Supplementary Information

Cellular Regeneration and Proliferation on Polymeric 3D Inverse-Space Substrates and Effect of Doxorubicin

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Fig.S1. Average pore size of 3DIS polymer films (PS, PLA and PLGA) increases from 1 to 18 μm with increase in A. relative humidity from 60 % to 100 %, B. polymer concentration from 1 to 5 mg/ml.

Fig.S2. A. Confocal microscopy image showing phalloidin labeled (green) actin fibers in A549 cells cultured on glass cover slips. Nuclear DAPI is indicated in blue. Scale bar indicates 20 μm. B. DOX exhibited a slow release from the 3DIS(+) with ~66 % release in 48 h. In the initial 6 h, there was rapid release of DOX with ~36.7 % release.
Fig. S3 Cell viability assay showed 47 % to 50 % cell viability on PLGA 2D and 3DIS substrates when exposed to IC$_{50}$ concentration of DOX as determined on glass-surface cell culture.

**Video files**: 

A549 Cell on Glass_2D.avi

**Video 1.** A549 Cell(s) cultured on Glass surface, magnified view.

A549 Cell on PLGA 3DIS Pore size_20 um.avi

**Video 2.** A549 Cell cultured on 3DIS substrate with mean pore size = 20 µm, magnified view. Cell demonstrates significant cell thickness with bulk of the cytoplasm (green) within the pore and the nucleus (blue) positioned towards the pore opening. The cell also displays processes extending over the pore opening.

A549 Cell on PLGA 3DIS Pore size_12 um.avi

**Video 3.** A549 Cell cultured on 3DIS substrates with mean pore size = 12 µm, magnified view. The cell demonstrates its ability to occupy available pore capacity and extend its mass (green) beyond the primary housing pore (displaying the nucleus, blue) into neighboring pores.
Video 4. Orthogonal slice of two adjacent A549 cells on 3DIS substrate with mean pore size = 20 µm, magnified & cut-away view. The video displays the cytoplasmic masses (green) of two proximal cells sharing a pore, demonstrating the ability of the system to simulate tissue gap and subsequent lateral contact of the growing cells to mimic regenerative cell division, growth and gap-bridging. The nuclei (blue) can be seen at the apex of the pore system, towards the opening.

§ Supplementary video files can be viewed by double-clicking the video title boxes.