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


**Figure S1: Biaxial plot demonstrating the log-linear relation between protein molecular mass and migration distance ($R^2$ = 0.99).** Protein markers are SMMHC (blue, 227 kDa), α-SMA (magenta, 42 kDa) and CNN-1 (green, 34 kDa) (N = 47 single SMCs from one mouse).

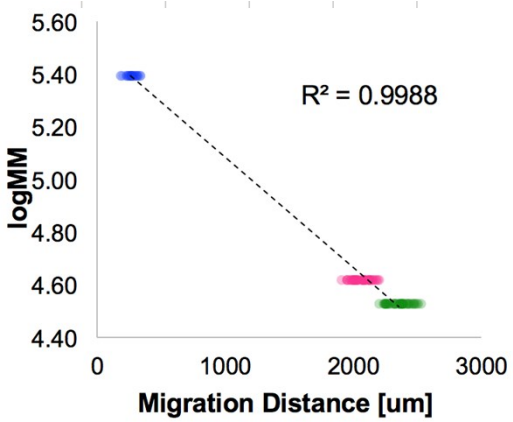


**Figure S2: Empty microwells serve as negative controls.** (A) False-colored fluorescence micrograph of single-cell immunoblot probed for α-SMA, CNN-1 and SMMHC. (B) False-colored micrograph of three adjacent single-cell immunoblots and corresponding fluorescence intensity profiles. Microwells loaded with SMCs (left and right) show immunoblot signal for all three markers (solid lines), while the negative control (empty microwell) shows no immunoblot signal for either marker (dashed lines). Black arrows mark position of protein peaks.

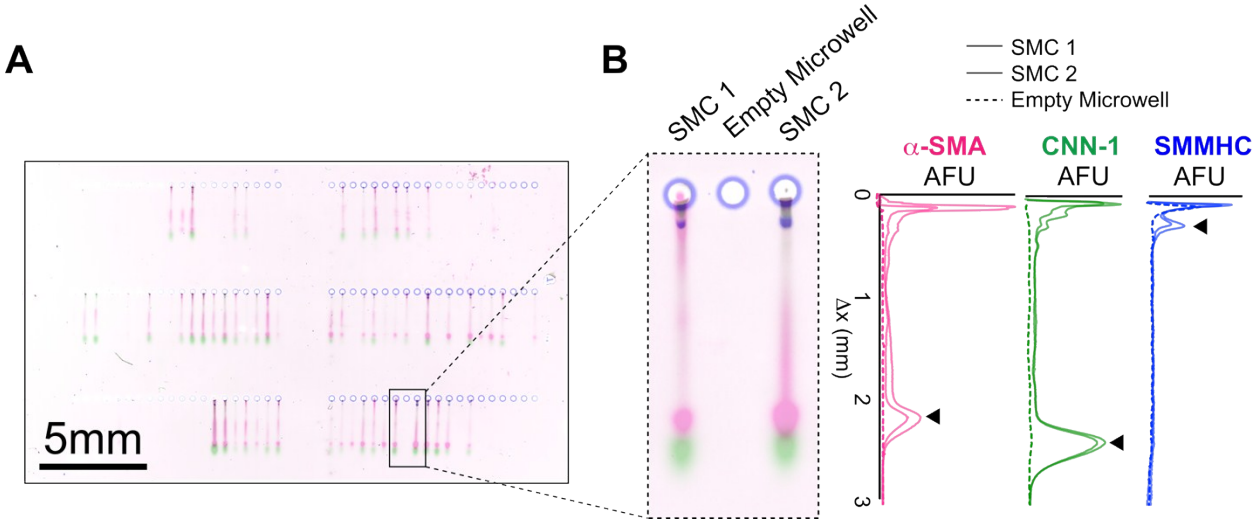

**Figure S3: Experimental determination of SMC lysis.** False-colored micrographs of probed α-SMA after lysis with RIPA-like lysis buffer (left) and lysis buffer optimized for lysis-hardy cells (right). On bottom, fluorescence intensity profiles are shown for representative samples of each. RIPA-like lysis buffer demonstrates high signal at edge of microwell (left micrograph, black arrow), indicating poor solubilization of α-SMA. The buffer optimized for lysis-hardy cells, however, showed a band in separation lane (right, black arrow) and signal at edge of microwell. For both buffers, running conditions were: lysis duration 25 s, electromigration duration 25 s and E = 40 V/cm, 55°C.

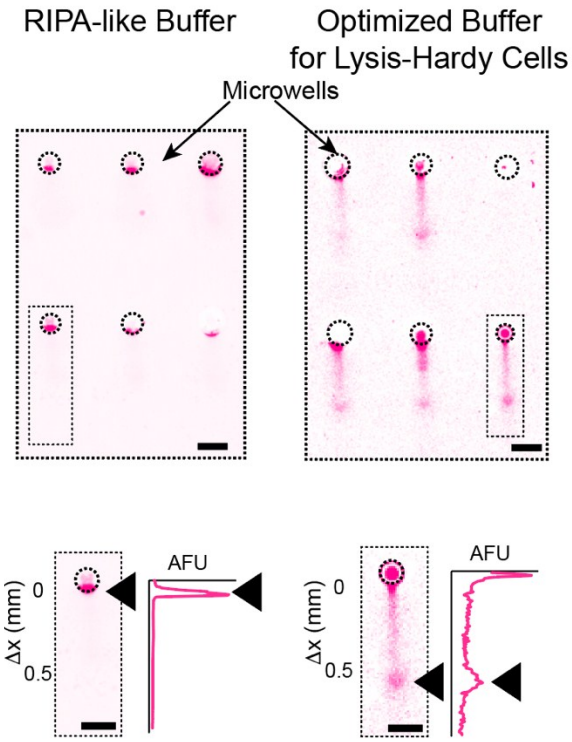

**Figure S4: Hierarchical clustering of individual SMCs by expression of α-SMA, CNN-1 and SMMHC identifies subpopulations of SMCs.** Heat map of hierarchical clustering of SMCs by expression of α-SMA, CNN-1 and SMMHC, where cells are plotted on the y-axis and protein markers are plotted on the x-axis (cosine similarity, N = 45 single SMCs). The majority (96 %) of SMCs are clustered into two major populations, one displaying generally high (*) and the other generally low (**) expression of all three markers. Inside the latter group, a subpopulation (4%) of SMCs show high α-SMA and low CNN-1 and SMMHC expression (red rectangle, ***), corresponding with the phenotype of immature-like SMCs.