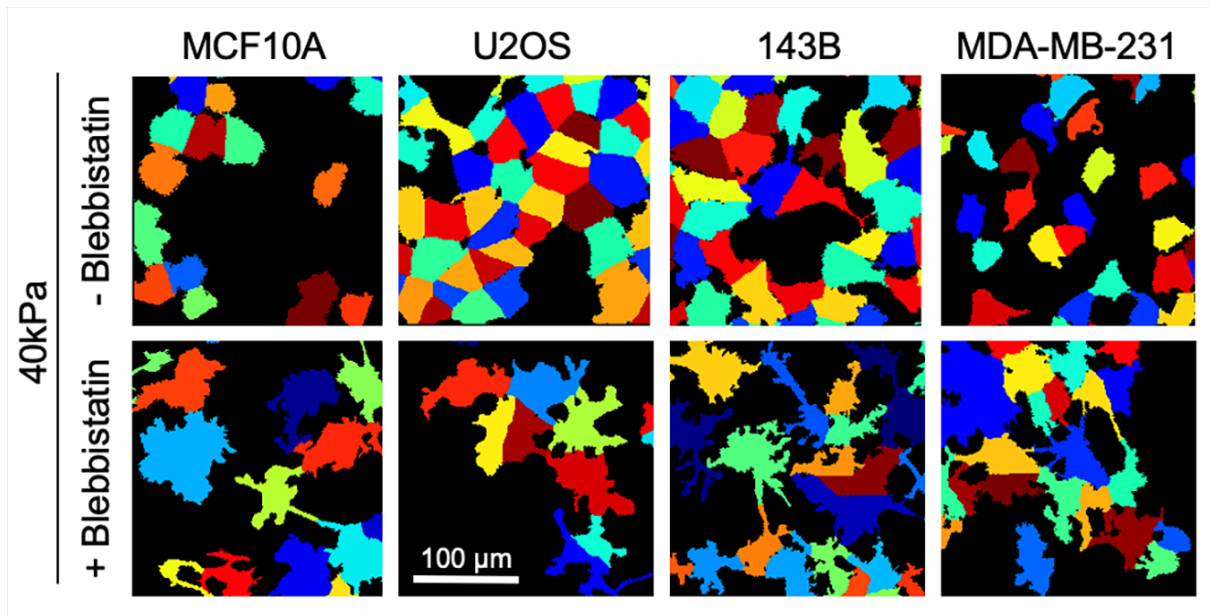


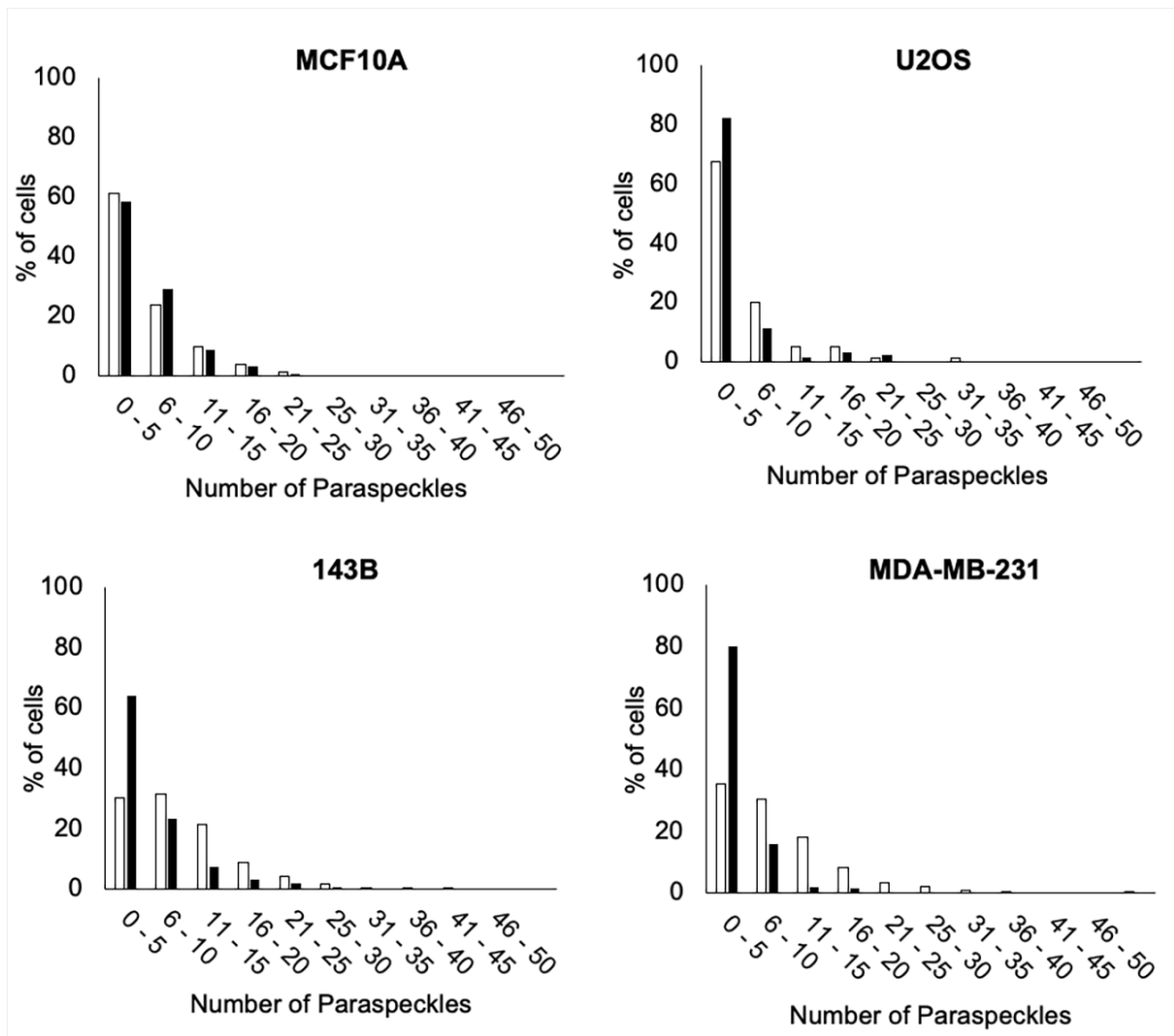
Supplemental Materials

Molecular Biology of the Cell

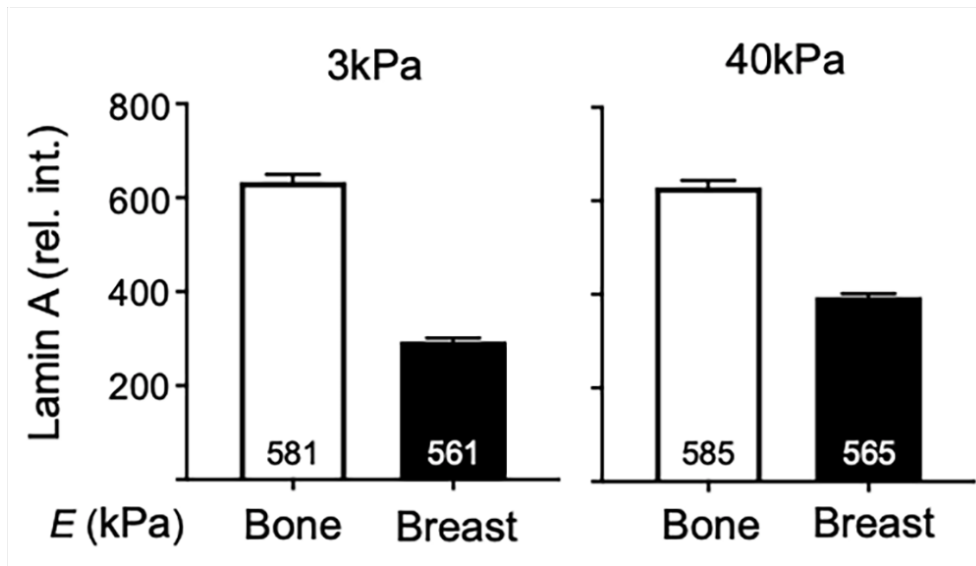
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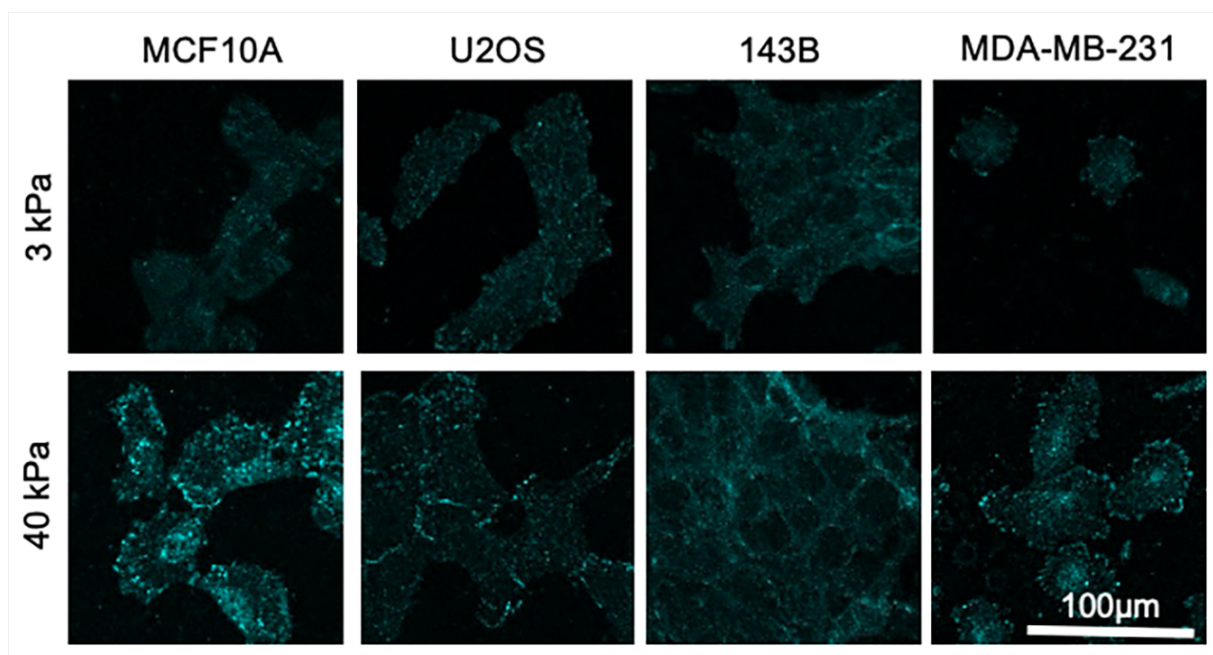
Supplementary Figure 1. Representative images of cells showing differences in size and morphology on blebbistatin treatment compared to control. Images were taken at 20x magnification and visualized in multicolour images by CellProfiler using F-actin for cell boundary recognition. Scale bar = 100μm.



Supplementary Figure 2. Distribution of the number of cells with paraspeckles in MCF10A, U2OS, 143B and MDA-MB-231 cells cultured on 3 kPa and 40 kPa hydrogels grouped in bins of how many paraspeckles were per cell. The distribution of paraspeckles in the breast epithelial MCF10A cell line did not change pending on the stiffness cells were cultured on however paraspeckles in the U2OS, 143B and MDA-MB-231 cells appeared more heterogeneous in distribution when cultured on 3 kPa hydrogels compared to 40 kPa hydrogels.



Supplementary Figure 3. Lamin A intensity in cells of bone origin (143B and U2OS) was collectively greater than in cells of breast (MCF10A and MDA-MB-231) origin. Data was presented for cells cultured on 3 kPa and 40 kPa hydrogels.



Supplementary Figure 4. Immunofluorescence staining of Vinculin in MCF10A, U2OS, 143B and MDA-MB-231 cultured on 3 kPa and 40 kPa hydrogels. Focal adhesions located at the edges of individual cells appeared more prominent in cells culture on 40 kPa hydrogels compared to 3 kPa hydrogels. Images were taken using confocal microscopy at 20x magnification. Scale bar = 100 μ m.