

Supplementary Figures

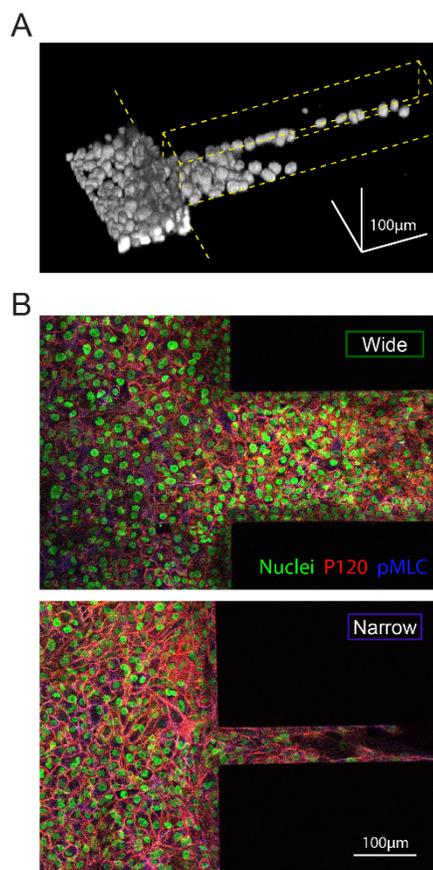


Figure. S1: Representative image of wild type MCF10A cell monolayer entering channels. (A) 3D reconstruction of cell nuclei inside and outside the channels. Yellow dash lines indicate the PDMS channel. **(B)** Immunofluorescence image of nuclei (green), P120 (red), and pMLC (blue) in wide and narrow channels at 24h.

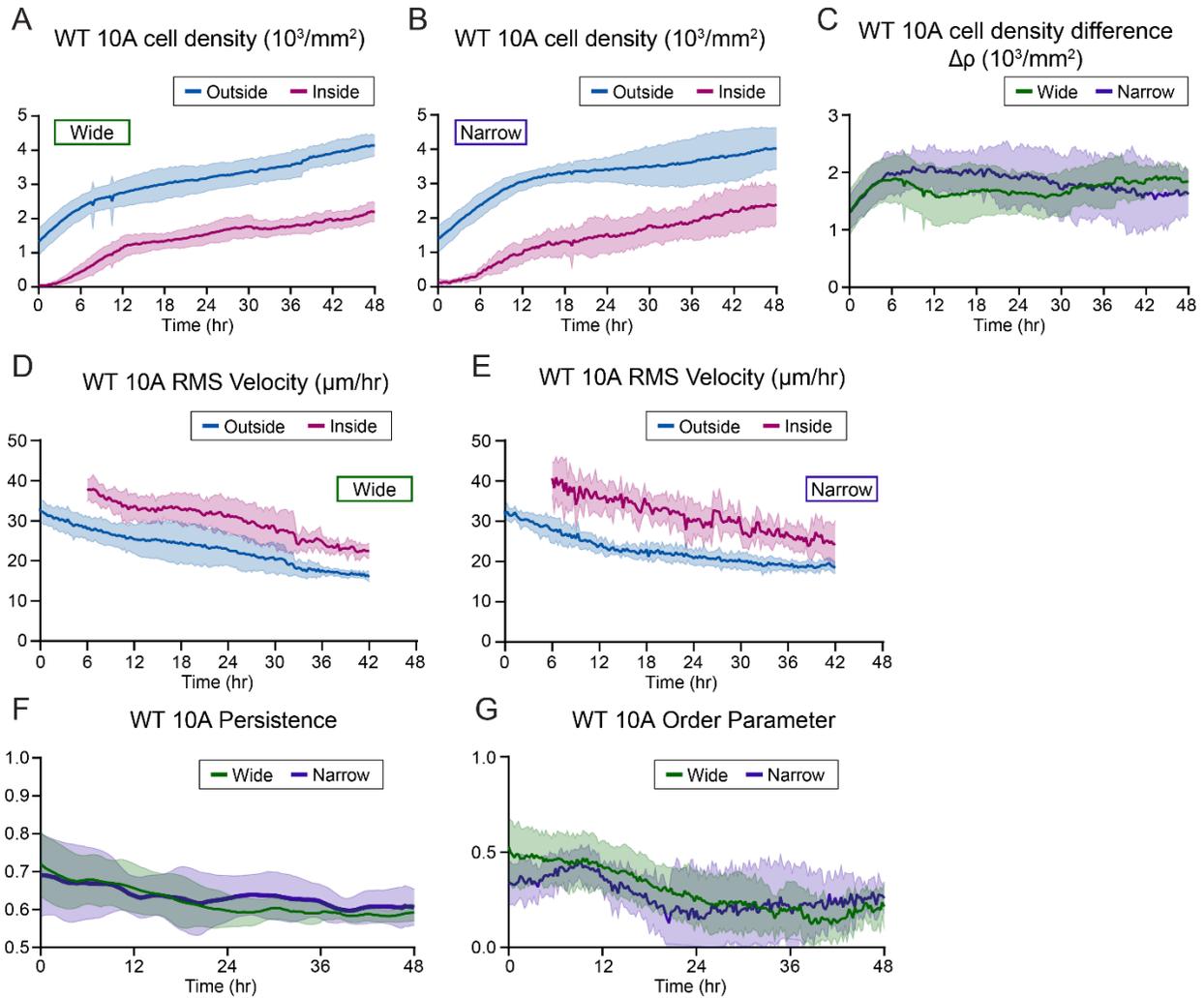


Figure. S2: Cell density and migration across confinement vary with cell types over 48 hours. (A, B) Cell density curves of wild type (WT) monolayers entering wide and narrow microchannels over 48 hours, where blue and magenta curves represent average cell density outside and inside the microchannels, respectively. (C) Temporal variation of difference in cell density ($\Delta\rho$) between inside and outside the microchannels for WT 10A cells. (D, E) Root-mean-squared (RMS) velocities of WT 10A cells outside (blue) and inside (magenta) channels. (F, G) Curves of persistence and order parameter within the monolayers in WT 10A cells. Data expressed as mean \pm SD and $n \geq 6$.

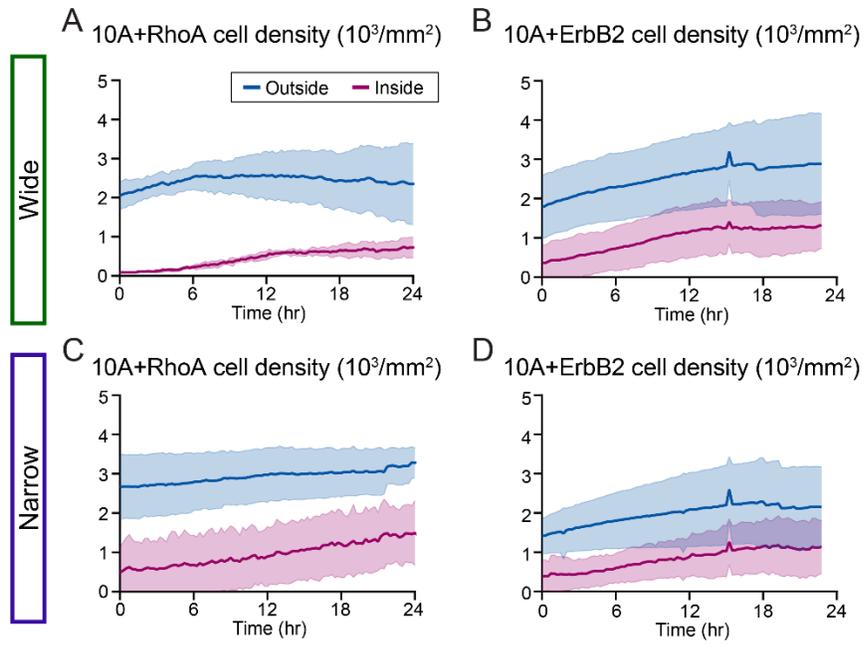


Figure. S3: Root-mean-squared (RMS) velocities of 10A+RhoA. (A, C), and 10A+ErbB2 (B, D) outside (blue) and inside (magenta) channels.

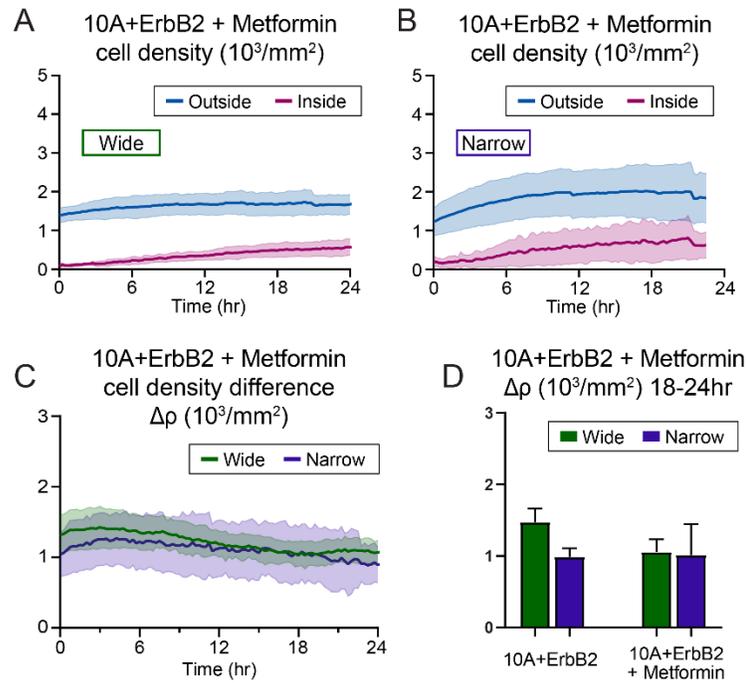


Figure. S4: Cell density of metformin treated +ErbB2 cells. (A, B) Cell density curves of metformin treated +ErbB2 monolayers entering wide and narrow microchannels over 24 hours, where blue and magenta curves represent average cell density outside and inside the microchannels, respectively. (C) Temporal variation of difference in cell density ($\Delta\rho$) between inside and outside the microchannels for metformin treated +ErbB2 cells. (D) Cell density difference averaged from 18-24 hour. Data expressed as mean \pm SD and $n \geq 6$.

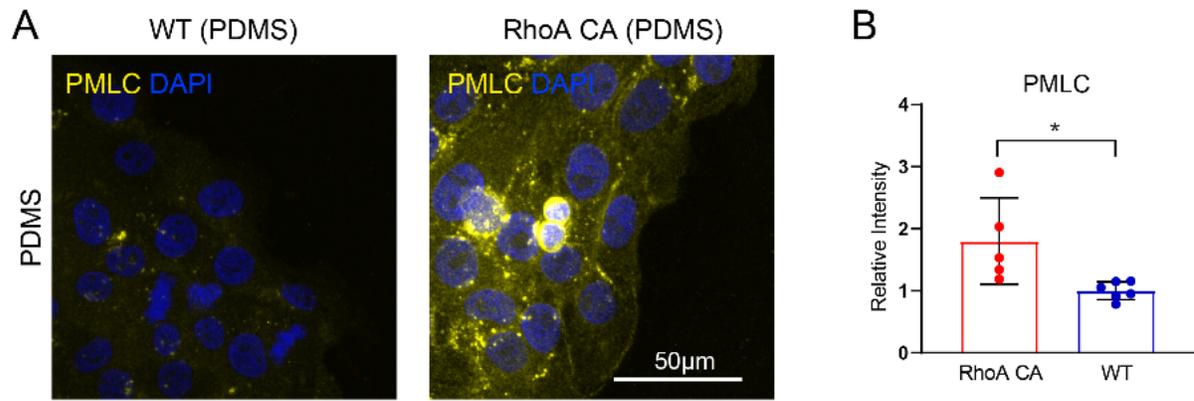


Figure. S5. Immunofluorescence image of pMLC for WT 10A and 10A+RhoA cell on PDMS substrates.