Blockade of vascular endothelial growth factor receptors by tivozanib has potential anti-tumour effects on human glioblastoma cells

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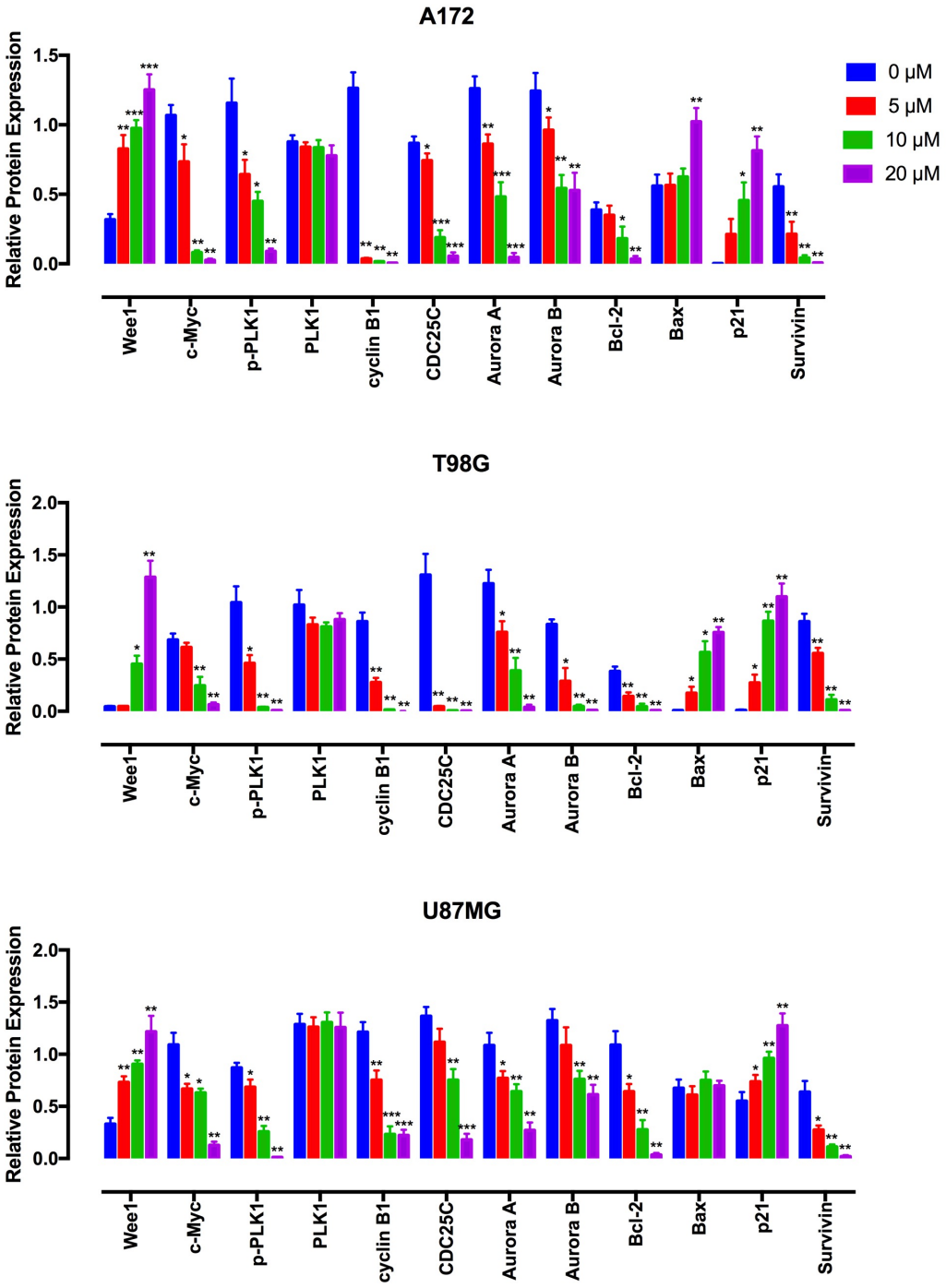
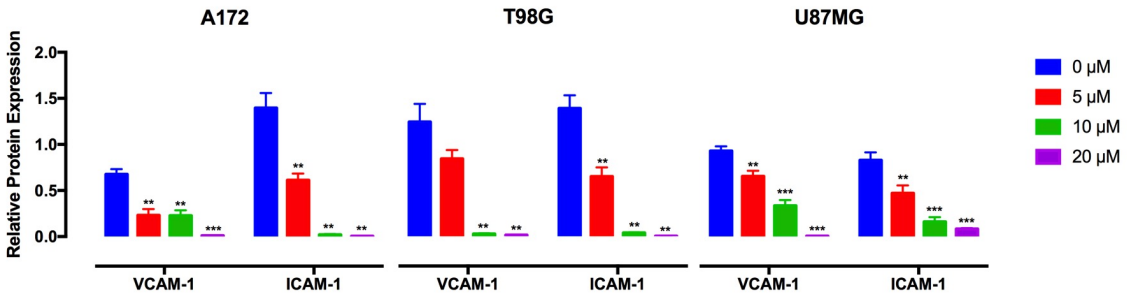
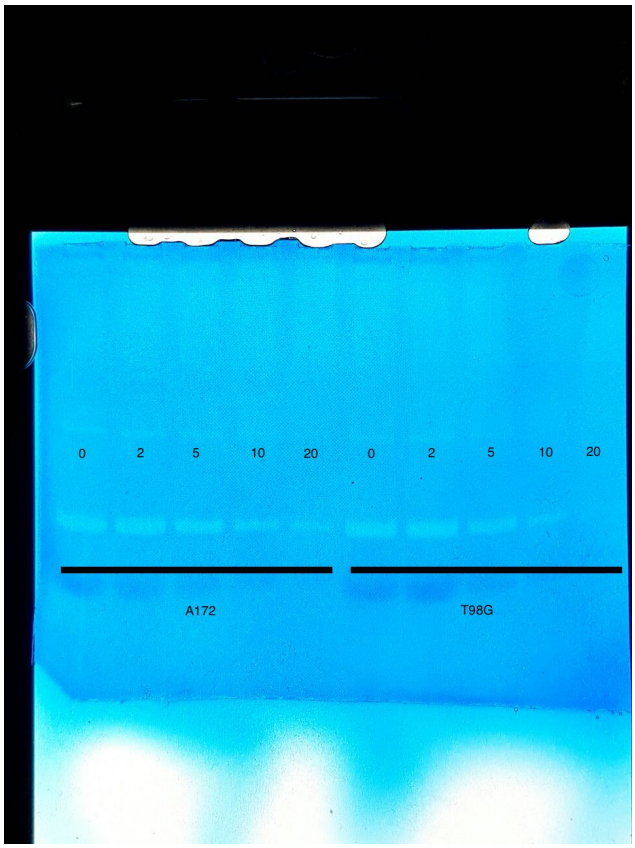
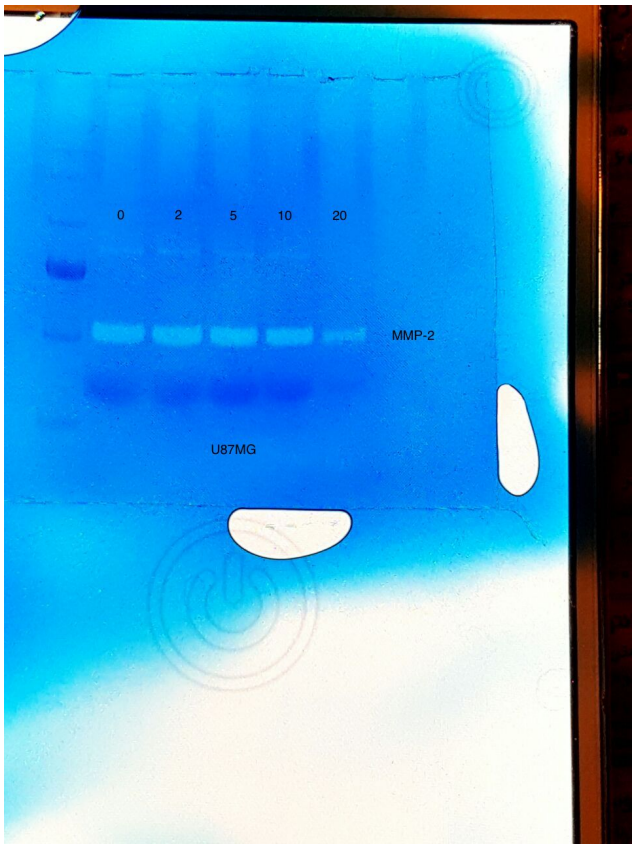


Supplementary Fig. 1



Supplementary Fig. 2





Supplementary Fig. 3

Supplementary Table 1

Concentrations		fa	CI	DRI	
Tivozanib (μM)	Irinotecan (μg/mL)			Tivozanib	Irinotecan
		T98G			
10	0.1	0.5	0.73	1.37	645.91
10	0.5	0.5	0.72	1.41	134.43
10	1	0.52	0.69	1.48	72.5
10	2.5	0.5	0.69	1.51	30.1
10	5	0.53	0.69	1.59	16.22
10	10	0.54	0.72	1.65	8.65
10	25	0.61	0.65	2.17	5.34
		U87MG			
10	0.1	0.31	1	1	97.77
10	0.5	0.34	0.95	1.1	23.41
10	1	0.37	0.89	1.22	14.18
10	2.5	0.39	0.94	1.29	6.24
10	5	0.45	0.87	1.55	4.4
10	10	0.56	0.71	2.14	4
10	25	0.65	0.73	2.79	2.66

Supplementary Table 2

Concentrations (μM)		fa	CI	DRI	
Tivozanib	Temozolomide			Tivozanib	Temozolomide
		T98G			
10	50	0.42	0.97	1.1	16.98
10	100	0.44	0.92	1.24	8.92
10	200	0.47	0.88	1.5	4.82
10	500	0.51	1	1.88	2.12
10	1000	0.66	0.85	4.85	1.55
10	2500	0.72	1.47	7.61	0.75
		U87MG			
10	50	0.37	0.69	1.7	9.94
10	100	0.39	0.73	1.83	5.34
10	200	0.4	0.87	1.94	2.83
10	500	0.44	1.1	2.45	1.43
10	1000	0.58	0.89	4.96	1.45
10	2500	0.73	0.79	12.11	1.42

Supplementary legends

Supplementary Fig. 1: Quantification of protein expression. Quantification of protein band intensities was done using the ImageJ software after normalising to the corresponding β-actin levels. Data are given as mean ± SD. Statistically significant values of $*p < 0.05$, $**p < 0.01$, and $***p < 0.001$ were determined compared with the control.

Supplementary Fig. 2: Quantification of VCAM-1 and ICAM-1 protein expression. Relative protein expression was obtained by normalisation against β-actin. Data are given as mean ± SD. Statistically significant values of $**p < 0.01$ and $***p < 0.001$ were determined compared with the control.

Supplementary Fig. 3: The effects of tivozanib on MMP-2 enzymatic levels. Gelatinolytic activities are visualized as clear bands against the blue background of stained gelatin. The zymograms are representative of three independent experiments with similar results.

Supplementary Table 1: Combination index (CI) and dose reduction index (DRI) of tivozanib and irinotecan combination in T98G and U87MG cells. DRI represents the order of magnitude of dose reduction that is allowed in combination for a given degree of effect as compared with the dose of each drug alone. ‘‘fa’’ denotes fraction affected.

Supplementary Table 2: CI and DRI values of tivozanib and temozolomide combination in T98G and U87MG cells.