Electronic Supplementary Material (ESI) for Lab on a Chip.

Supplemental document

Table S1. Model parameters for oxygen transport and consumption at 37 °C.

Description	Parameters	Value	References
Henry's constant for oxygen	K_{H,O_2}	1.32×10^{-3} mol·m^{-3}·mmHg^{-1}	[1]
Oxygen partial pressure in atmosphere	P_{O_2}	159 mmHg	[1,2]
Oxygen concentration in medium entering the system	C_{in,O_2}	0.21 mol/m^3	[3,4]
Oxygen diffusion in medium	D_{O_2}	3×10^{-9} m^2/s	[3,4,9]
Oxygen permeability in PDMS	P_{PDMS}	3.786×10^{-11} mol·m·m^{-2}·s^{-1}·mmHg^{-1}	[5,6]
Hepatocyte maximum oxygen consumption rate	V_{max,O_2}	4.8×10^{-17} mol·cell^{-1}·s^{-1}	[3,7,8]
Michaelis-Menten constant for hepatocyte oxygen consumption	K_{m,O_2}	0.5 mmHg	[3,8]

Table S2. The primers used for real-time RT-PCR in this study.

Gene Symbol	Primers (forward/reverse; 5' to 3')
CYP1A1	GATGGTCAAGGAGCACTACA/AAAGAGGTCCAAGACGATGT
CYP1A2	TCAATGACATCTTTGGAGCAG/CTCTGTATCTCAGGCTTGGTC
CYP2B6	GGGAGATTGAACAGGTGATTG/GATGATGTACCCTCGGAAGC
CYP2C9	GGATTTGTGTGGGAGAAGC/TGAAGCACAGCTGGTAGAAG
CYP2D6	CGCATCCCTAAGGGAACGACA/CAGGAAGTGTTCGGGGTGGAA
CYP2E1	CCATCAAGGATAGGCAAGAG/TCCAGAGTTGGCACTACGAC
CYP3A4	TGTCCTACCATAAGGGCTT/GGCTGTTGACCATCATAAAAG
CYP3A5	ATATGGGACCCGTACACATG/CAGAGACCCTGACGATAGGAC
UGT1A1	GAATCAACTGCCTTCACCAA/GACTGTCTGAGGGATTTTGC
UGT2B4	TGTCTACAGCCTCCGCTTCT/GAACTGATCCCACTTCTTCATG
SULT1A1	GAGCCGCACCCACCCTGTT/TGAACGACGTGTGCTGAACCAC
SULT2A1	AAAGACGTTAGAACCCGAAGA/TTTCCAGTCCCCAGATACACC
OAT2	GTGATGCTGCTGGCACTGGTT/CTCTTTCACATGGCCTTGGGTC
OCT1	AAGAGGATGTCACCGAAAAGC/GGATGAGCCCCTGATAGAGCA
SLCO1B3	GCCTAACCTTGACCTATGAT/CAGGTAAGTTATTCCATTGTTC
SLCO2B1	GGGAGTCCACGAAGAAGCAG/GACAGGACCACCAGCAGGAA
AHR	GGTTGTGATGCCAAAGGAAGA/TCATTCGGATATGGGACTCG
RXRα	TCGTCCTCTTTAACCCTGACTC/GCTGCTCTGGGTACTTGTGCT
PXR	GGTCCCCAAATCTGCCGTGTA/CCGGGCGTTGCGTTTCATG
CAR	TTGCAGAAGTGCTTAGATGCT/TCAGCTCTTCTTGCTCCTTACT
MRP2	GACAATTCTAATCTAGCCTACTCC/CATCAACTTCCCAGACATCC
BCRP	GTTCTTGGATGAGCCTACA/CTGAGGCCAATAAGGTCA
MDR1	GCTCGTGCCCTTGTTAGAC/GTGCCATGCTCCTTGACTC
BSEP	CCCTCATCCGAAATCCCAAGA/TGCAGTGCCATGTTCAAAACC
Albumin	ACCCCAAGTGTCAACTCCAA/GGTTCAGGACCACGGATAGA

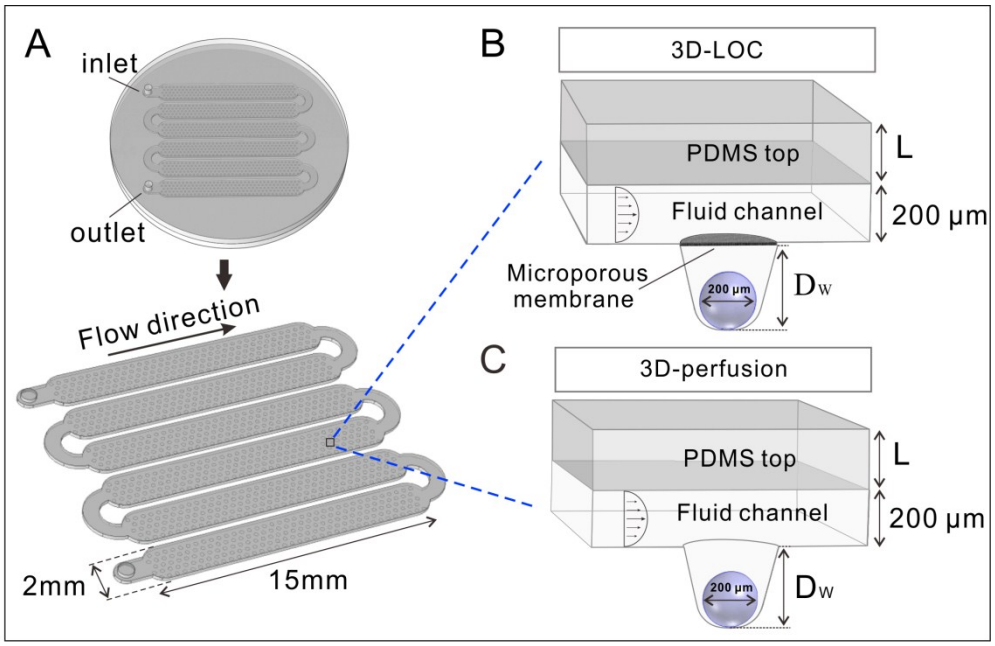

Figure S1. FEM model geometry of the 3D-LOC for the simulation analysis of flow and oxygen mass transfer. (A) 3D geometry of the modeled microfluidic channel, which contained 180 × 6 arrays of microwells. (B) A single microwell geometry of 3D-LOC, which contained a microporous membrane and a Ø 200 μm cell spheroid. The microwell had a V-shape cross section with round bottom (Ø 250 μm × 25° Angle). The fluid channel height, top PDMS layer thickness and microwell depth were 200μm, L and D_W, respectively. (C) For comparison, a single microwell geometry with open microwell (Conventional perfusion method, 3D-perfusion) was also constructed and the dimension parameters were the same as those in (B).

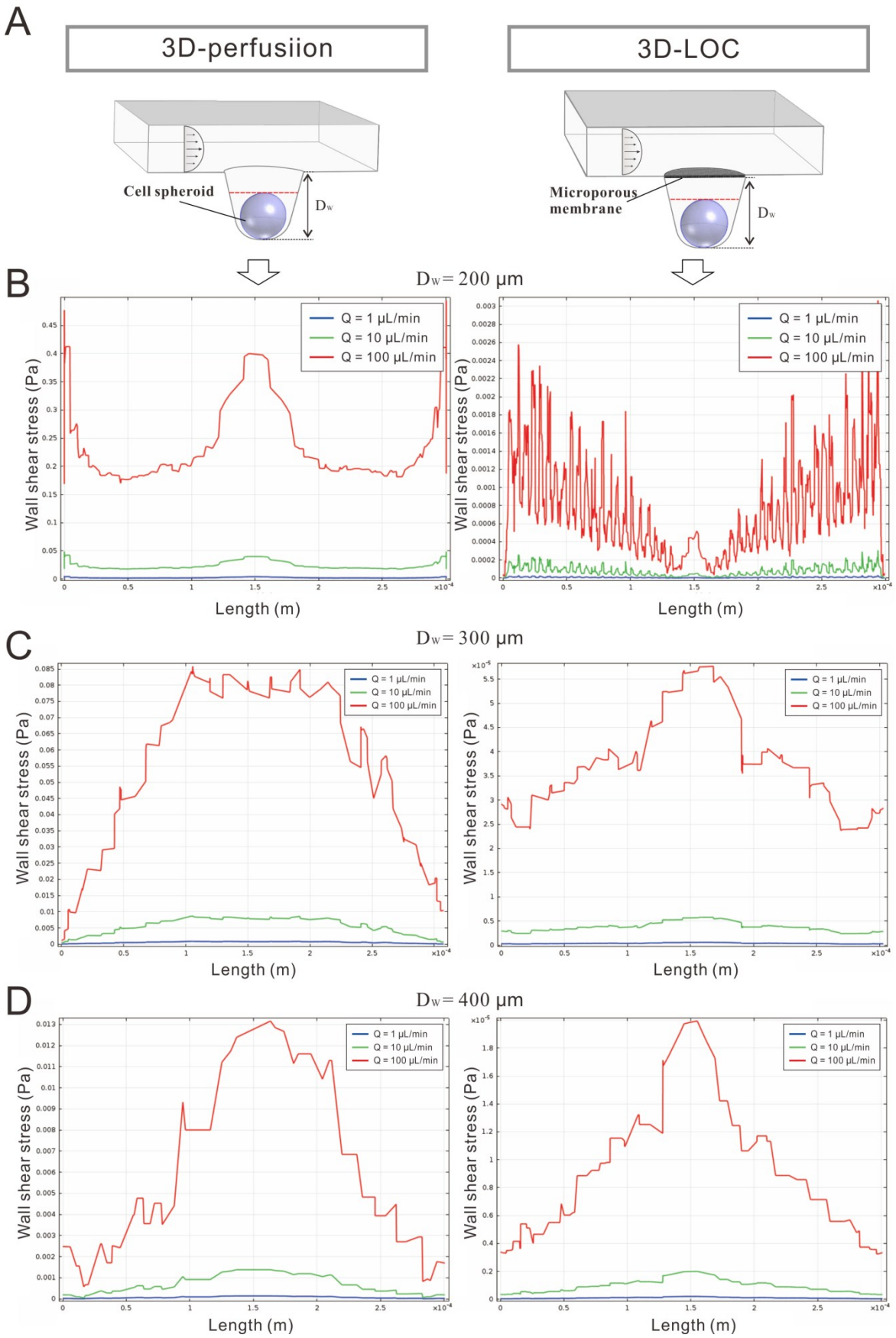

Figure S2. A comparison of wall shear stress in two different perfusion methods (3D-perfusion and 3D-LOC). The wall shear stress distribution along the top red cut lines of the cell spheroid, described in (A), were plotted in different microwell depth configurations including (B) D_W = 200 μm, (C) D_W = 300 μm, and (D) D_W = 400 μm.

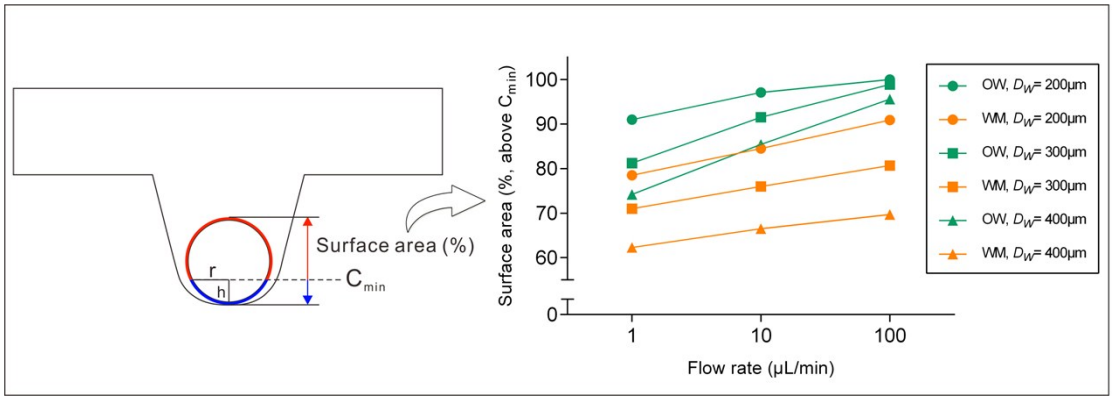

Figure S3. The percentage of the surface area of the cell spheroid (Oxygen concentration > C_{min}) in the 3D-perfusion method with open microwell (OW) and in the 3D-LOC method with membrane (MW) at different microwell depths (D_W = 200, 300 and 400 µm) and flow rates (Q = 1, 10 and 100 µL/min).

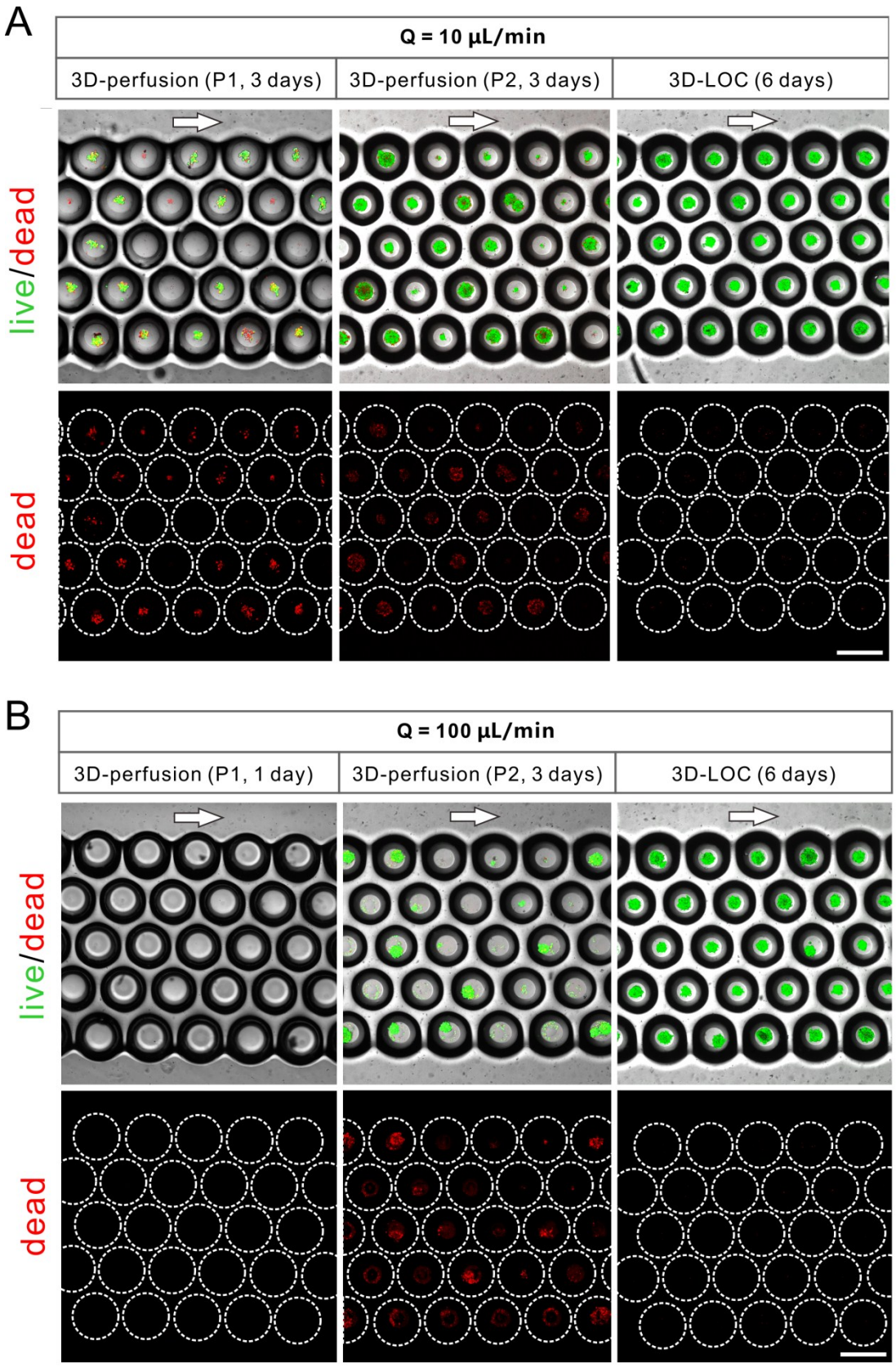

Figure S4. A comparision of cell spheroids viability and loss status in three different perfusion methods (3D-perfusion (P1), 3D-perfusion (P2), and 3D-LOC) perfusion cultured for 1-6 days at different flow rates including (A) Q = 10 µL/min, and (B) Q = 100 µL/min. White arrows indicate the direction of fluid flow. Scale bars = 400 µm.

Supplementary References

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