Supporting Information

High throughput drug screening platform utilizing capillary and artery cell layered models based on tumor-vascular cell interactions

Jihyeon Song[1†], Yeji Lee[1†], Min-Seok Kim[2], Giheon Ha[1], WonJun Jang[2,3], Ulziituya Batjargal[2,3], Young Gyun Kim[4], Han-Jun Kim[2,3]*, Junmin Lee[1]*

[1]Department of Materials Science and Engineering, Pohang University of Science and Technology (POSTECH), Pohang 790-784, Republic of Korea

[2]College of Pharmacy, Korea University, Sejong, 30019, Republic of Korea

[3]Interdisciplinary Major Program in Innovative Pharmaceutical Sciences, Korea University, Sejong 30019, Republic of Korea

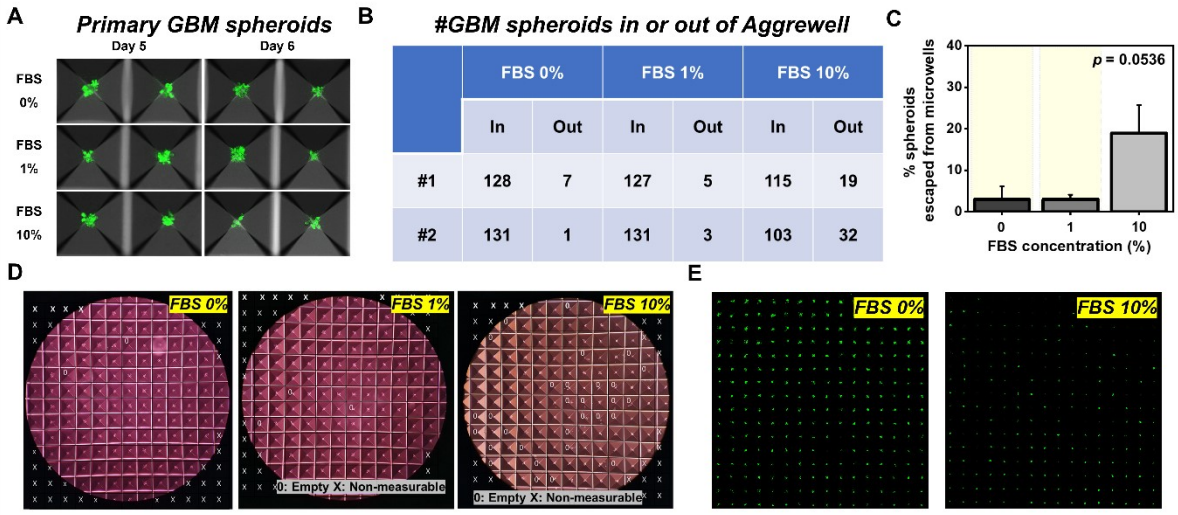
[4]Department of Bioengineering, University of Southern California, Los Angeles, CA 90089, USA

[†]These authors equally contributed to this work.

***Authors to whom any correspondence should be addressed.**

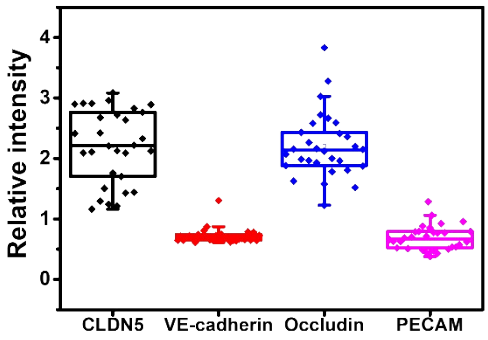
E-mail: hanjun@korea.ac.kr, junmin@postech.ac.kr

Keywords: High-throughput screening platform; Tumor-vascular cell interaction; Tumor-capillary model; Tumor-artery model; Personalized medicine



	FBS 0%		FBS 1%		FBS 10%	
	In	Out	In	Out	In	Out
#1	128	7	127	5	115	19
#2	131	1	131	3	103	32

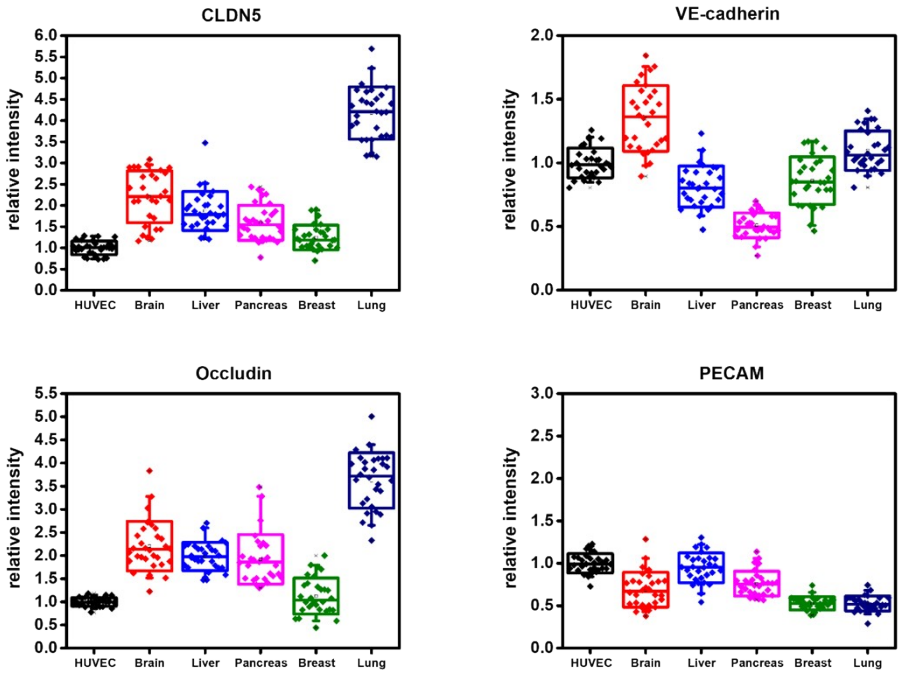
SI. Figure 1. Fabrication of GBM-only spheroids. (**A**) Representative confocal 2D image of GBM spheroids by FBS concentration. (**B**) Number of spheroids in aggrewell according to FBS concentration. (**C**) Spheroid escape rate according to FBS concentration (**D**) Optical microscopy images of spheroids in aggrewell according to FBS concentration. (**E**) 2D confocal images of spheroids in aggrewell according to FBS concentration.



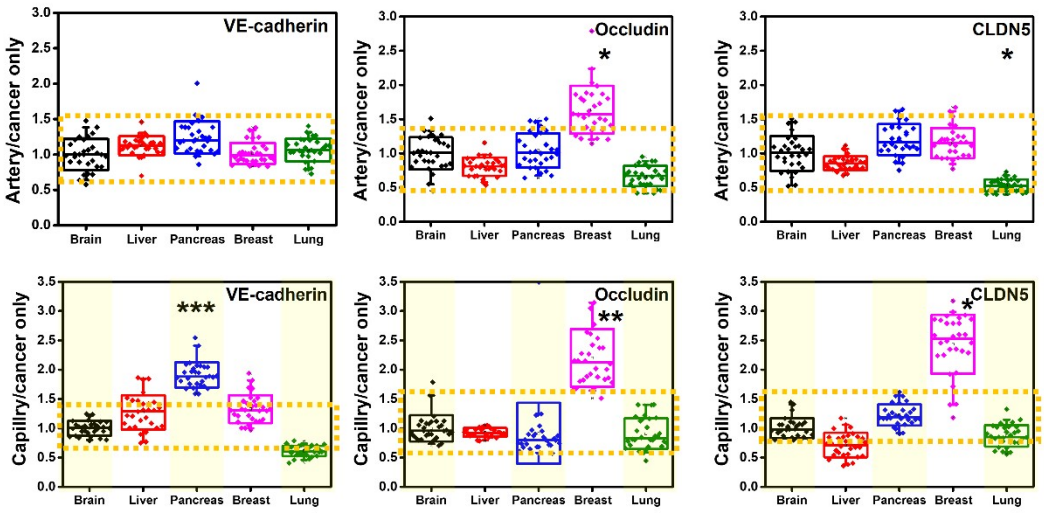
SI. Figure 2. Relative expression levels of junction markers in GBM only spheroids

Gene		Forward Primer	Reverse Primer
Oncogene	*BRAF*	AACGAGACCGATCCTCATCAGC	GGTAGCAGACAAACCTGTGGTTG
	EGFR	AACACCCTGGTCTGGAAGTACG	TCGTTGGACAGCCTTCAAGACC
	HER2	GGAAGTACACGATGCGGAGACT	ACCTTCCTCAGCTCCGTCTCTT
	HER3	CTATGAGGCGATACTTGGAACGG	GCACAGTTCCAAAGACACCCGA
	KRAS	CAGTAGACACAAAACAGGCTCAG	TGTCGGATCTCCCTCACCAATG
	MYC	CCTGGTGCTCCATGAGGAGAC	CAGACTCTGACCTTTTGCCAGG
	AKT	TGGACTACCTGCACTCGGAGAA	GTGCCGCAAAAGGTCTTCATGG
	PI3K	GAAGCACCTGAATAGGCAAGTCG	GAGCATCCATGAAATCTGGTCGC
Tumor Suppressor	*BRCA1*	CTGAAGACTGCTCAGGGCTATC	AGGGTAGCTGTTAGAAGGCTGG
	BRCA2	GGCTTCAAAAAGCACTCCAGATG	GGATTCTGTATCTCTTGACGTTCC
Rho	*CDC42*	TGACAGATTACGACCGCTGAGTT	GGAGTCTTTGGACAGTGGTGAG
	RAC1	CGGTGAATCTGGGCTTATGGGA	GGAGGTTATATCCTTACCGTACG
	RhoA	TCTGTCCCAACGTGCCCATCAT	CTGCCTTCTTCAGGTTTCACCG
Metastasis	*CXCR4*	CTCCTCTTTGTCATCACGCTTCC	GGATGAGGACACTGCTGTAGAG
	MMP3	CACTCACAGACCTGACTCGGTT	AAGCAGGATCACAGTTGGCTGG
Housekeeping genes	*β-actin*	TGCCCATCTACGAGGGGTATG	TCCTTAATGTCACGCACGATTTC
	GAPDH	CCTCCTGCACCACCAACTGCTT	GAGGGGCCATCCACAGTCTTCT
	18s rRNA	GGCCCTGTAATTGGAATGAGTC	CCAAGATCCAACTACGAGCTT

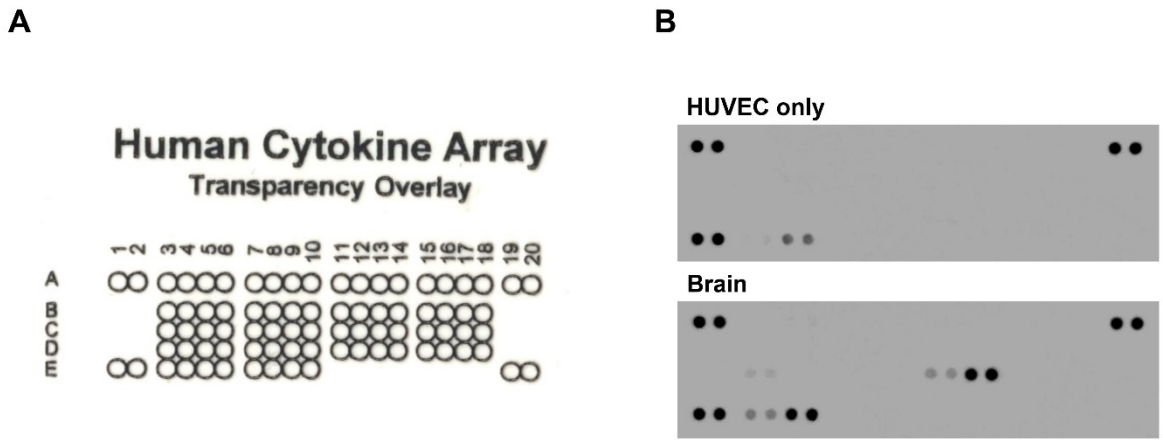
Table S1. Human primer sequences for qRT-PCR assay



SI. Figure 3. Relative expression levels of junction markers in each cancer only spheroids



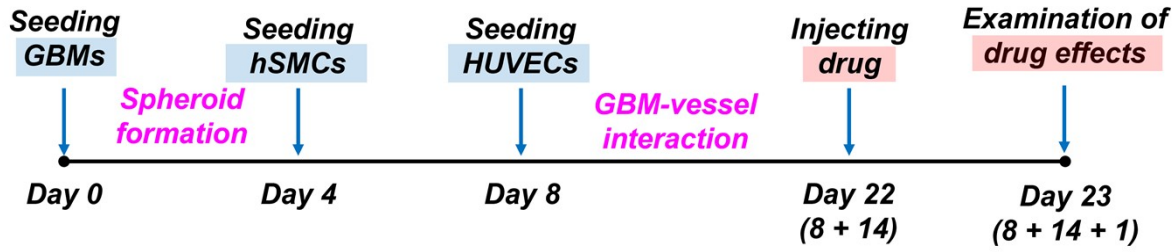
SI. Figure 4. Relative junction marker expression levels in artery and capillary models.



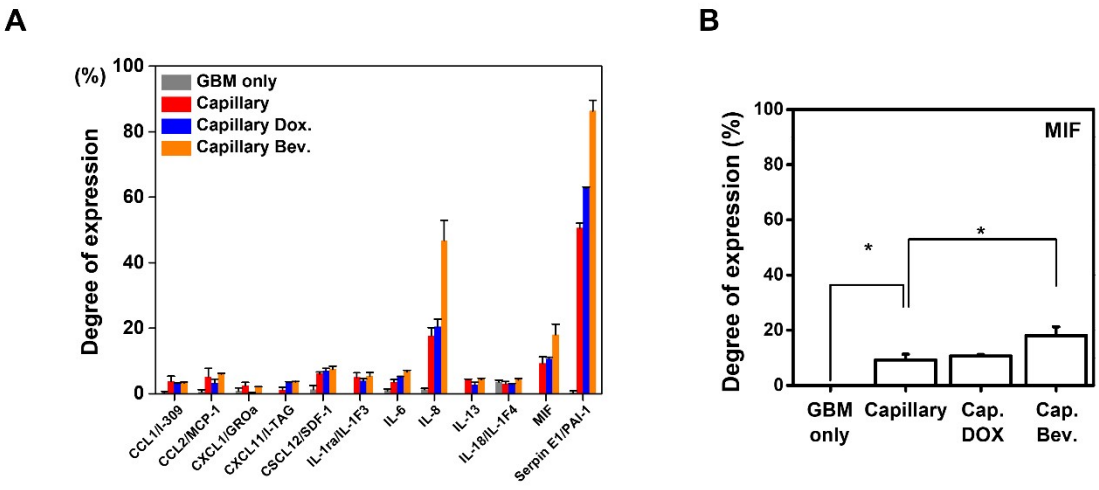
SI. Figure 5. The overlay and raw image of human cytokine assay.

coordinate	Target/Control	coordinate	Target/Control
A1, 2	Reference Spots	C7, 8	IL-4
A3, 4	CCL1/I-309	C9, 10	IL-5
A5, 6	CCL2/MCP-1	C11, 12	IL-6
A7, 8	MIP-1α/MIP-1β	C13, 14	IL-8
A9, 10	CCL5/RANTES	C15, 16	IL-10
A11, 12	CD40 Ligand/TNFSF5	C17, 18	IL-12 p70
A13, 14	Complement Component C5/C5a	D3, 4	IL-13
A15, 16	CXCL1/GROα	D5, 6	IL-16
A17, 18	CXCL10/IP-10	D7, 8	IL-17A
A19, 20	Reference Spots	D9, 10	IL-17E
B3, 4	CXCL11/I-TAC	D11, 12	IL-18/IL-1F4
B5, 6	CXCL12/SDF-1	D13, 14	IL-21
B7, 8	G-CSF	D15, 16	IL-27
B9, 10	GM-CSF	D17, 18	IL-32α
B11, 12	ICAM-1/CD54	E1, 2	Reference Spots
B13, 14	IFN-γ	E3, 4	MIF
B15, 16	IL-1α/IL-1F1	E5, 6	Serpin E1/PAI-1
B17, 18	IL-1β/IL-1F2	E7, 8	TNF-α
C3, 4	IL-1ra/IL-1F3	E9, 10	TREM-1
C5, 6	IL-2	E19, 20	Negative Control

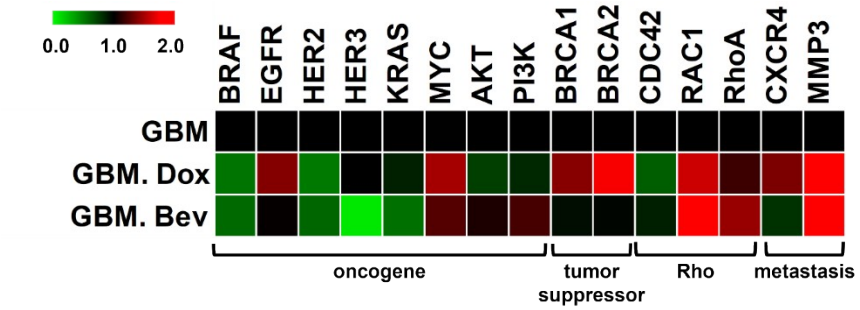
Table S2. Coordinates of the spots and list of the 40 different cytokine proteins on the Human Cytokine Array membrane.



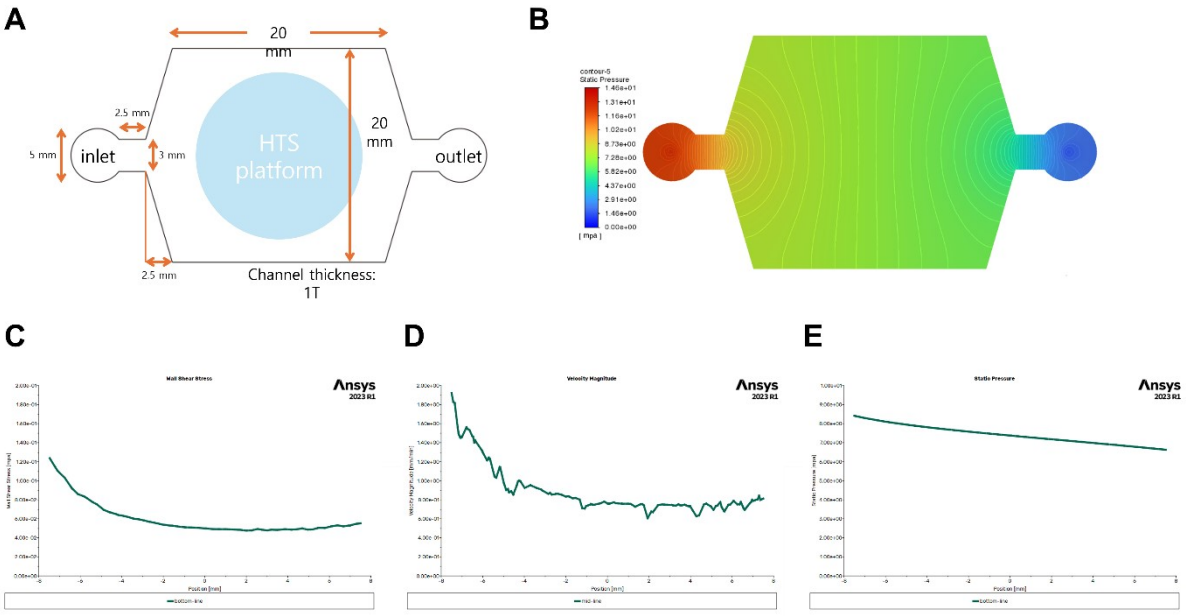
SI. Figure 6. The time point for drug test.



SI. Figure 7. Comparison of cytokines in vascular models according to drugs. (A) The degree of expression of human cytokine in drug-treated conditions. (B) The degree of expression of MIF in drug-treated conditions.



SI. Figure 8. qRT-PCR data for drug-treated GBM only spheroids plotted as a heatmap.



SI. Figure 9. Device design and simulation data. (A) Specific values for device design. (**B**) Simulation of the static pressure of the fluidic device. Plot of (**C**) shear stress, (**D**) velocity, and (**E**) pressure within the HTS platform range