

# ADVANCED BIOSYSTEMS

## Supporting Information

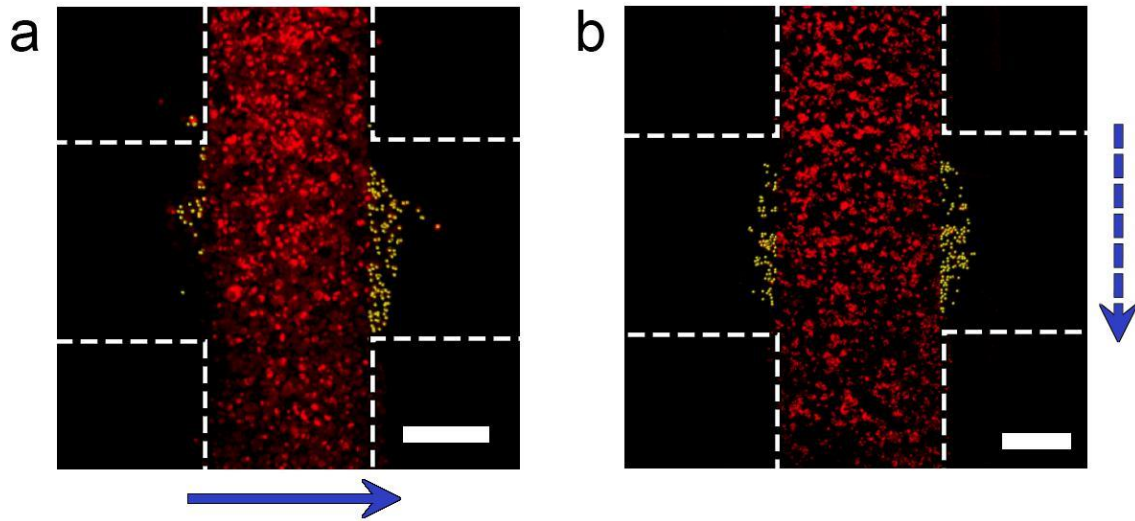
for *Adv. Biosys.*, DOI: 10.1002/adbi.201900273

Multi-Domain Photopatterned 3D Tumor Constructs in a  
Micro-Physiological System for Analysis, Quantification, and  
Isolation of Infiltrating Cells

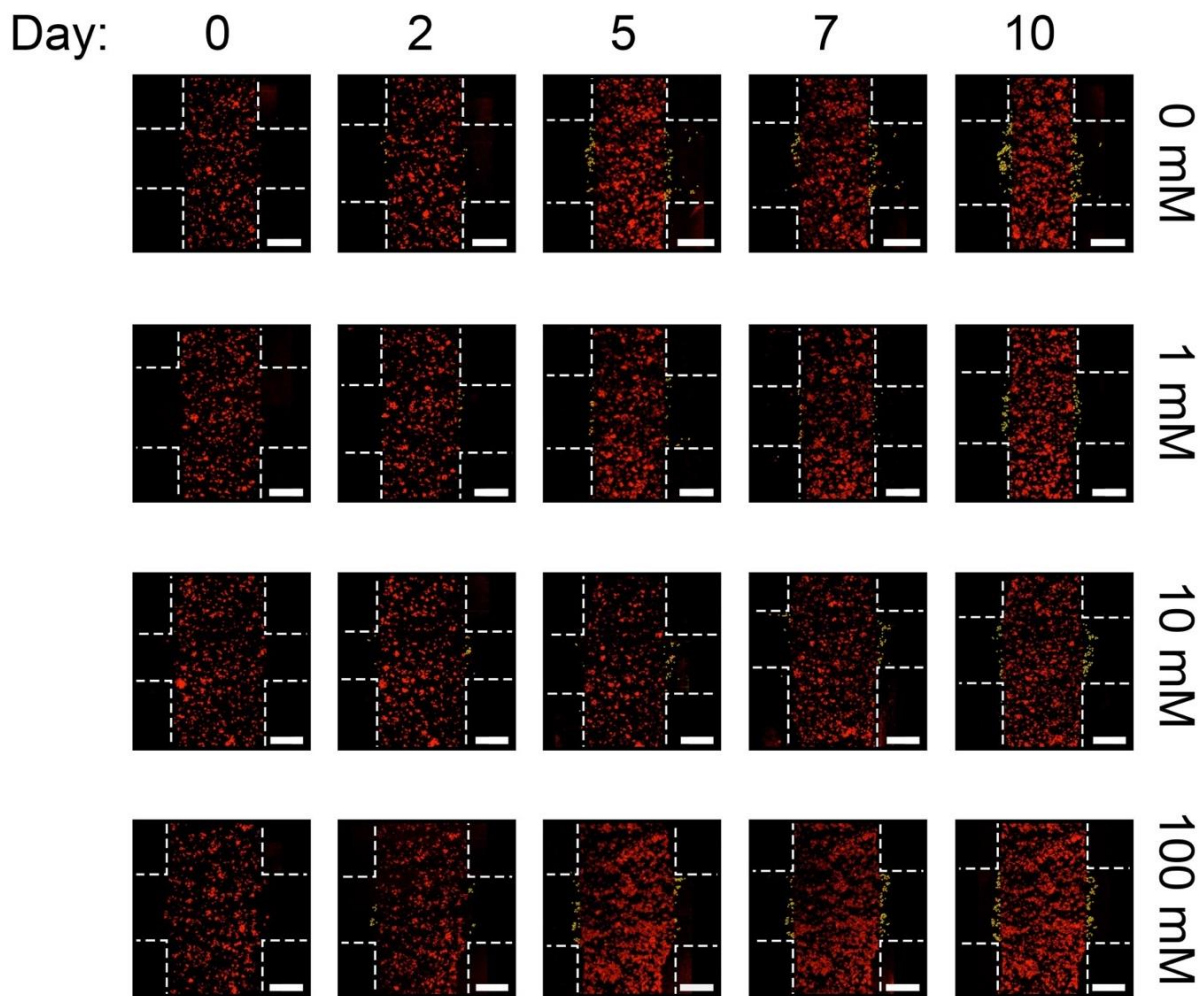
*Shiny A. P. Rajan, Aleksander Skardal,\* and Adam R. Hall\**

*Supporting Information*

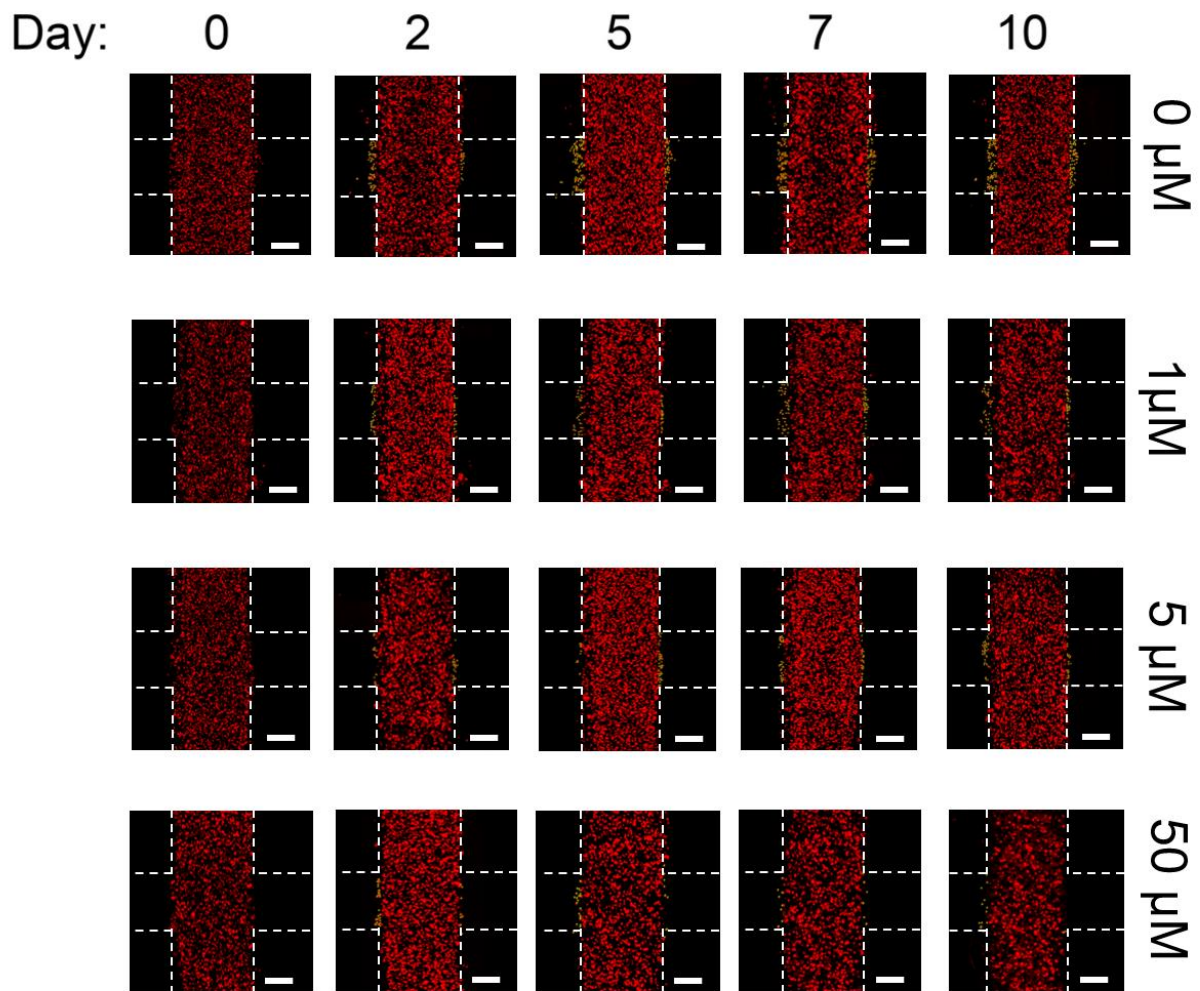
**Multi-domain photopatterned 3D tumor constructs in a micro-physiological system for analysis, quantification, and isolation of infiltrating cells**



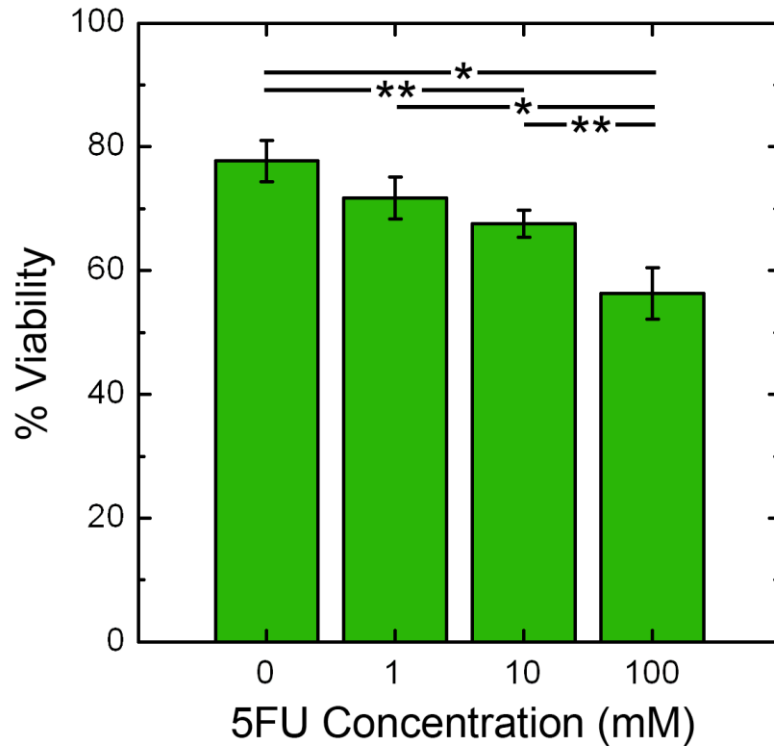
**Supplemental Figure S1** Processed maximum projection confocal micrographs of HCT-116 cells after 10 days of continuous flow (a, 4  $\mu\text{L}/\text{min}$ ) and 10 days of intermittent flow (b, 4  $\mu\text{L}/\text{min}$  for 10 min, no flow for 120 min). Positions of invading cells are marked digitally with yellow points overlaid on the micrograph. Blue arrow indicates direction of flow. Construct borders are roughly indicated by white dashed lines (inter-region border not indicated for clarity). Scale bars are 300  $\mu\text{m}$ .



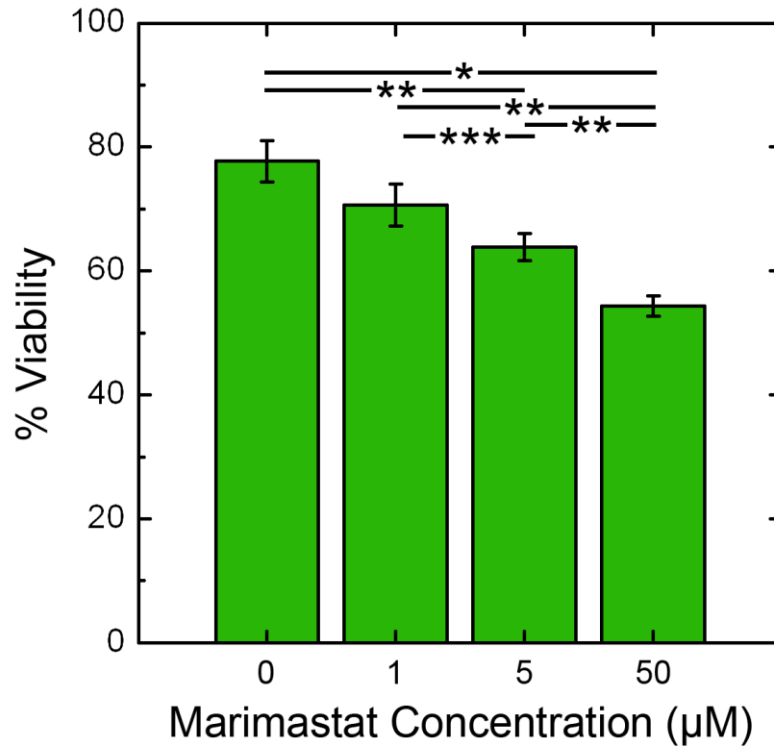
**Supplemental Figure S2.** Expanded view of the typical maximum projection confocal micrographs of cell invasion for all 5FU conditions and time points. Images are processed to show positions of invading cells with yellow points overlaid on the micrograph. Construct borders are roughly indicated by white dashed lines (inter-region border not indicated for clarity). All scale bars are 300  $\mu\text{m}$ .



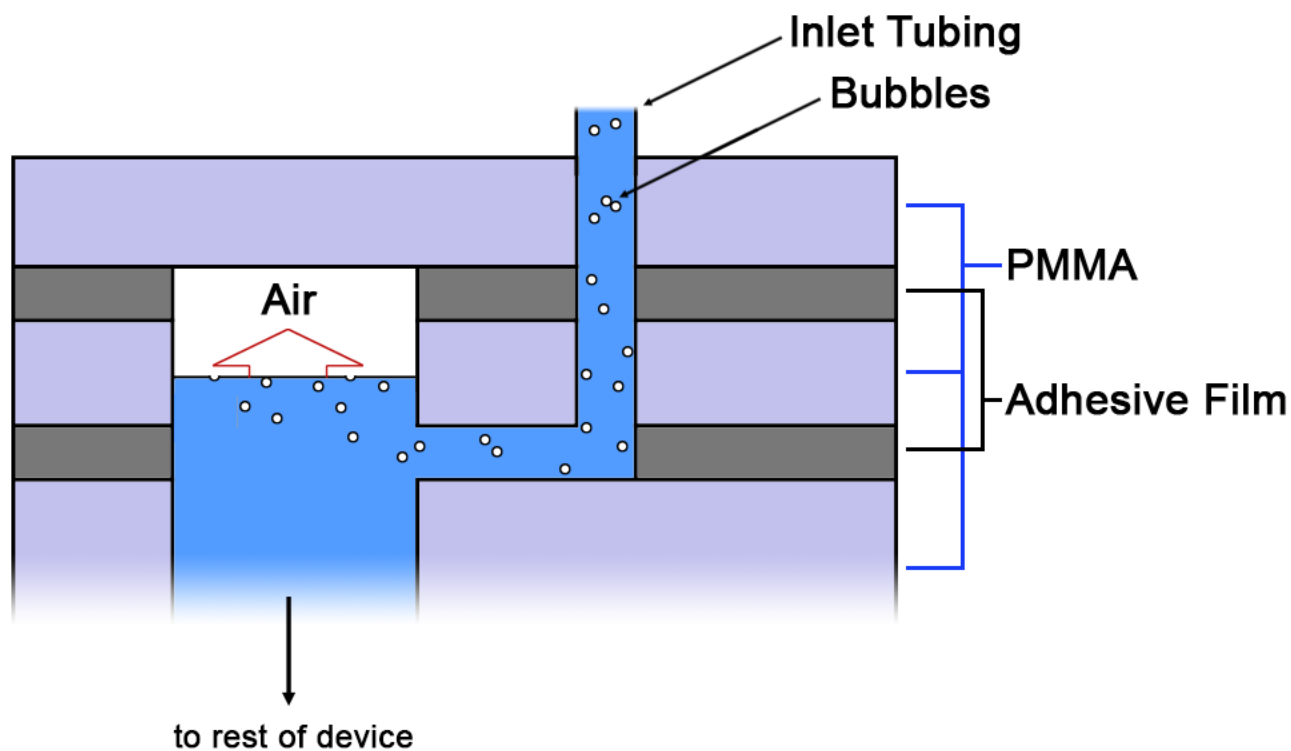
**Supplemental Figure S3.** Expanded view of the typical maximum projection confocal micrographs of cell invasion for all Marimastat conditions and time points. Images are processed to show positions of invading cells with yellow points overlaid on the micrograph. Construct borders are roughly indicated by white dashed lines (inter-region border not indicated for clarity). All scale bars are 300  $\mu\text{m}$ .



**Supplemental Figure S4. LIVE/DEAD viability of HCT-116 under 5FU insult.** Viability on day 10 calculated as the ratio of live cells under a given condition to total number of live and dead cells in the same construct (i.e. *not* scaled to control construct as in the main text). Significance: \*  $p < 0.01$ , \*\*  $p < 0.05$ .



**Supplemental Figure S5. LIVE/DEAD viability of HCT-116 under Marimastat insult.** Viability on day 10 calculated as the ratio of live cells under a given condition to total number of live and dead cells in the same construct (i.e. *not* scaled to control construct as in the main text). Significance: \*  $p < 0.001$ , \*\*  $p < 0.01$ , \*\*\*  $p < 0.05$ .



**Supplemental Figure S6. Schematic of bubble trap.** Cross-sectional view of the top four layers shown in Figure 1a of the main text, illustrating the functioning of the bubble trap. As bubbles in the fluid pass, they are collected in the air cavity due to buoyancy, removing them from the flowing medium. This structure effectively eliminated small and large bubbles from the system, limited ultimately by the volume of the cavity.