On-chip-angiogenesis based on a high-throughput biomimetic three-

dimensional cell spheroid culture system

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Fig. S1 HUVECs seeding results on the lateral face of meshes. (a-c) The schematic diagram of HUVEC cell seeding process principle, the yellow star represents collagen; (d) The results of the cell seeded into each mesh; (e) 3D reconstructed cell confocal images of cells seeded in chip; (f) Bright field image of cells adhered on the SU-8 mesh lateral faces; (g) Fluorescence image of cells adhered on the SU-8 mesh lateral faces; (h) Overlooking fluorescence field image of cell aggregates adhered on the SU-8 mesh lateral faces.



Fig. S2 HUVECs embedded into collagen hydrogel.(a-c)The diagrammatic sketches of the processes of filling collagen hydrogel into meshes and the angiogenesis processes in collagen;(d) HUVECs were unevenly distributed in the chip grid before filling the hydrogel; (e) A more uniform and smooth multilayer cellular structure in hydrogels, the collagen labeled with FITC-collagen (Green) and the HUVECs labeled with cell Tracker[™] Orange CMRA Dye (Red); (f) HUVECs began migrate and sprout to collagen hydrogel.



Fig. S3 The on-chip-angiogenesis processes under different concentrations of VEGF (Ong/mL, 50ng/mL, 100ng/mL).



Fig. S4 Enlarged images of the sprouts at 50 and 100ng/mL VEGF concentration. The white arrow denotes the nascent sprouts from the 3D multilayer HUVECs in the well, the green arrow indicates the microtubules that grown in the presence of VEGF.