Electronic Supplementary Material (ESI) for Lab on a Chip.

Supplementary Information

Acoustic formation of multicellular tumor spheroids enabling functional and structural characterization with single cell resolution

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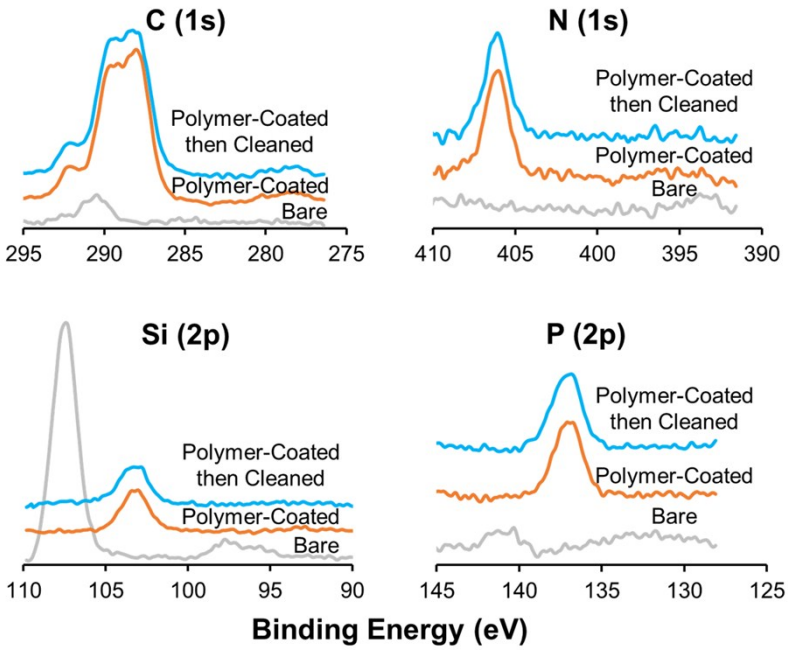

Figure S1 Plots show elemental surface composition on the multi-well microplate, measured by X-ray photoelectron spectroscopy (XPS), of an uncoated (grey line), coated (orange line) and coated and subsequently cleaned microplate (blue line). The cleaned microplate was exposed to 10 min in the ultrasound bath submerged in 70% ethanol and further incubated in 70% ethanol for 1 hour. Charge corrections was applied for better comparison.

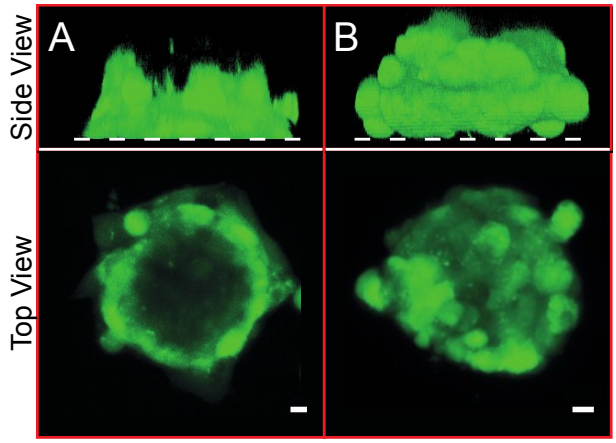

Figure S2: Side and top views of a 3D reconstruction from confocal z-stacks of calcein-stained HepG2 MCTS formed after 24 hours active + 24 hours passive incubation in an uncoated microplate (A) and a coated microplate (B). Note the difference in shape in the side view where the MCTS in (A) adheres to the substrate and reassembles a hemisphere structure compared to the unanchored micro-tumor in (B) that rests on the substrate and exhibits a more spherical formation. Scale bars are 10 µm. The dotted line in the side view marks the micro-well bottom.

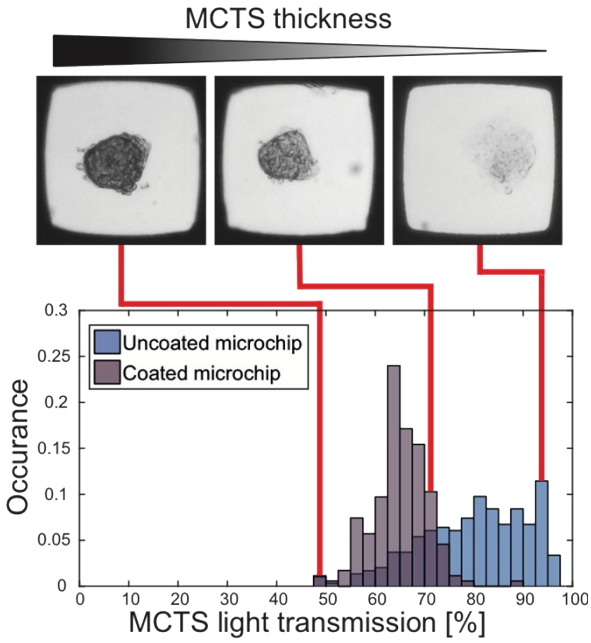

Figure S3: An automatic image-based method was developed to find and analyze the shape and light transmission through the MCTS. Image analysis of the light transmission through the MCTS was used to compare the MCTS thickness in the coated microplate (N = 175) relative to the MCTS in the uncoated microplate (N = 297) after 24 hours active + 24 hours passive incubation.

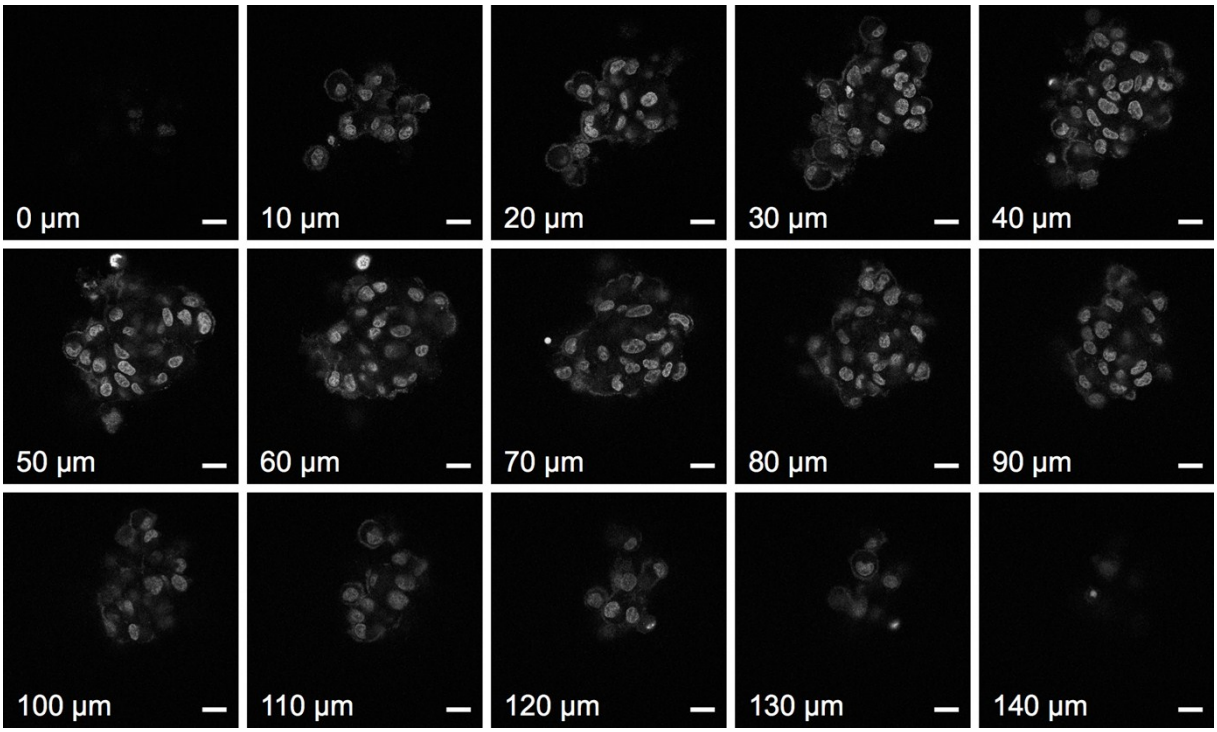

Figure S4: Representative unprocessed images at different z-positions, from 0 µm to 140 µm, in a confocal z-stack, acquired with a 63x oil-immersion objective, of a RIMS treated A498 renal carcinoma MCTS stained with the nuclear dye NucBlue Live based on Hoescht 33342. The laser intensity used to excite the staining was modulated as images were acquired deeper into the sample. Scale bar is 20 µm in all sections.