

Figure 1E

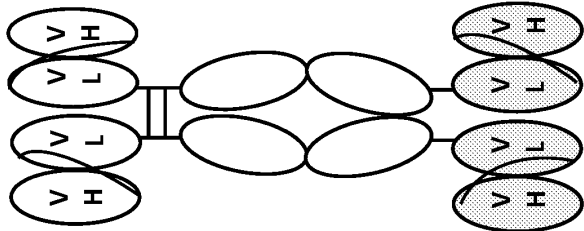


Figure 1D

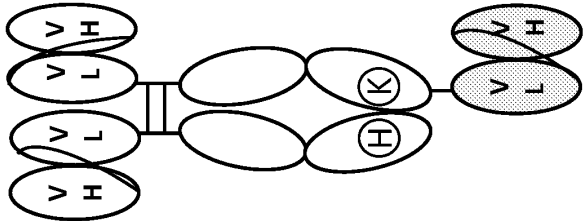


Figure 1C

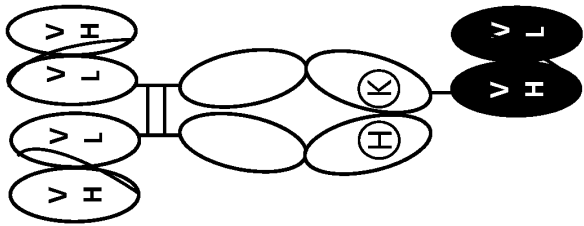


Figure 1B

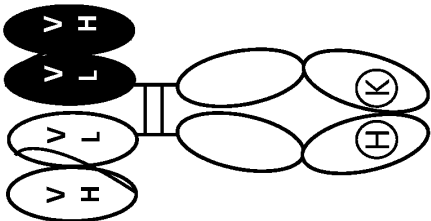
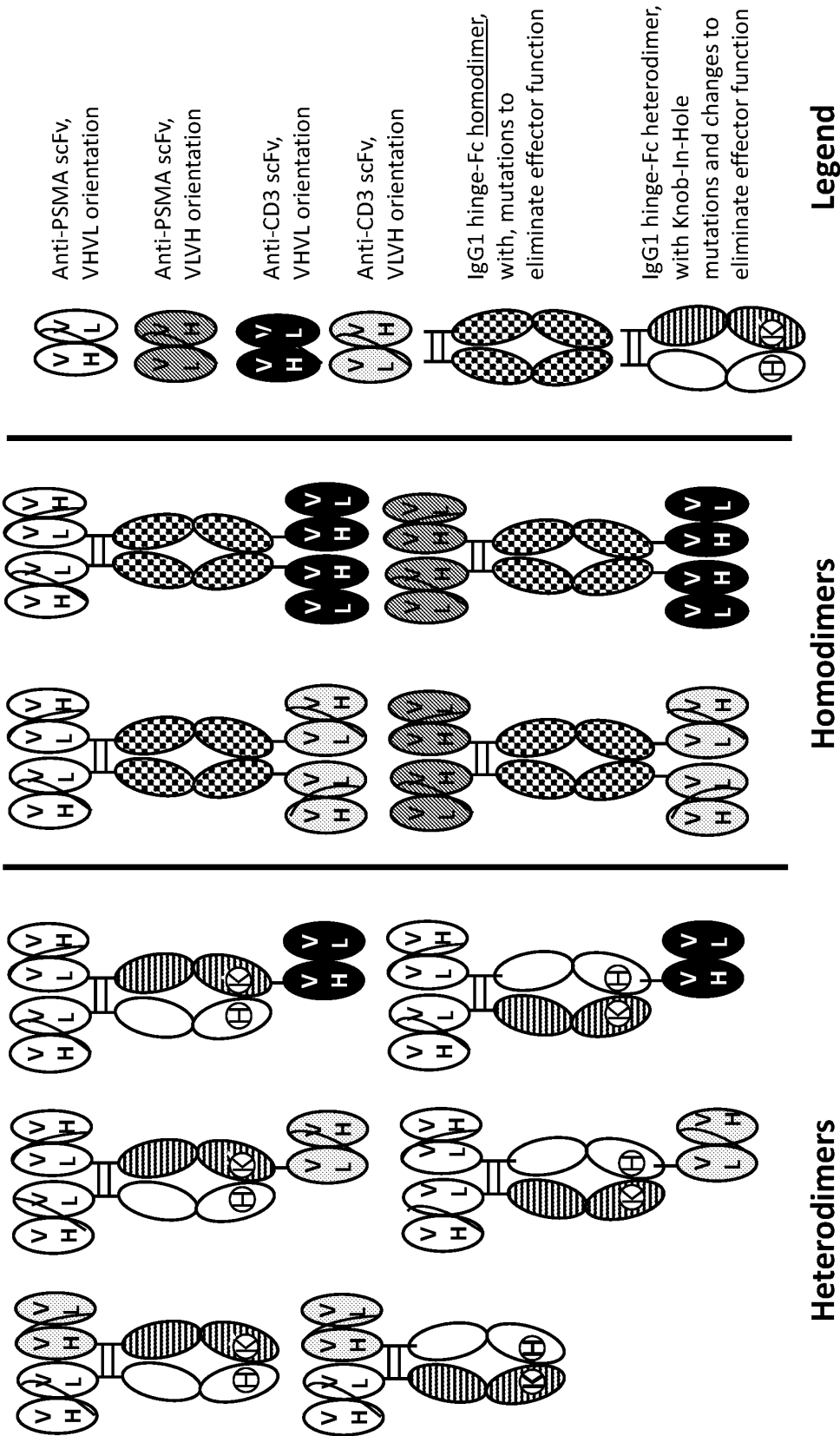


Figure 1A



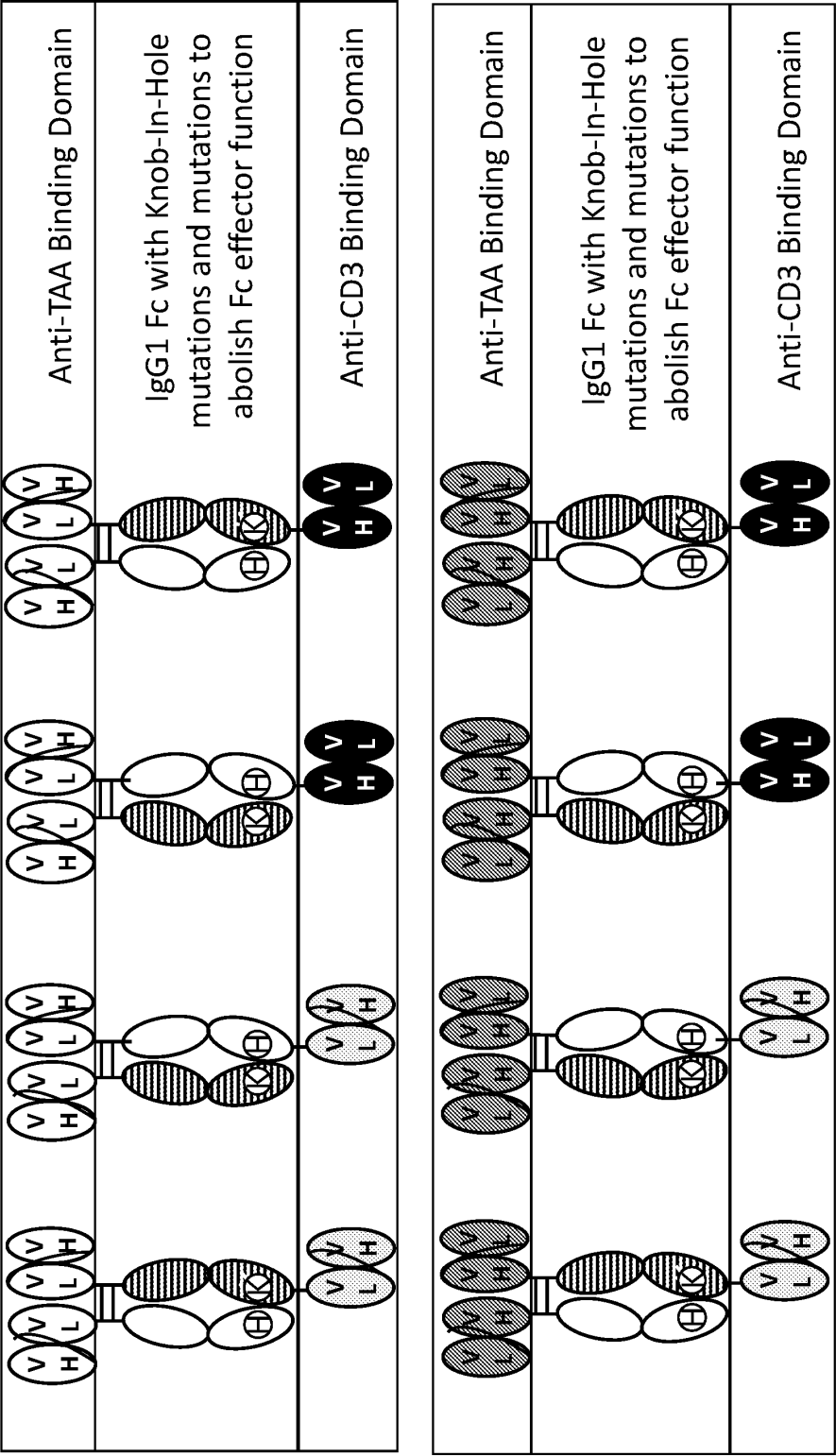


Figure 1G

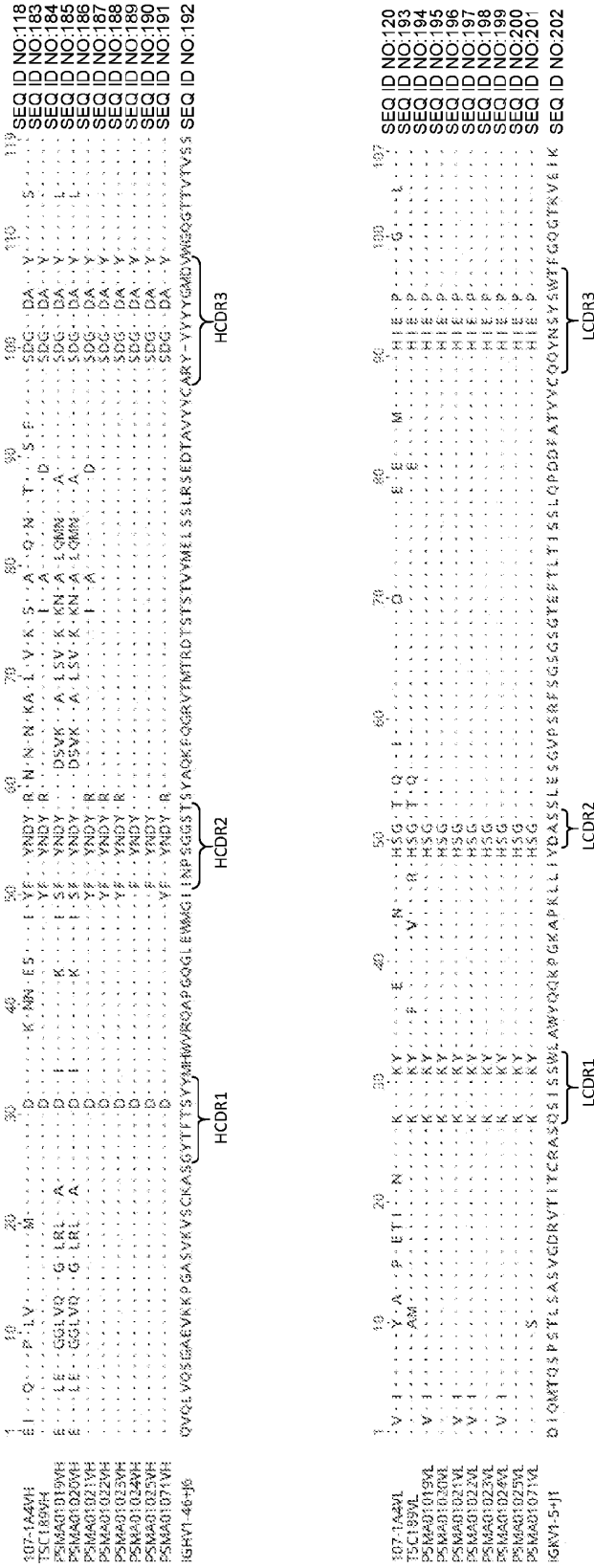


Figure 2

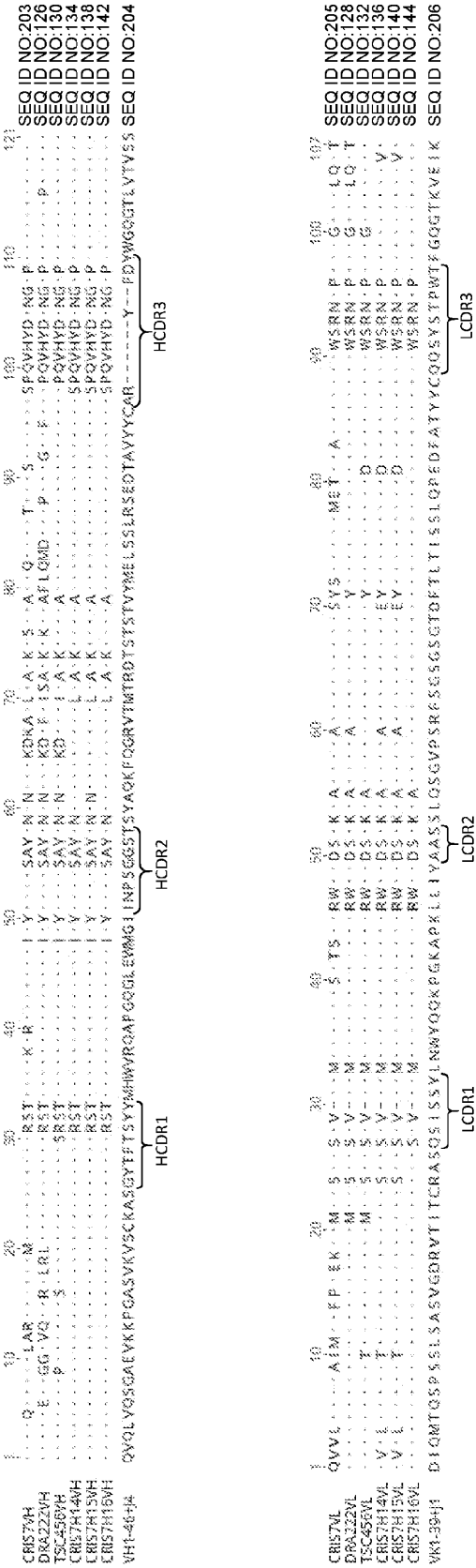
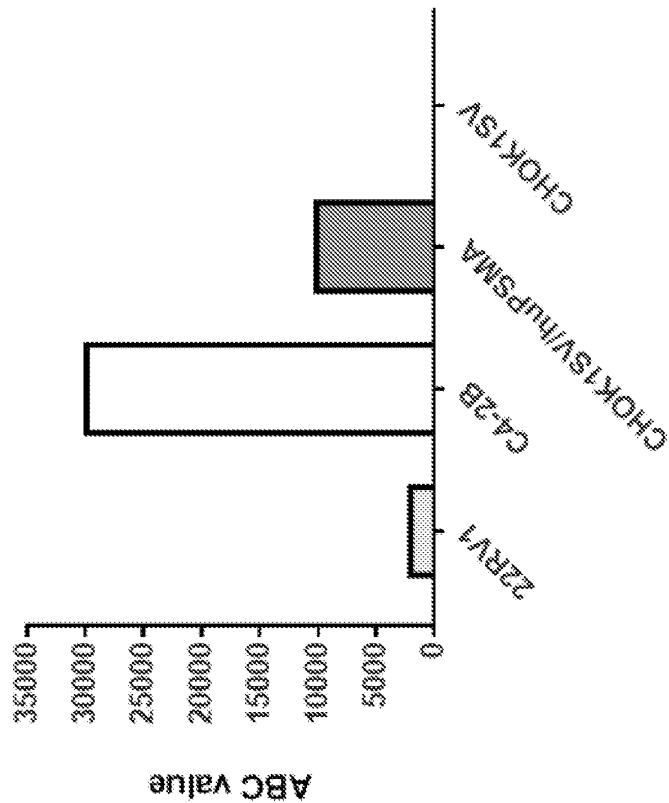


Figure 3

Figure 4: Expression levels of surface PSMA on various cell lines



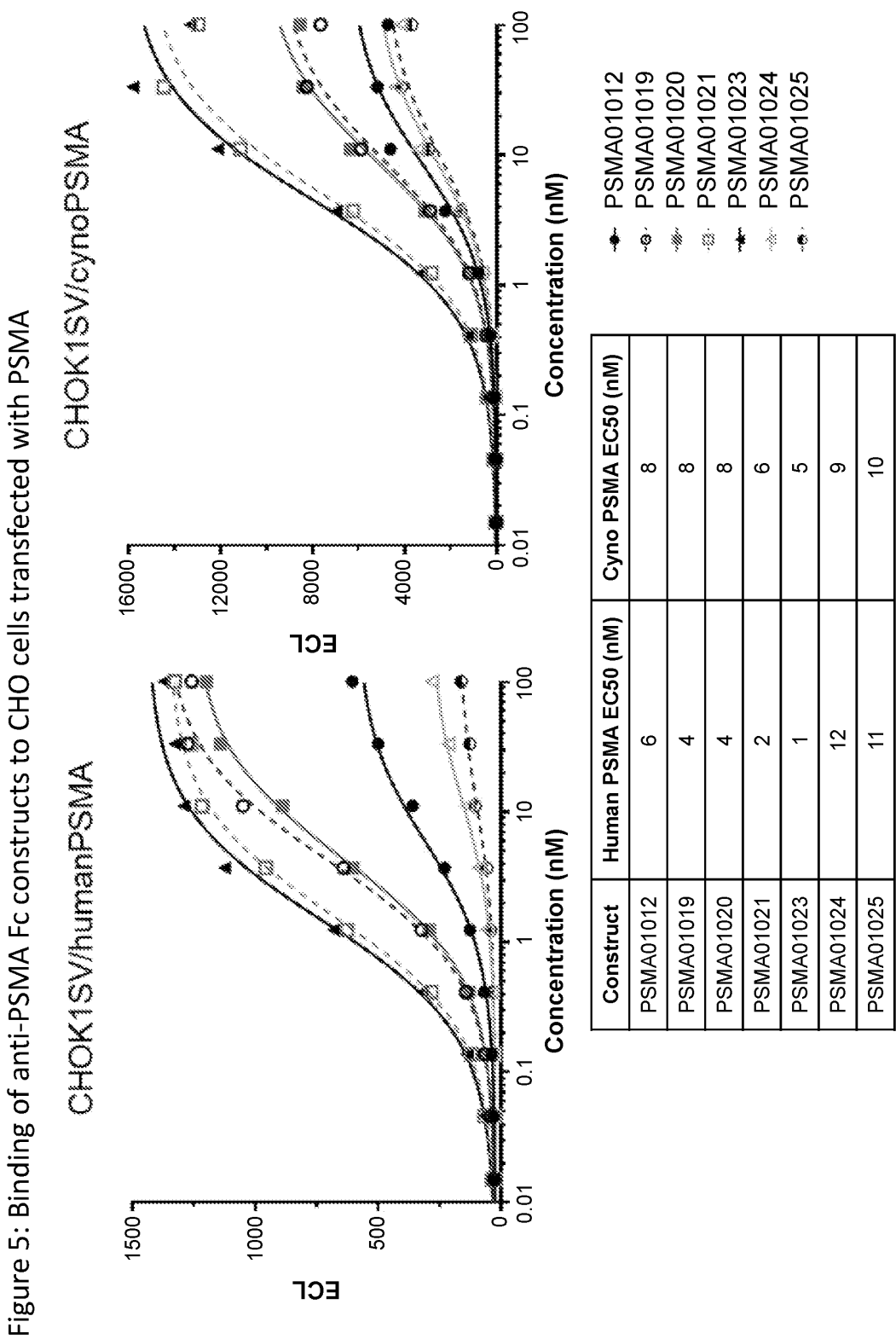


Figure 6: Binding of anti-PSMA Fc constructs in different orientations to PSMA expressing cell lines

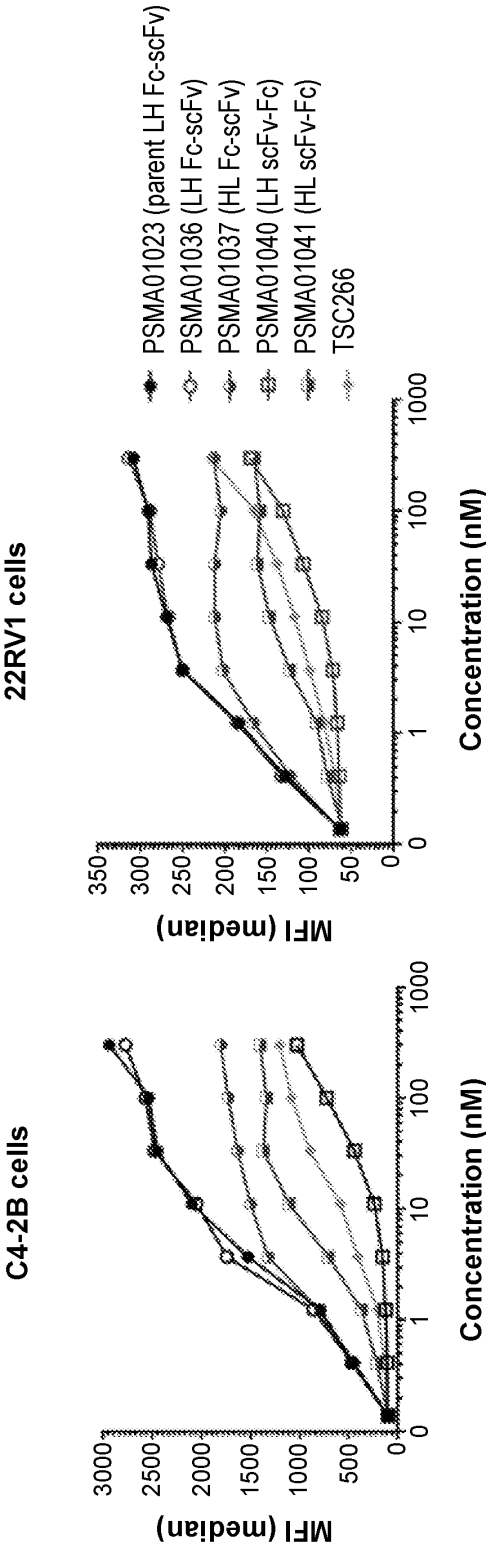


Figure 7: Binding of bivalent and monovalent anti-TA x anti-CD3ε constructs to CD3+ cells

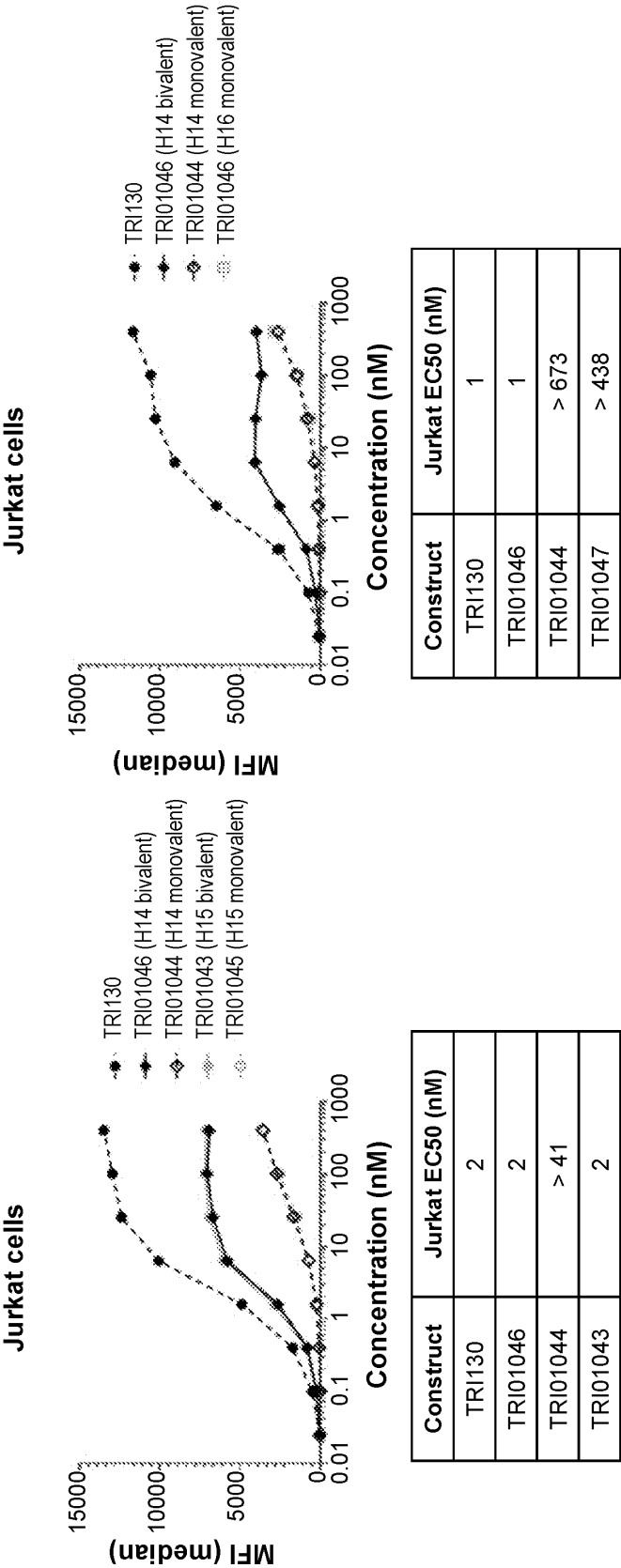


Figure 7B

Figure 7A

Figure 8: T-cell activation induced by monovalent and bivalent anti-TA x anti-CD3ε constructs at 24 hrs with TA+ target cells

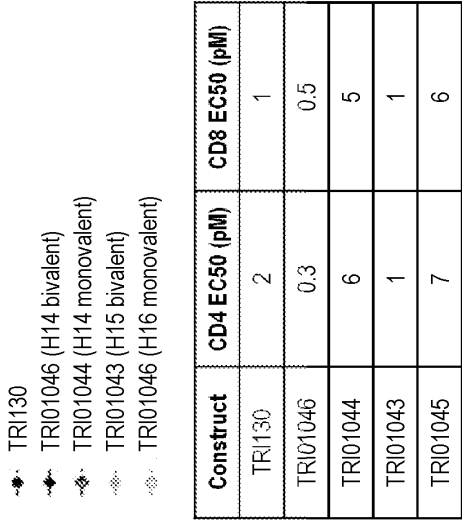


Figure 8A

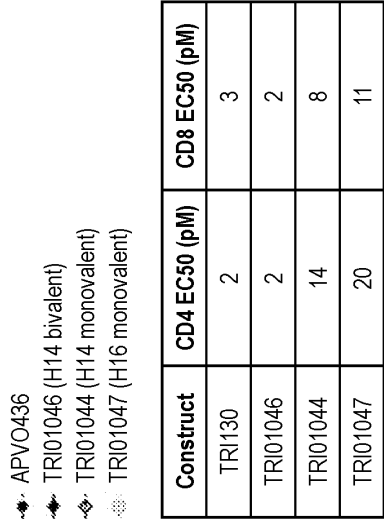
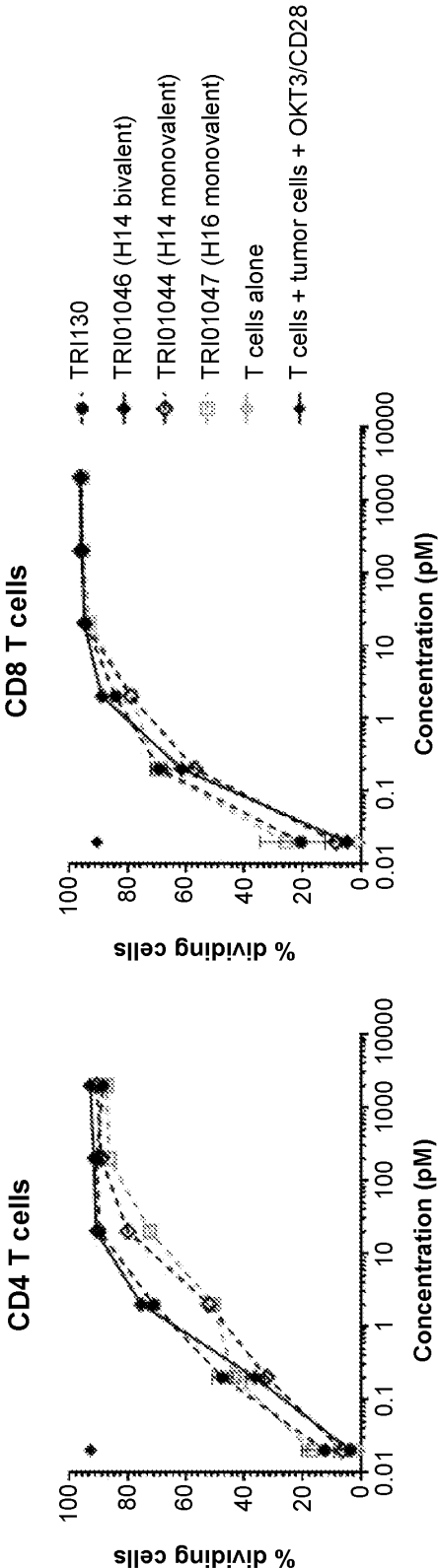


Figure 8B

Figure 9: T-cell proliferation induced by monovalent and bivalent anti-TA x anti-CD3ε constructs at 96 hrs with TA+ target cells



Construct	CD4 EC50 (pM)	CD8 EC50 (pM)
TRI130	0.1	0.01
TRI01044	0.5	0.01
TRI01046	0.3	0.05
TRI01047	0.01	0.01

Figure 10: Cytotoxicity of tumor target cells induced by monovalent and bivalent anti-TA x anti-CD3ε constructs at 96 hrs

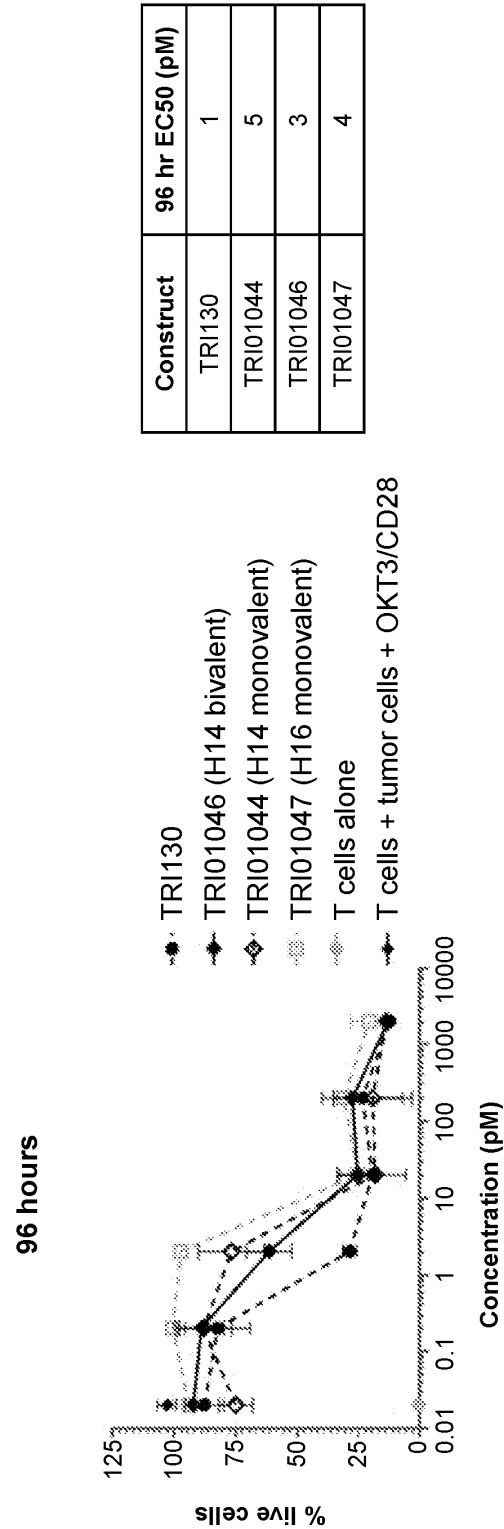
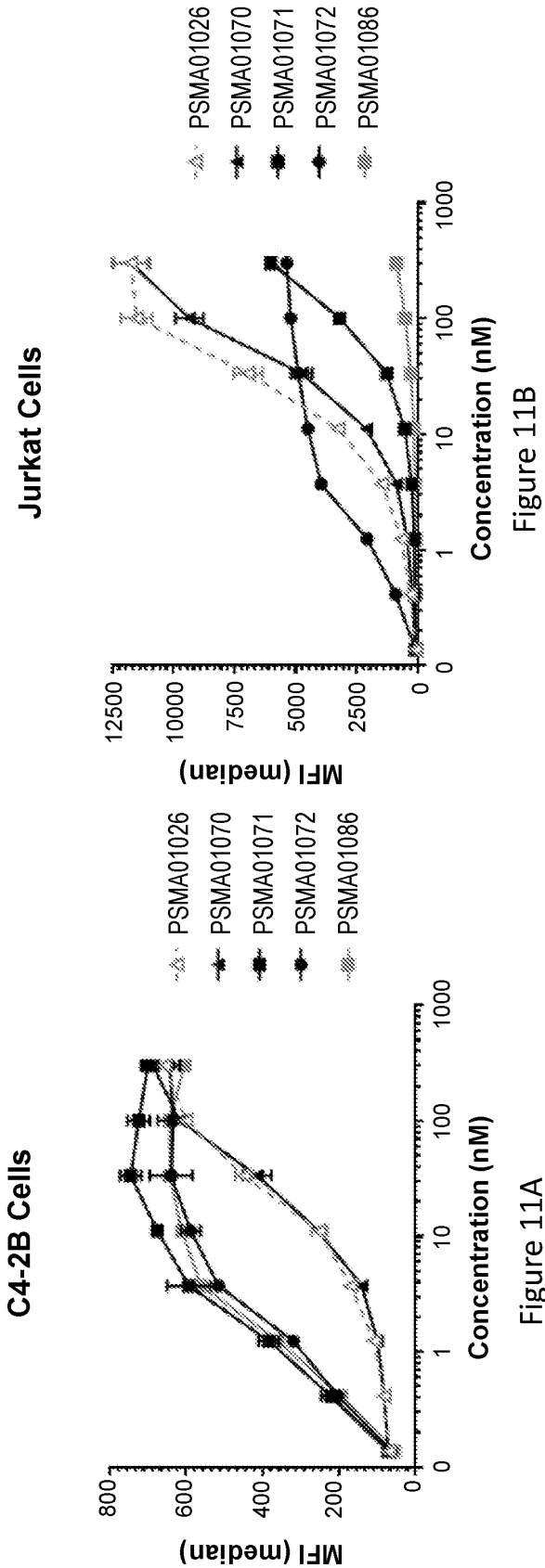


Figure 11: Binding of anti-PSMA x anti-CD3ε constructs in various formats to PSMA+ and CD3+ cells



Construct	C4-2B EC50 (nM)	Jurkat EC50 (nM)
PSMA01026	23	26
PSMA01070	30	52
PSMA01071	1	> 204
PSMA01072	1	1
PSMA01086	1	>> 261

Figure 12: T-cells activation induced by anti-PSMA x anti-CD3ε constructs in various formats at 24 hrs
(A) with C4-2B target cells and (B) without PSMA+ target cells

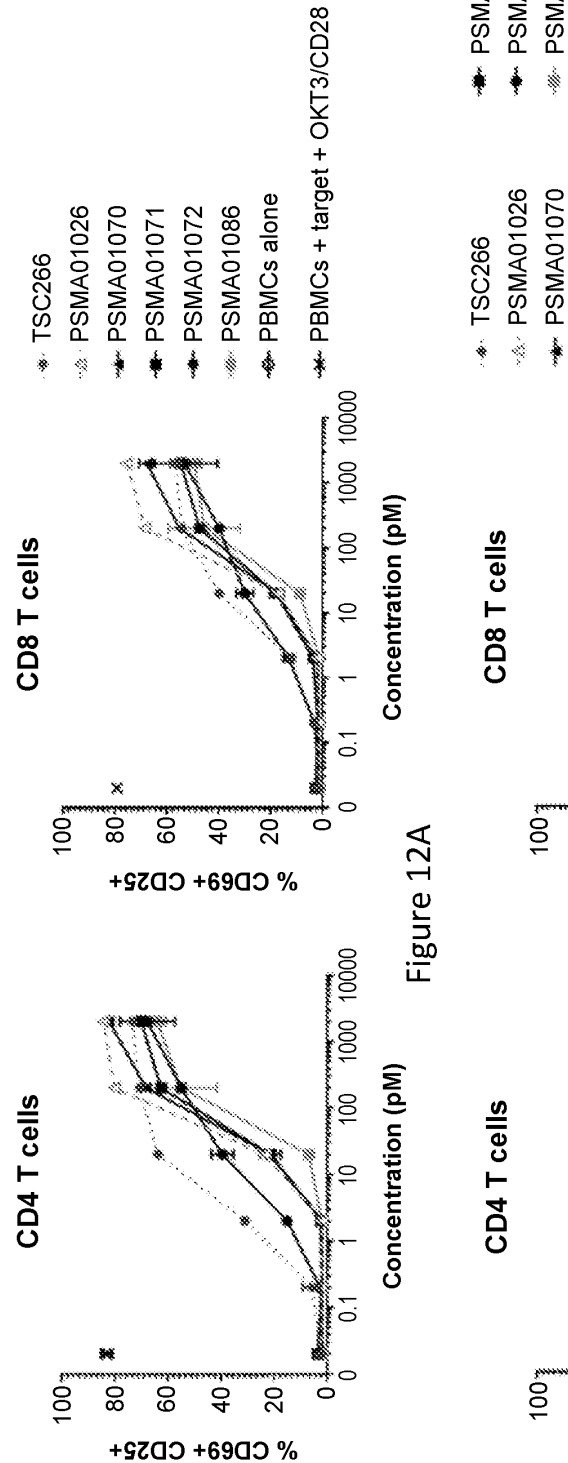


Figure 12A

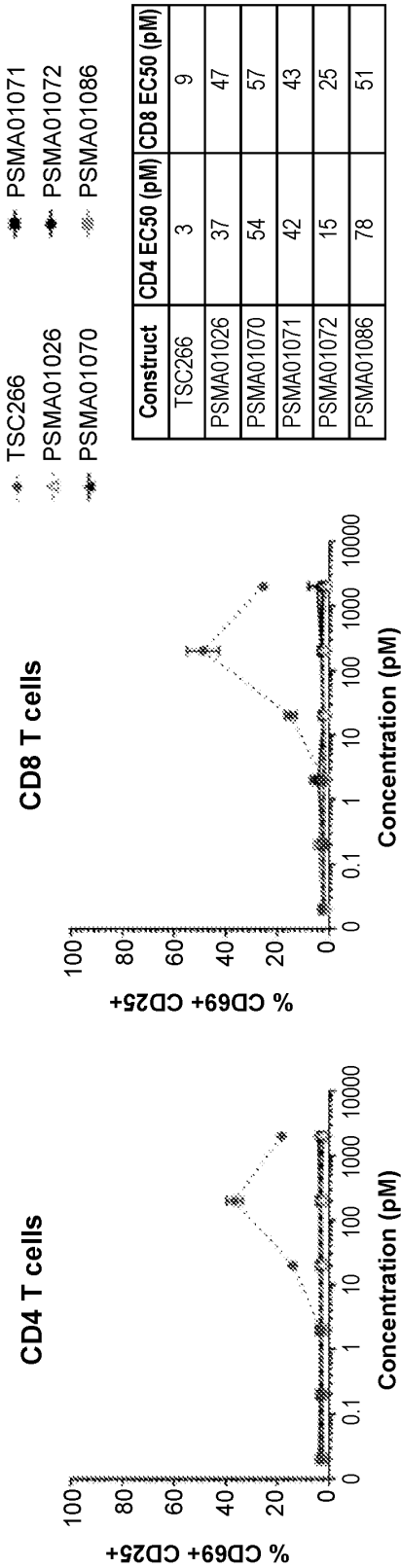


Figure 12B

Figure 13: Cytokine secretion induced by PSMA x CD3 constructs after 24 hrs of stimulation with PSMA+ target cells

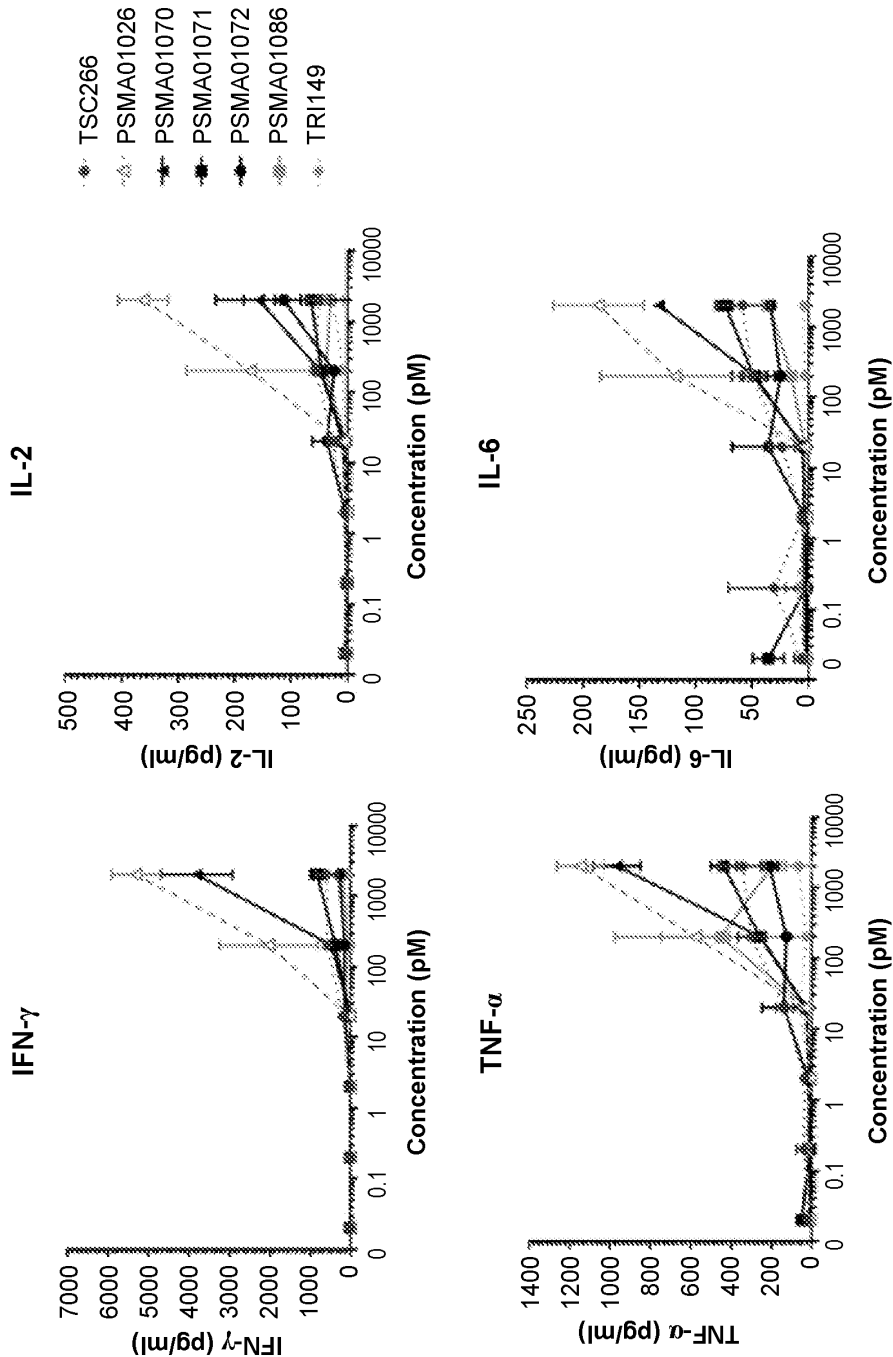


Figure 14: T-cell proliferation induced by PSMA x anti-CD3ε constructs at 96 hours in the presence of PSMA+ tumor cells

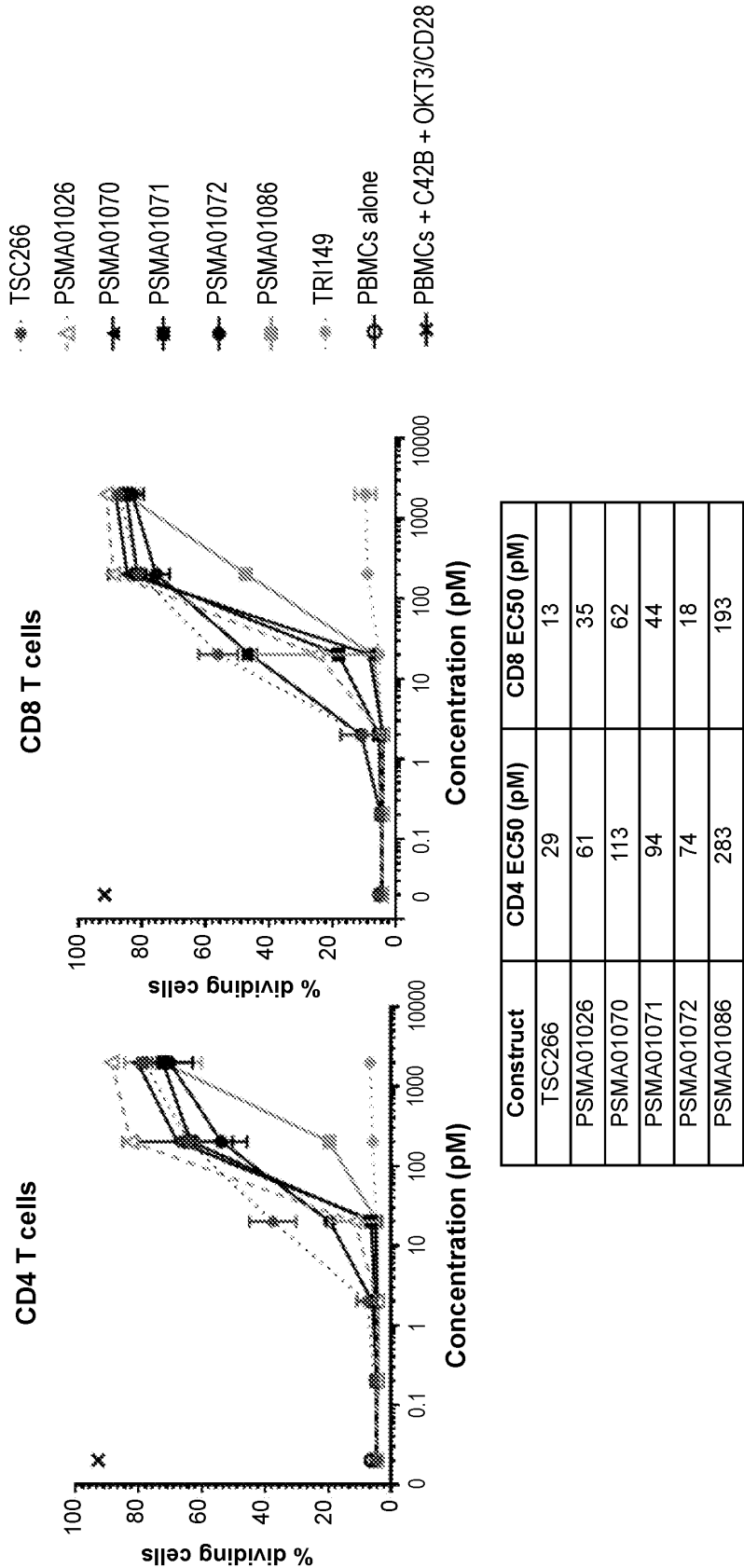


Figure 15: Cytotoxicity of PSMA+ target cells induced by anti-PSMA x anti-CD3ε constructs at 72 and 96 hrs

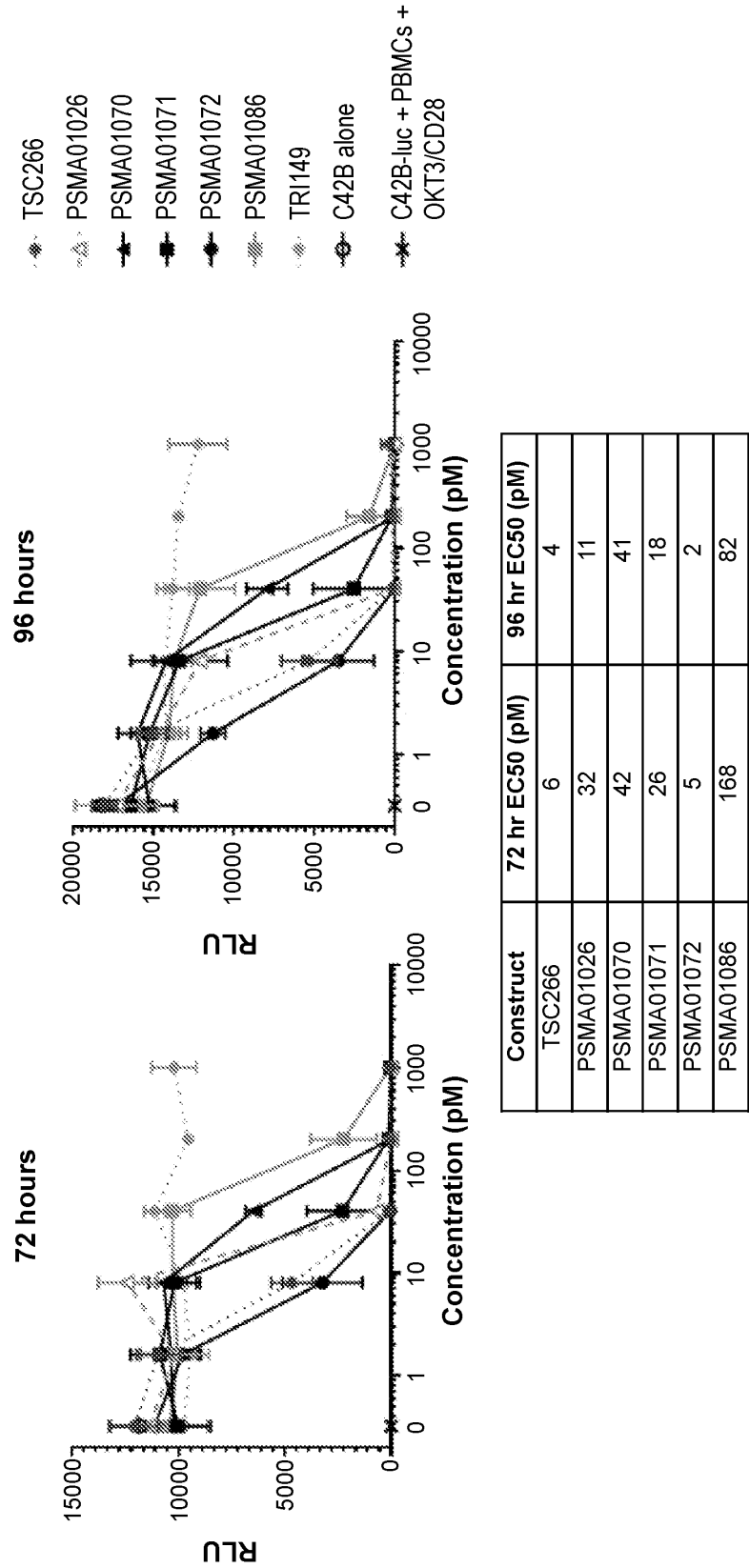


Figure 16: Growth of C4-2B-luciferase tumor cells in NOD/SCID mice following treatment with CD3 x PSMA constructs

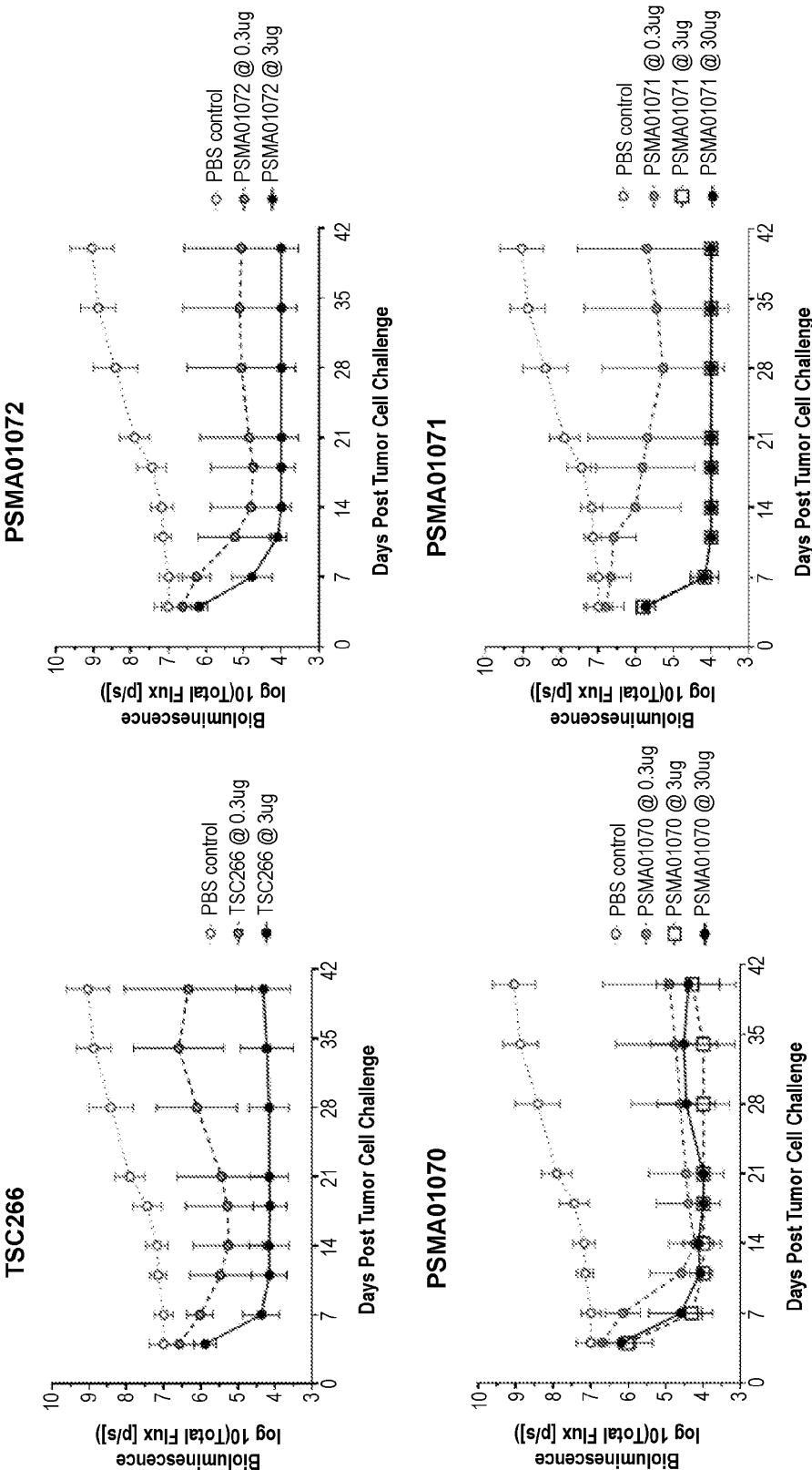
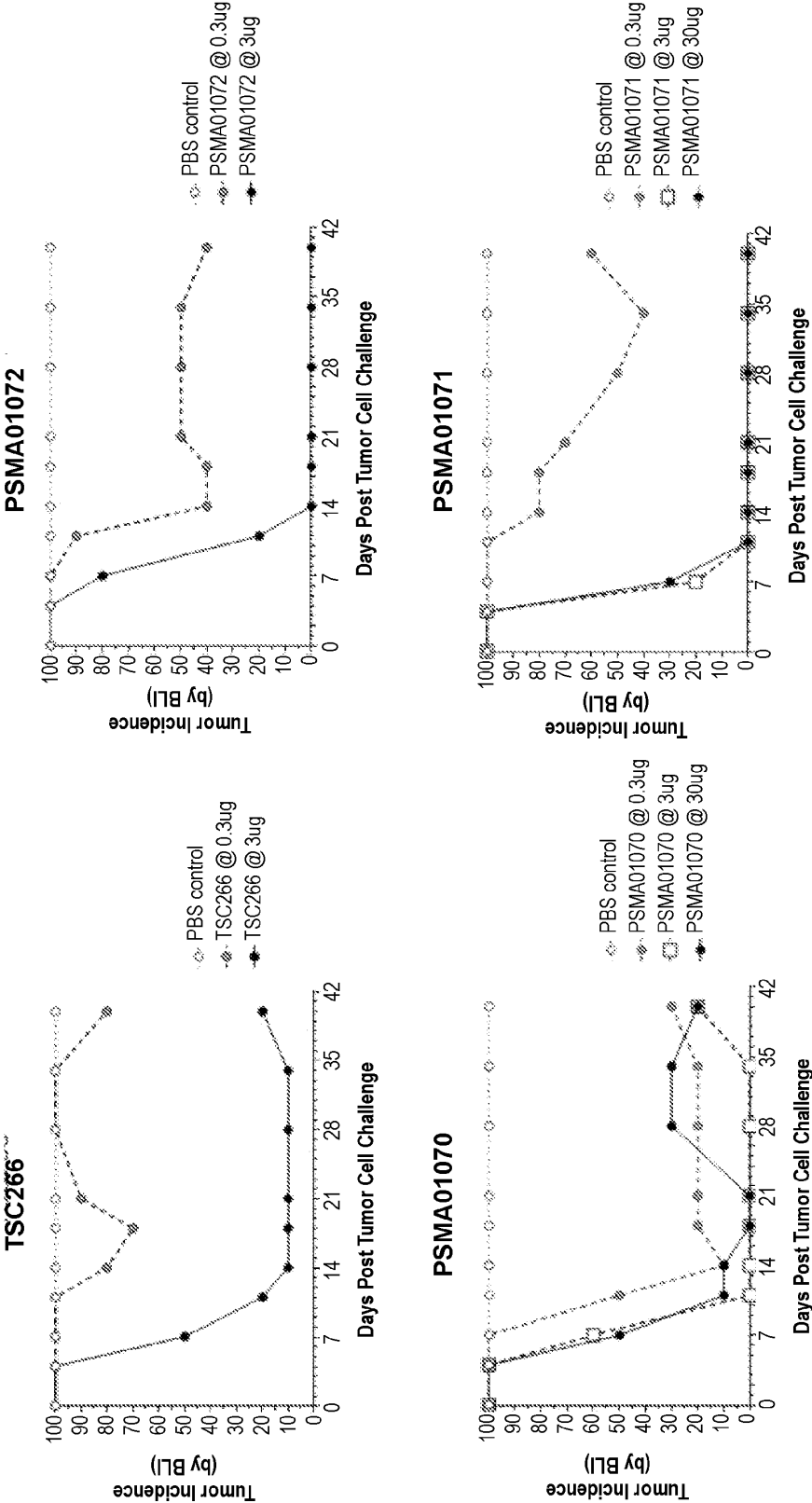


Figure 17: C4-2B-luciferase tumor incidence cells in NOD/SCID mice following treatment with CD3 x PSMA constructs



Figures 18A-B: Binding of anti-PSMA x anti-CD3ε constructs in various formats to (A) PSMA+ and (B) CD3+ cells

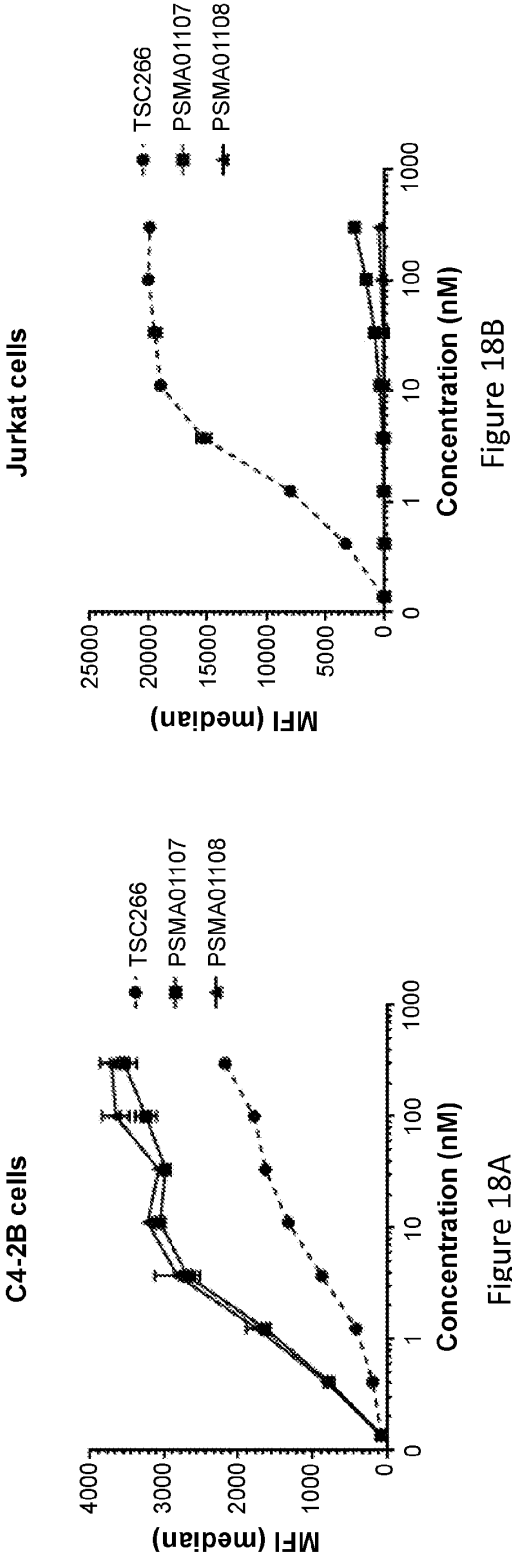
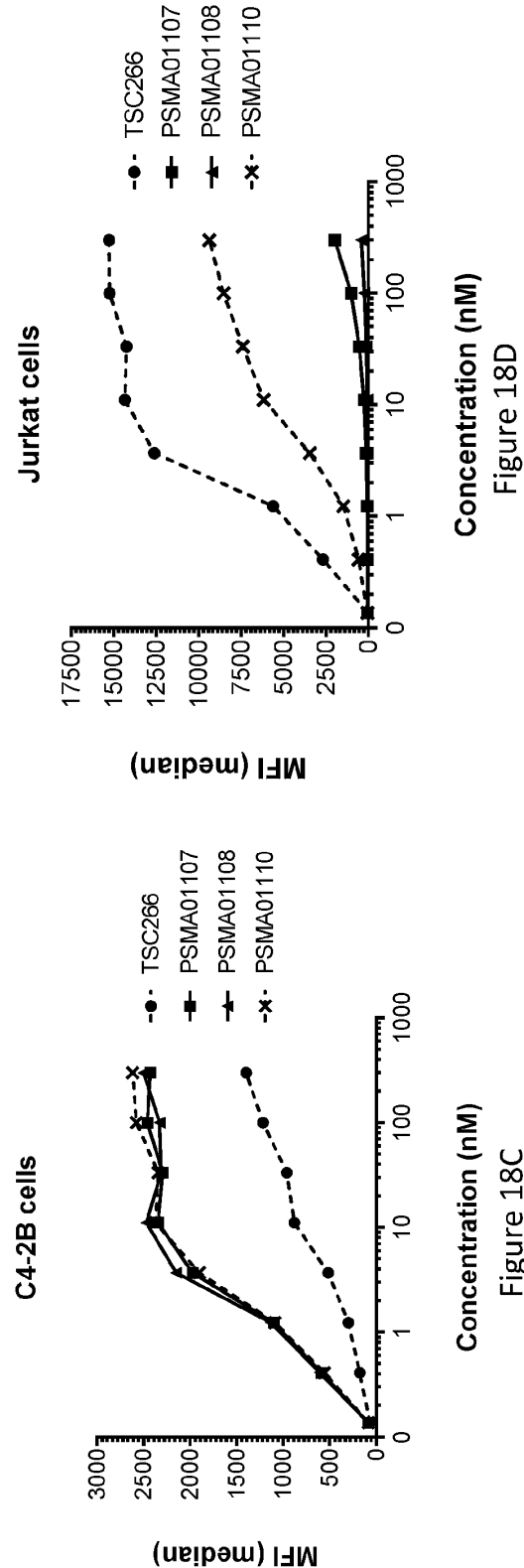


Figure 18A

Figure 18B

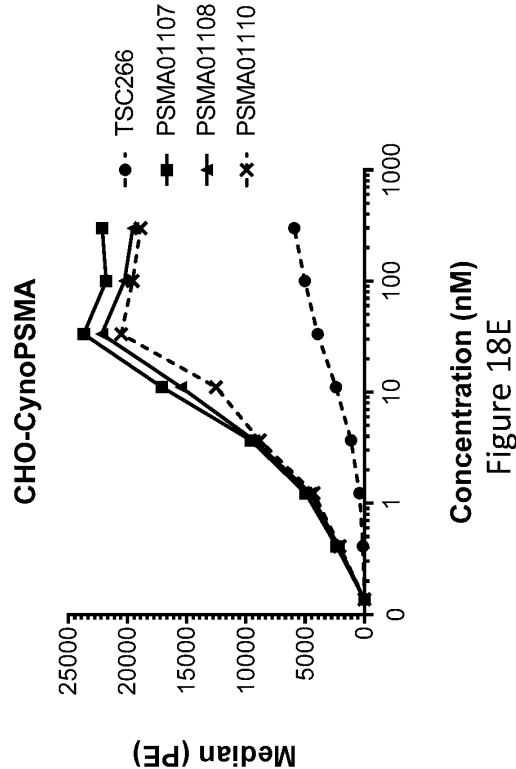
Construct	C4-2B EC50 (nM)	Jurkat EC50 (nM)
TSC266	7	2
PSMA01107	1	> 208
PSMA01108	1	>> 210

Figures 18C-D: Binding of anti-PSMA x anti-CD3ε constructs in various formats to (C) PSMA+ and (D) CD3+ cells



Construct	C4-2B EC50 (nM)	Jurkat EC50 (nM)
TSC266	7	2
PSMA01107	1	> 200
PSMA01108	1	>> 200
PSMA01110	1	6

Figure 18E: Binding of anti-PSMA x anti-CD3ε constructs in various formats to (E) CHO-CynoPSMA cells



On-Cell Binding Parameters	PSMA01108		PSMA01107		PSMA01110		TSC266	
	EC50 (nM)	Max	EC50 (nM)	Max	EC50 (nM)	Max	EC50 (nM)	Max
Human PSMA binding affinity	1	2500	1	2400	1	2600	10	1400
Cyno PSMA binding affinity	4	19500	5	22000	5	19000	20	6000
Human CD3 binding affinity	> 200	409	> 200	2000	7	9000	2	15000

Figure 19: T-cell activation induced by monovalent and bivalent anti-PSMA x anti-CD3ε constructs at 24 hrs (A) with PSMA+ target cells or (B) without target cells

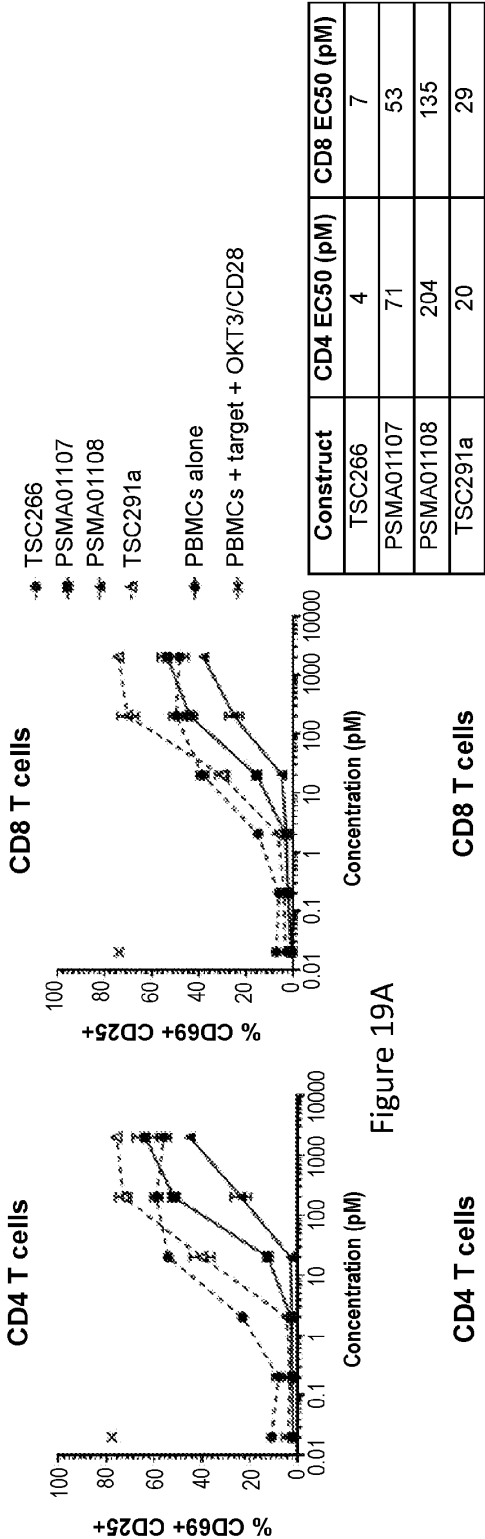


Figure 19 C-E: T cell activation induced by monovalent and bivalent anti-PSMA x anti-CD3ε constructs at 24hrs (C) with PSMA+ target cells (D) without PSMA+ target cells and (E) single concentration (200 pM)

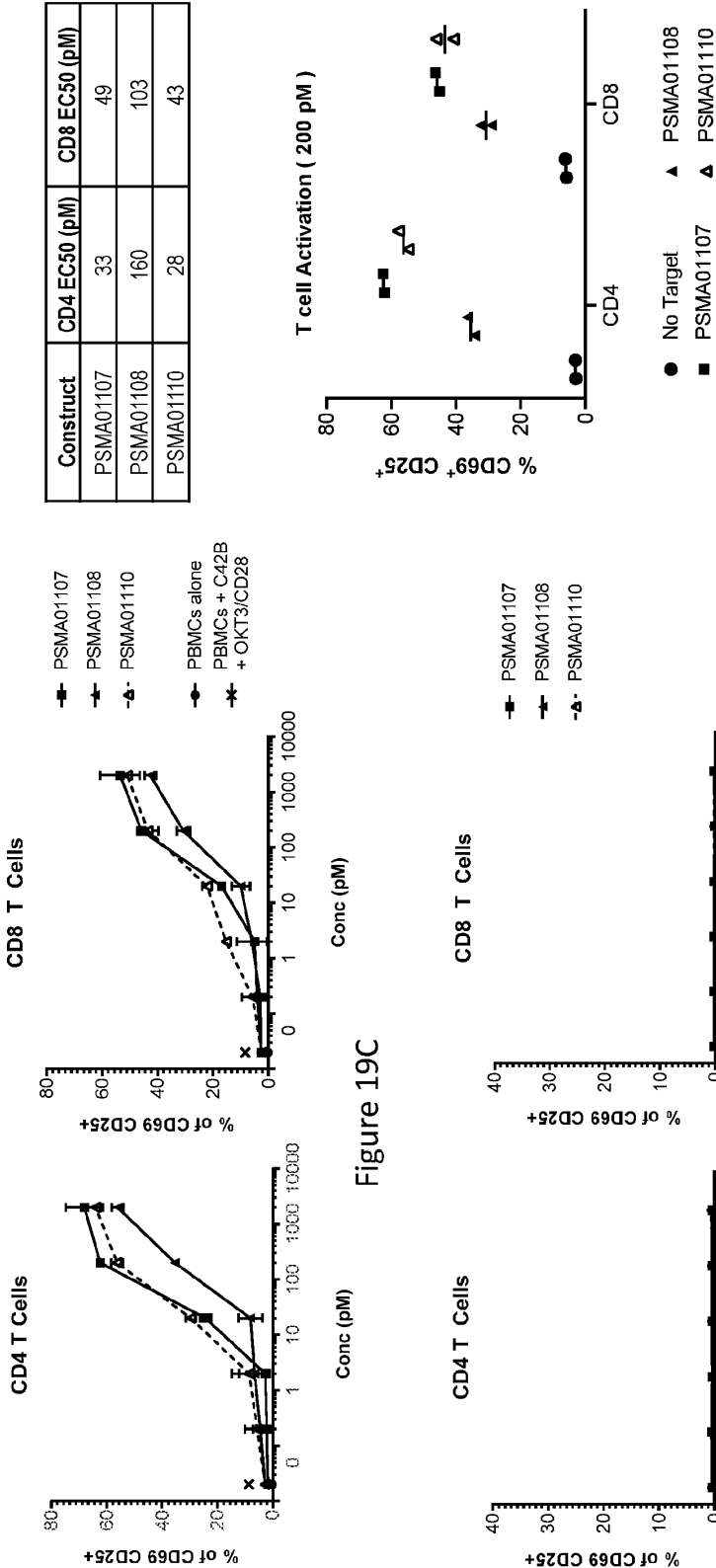
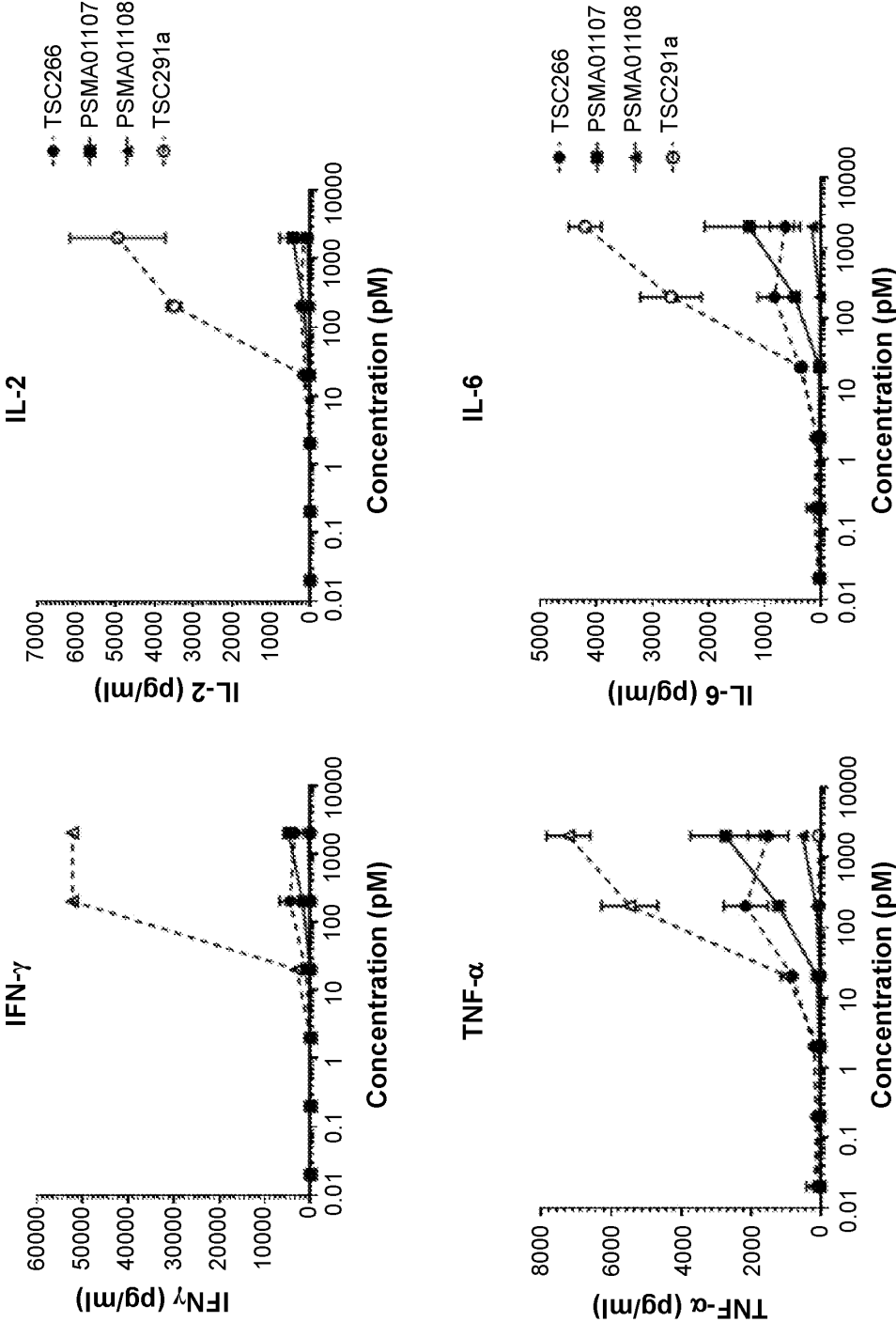


Figure 20A: Cytokine secretion induced by monovalent and bivalent anti-PSMA x anti-CD3ε constructs at 24hrs



Figures 20 B-E: Cytokine secretion induced by monovalent and bivalent anti-PSMA x anti-CD3ε constructs at 24hrs (C) with PSMA+ target cells (D) without PSMA+ target cells and (E) single concentration (200 nM)

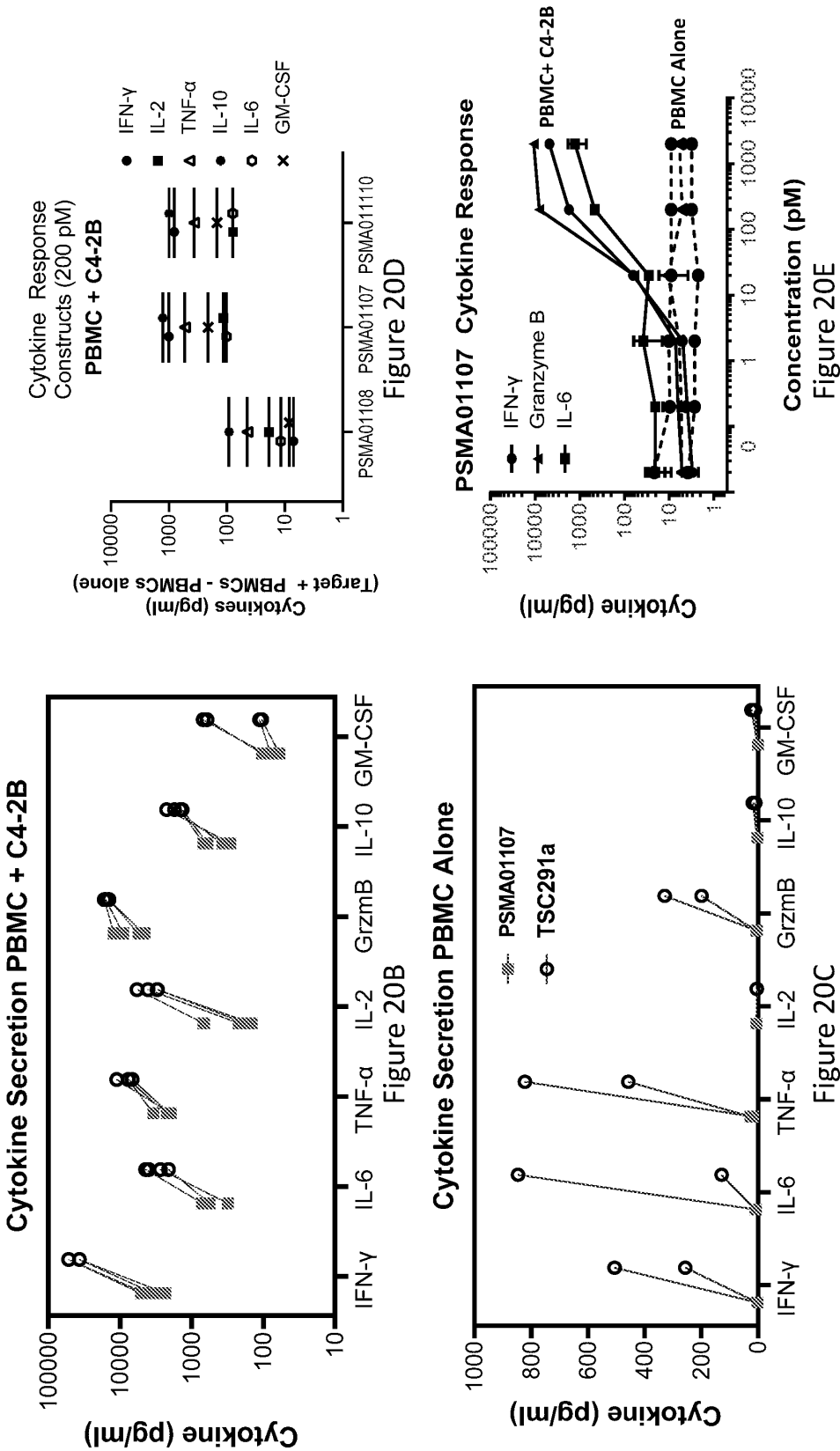
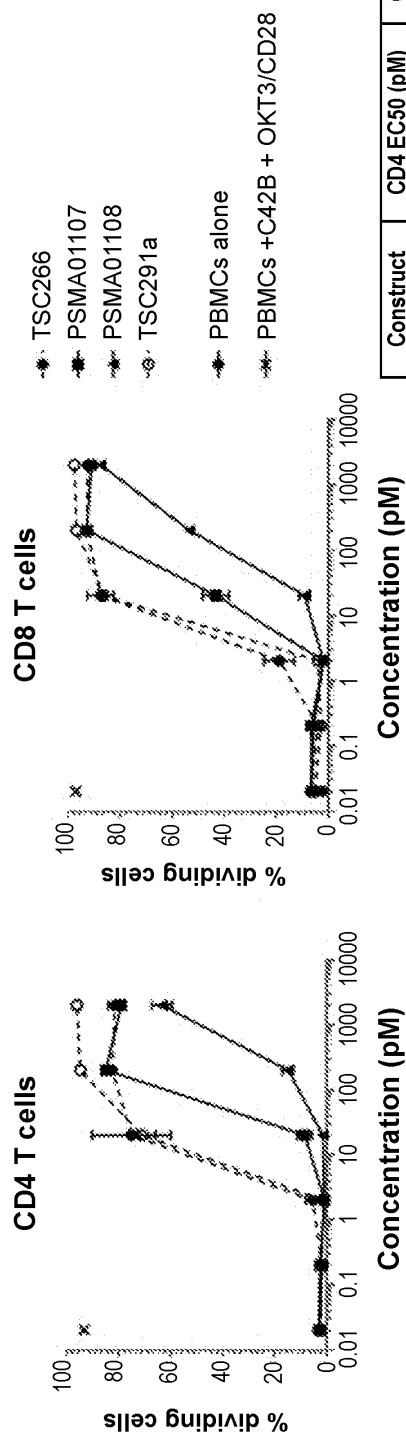


Figure 21: T-cell proliferation induced by monovalent and bivalent anti-PSMA x anti-CD3ε constructs at 24 hrs (A) with PSMA+ target cells or (B) without target cells



Construct	CD4 EC50 (pM)	CD8 EC50 (pM)
TSC266	8	5
PSMA01107	27	21
PSMA01108	265	164
TSC291a	17	14

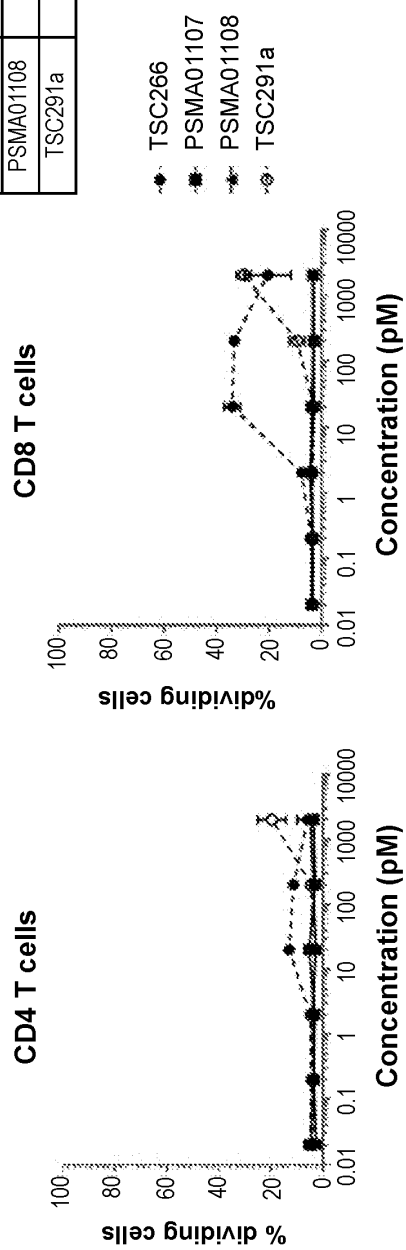


Figure 21B

Figure 22: (A) T-cell proliferation induced by monovalent and bivalent anti-PSMA x anti-CD3ε constructs at 96 hrs with PSMA+ target cells dose curve and (B) at a single concentration (200 pM)

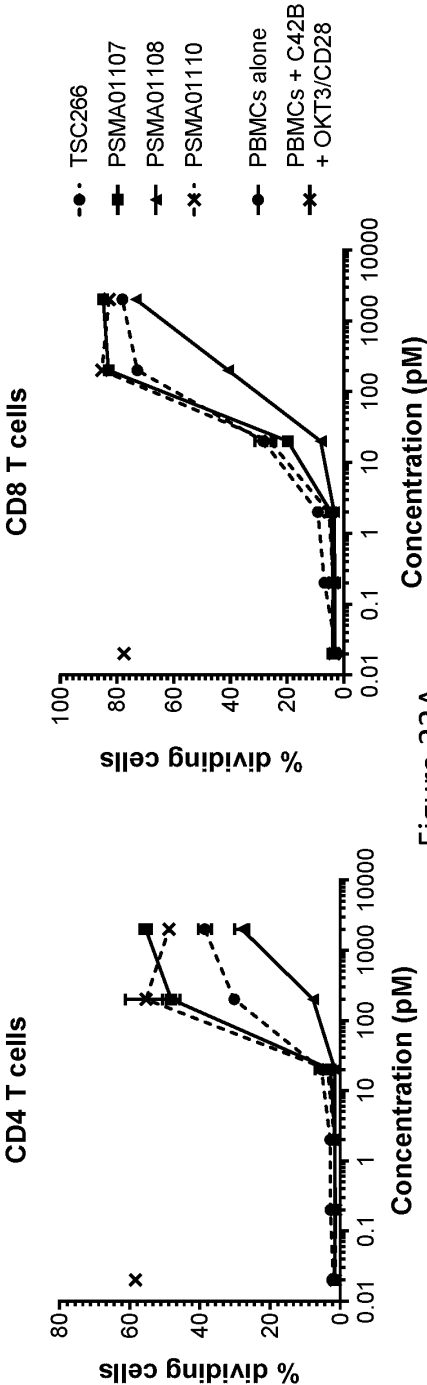


Figure 22A

Construct	CD4 EC50 (pM)	CD8 EC50 (pM)
TSC266	8	5
PSMA01107	27	21
PSMA01108	265	163
PSMA01110	17	14

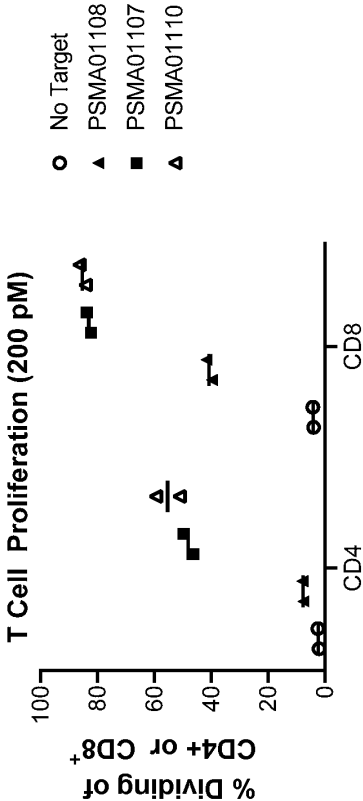


Figure 22B

Figure 23: Cytotoxicity of PSMA+ target cells induced by anti-PSMA x anti-CD3ε constructs at 72 and 96 hrs

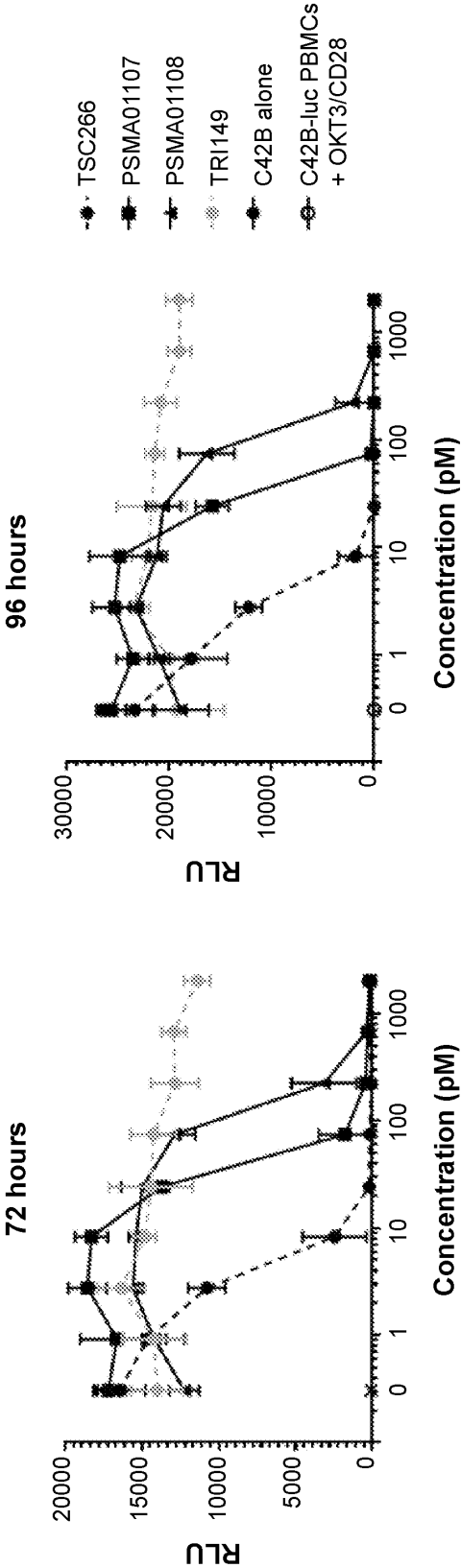
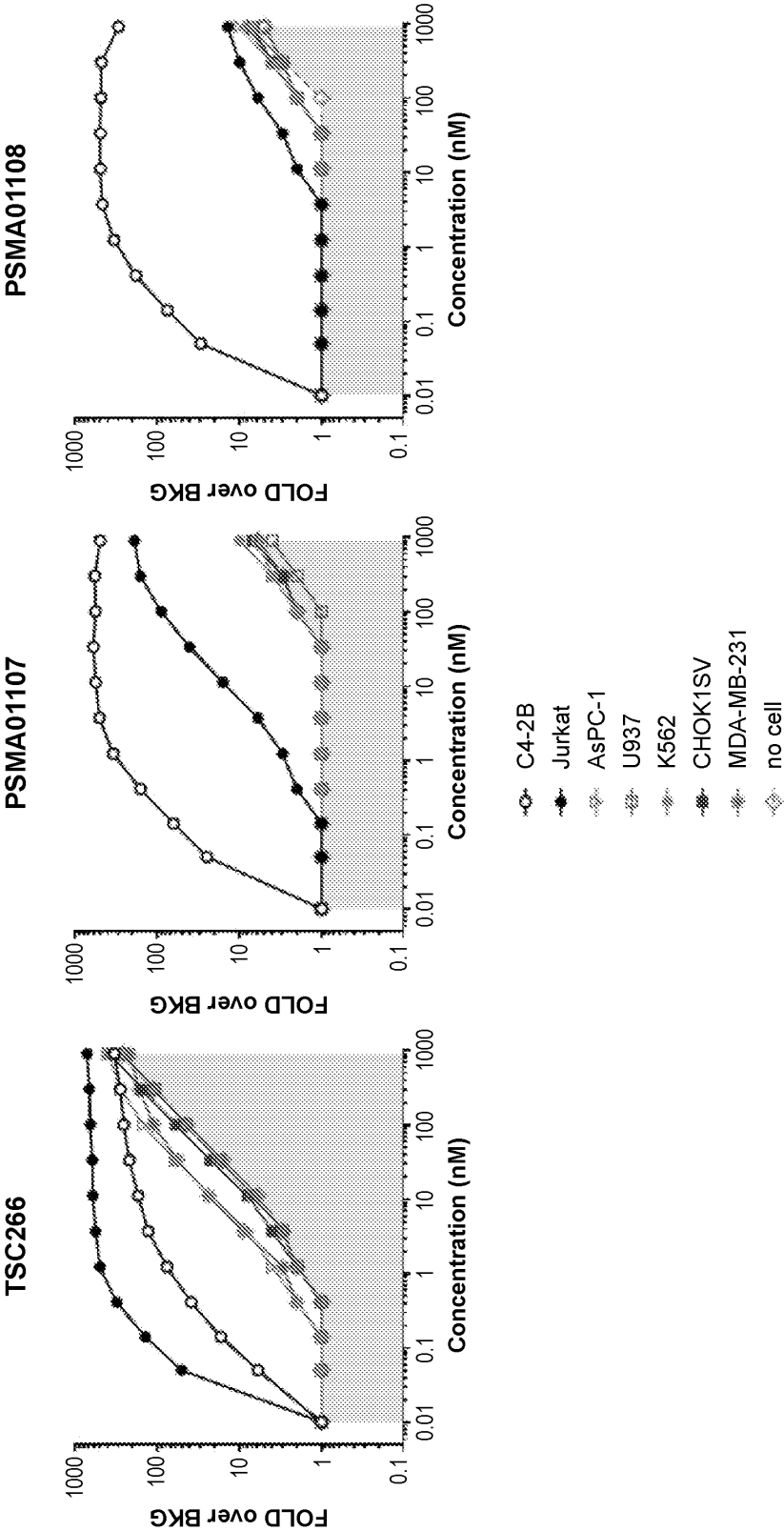


Figure 24: Non-specific binding analysis of anti-PSMA x anti-CD3ε constructs



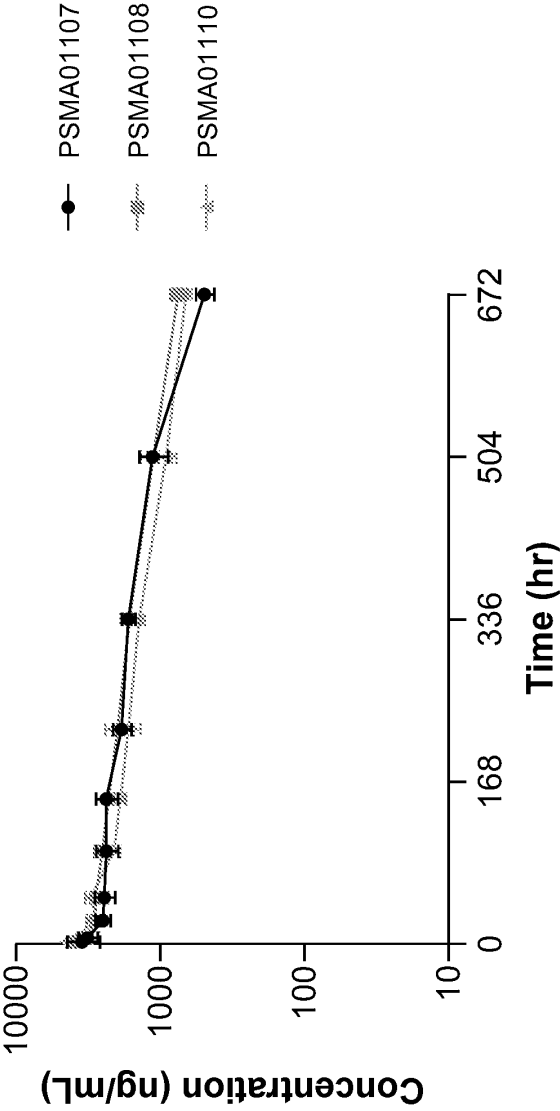
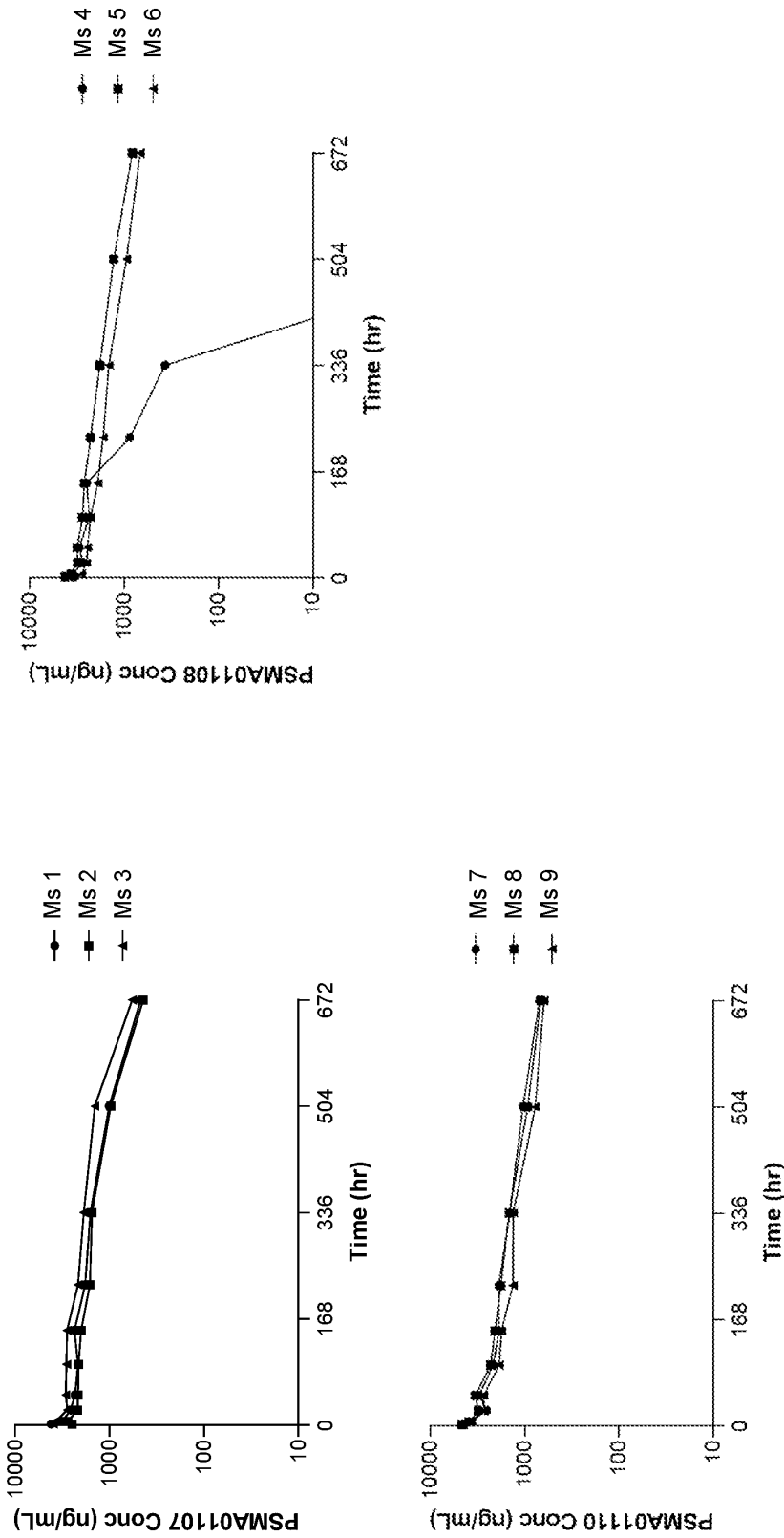


Figure 25

Figure 26



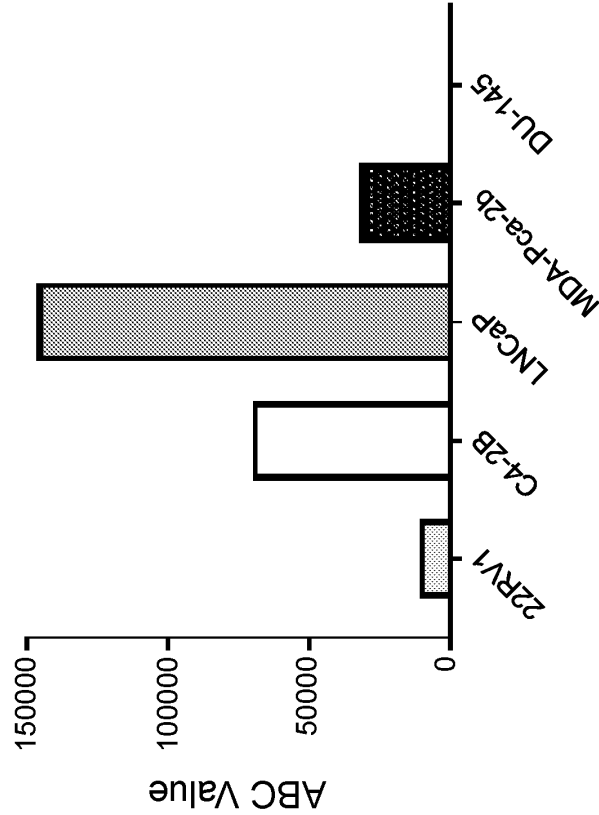


Figure 27

Figure 28

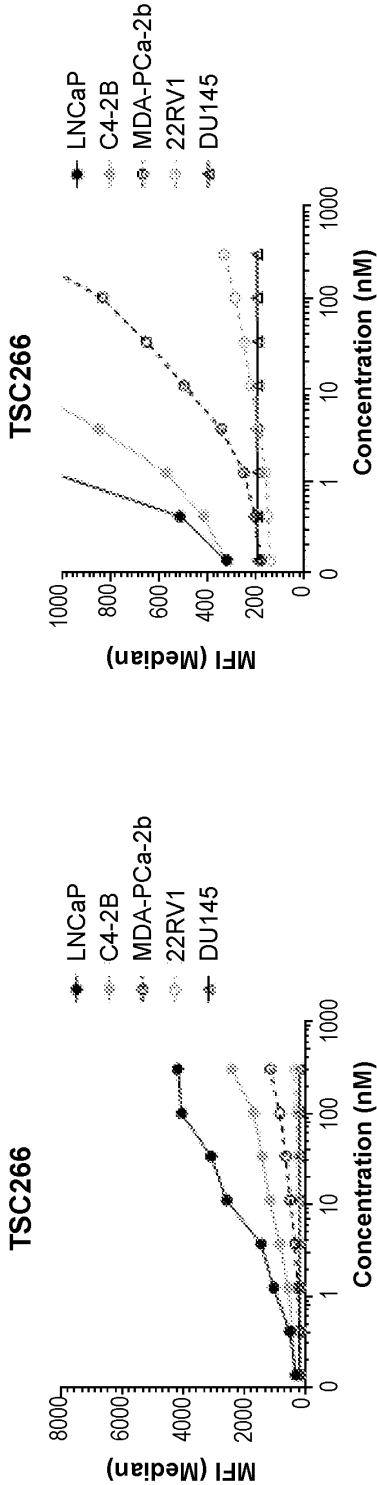


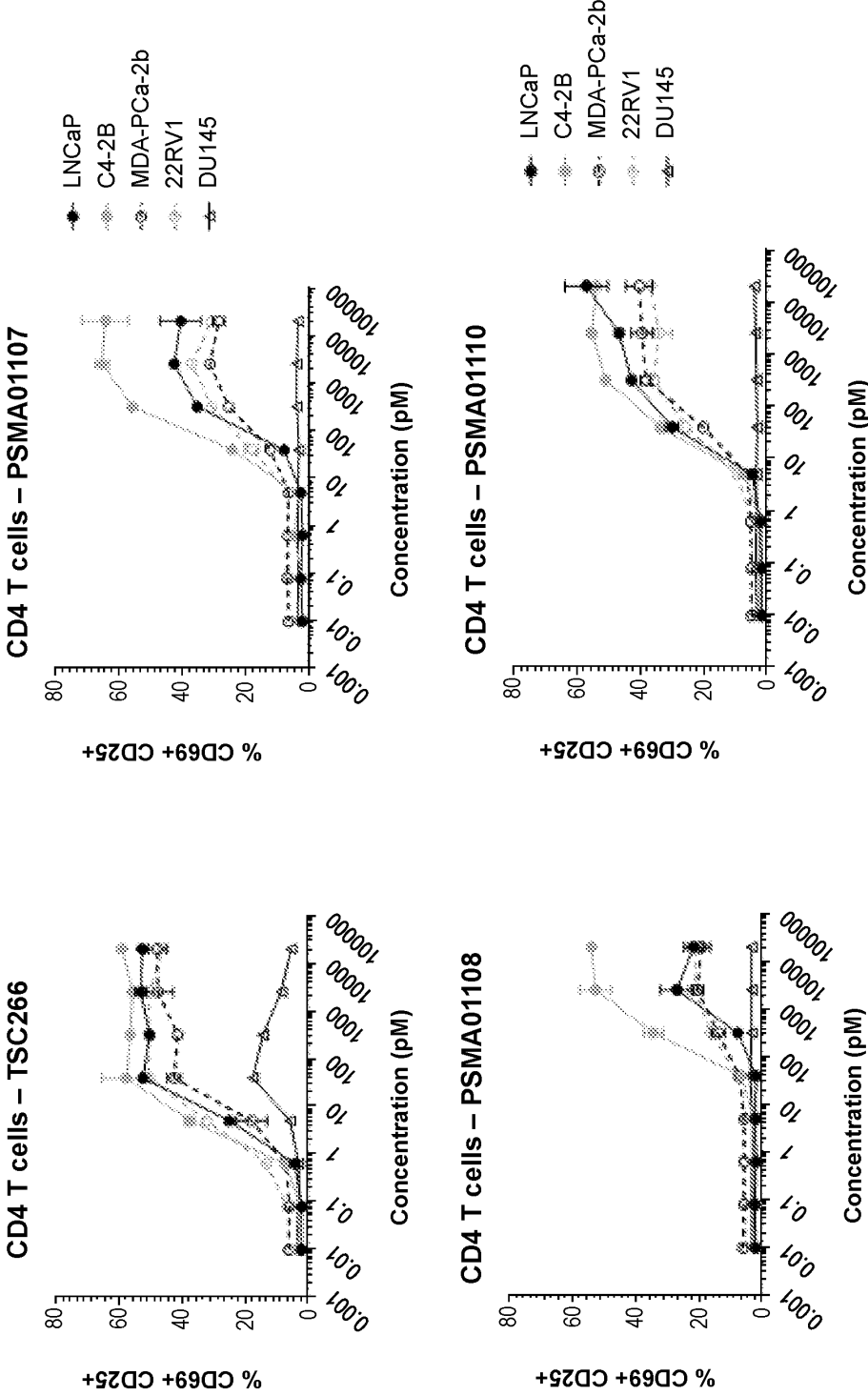
Figure 28A

Figure 28B

Figure 28C

Figure 28D

Figure 29



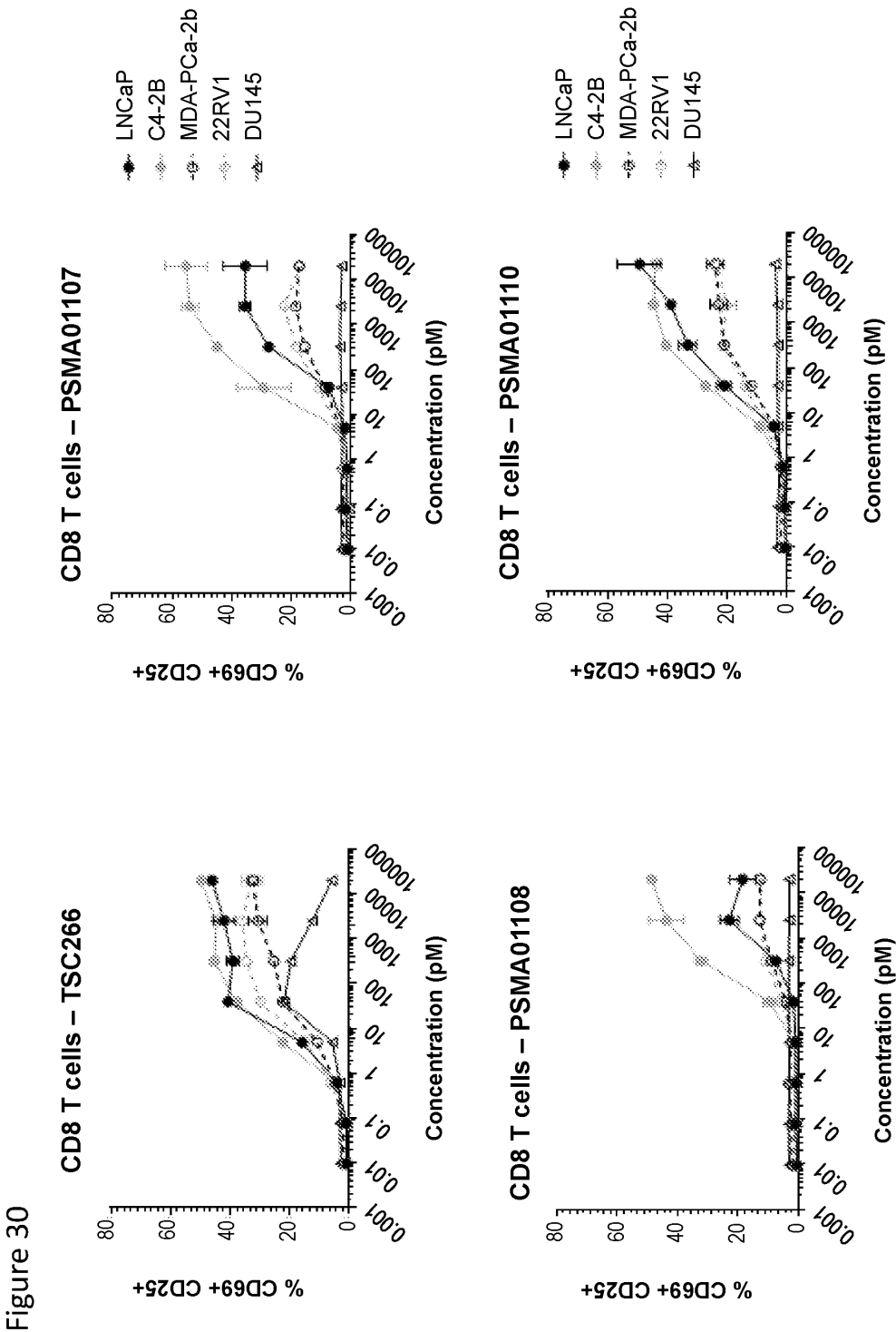


Figure 31: Cytotoxicity on tumor targets expressing a range of PSMA expression with anti-PSMA x anti-CD3ε constructs

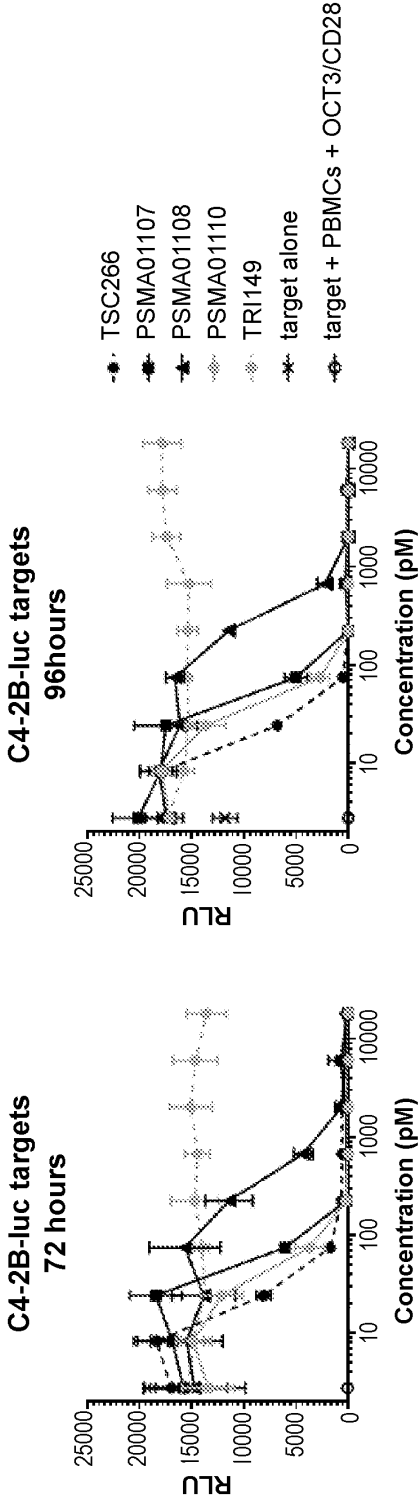


Figure 31A

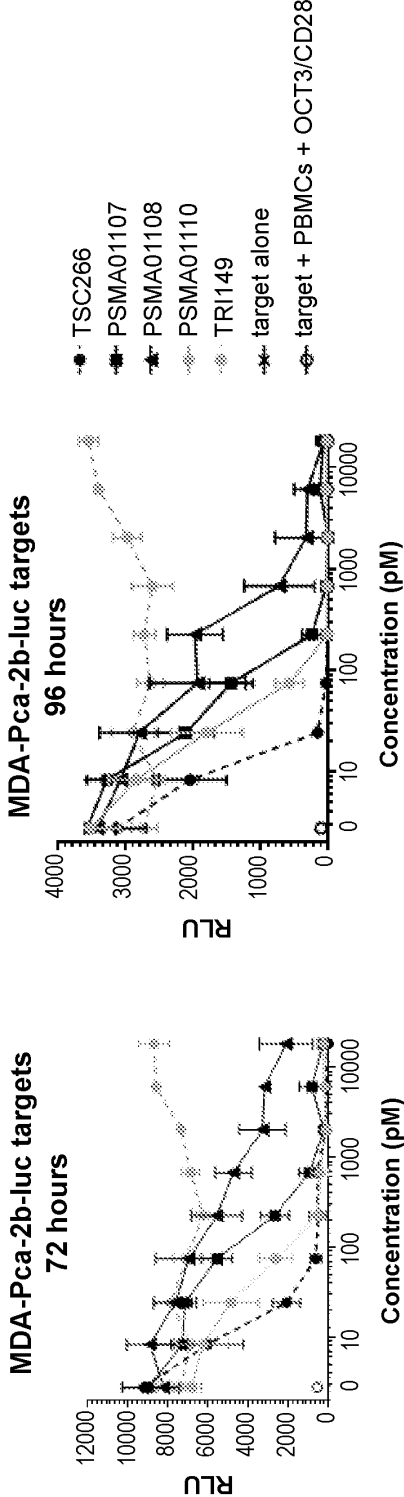


Figure 31B

Figure 32: Total PSMA receptor quantification and (A) and T cell mediated cytotoxicity versus binding of anti-PSMA x anti-CD3ε constructs to tumor cell lines expressing a range of PSMA (B)

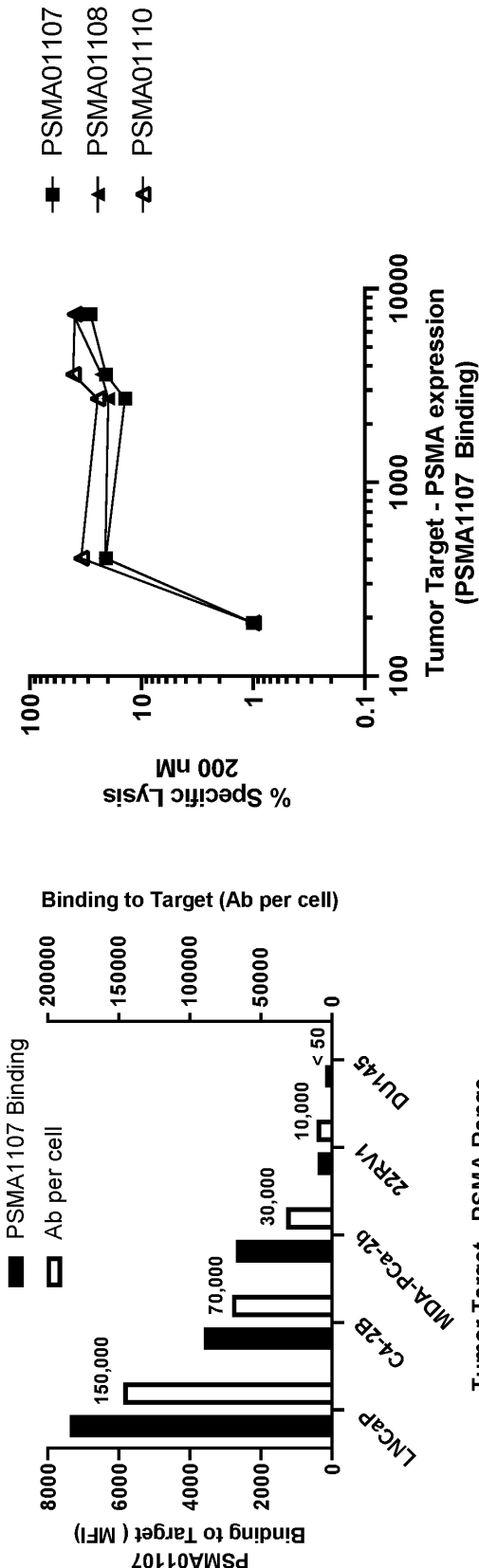


Figure 32B

Figure 32A

Figure 33

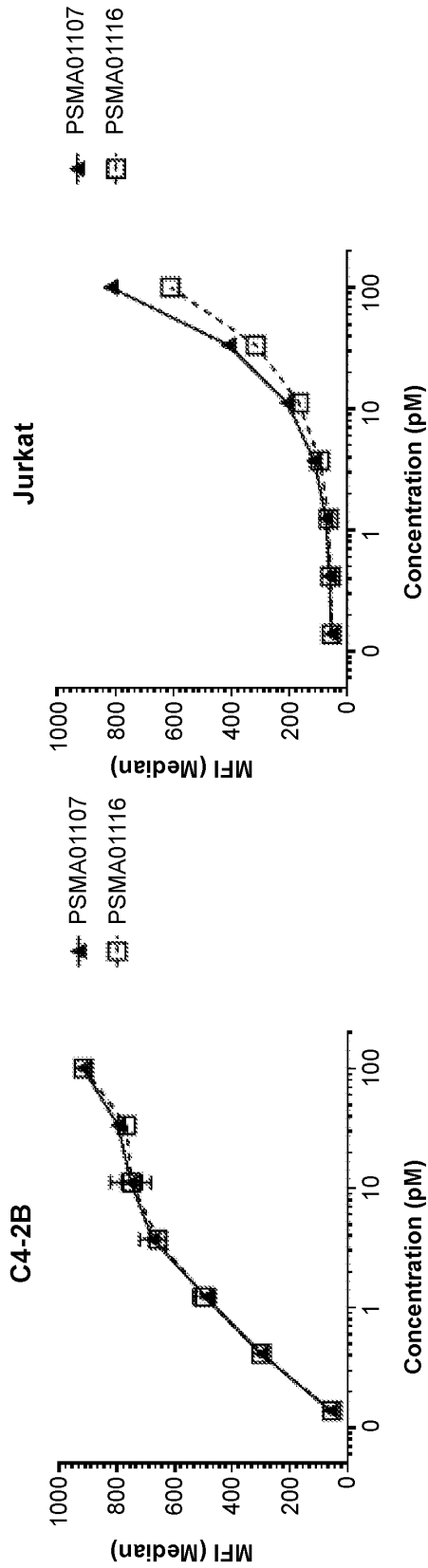


Figure 33A

Figure 33B

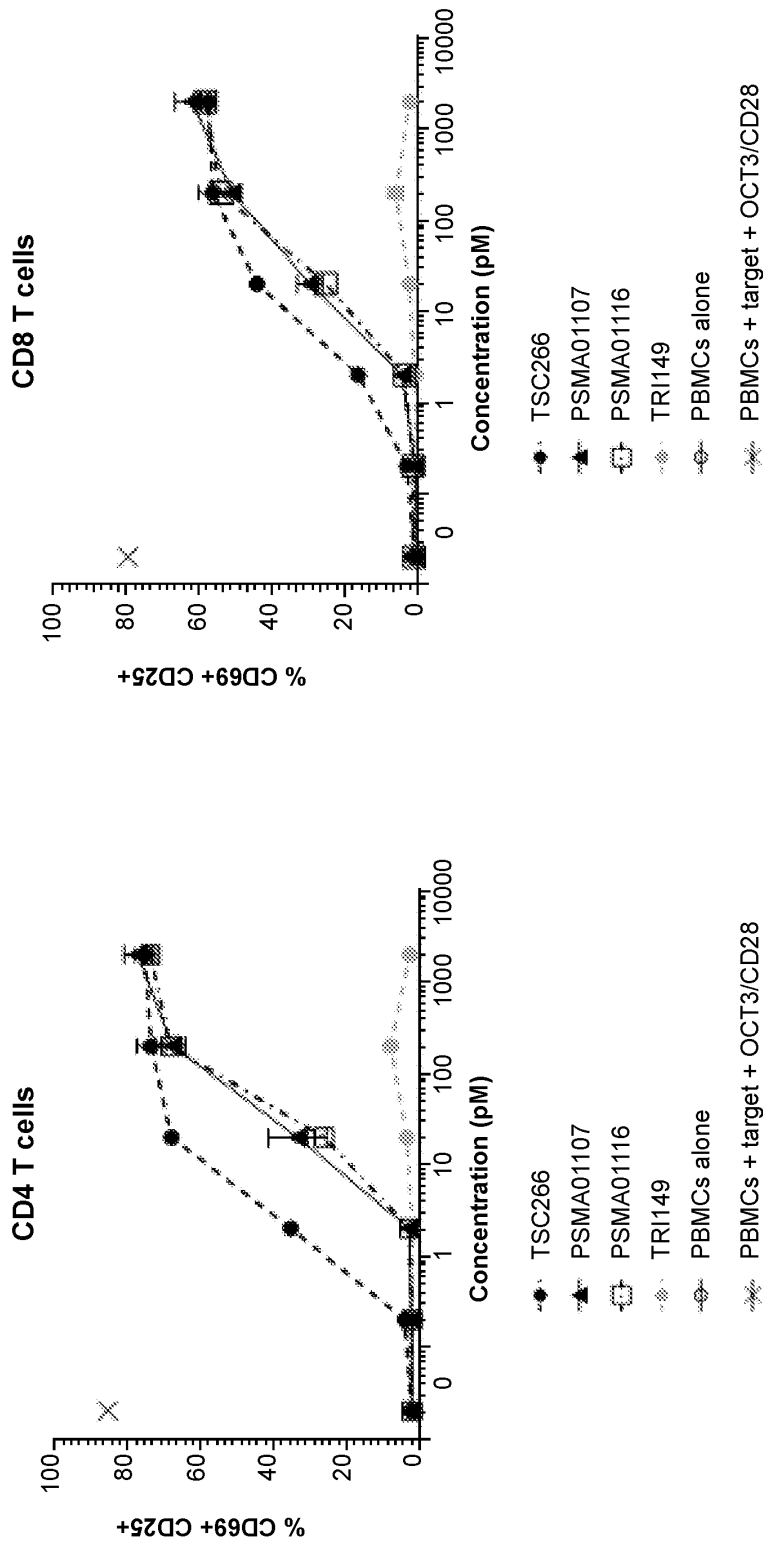


Figure 34

Figure 35

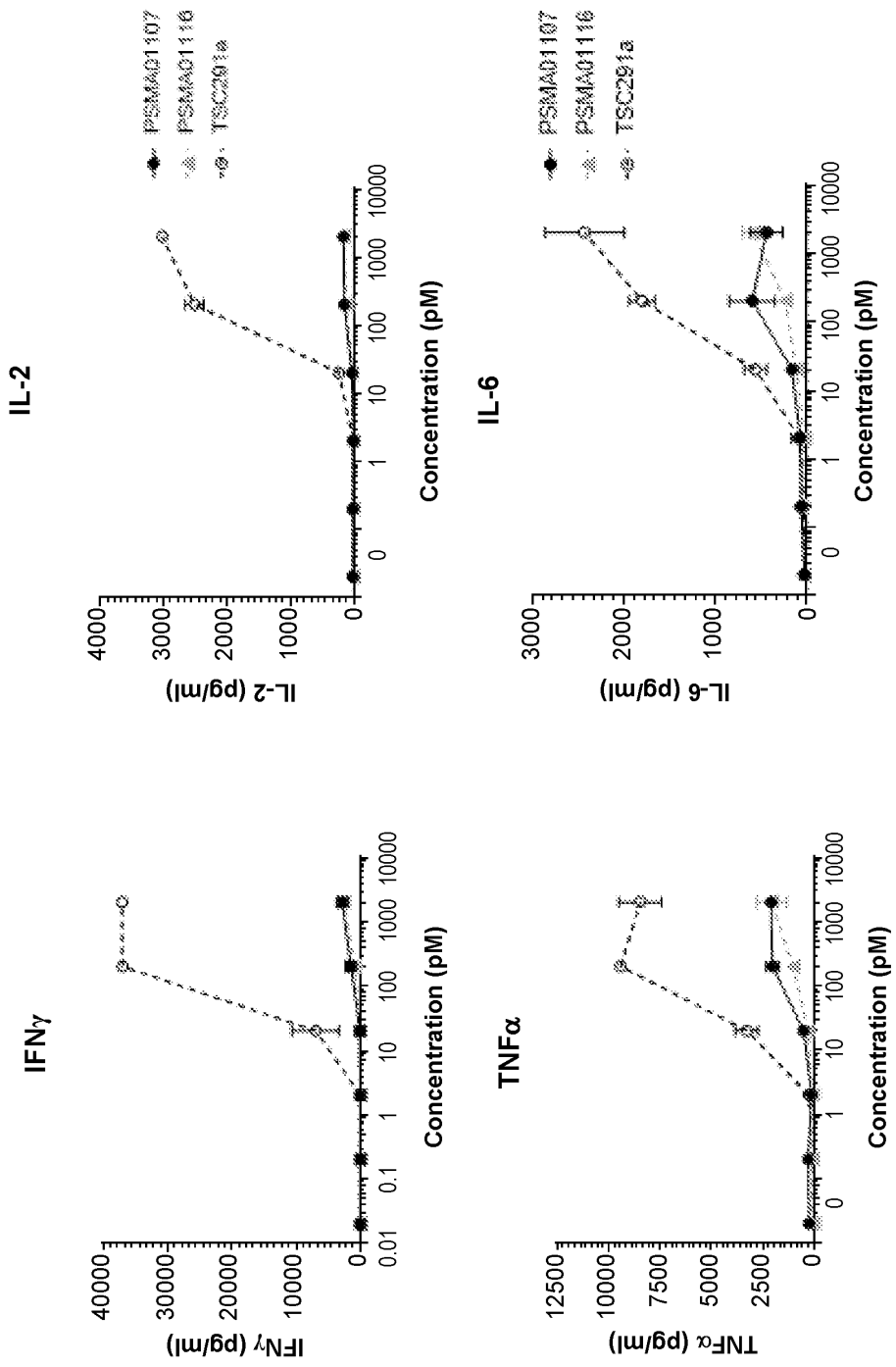


Figure 36

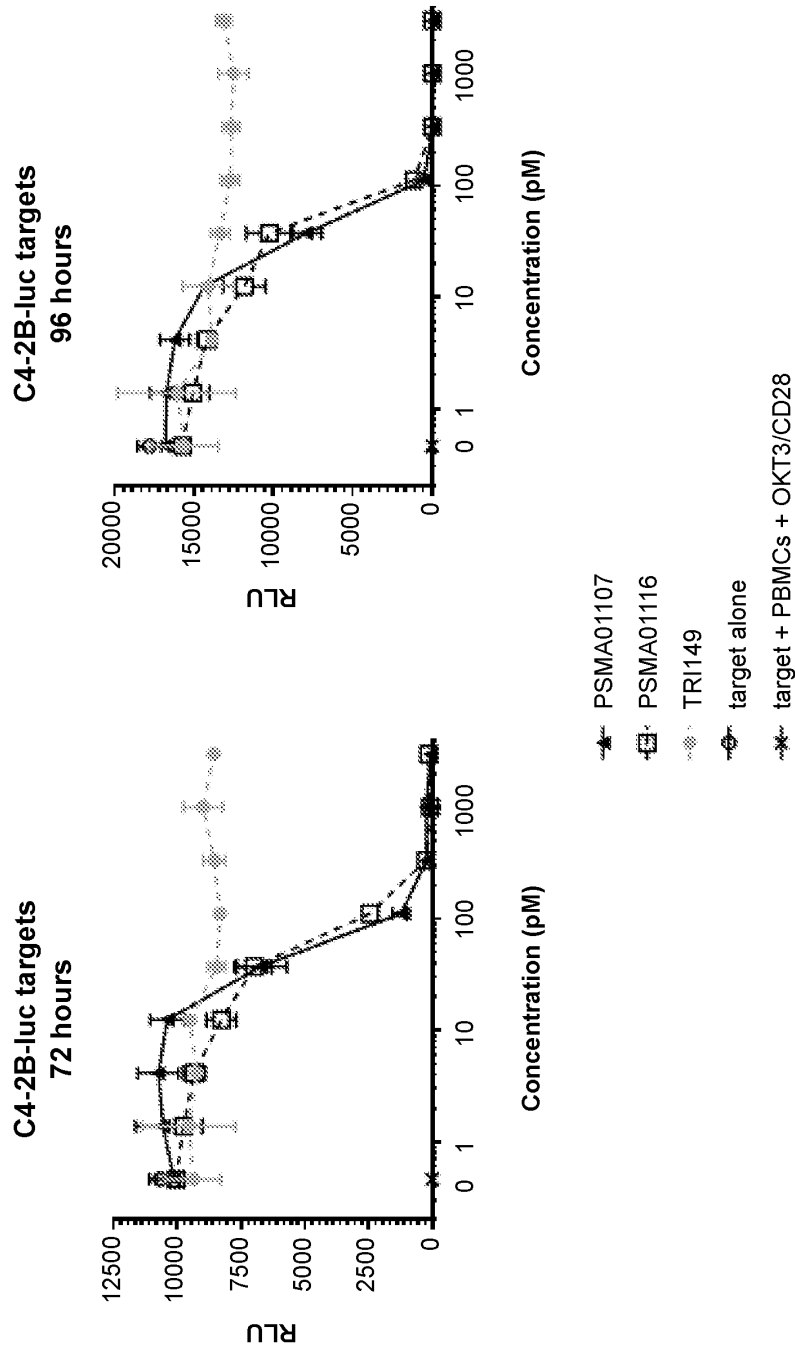


Figure 37

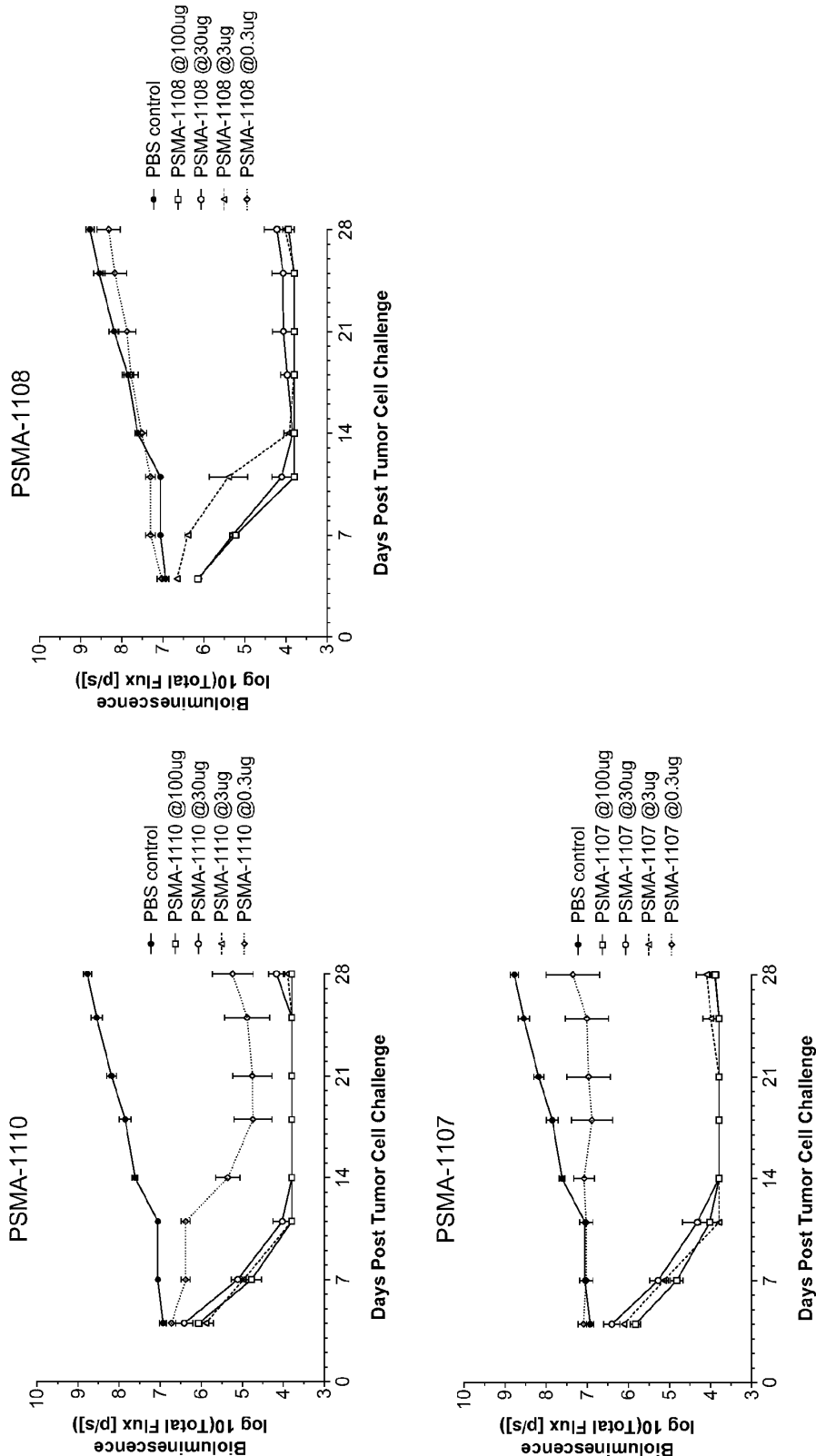


Figure 38

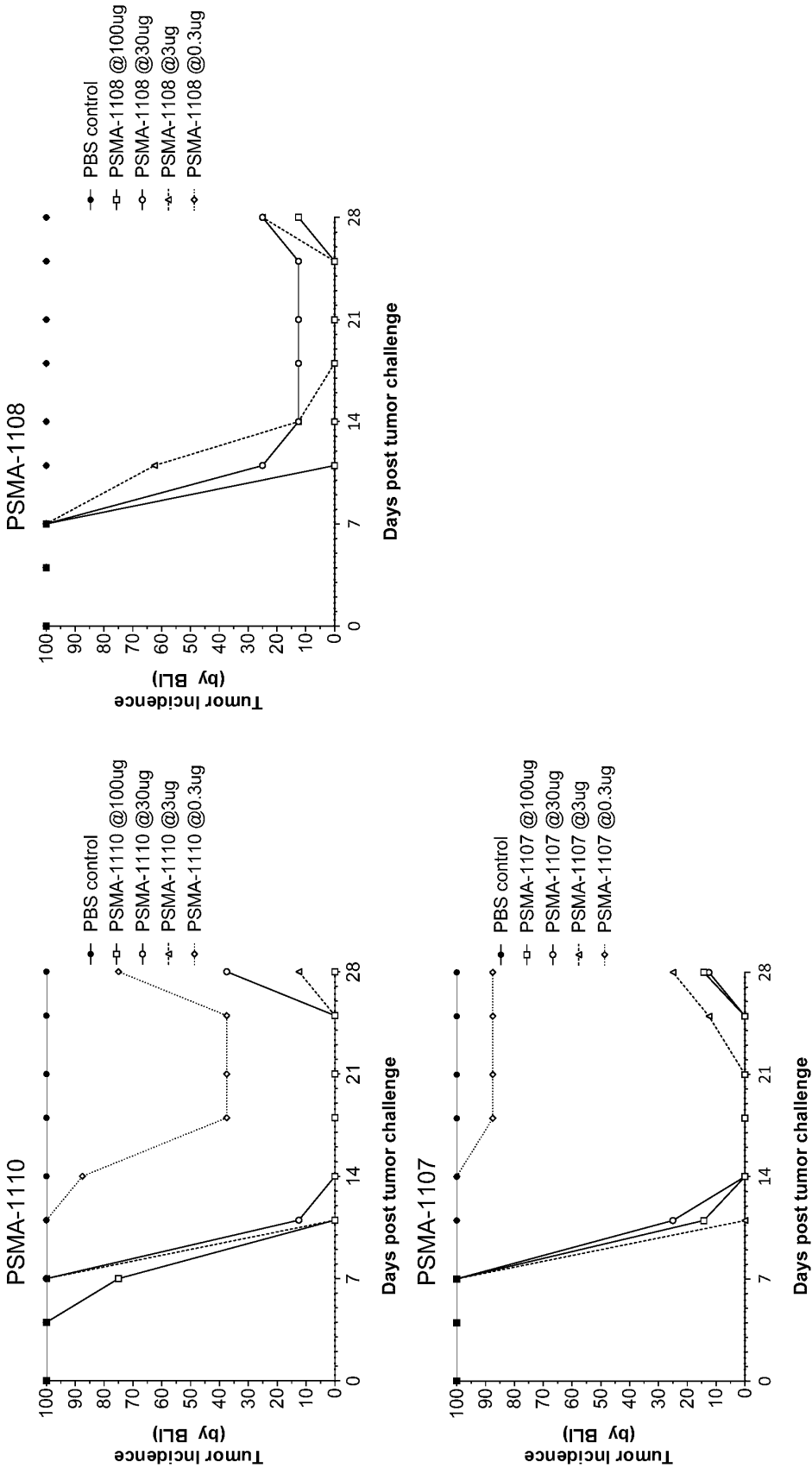


Figure 39A: Growth of C4-2B luciferase tumor cells in NOD/SCID mice following treatment with CD3 x PSMA constructs

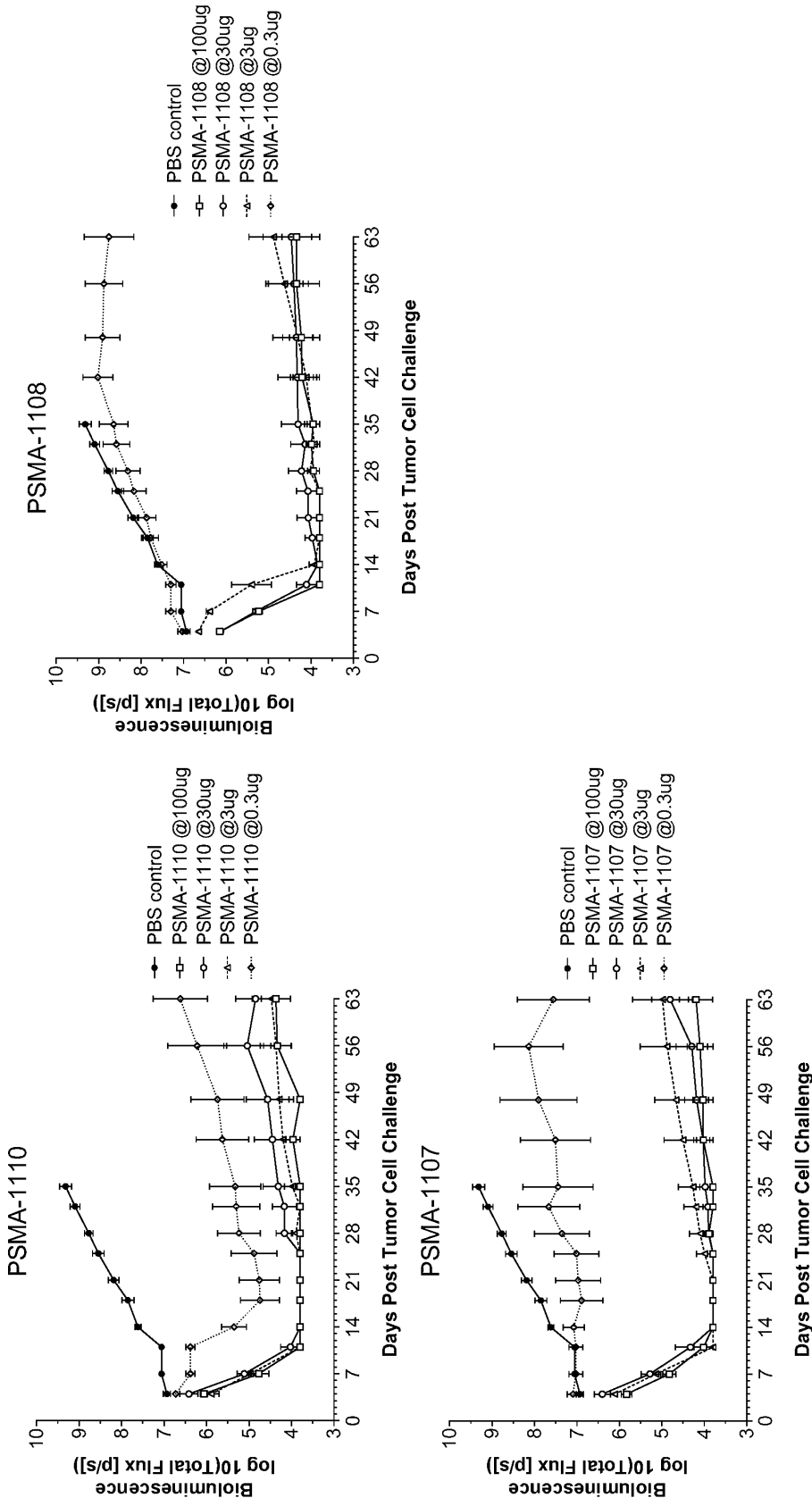


Figure 39B: Growth of C4-2B luciferase tumor cells in NOD/SCID mice following treatment with CD3 x PSMA constructs

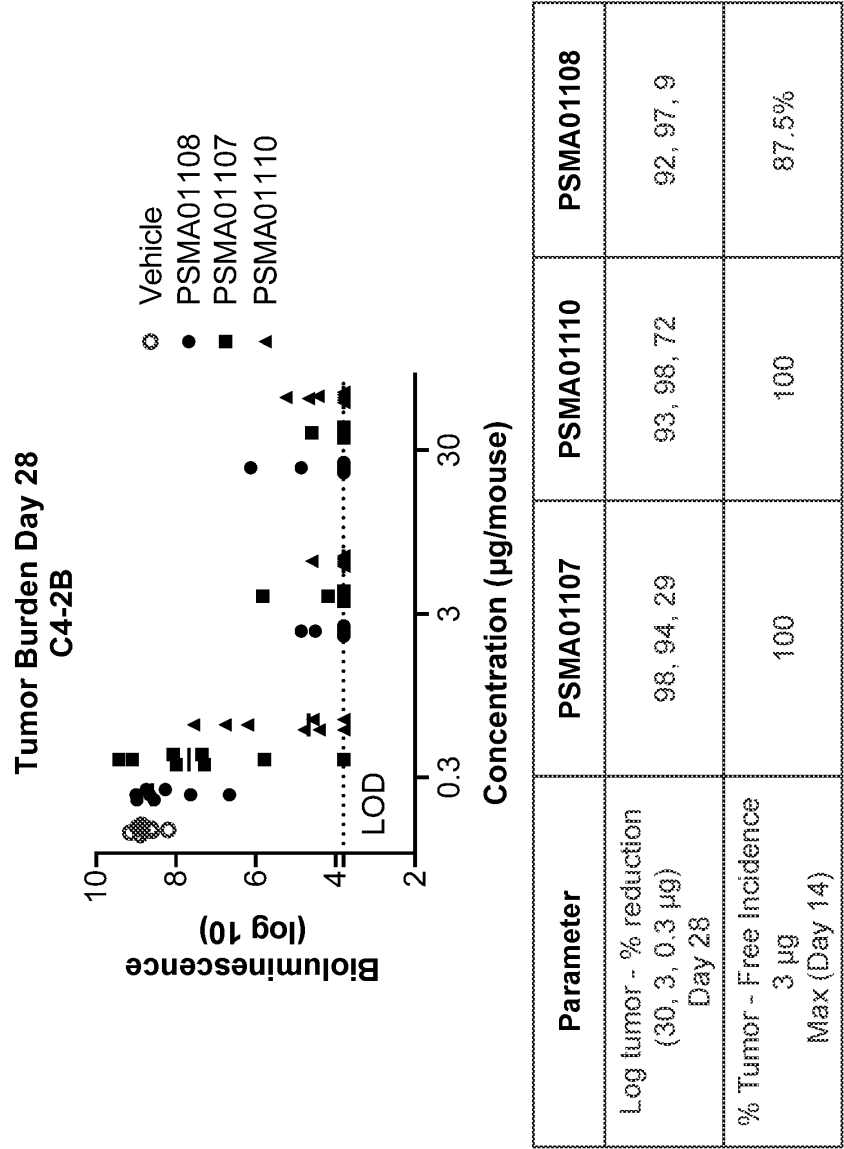


Figure 40: Visualization of tumor cells in NOD/SCID mice following treatment with CD3 x PSMA constructs

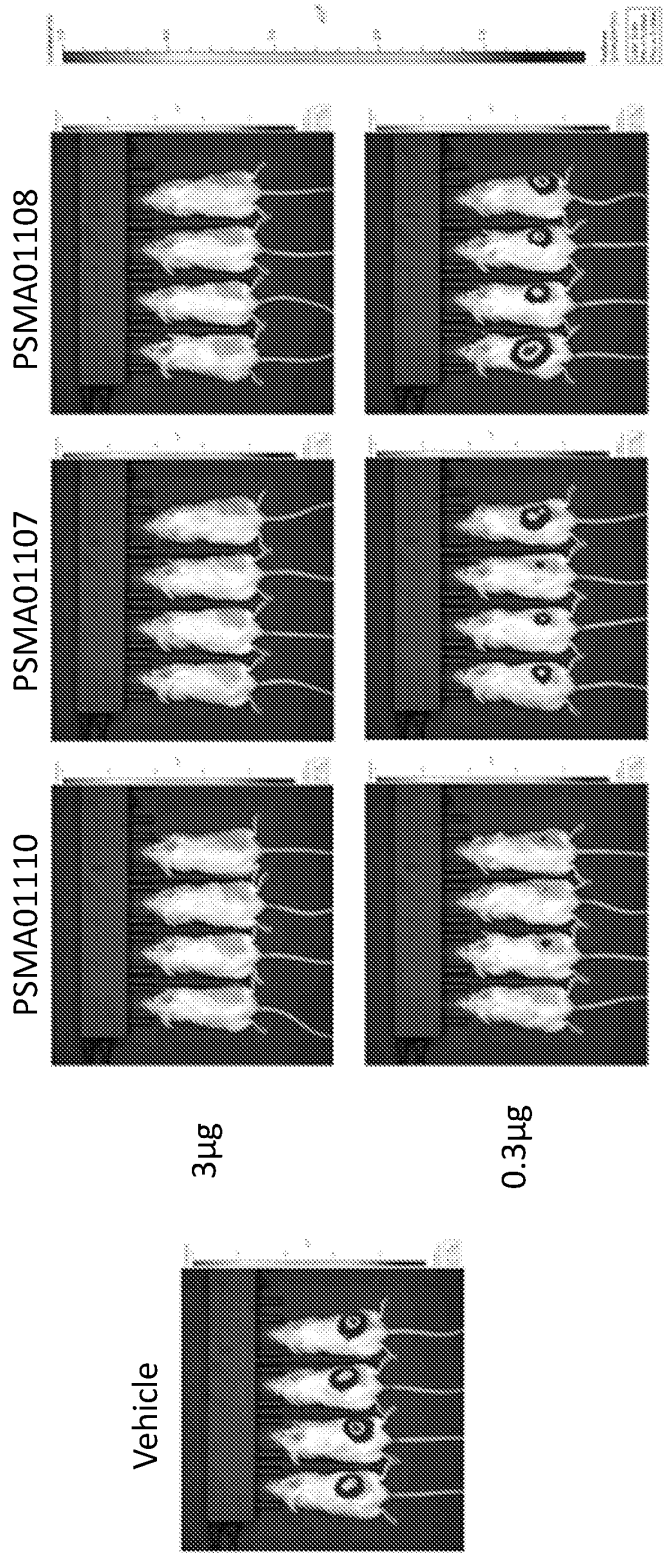


Figure 41

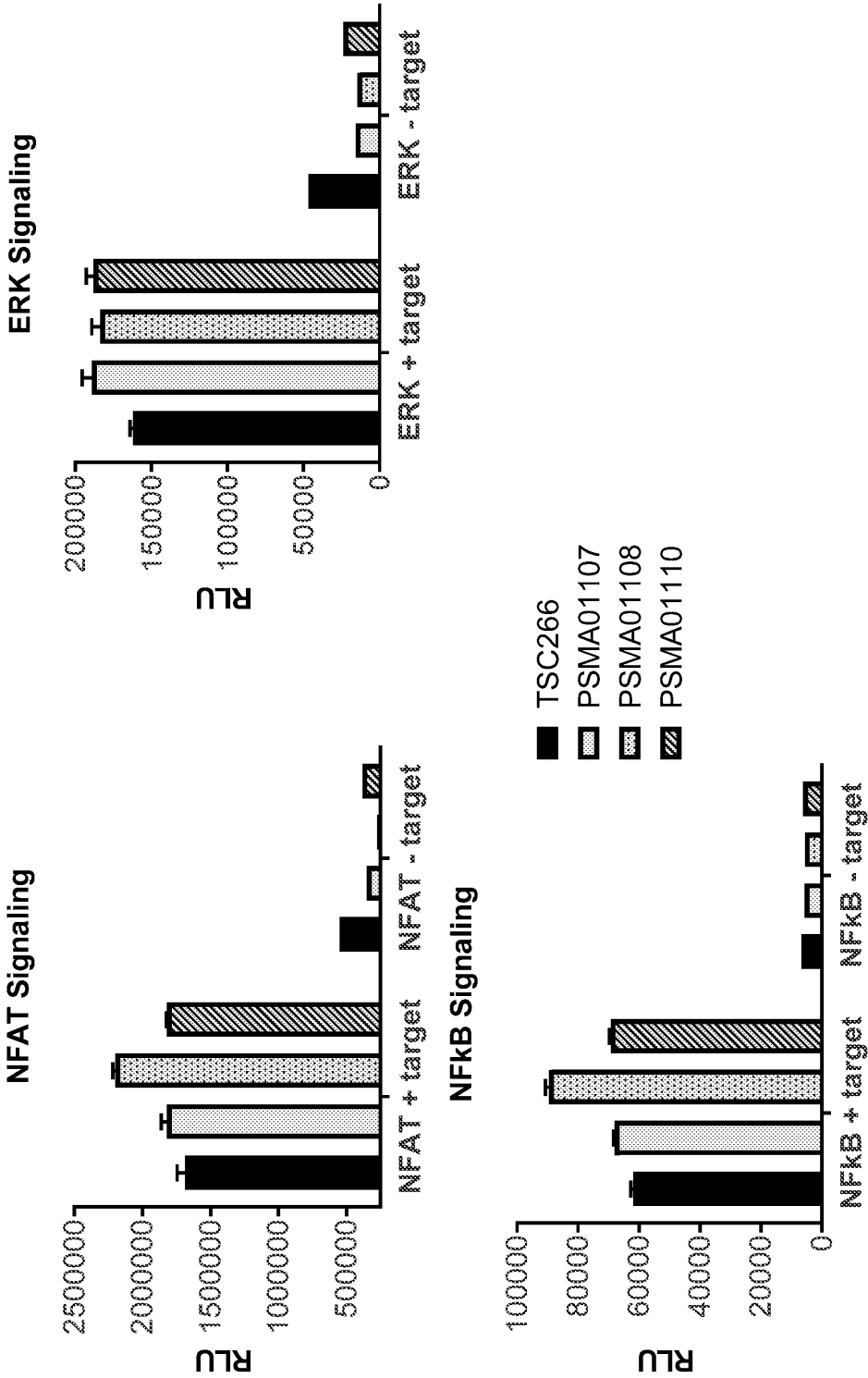


Figure 42A

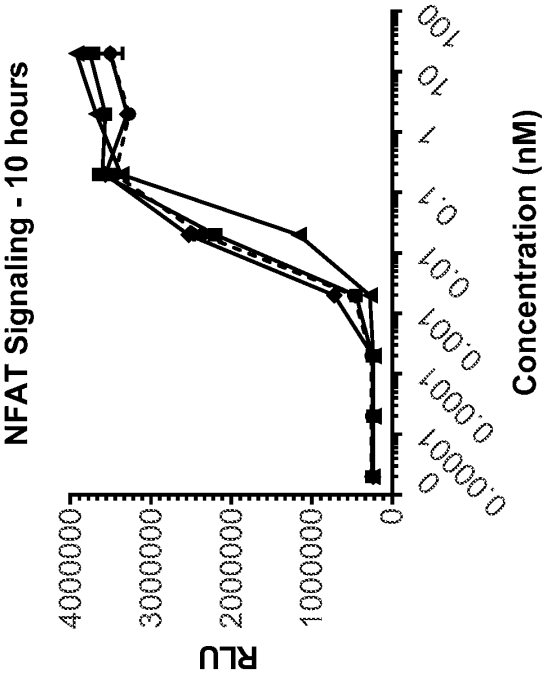


Figure 42B

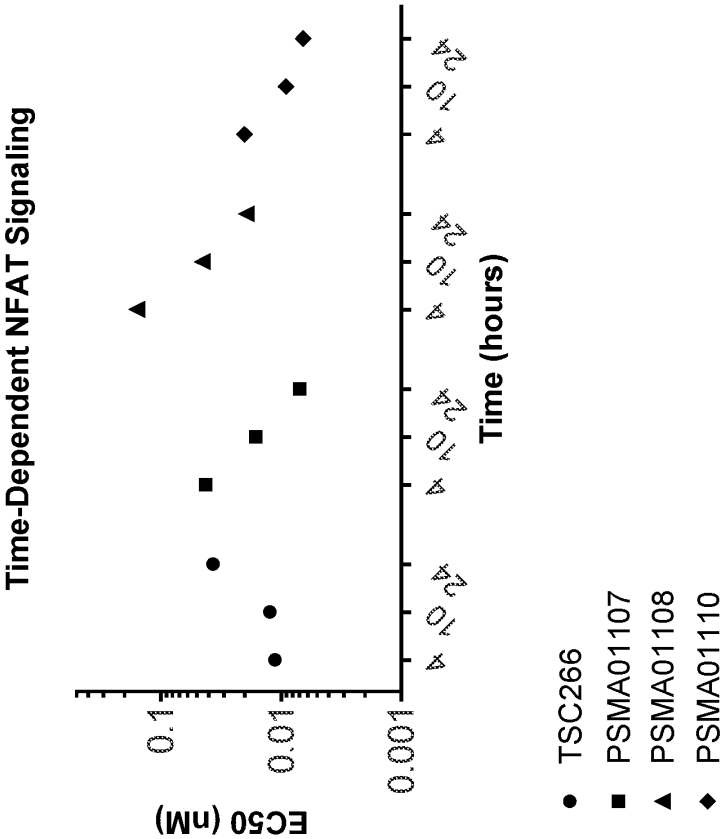


Figure 43

NFAT	4 hour EC50 (nM)	10 hour EC50 (nM)	24 hour EC50 (nM)
TSC266	0.011	0.012	0.037
PSMA01107	0.043	0.016	0.007
PSMA01108	0.159	0.045	0.019
PSMA01110	0.020	0.009	0.007
ERK	4 hour EC50 (nM)	10 hour EC50 (nM)	24 hour EC50 (nM)
TSC266	0.0012	0.0010	0.0193
PSMA01107	0.0107	0.0023	0.0078
PSMA01108	0.0241	0.0026	0.0077
PSMA01110	0.0039	0.0030	0.0320
NFkB	4 hour EC50 (nM)	10 hour EC50 (nM)	24 hour EC50 (nM)
TSC266	0.023	0.042	0.066
PSMA01107	0.162	0.077	0.065
PSMA01108	0.585	0.181	0.097
PSMA01110	0.070	0.051	0.045

Figure 44A

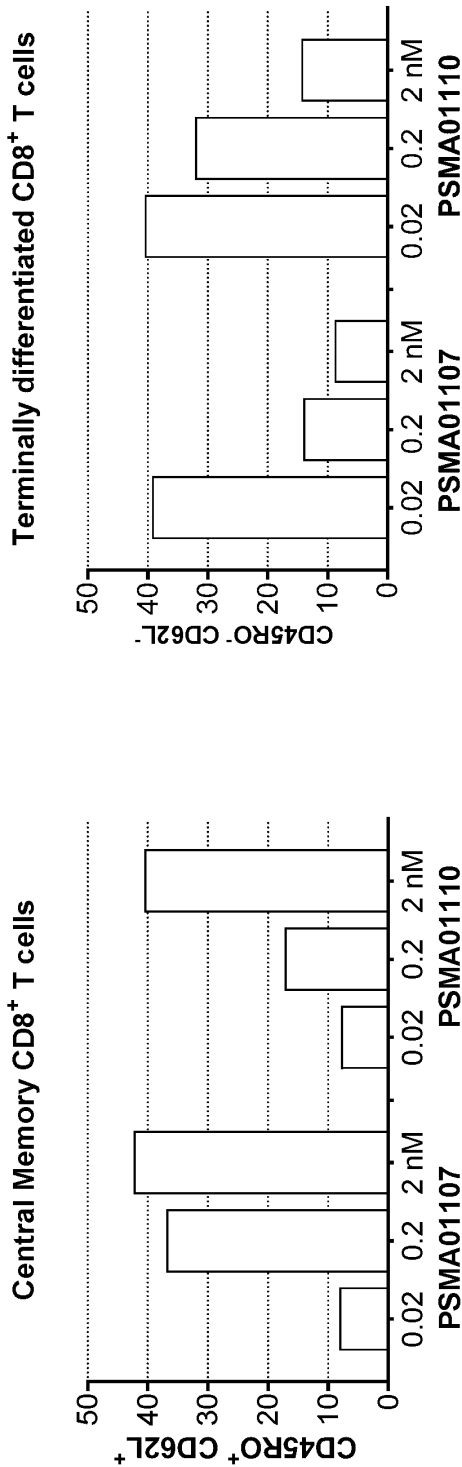


Figure 44B

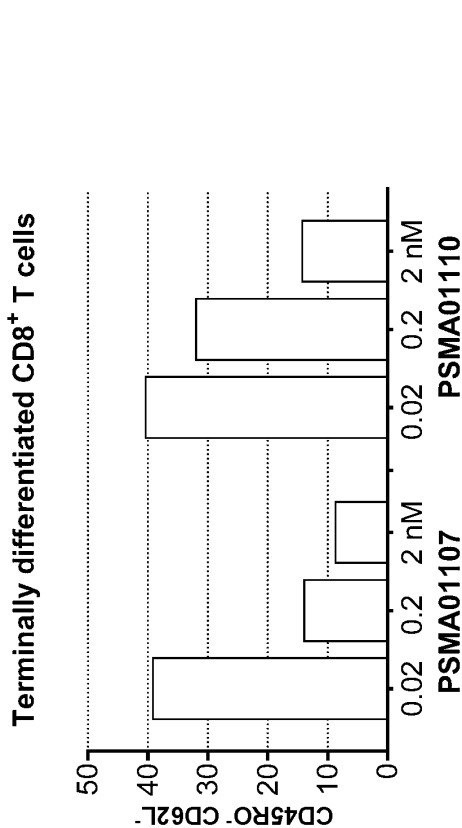


Figure 44C

