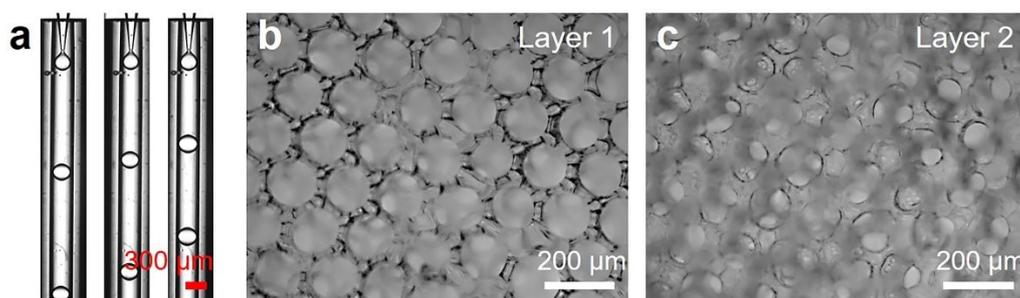


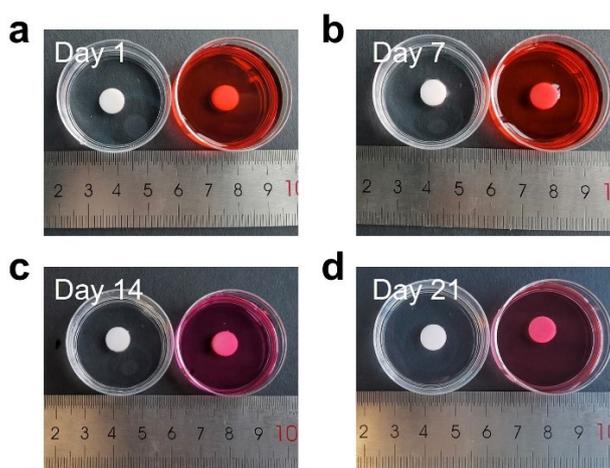
## Supplementary materials

### In vitro vascularized liver tumor model based on microfluidic inverse opal scaffold for immune cell recruitment investigation

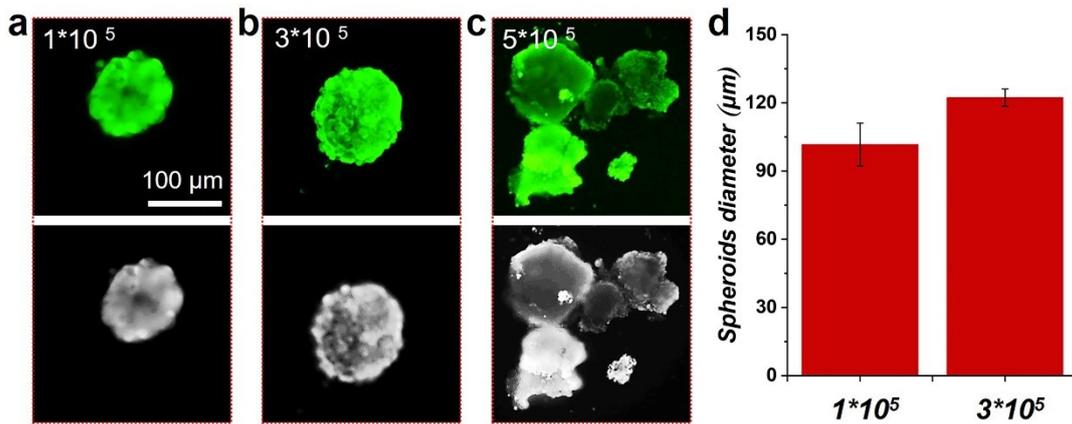
Pingwei Xu, Junjie Chi\*, Xiaochen Wang, Meng Zhu, Kai Chen, Qihui Fan\*, Fangfu Ye\*,  
Changmin Shao\*



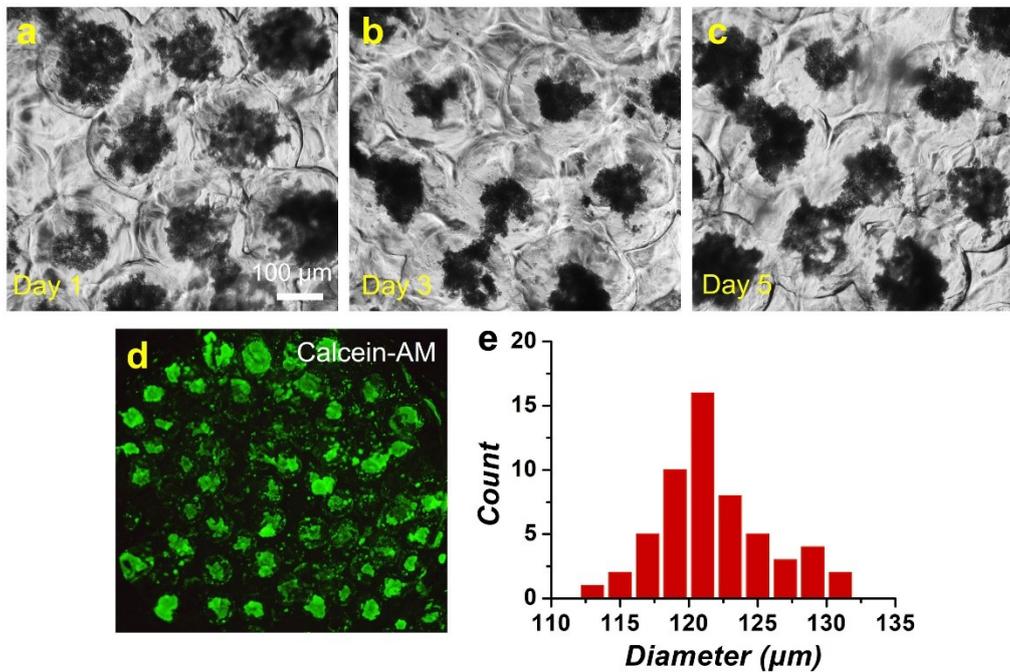
**Figure S1.** (a) Microscopic images of droplets generation in real time. (b and c) Images of the inverse opal hydrogel scaffold under different focal planes of optical microscopy.



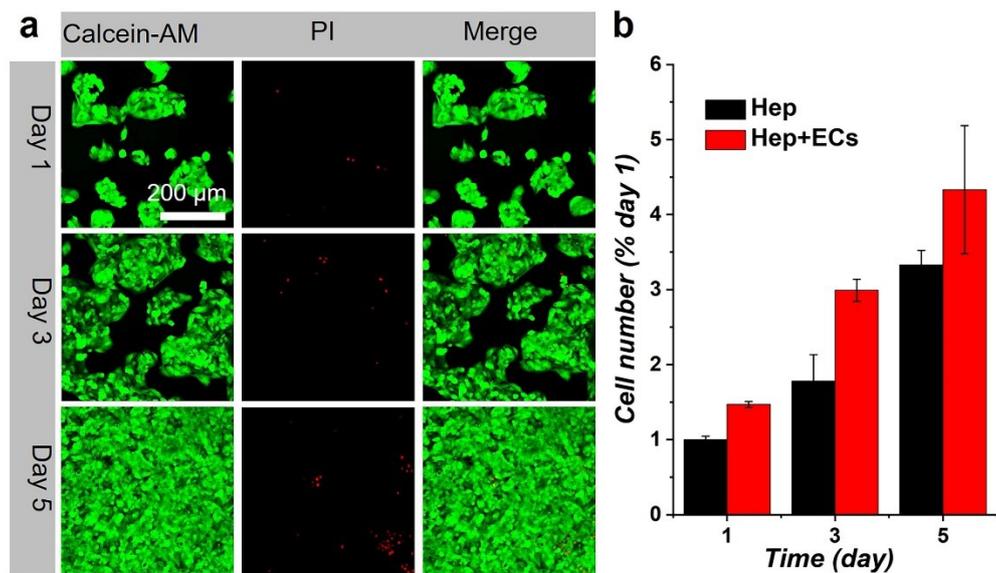
**Figure S2.** Images of the inverse opal hydrogel scaffold cultured in PBS a) and medium b) for 21 days.



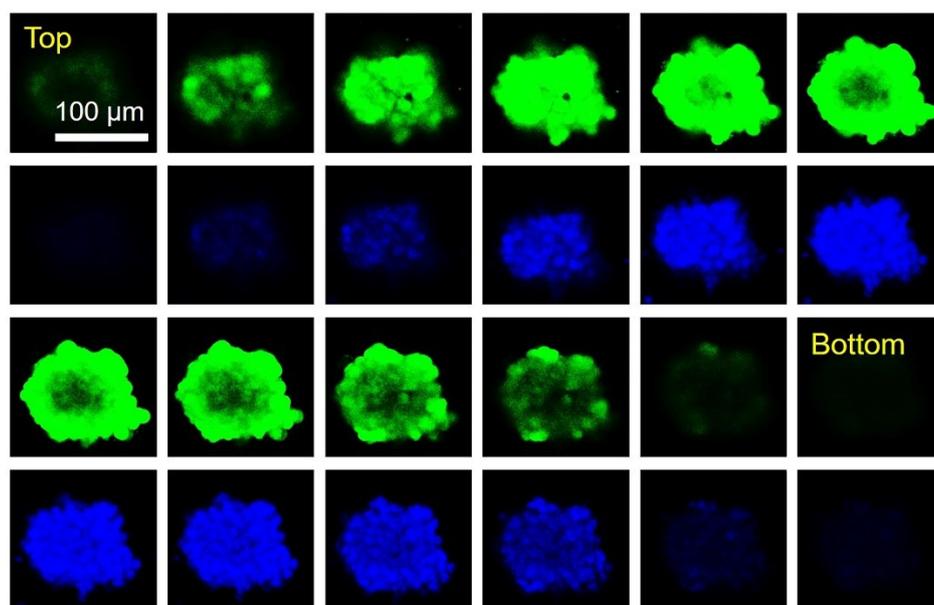
**Figure S3.** (a-c) Fluorescent images of the HepG2 spheroids formed at different cell concentrations ( $1 \times 10^5$ ,  $3 \times 10^5$  and  $5 \times 10^5$  cells/mL). (d) Statistical results of the HepG2 spheroids diameter.



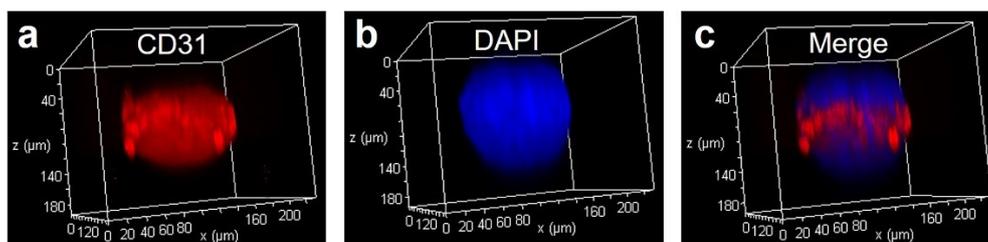
**Figure S4.** (a-c) Microscopic images of the Hep+ECs spheroids cultured in the inverse opal hydrogel scaffold at day 1 a), 3 b) and 5 c). (d) Fluorescent image of the Hep+ECs spheroids formed in the inverse opal hydrogel scaffold. (e) The size distribution of the spheroids.



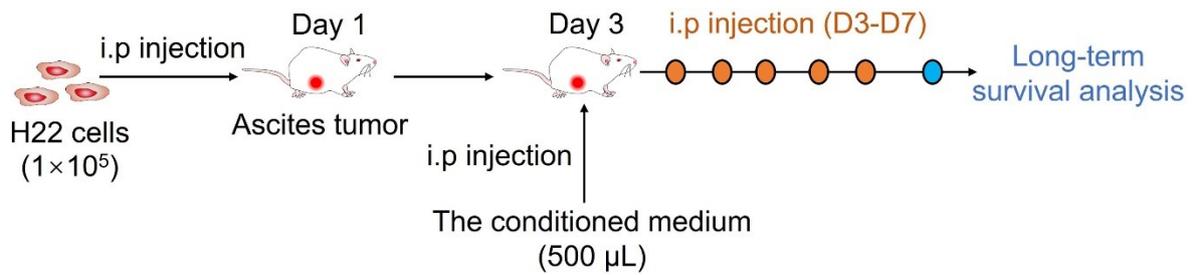
**Figure S5.** (a) Fluorescence images of 2D cells stained with calcein-AM and propidium iodide at day 1, 3 and 5. (b) CCK-8 results of 2D cells.



**Figure S6.** The CLSM z-stack scanning images of the Hep+ECs spheroids. The z-stack scanning with a step size of 10  $\mu\text{m}$  from the top of a spheroid.



**Figure S7.** 3D reconstruction images of CD31 stained Hep+ECs spheroid.



**Figure S8.** Flow chart of long-term survival rate analysis of mice. BALB/c mice were i.p. injected with  $1 \times 10^5$  H22 tumor cells, following the conditioned medium (500  $\mu$ L) i.p. injection daily for 5 days.

### Supplementary Table 1 Primer sequence

Gene	Primer	Primer sequence
Homo <i>CXCL1</i>	FW	AGCTTGCCTCAATCCTGCATCC
	RV	TCCTTCAGGAACAGCCACCAGT
Homo <i>CXCL2</i>	FW	GGCAGAAAGCTTGTCTCAACCC
	RV	CTCCTTCAGGAACAGCCACCAA
Homo <i>Actb</i>	FW	CACCATTGGCAATGAGCGGTTC
	RV	AGGTCTTTGCGGATGTCCACGT