Electronic Supplementary Information

Influence of Microenvironment Topography and Stiffness on Mechanics and Motility of Normal and Cancer Renal Cells

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Table 1 Young moduli of polyacrylamide (PA) gels presented in Figure 1A. Values reported are the median of the Young's modulus from all force curves in Pascal. The width of the distribution is quantified by given the distance to the 75^{th} (+) and 25^{th} percentiles (-).

PA gels	**E $+75^{th}$ -25^{th}**
3 kPa	3009 +346 -587
17 kPa	16527 +4452 -3258
31 kPa	31357 +9291 -6367

Table 2 Young moduli of normal and cancer renal cells on different stiffness materials (3, 17 and 31 kPa PA gels and Petri dish), presented in figure 2A of the main text. Values reported are the median of the Young's modulus from all force curves in Pascal. The width of the distribution is quantified by given the distance to the 75^{th} (+) and 25^{th} percentiles (-).

		Substrate			
		3 kPa gel	**17 kPa gel**	**31 kPa gel**	**Petri dish**
Cell Type	**ACHN**	1947 +455 -786	1474 +1027 -745	1030 +867 -450	1177 +833 -508
	A498	1233 +674 -761	1276 +936 -606	1011 +709 -405	906 +604 -342
	RC142	1225 +545 -810	1569 +750 -494	2094 +853 -561	2209 +1097 -832

Table 3 Young moduli of normal and cancer renal cells on different structured materials (Petri dish, flat and patterned PDMS) as reported in figure 4A of the main text. Values reported are the median of the Young's modulus from all force curves in Pascal. The width of the distribution is quantified by given the distance to the 75^{th} (+) and 25^{th} percentiles (-).

		Substrate		
		Petri dish	**Flat PDMS**	**Patterned PDMS**
Cell Type	**ACHN**	1839 +934 -714	1160 +754 -424	1647+1116 -841
	A498	1040 +765 -344	853 +583 -429	740 +689 -326
	RC142	1960 +1247 -743	1993 +1453 -813	2480 +1280 -974

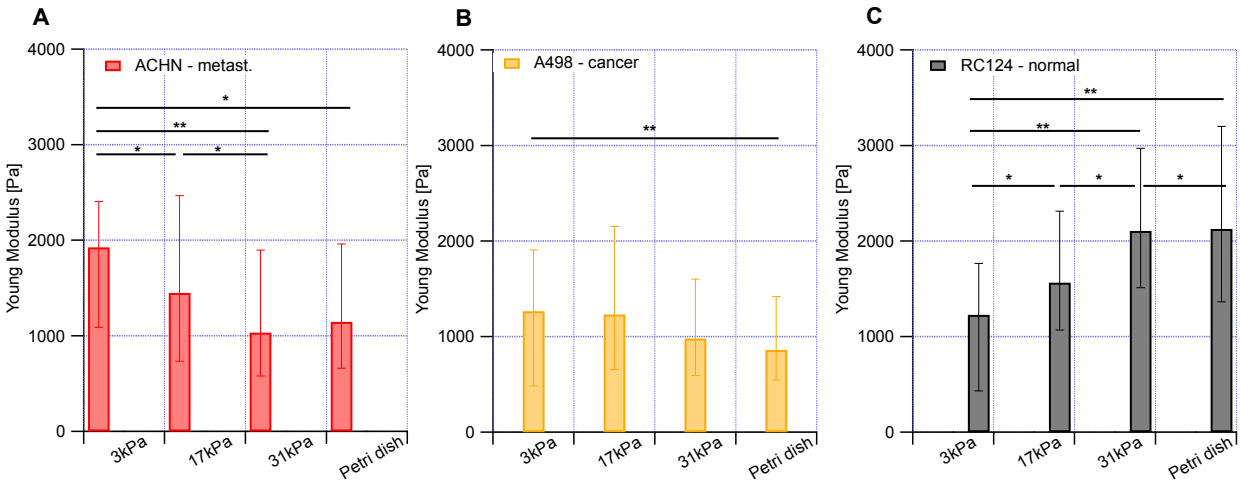

Figure S1. Statistical differences between Young modulus values calculated among the same cell type on different substrates using a Mann-Whitney (or Wilcoxon) test. (A-C) represent Young modulus values of metastatic, cancer and normal renal cells on different stiffness gels (3, 17 and 31 kPa) and conventional Petri dishes (*$p<0.05$, **$p<0.005$). The bars correspond to the median and the 25th and 75th percentile to depict the width of the distribution.

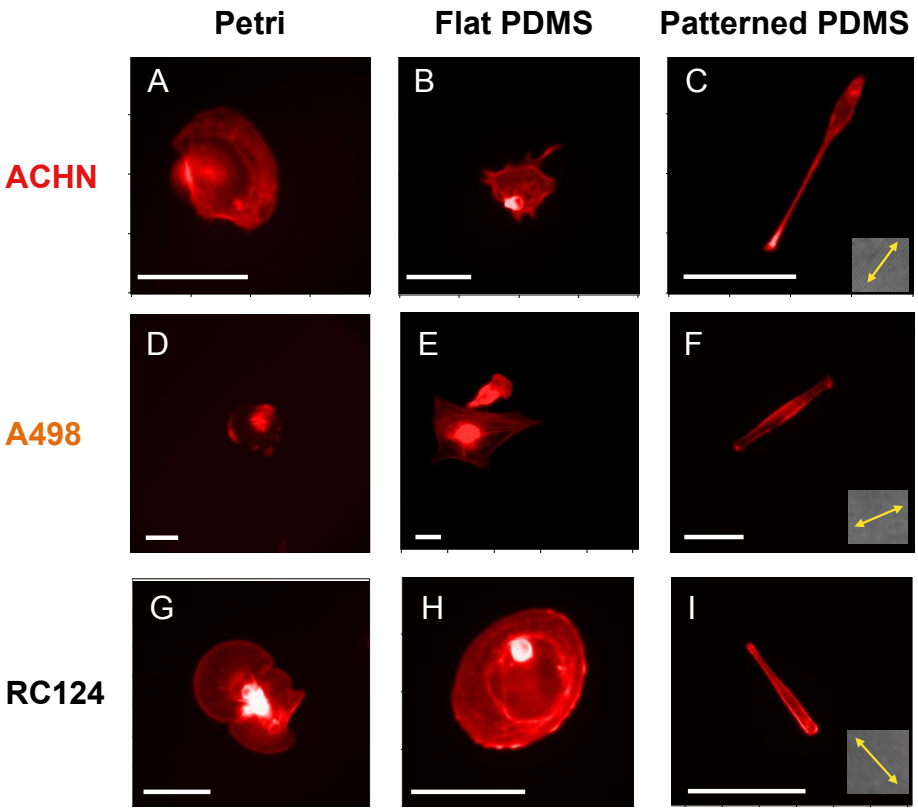

Figure S2. Fluorescent images of normal and cancer renal cells on substrates with different topography. Cells were fixed and labeled with rhodamine phalloidin to visualize actin cytoskeleton. From top to the bottom, panels show metastatic (A-C), cancer (D-F) and normal cells (G-I), seeded on Petri dish, flat and patterned PDMS, respectively. Transmission images of the patterned substrates are shown on the bottom right corner of panels C, F and I, indicating the direction of the pattern with a yellow arrow. Scale bars are 100 μm.

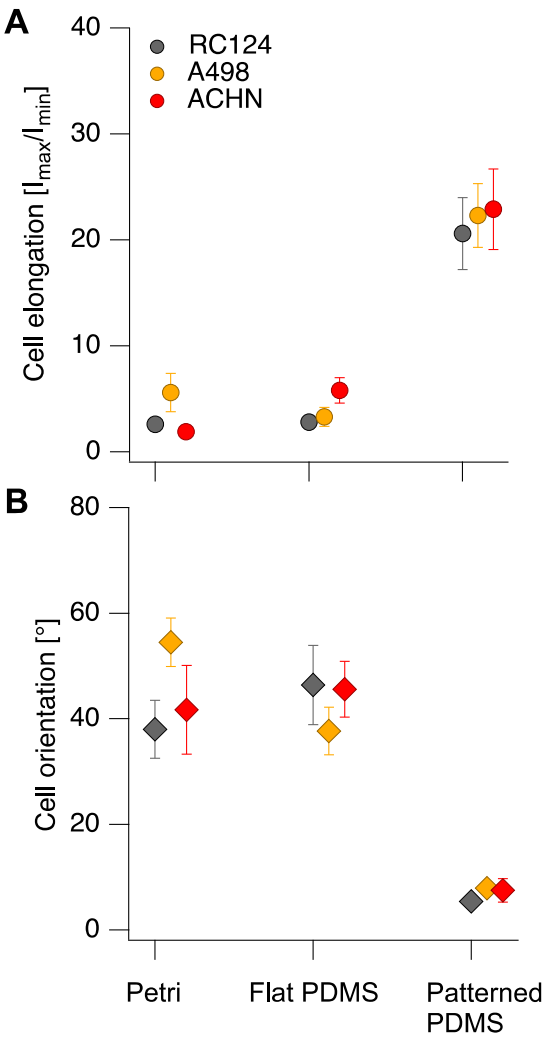

Figure S3. Quantitative analysis of cell elongation index (A) and cell orientation (B) on conventional Petri dishes, flat and patterned PDMS substrates. Black, orange and red refer to normal, cancer and metastatic cells, respectively. Bars refer to standard error of the mean.

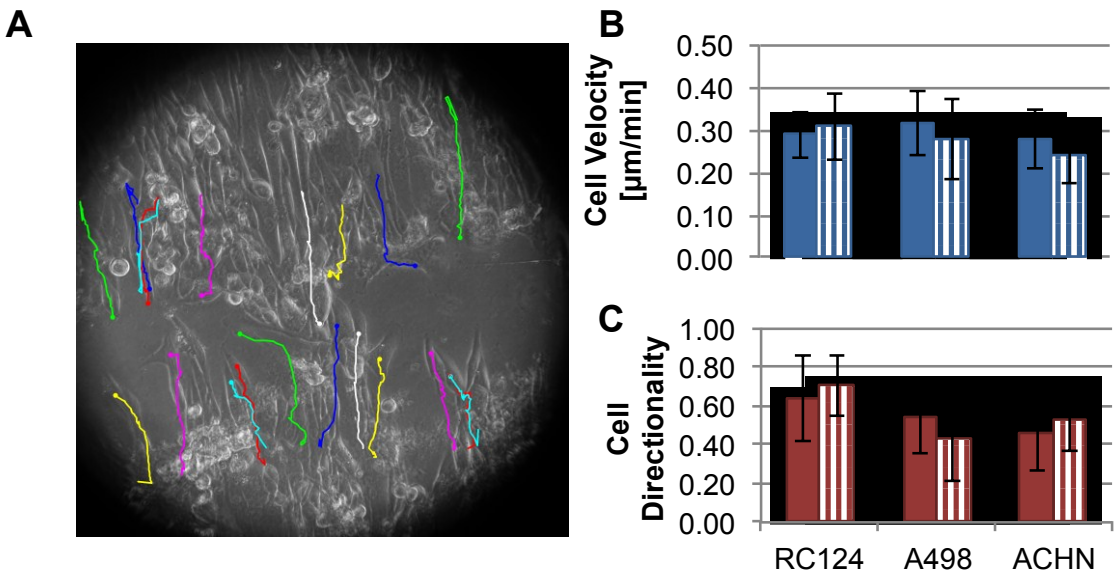

Figure S4. Some of the cell trajectories tracked with manual tool of ImageJ (A). Cell velocity (B) and directionality (C) of RC124, A498 and ACHN cells. Filled and striped bars stand for flat and patterned PDMS, respectively. Error bars are standard deviations

Video1. Wound healing video of Normal cells (RC124) migrating for 24 hours on flat PDMS. Images were taken every two minutes.

Video2. Wound healing video of Normal cells (RC124) migrating for 24 hours on patterned PDMS. Scratch was created orthogonally to the pattern direction. Images were taken every two minutes.

Video3. Wound healing video of Cancer renal cells (A498) migrating for 24 hours on flat PDMS. Images were taken every two minutes.

Video4. Wound healing video of Cancer renal cells (A498) migrating for 24 hours on patterned PDMS. Scratch was created orthogonally to the pattern direction. Images were taken every two minutes.

Video5. Wound healing video of cancer methastatic cells (ACHN) migrating for 24 hours on flat PDMS. Images were taken every two minutes.

Video6. Wound healing video of cancer metastatic cells (ACHN) migrating for 24 hours on patterned PDMS. Scratch was created orthogonally to the pattern direction. Images were taken every two minutes.