

## Supplementary Materials:

### 3D Bioprinting of Model Tissues that Mimic the Tumor Microenvironment

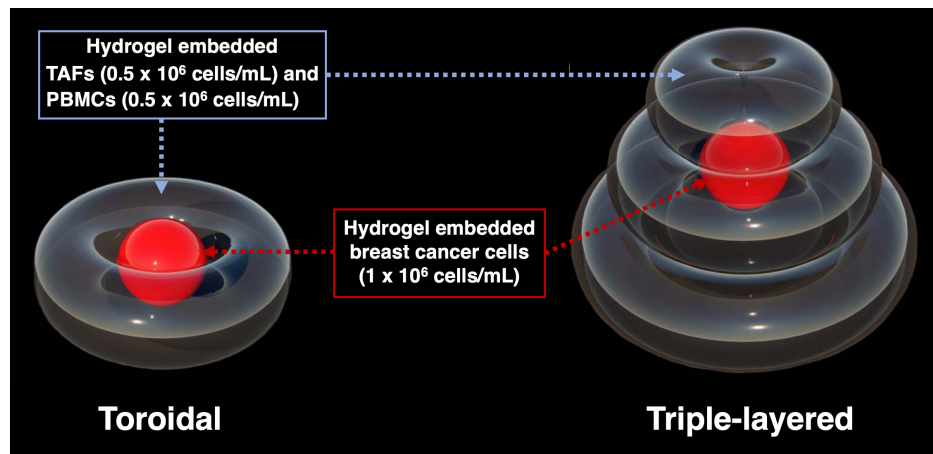
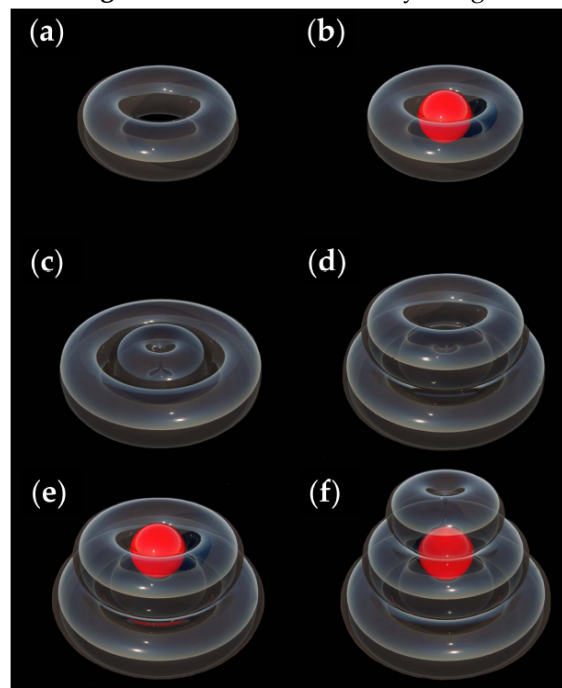
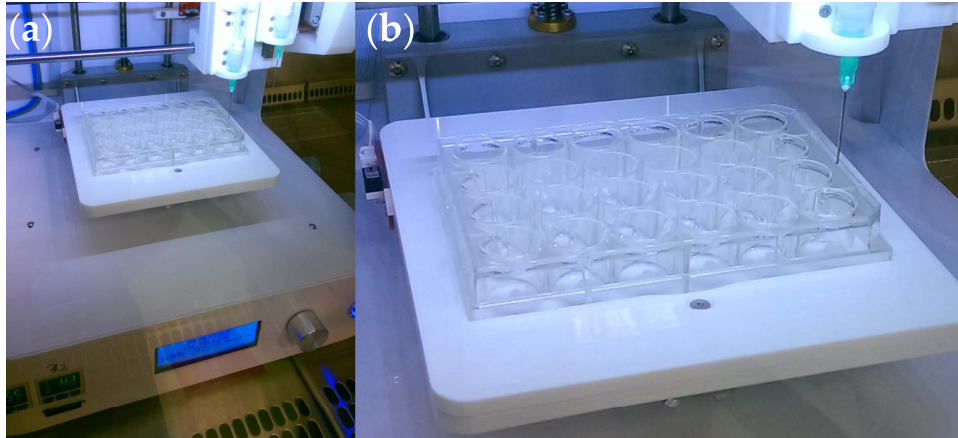


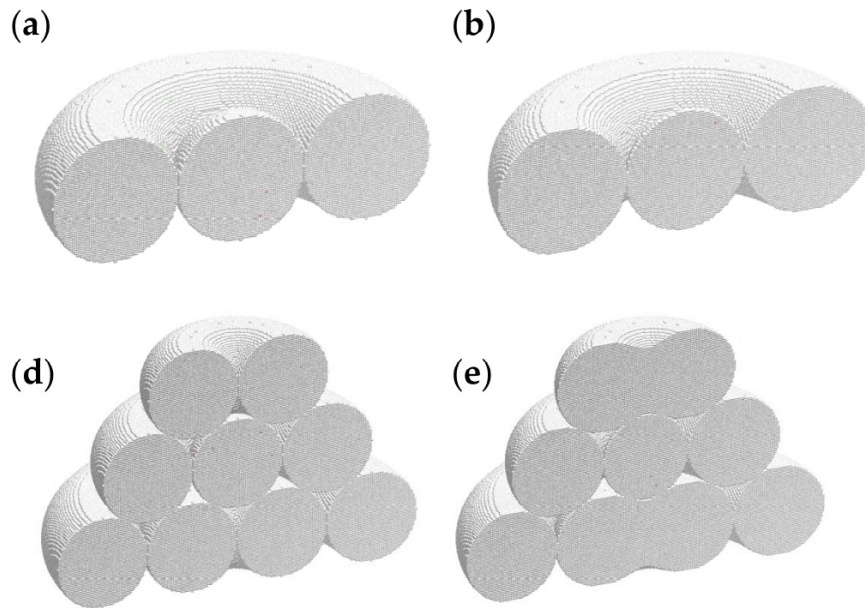
Figure 1. Scheme of the study design.



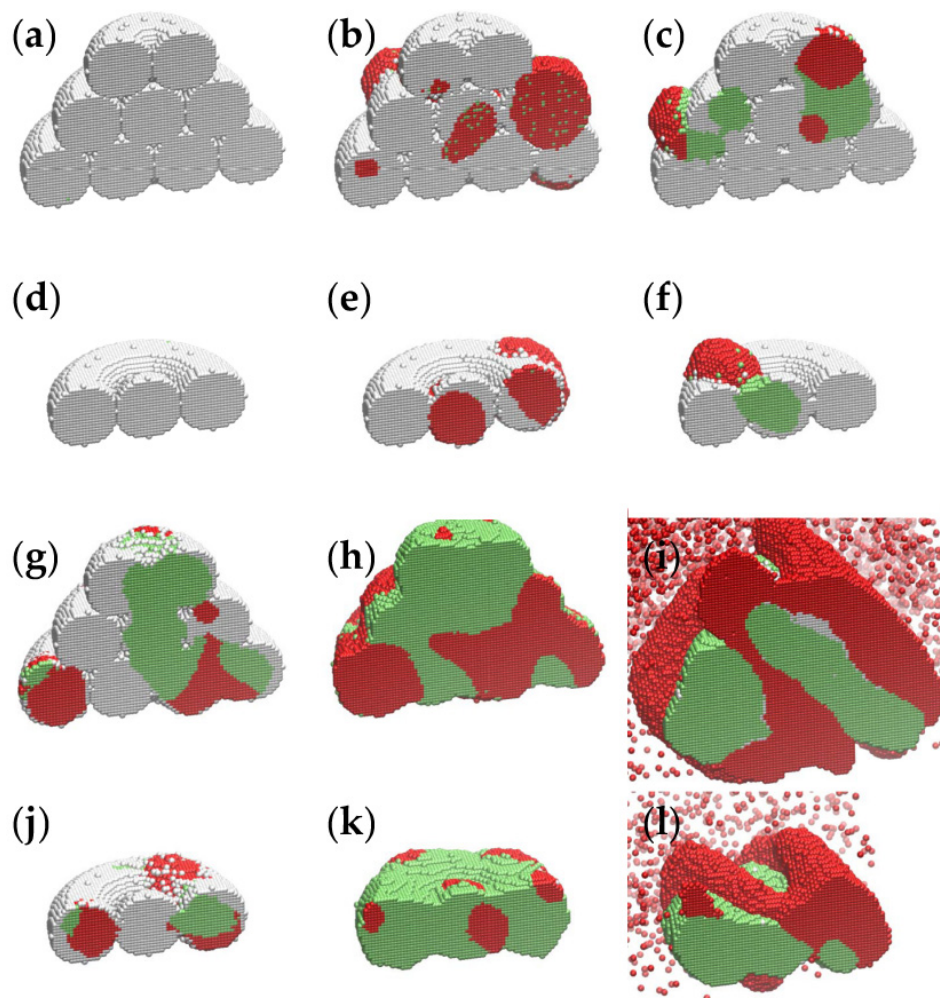
**Figure S2.** Digital models of tissue constructs that mimic the tumor microenvironment: the toroidal structure (b) and the triple-layered structure (f); here, transparent tori represent rings of hydrogel loaded with peritumoral cells, whereas the red sphere stands for a cancer-cell-laden hydrogel droplet. (a) The model of the structure delivered by the first print head; (b) The model of the toroidal structure completed by the second print head; (c) The base layer; (d) The first item of the second layer; (e) The first two layers completed; (f) The triple-layered structure.



**Figure S3.** Picture of the actual bioprinting process. (a) Digital photograph of the INKREDIBLE bioprinter (CELLINK, Sweden) placed in a sterile cell culture hood, taken during the bioprinting of the hydrogel loaded with peritumoral cells; the digital displays from the lower-left corner show the pressures applied to the print heads (PH) – 55 kPa for PH<sub>1</sub> and 42 kPa for PH<sub>2</sub>; (b) A zoom-in picture of the 24-well cell culture plate during the printing of the base layer (according to the model depicted in Figure S2d).



**Figure S4.** Computational models of the tissue constructs shown in cross-section to illustrate the relatively low number of cells in each section. Visualization was done in VMD [40]. Here, silver spheres depict hydrogel volume elements, whereas green and red spheres represent peritumoral cells and cancer cells, respectively. These models were built on a 3D cubic lattice and represent the corresponding biological systems at a scale of 1:1, at a cell density of  $10^6$  cells/mL, used in experiments (see main text for details). (a) Axial cross-section of the toroidal structure; (b) A representative vertical cross-section of the toroidal structure; (c) Axial cross-section of the triple-layered structure; (d) A typical vertical cross-section of the triple-layered structure.



**Figure S5.** Snapshots of representative computer simulations of the evolution of bioprinted model tissues shown in axial cross-section. (a) The computational model of the initial state of the triple-layered structure (scale 1:3); (b, c, g, h, i) The outcome of  $5 \times 10^4$  MCS performed on the model of the triple-layered structure with parameter sets 1-5, respectively (listed in Table 1); (d) The initial state of the toroidal structure (scale 1:3); (e, f, j, k, l) The result of  $5 \times 10^4$  MCS conducted on the model of the toroidal structure with parameter sets 1-5, respectively. Visualization is described in the caption of Figure S2.

**Video S1:** The bioprinting process illustrated by printing ultrasound transmission gel onto the lid of a 24-well cell culture plate. Here, the transparent gel – Konix ultrasound transmission gel (Turkuaz, Turkey) – represents the peritumoral medium, whereas the same gel stained with Neutral Red dye (Sigma-Aldrich, Germany) represents the droplet of hydrogel loaded with cancer cells.