## Supporting Information

Tailoring esophageal tumor spheroids on a chip with inverse opal scaffolds for drug screening

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**Figure S1.** Hydrogel molding experiments with different concentrations (20%, 40%, 60%, 80% from left to right). a: Moment of solidification; b: After natural withering.



**Figure S2.** (a) Real-time microscopic images of the generation of single emulsion droplets. (b) The size distribution of the droplets. (c) The relationship between the droplet diameter and the inner flow rate (F1). (d) The relationship between the droplet diameter and the outer flow rate (F2).



**Figure S3.** Biocompatibility test of materials (green: Calcein AM, red: PI). The scale bar is 100µm.



**Figure S4.** The confocal images of KYSE-70 spheroid in different Z-planes. The scale bar is100µm.



**Figure S5.** HE staining of esophageal cells. (a) Cell morphology in two-dimensional culture mode. (b) Cell spheroplast morphology in different layers. The scale bar is 100µm.



**Figure S6.** Immunofluorescence images of two-dimensional cell models and three-dimensional spherical polymers. (a) The fluorescence pattern of Ki67 proliferation signal in the two-dimensional model. (b) The fluorescence pattern of TUNEL apoptosis signal in the two-dimensional model. (c) The fluorescence pattern of Ki67 proliferation signal in the three-dimensional model. (d) The fluorescence pattern of TUNEL apoptosis signal in the three-dimensional model. The scale bar is 100µm.