

## Supplementary Information

Self-Aligning Tetris-Like (TILE) Modular Microfluidic Platform for Mimicking Multi-Organ Interactions (Ong et al., 2019)

**SI Table 1:** List of all immunostaining reagents with the working concentration.

**SI Table 2:** List of all primers used for RT-qPCR.

**SI Figure 1.** Setting up and performing perfusion culture in a single TILE tissue module.

**SI Figure 2.** Establishment of an endothelium inflammatory response assay by NF- $\kappa$ B nuclear translocation in human coronary artery endothelial cells (HCAEC).

**SI Figure 3.** Patient derived primary (HN137-pri) and metastatic (HN137-met) oral cell squamous carcinoma (OSCC) tumor spheroids exhibited differential susceptibility to cyclophosphamide induced cytotoxicity.

**SI movie.** Fluidic network setup and operation with TILE microfluidic modules.

**SI script.** Python script to for pressure drop estimation from CFD.

### SUPPLEMENTARY TABLES

Antibodies	Cat no	Working concentration ( $\mu$ g/ml)	Source
NF- $\kappa$ B (rabbit)	AB32536	20	Abcam
CYP3A4 (rabbit)	AB135813	20	Abcam
CK19 (mouse)	AB7754	20	Abcam
Vimentin (rabbit)	AB92547	40	Abcam
E-CAD (mouse)	AB1416	40	Abcam
CD31 (mouse)	AB24590	20	Abcam
Donkey anti-mouse Alexa Fluor <sup>®</sup> 555	A-31570	10	Life Technologies
Donkey anti-rabbit Alexa Fluor <sup>®</sup> 647	A-31573	10	Life Technologies

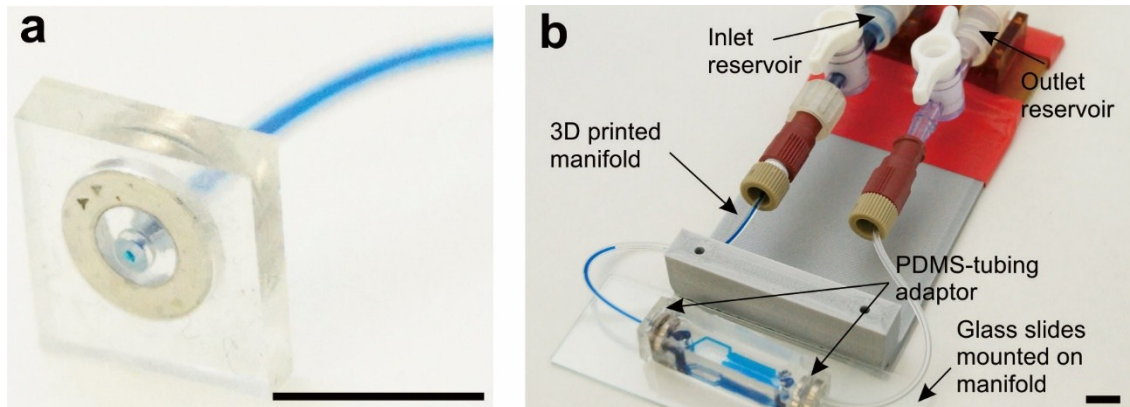
**SI Table 1:** List of all immunostaining reagents with the working concentration.

Name	Forward (5' to 3')	Reverse (5' to 3')
<i>GAPDH</i>	GAGTCAACGGATTTGGTCGT	GACAAGCTTCCCGTTCTCAG
<i>CYP1A2</i>	GGGCCGGCCTGACCTCTACA	CAGGGGGTTCCCGGAGGAGG
<i>CYP3A4</i>	AAGTCGCCTCGAAGATACAC A	AAGGAGAGAACAACACTGCTCGTG
<i>CYP2B6</i>	AGACGCCTTCAATCCTGACC	CCTTCACCAAGACAAATCCG
<i>ICAM1</i>	TCTGTGTCCCCCTCAAAGT C	GGGGTCTCTATGCCCAACAA

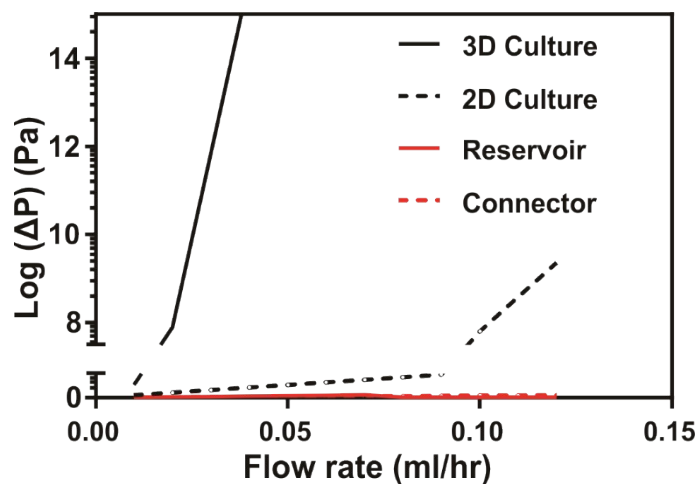
<i>EphB4</i>	GAGAGGTACCTCCTGCAGTGT TC	CCATGTCCGATGAGATACTGTCC G
<i>eNOS</i>	TGATGGCGAAGCGAGTGAA G	ACTCATCCATACACAGGACCC
<i>NOTCH1</i>	CACGCGGATTAATTTGCATC	TCTTGGCATAACACTCCG

**SI Table 2:** List of all primers used for RT-qPCR. All primers were obtained from Integrated DNA Technologies (USA).

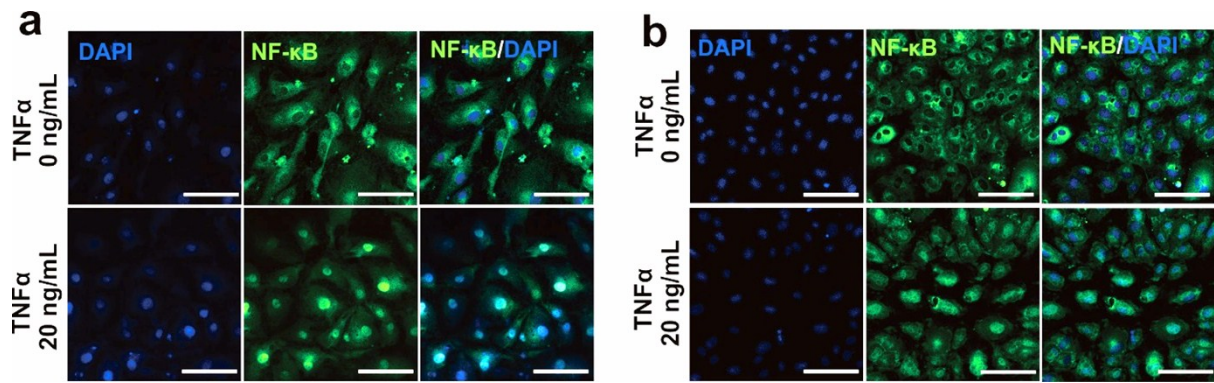
## SUPPLEMENTARY DATA



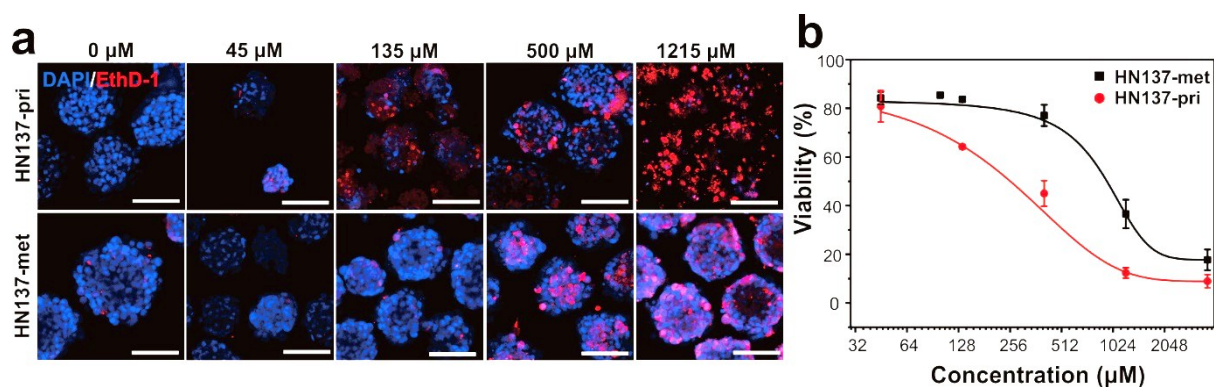
**SI Figure 1.** Setting up and performing perfusion culture in a single TILE tissue module. **(a)** Magnet-embedded PDMS-tubing adaptor to connect inlet and outlet of a module to tubing. **(b)** 3D printed manifold for performing pump-free perfusion culture. The inlet and outlet of the module was connected to a pair of reservoirs that were placed horizontally at different heights to generate a constant hydrostatic pressure to drive medium perfusion. The height difference between the inlet and outlet reservoirs was controlled by the 3D printed manifold. Scale bars = 1 cm.



**SI Figure 2.** CFD simulation for pressure drop across individual TILE modules at different perfusion flow rates. The 3D tissue culture module had the largest pressure drop due to a more complex device geometry as compared to other modules.



**SI Figure 3.** Establishment of an endothelium inflammatory response assay by NF- $\kappa$ B nuclear translocation in human coronary artery endothelial cells (HCAEC). HCAECs were seeded and cultured for 48 hours in a 2D culture module or conventional tissue culture polystyrene plate. Fluorescence immunostaining of NF- $\kappa$ B (green) / DAPI (blue) in HCAECs without (top panel) and with (bottom panel) 20 ng/ml TNF $\alpha$  treatment in a (a) PDMS 2D culture module and (b) tissue culture polystyrene (TCPS) well plate. It was observed that the extent of NF- $\kappa$ B nuclear translocation after TNF $\alpha$ -treatment for HCAECs grown in the 2D culture module was comparable to that of cells cultured on conventional TCPS surfaces. Scale bars = 20  $\mu$ m.



**SI Figure 4.** Patient derived primary (HN137-pri) and metastatic (HN137-met) oral cell squamous carcinoma (OSCC) tumor spheroids exhibited differential susceptibility to cyclophosphamide induced cytotoxicity. Cell viability was determined by double staining for the nuclei of the necrotic and total cell population with EthD-1 and DAPI respectively after 48 hours of drug treatment. (a) Confocal images showing necrotic (red) and total cell (blue) population at varying drug concentrations. (b) Viability quantification. Data are averages  $\pm$  s.e.m of 3 independent experiments. The IC<sub>50</sub> values of cyclophosphamide for HN137-pri and HN137-met spheroids were 256 and 1000  $\mu$ M respectively. Scale bars in (a) = 20  $\mu$ m.