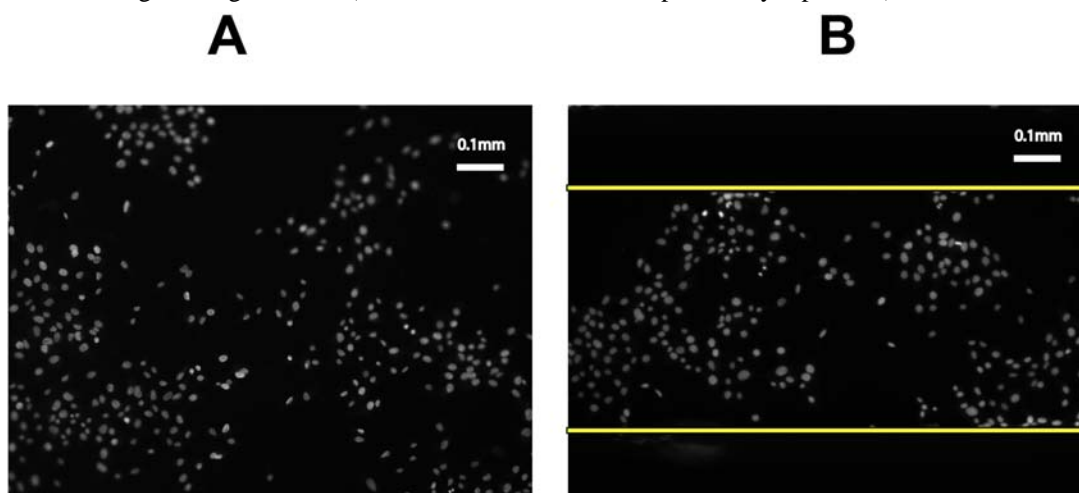
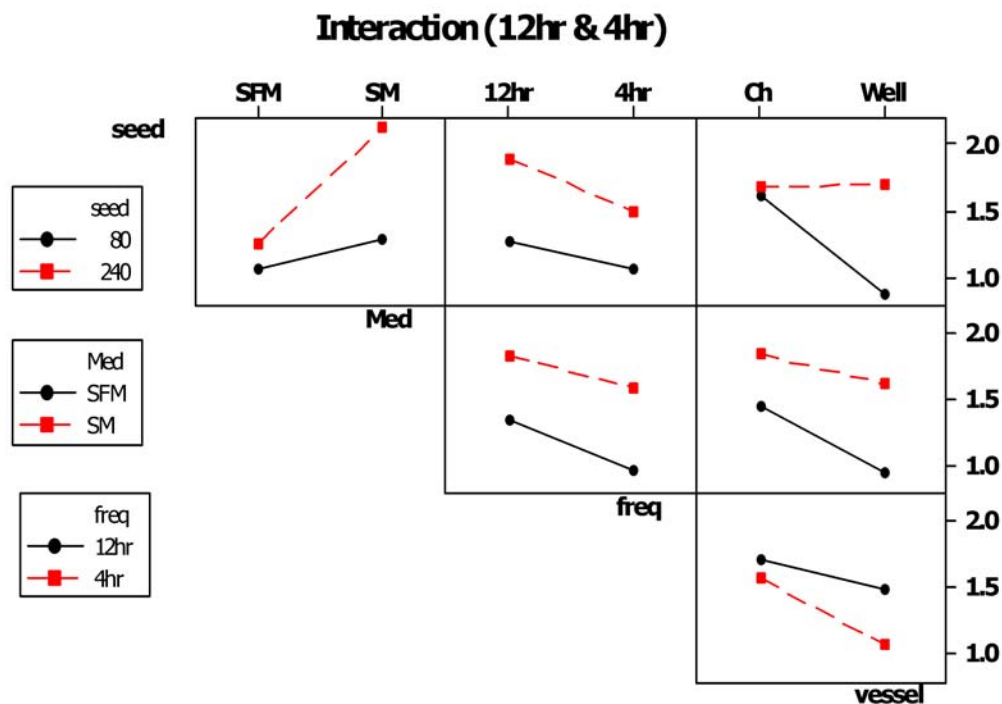


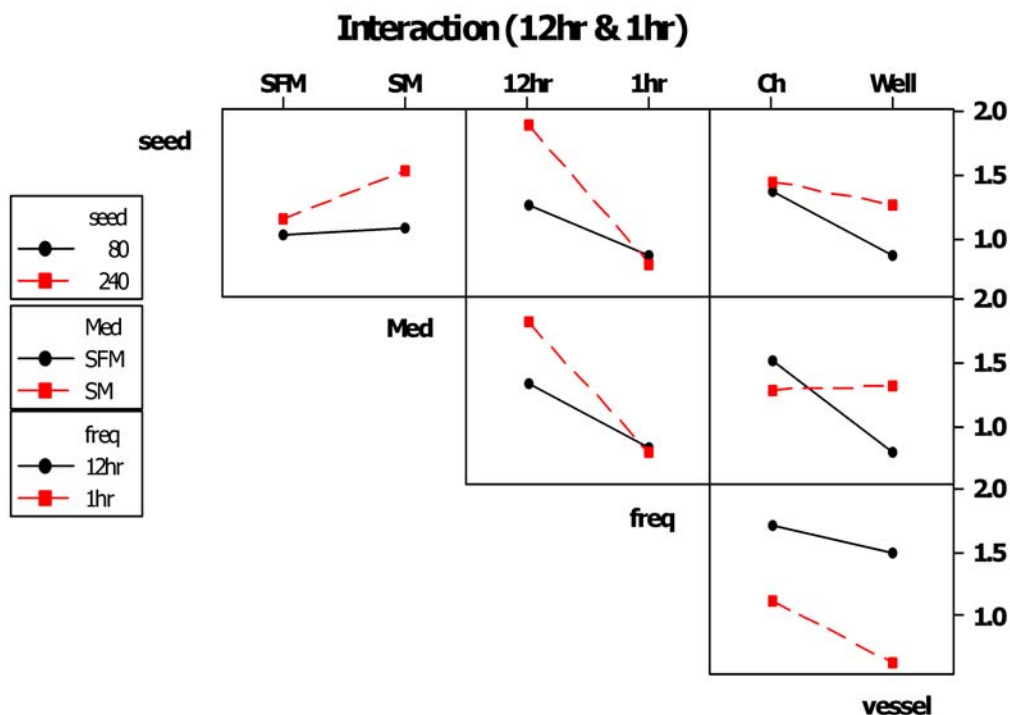
Supplementary Fig. S1 The 96-well format microchannel arrays used in this study are shown. (A) The layout shows that each of the 48 channels expand into a 2-well region of a standard 96-well plate, such that the total surface area of every microchannel is the same as one well (30 mm^2) of the 96-well plate. (B) The fabricated PDMS microchannel arrays are spontaneously attached to an Omnitray and filled with food color dye to compare with the 96-well plate (shown underneath). Due to the same layout of the channel arrays and the wells and the plate dimension, both can be scanned with common plate readers for high through readout (additional details have been previously reported⁹).



Supplementary Fig. S2 Typical fluorescence images of NMuMG cells after the experiments. After experiments, NMuMG cells are stained with Hoechst 33342 and imaged from the (A) 96-well plates and (B) microchannels.



Supplementary Fig. S3 The interaction between seeding density, culture medium, culture vessel and medium change frequency (every 12 h and 1 h). The values and the ranks of the significant interactions are shown in Table 2 of the manuscript. Three interactions out of the six 2-factor interactions are significant: the interaction between the medium and culture vessels, the interaction between the medium and medium change frequency, the interaction between medium and seeding density. The matrix plot shows: 1) the interaction between the medium and culture vessels is the strongest interaction, and also ranked as the 2nd most important factor among all the factors. The differences between the growth of NMuMG in SFM and SM were smaller in microchannels than in the wells, and SFM did not support the growth of NMuMG cells in the 96-well plates; 2) the interaction between the medium and medium change frequency is the 2nd strongest interaction and also ranked as the 4th most important factor in all the factors involved. The differences between SM and SFM became less when the medium was changed every 1h compared to every 12h medium change; 3) the interaction between seeding density and medium change frequency was the smallest significant interaction. The difference between high and low density cultures were reduced after the medium changed frequency was increased (12 h vs. 1 h).



Supplementary Fig. S4 The interaction between seeding density, culture medium, culture vessel and medium change frequency (every 12 h and 4 h). The values and the ranks of the significant interactions were shown in Table 3 of the manuscript. Two interactions out of the six 2-factor interactions are found to be significant: the interactions between the seeding density and culture vessel, and the interaction between the medium and seeding density. The matrix plot shows: 1) the differences between channel and wells were smaller for the high density cultures than for the low density cultures (120 cells mm⁻² vs. 80 cells mm⁻²) and the growth of the low density cultures was worse in wells than in channels. This effect was ranked as the 3rd most important effect; 2) high density cultures had increased overall growth as compared to the low density cultures, but this difference increases in the SM vs. in the SFM. This interaction is the least important factors of the five significant factors.