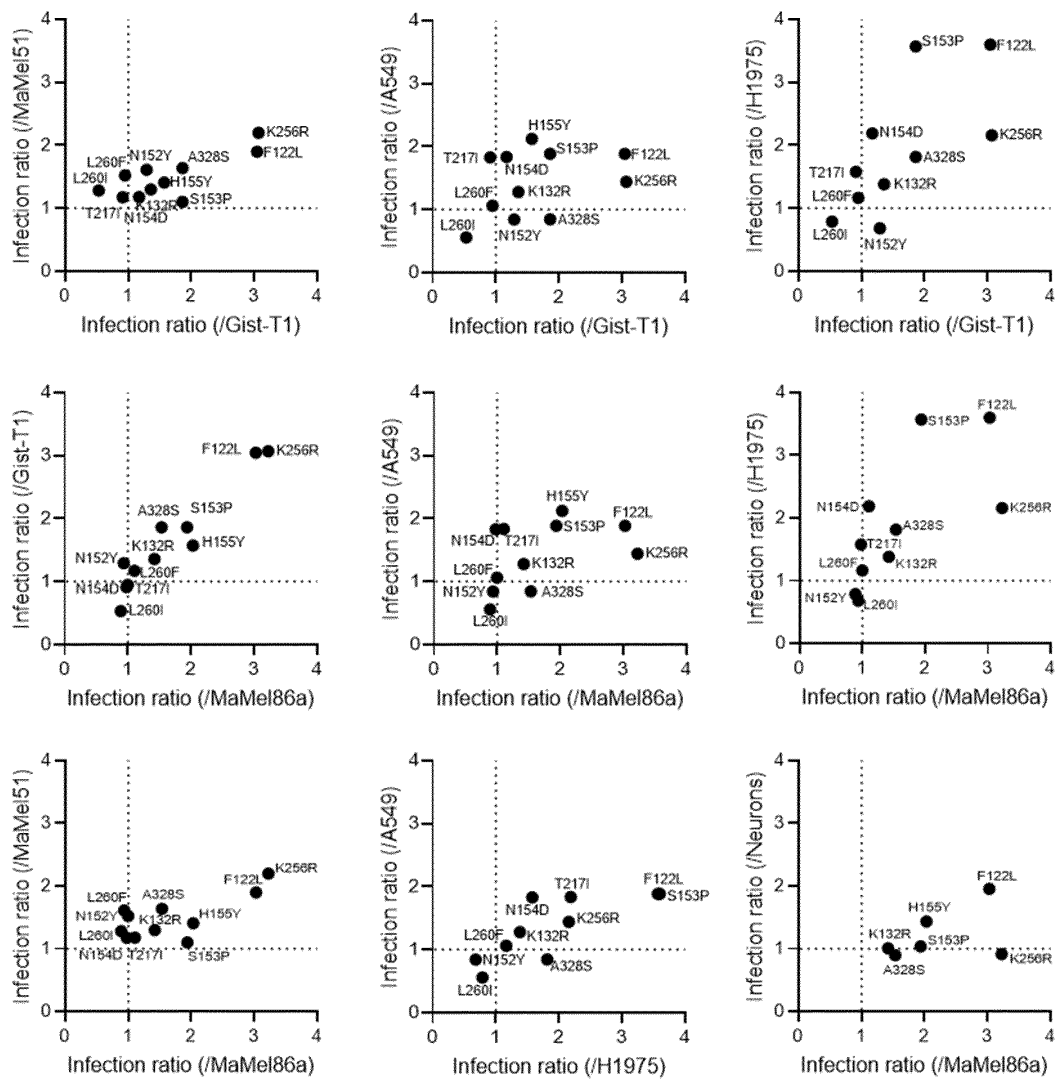
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Figure 8 (cont'd)

C

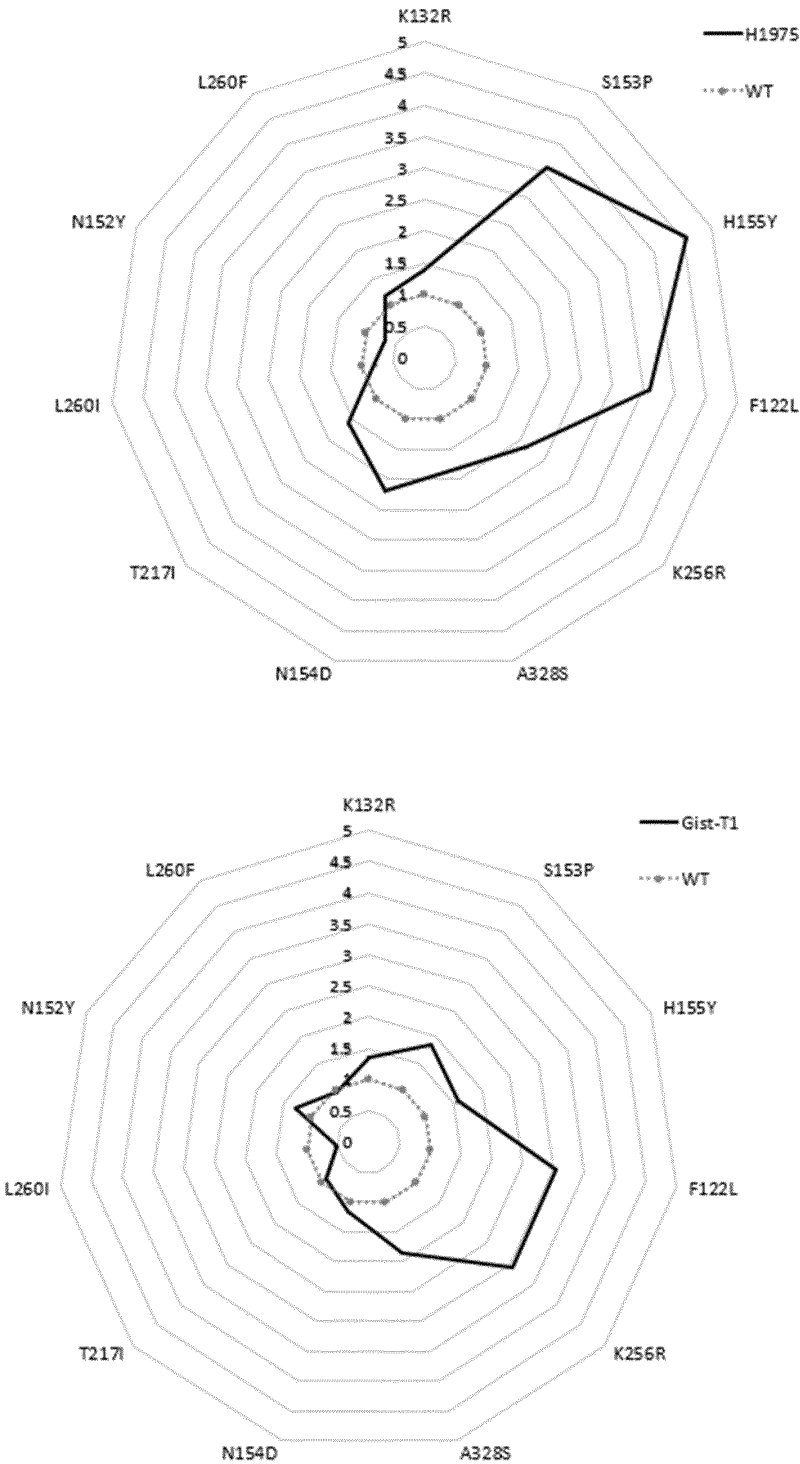


Figure 8 (cont'd)

C (cont'd)

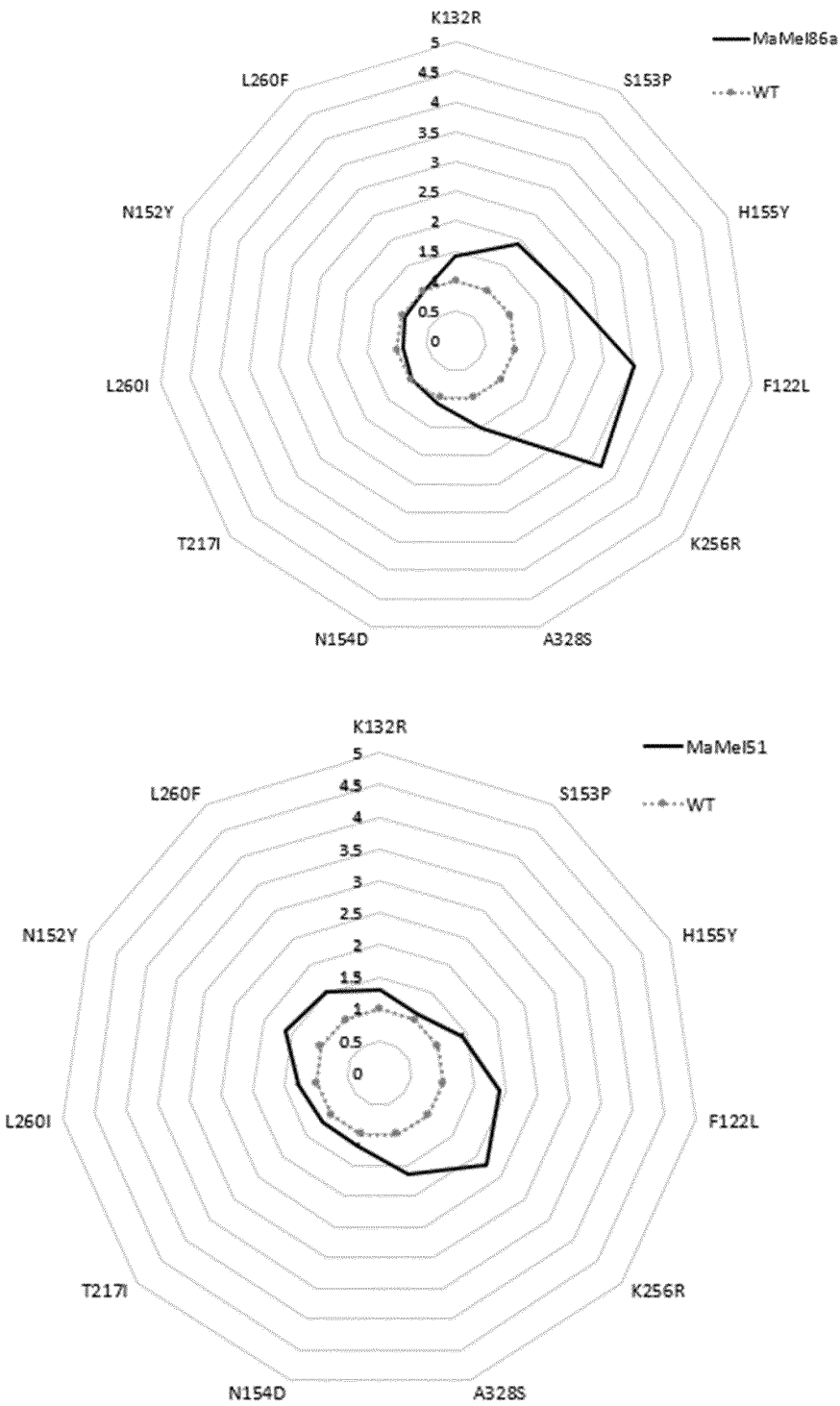


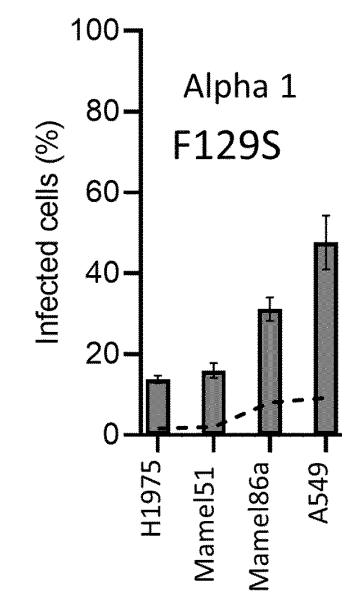
Figure 8 (cont'd)

C (cont'd)



Figure 8 (cont'd)

D



E

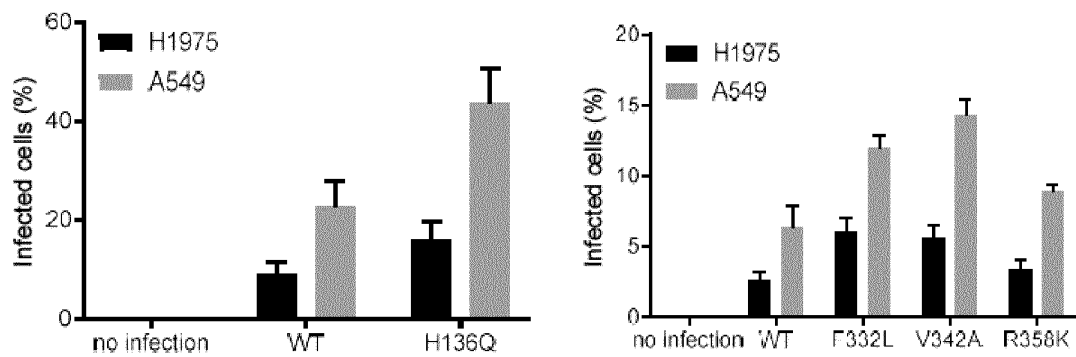
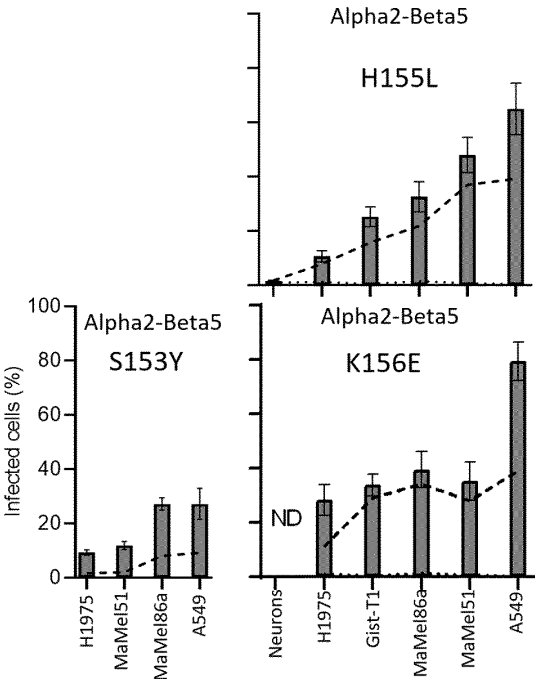
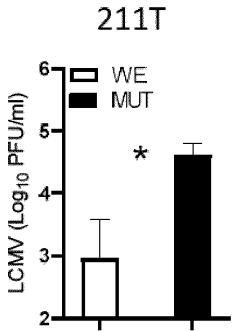


Figure 8 (cont'd)

F



G



H

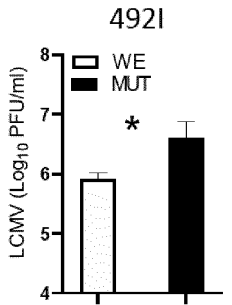


Figure 8 (cont'd)

I

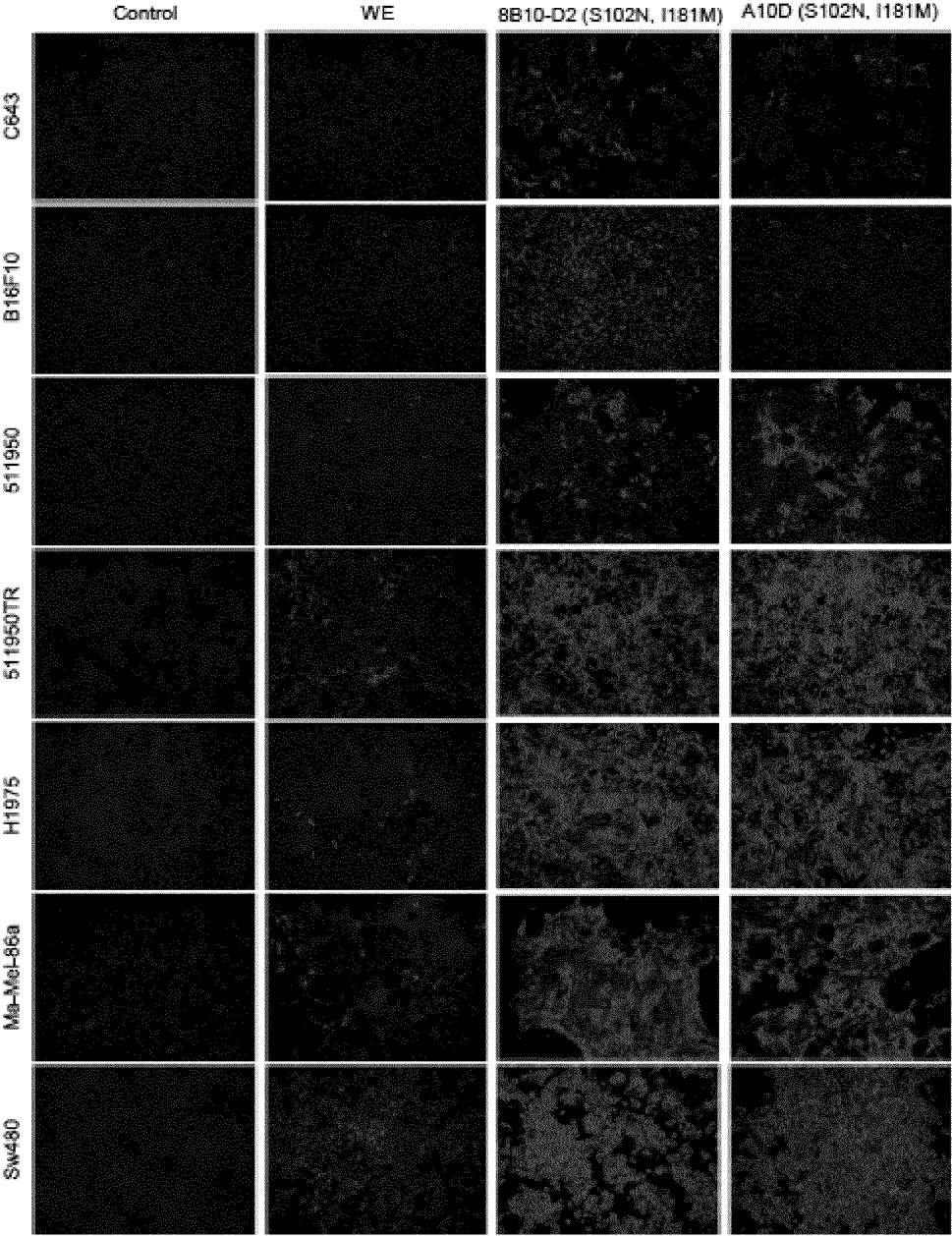


Figure 9

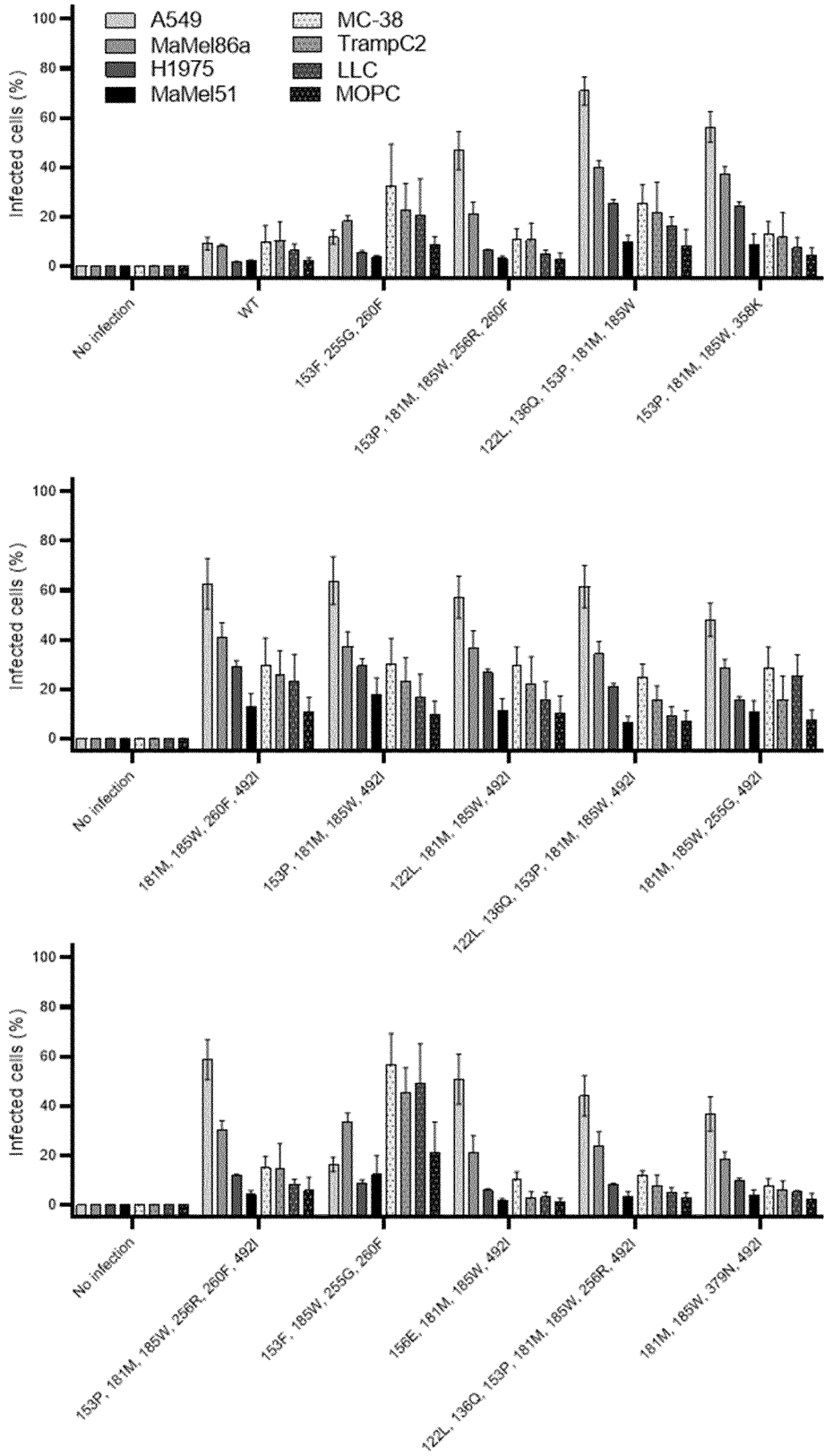


Figure 10

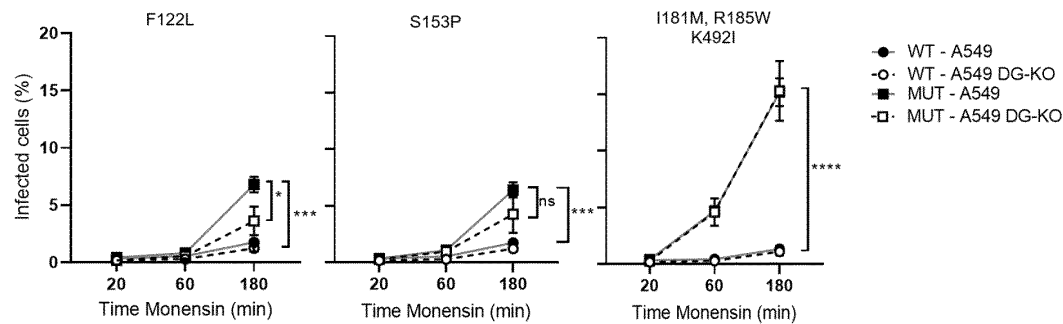
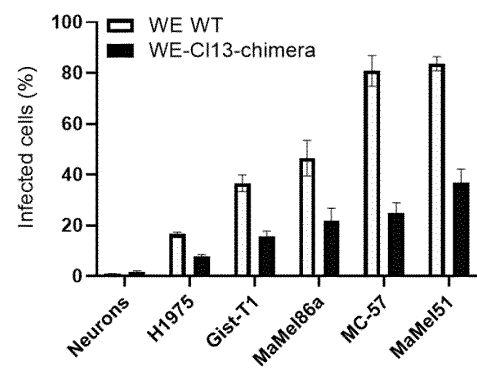


Figure 11

A



B

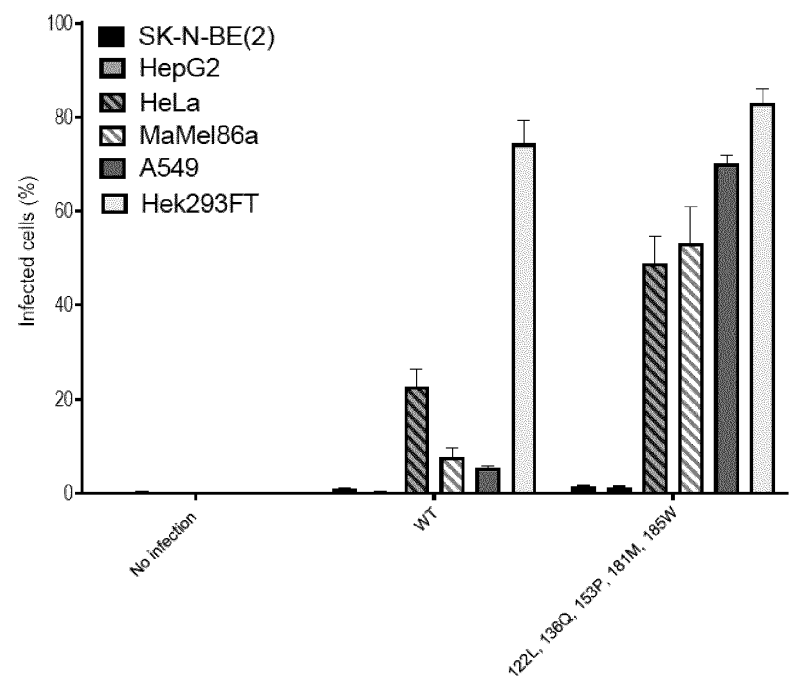
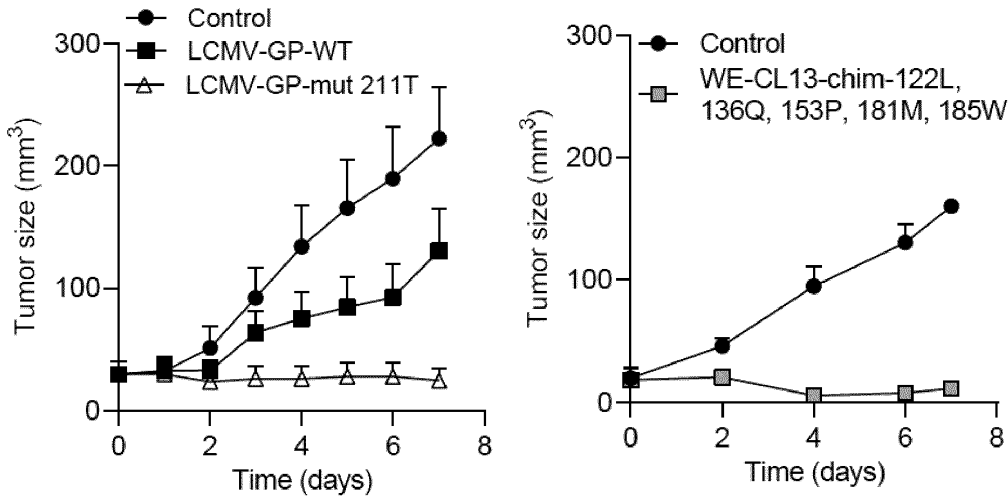


Figure 11 (cont'd)

C



D

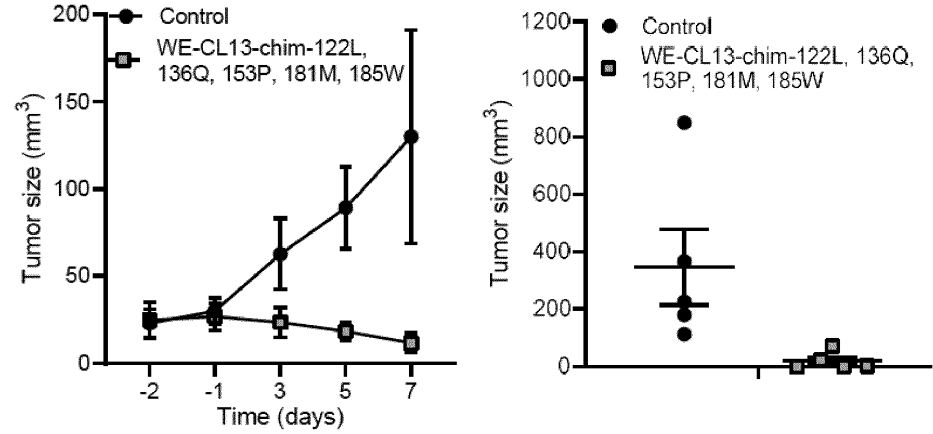


Figure 12

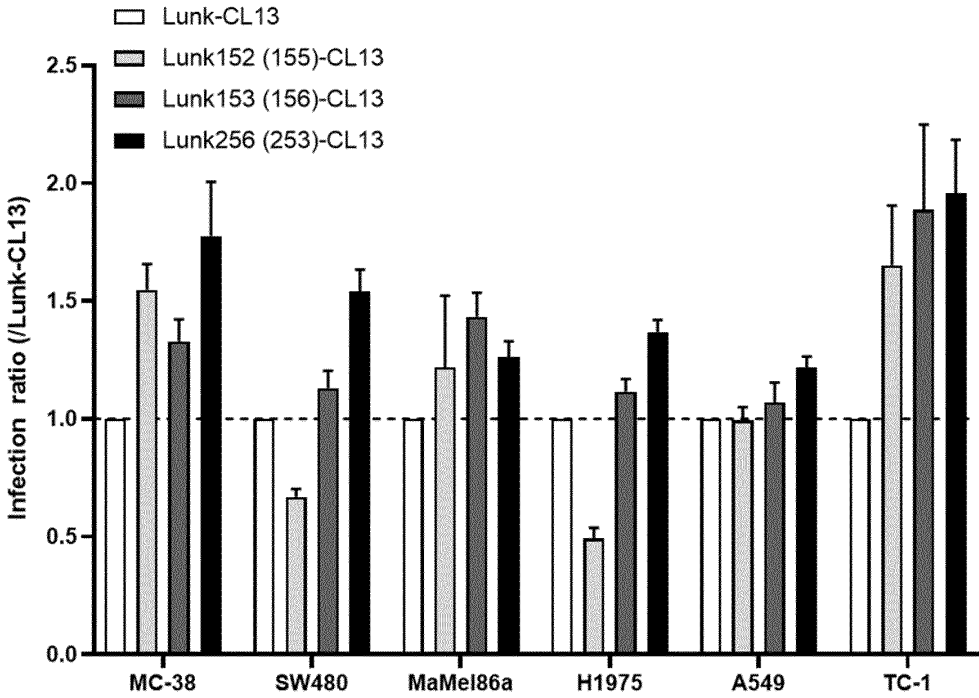


Figure 13

