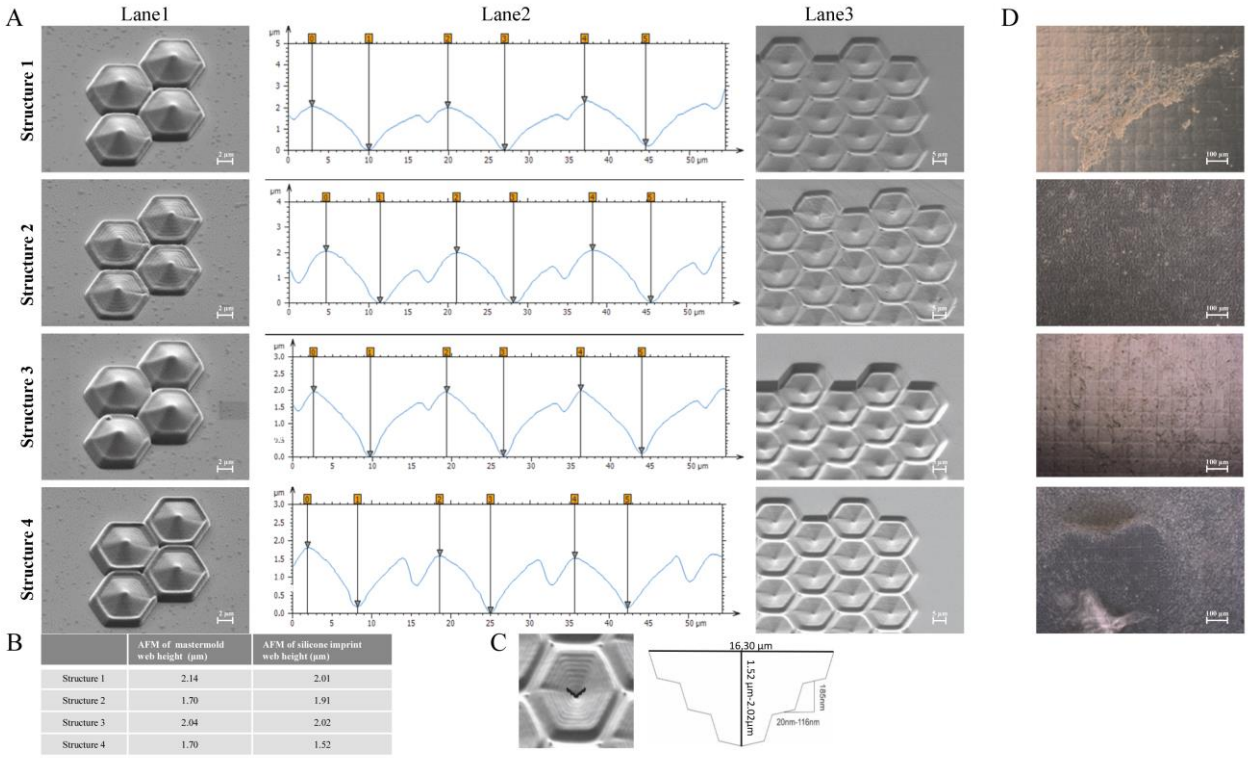
SDC: Engineering of an artificial Descemet´s membrane



	AFM of mastermold web height (µm)	AFM of silicone imprint web height (µm)
Structure 1	2.14	2.01
Structure 2	1.70	1.91
Structure 3	2.04	2.02
Structure 4	1.70	1.52

SDC Figure 1: Engineering of an artificial Descemet´s membrane (DM) substrate by molding the DM like topography (DLT) from an imprinted DM mastermold into silicone and characterization of its surface topography and corneal endothelial cell (CEC) differentiation impact.

A) SEM measurement of four, two-photonic laser polymerized master structures with defined inverted descemet like topography (DLT) differing in height and pitch angle in µm and nm scale (lane 1). The four DLT structures were analyzed with confocal microscopy (lane 2) and molded in silicone (lane 3).

B) Presentation of the molding accuracy of silicone from the master mold structures measured by atomic force microscopy

C) 1000-fold magnification of the molded structure. The silicone imprints differed in micro- and nanometer scale.

Adhesion of hMSC to each DLT structure induced changes of cell morphology from fibroblastic-like into a polygonal shape compared to control cells on smooth silicone between 2 and 5 days of cultivation. Further, in contrast to control cells cultured on the smooth substrate, the cells cultivated on structures decreased all more or less in size in dependency on the specific topography and changed their morphology. Interestingly, each micro- and nanotopography induced a different cellular morphology and a different macroscale behavior. Whereas the adhesion to structure 2 induced only marginal changes in morphology, hMSC cultured on structures 3 and 4 changed morphology to a polygonal shape and shrunk into 3D aggregates.

D) Only the adhesion on the silicone mold of Structure 1 enabled cells to build up 2D cell associations instead of maintaining their fibroblastic morphology (Structure 2) or staying in densely packaged 3D cell constructs (Structure 3 and 4).