Keratin 19 maintains E-cadherin localization at the cell surface and stabilizes cell-cell adhesion

Sarah Alsharif, Pooja Sharma, Karina Bursch, Rachel Milliken, Meagan Collins, Van Lam, Arwa Fallatah, Thuc Phan, Priya Dohlman, Georges Nehmetallah, Christopher B. Raub, and Byung Min Chung

Supplementary Data

Parameters	Full name	parental	*KRT19* KO	p-value
PhaseMean*	Average phase height	3950.0143	3638.2445	0.001
PhaseMean SD	Phase height standard deviation	1965.247	2032.7613	0.229
ku	Phase height kurtosis	2.2347166	2.2688439	0.649
sk*	Phase height skewness	-0.0868383	0.1012174	0
Area (um2)*	Segmented cell area	460.37573	562.81516	0
Perimeter (μm)*	Segmented cell perimeter	95.032022	128.03663	0
Eccentricity*	Segmented cell fit ellipse eccentricity	0.7245655	0.8664618	0
Form factor*	Segmented form factor	0.65458	0.462963	0
Contrast*	2nd order texture parameter	305.3581	329.5333	0.03
Correlation	2nd order texture parameter	0.943321	0.9389649	0.07
Energy	2nd order texture parameter	0.0001539	0.0001515	0.723
Homogenity	2nd order texture parameter	0.1841623	0.1816254	0.364
NuPhaseMx	Maximum nucleus phase height	9107.7722	9529.5549	0.103
NuPhaseAve	Average nucleus phase height	5727.4355	5454.9353	0.062
NuclearArea (um2)*	Segmented nucleus area	185.03516	225.39083	0
NuclearKu*	Phase height kurtosis	0.614367	0.6625169	0.001
NuclearSk*	Phase height skewness	1.6379479	1.8013399	0.05

Table S1. Cell parameters generated from DHM segmentation. Circularity is determined by eccentricity and form factor. A perfect circle has eccentricity equal to 0 and form factor equal to 1. Means from three experimental repeats are shown. Differences are not statistically significant unless denoted by *$p < 0.05$; **$p < 0.001$

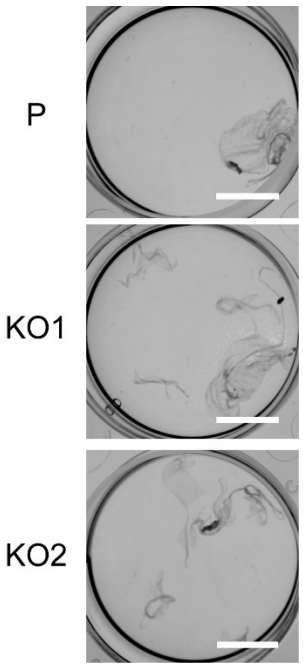
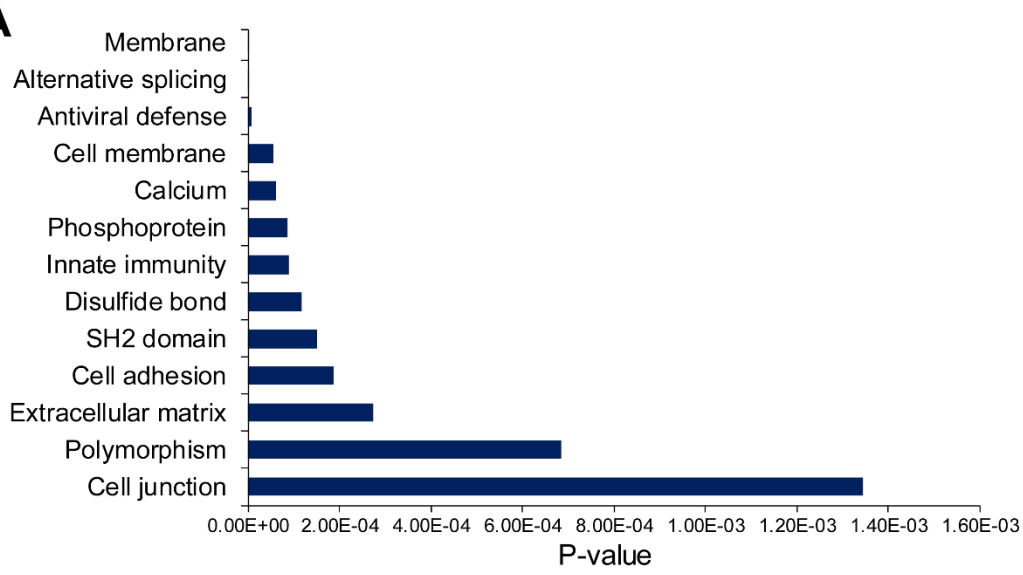


Figure S1. *KRT19* KO cells show weakened cell-cell adhesion. Phase contrast images of dispase treated cell fragments used for Fig 2E. Bar, 10 mm.

A



B

Gene symbol	Gene name	RPKM P	RPKM KO	Log2 (fold change)
ENG	Endoglin	10.5734	4.31752	-1.292165
ICAM1	Intercellular Adhesion Molecule 1	1.60904	0.810933	-0.988546
RND3	Rho Family GTPase 3	4.29229	2.21303	-0.955725
PCDH17	Protocadherin 17	1.80447	0.945641	-0.932211
WISP2	Cellular Communication Network Factor 5	16.1533	9.63394	-0.745631
JUP	Plakoglobin	288.177	177.411	-0.699860
ITGAV	Integrin Subunit Alpha 5	9.5714	12.4971	0.384791
PALLD	Palladin, cytoskeletal associated protein	15.3946	20.5519	0.416847
DSP	Desmoplakin	54.1604	74.638	0.462672
CLDN1	Claudin1	4.56072	6.68239	0.551103
CADM1	Cell Adhesion Molecule 1	3.58198	5.59959	0.644564
CDH3	P-cadherin	26.0338	42.2293	0.697858
ITGA6	Integrin Subunit Alpha 6	1.56782	2.54423	0.698469
FSCN1	Fascin Actin-bundling Protein 1	17.3017	28.4115	0.715561
PODXL	Podocalyxin Like	3.07798	5.14708	0.741770
COL18A1	Collagen Type XVIII Alpha 1 Chain	14.3142	26.2243	0.873457
CLDN9	Claudin9	2.43677	4.46757	0.874520
COL6A1	Collagen Type VI Alpha 1 Chain	4.91318	10.14	1.045329
ADAM9	ADAM Metallopeptidase Domain 9	9.97818	20.7437	1.055825
DNMBP	Dynamin Binding Protein	1.06723	2.31561	1.117521
ATP1B1	ATPase Na+/K+ Transporting Subunit Beta 1	7.76789	17.2509	1.151077
MCAM	Melanoma Cell Adhesion Molecule	1.89175	4.48657	1.245891
CXCL12	C-X-C Motif Chemokine Ligand 12	39.1316	97.7939	1.321410
GPNMB	Glycoprotein Nmb	1.16355	3.15029	1.436951
COL5A1	Collagen Type V Alpha 1 Chain	2.04502	7.10348	1.796411
MSLN	Pre-Pro-Megakaryocyte-Potentiating Factor	0.9121	4.1815	2.196752
TGFB2	Transforming Growth Factor Beta 2	0.43267	3.88347	3.166021

Figure S2. Increased expression of cell adhesion molecules in *KRT19* KO cells. (**A**) A list of top keywords in functional categories associated with genes upregulated in *KRT19* KO from the RNA sequencing data [32] using the DAVID functional annotation software. (**B**) List of major cell adhesion molecules that are differentially regulated in *KRT19* KO cells.

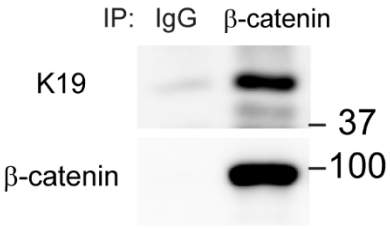


Figure S3. K19 interacts with β-catenin in MCF7 cells. IP with anti-β-catenin antibody or IgG control was performed. IP samples were subjected to SDS-PAGE and immunoblotting was performed with antibodies against K19 and β-catenin.

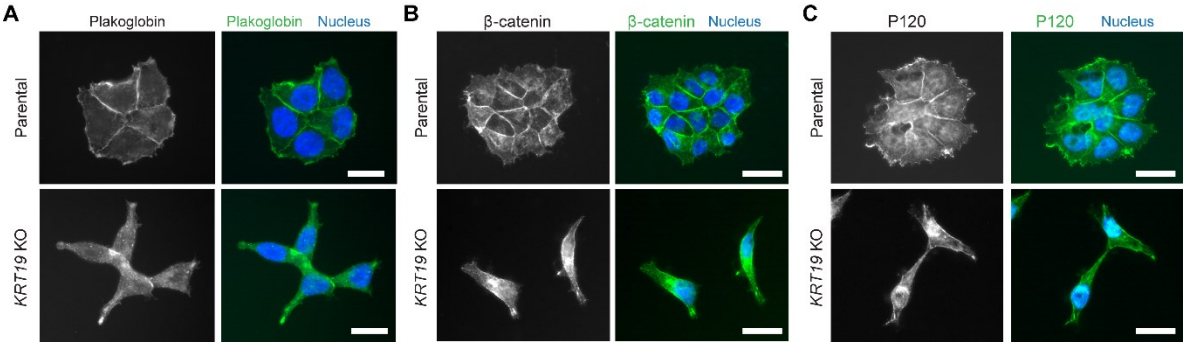


Figure S4. Decreased membrane-bound plakoglobin, β-catenin, and P120-catenin in *KRT19* KO Cells. Immunostaining of (**A**) plakoglobin, (**B**) β-catenin, and (**C**) P120-catenin in parental and *KRT19* KO (KO2) cells. Nuclei are shown in blue. Bar, 20 μm.

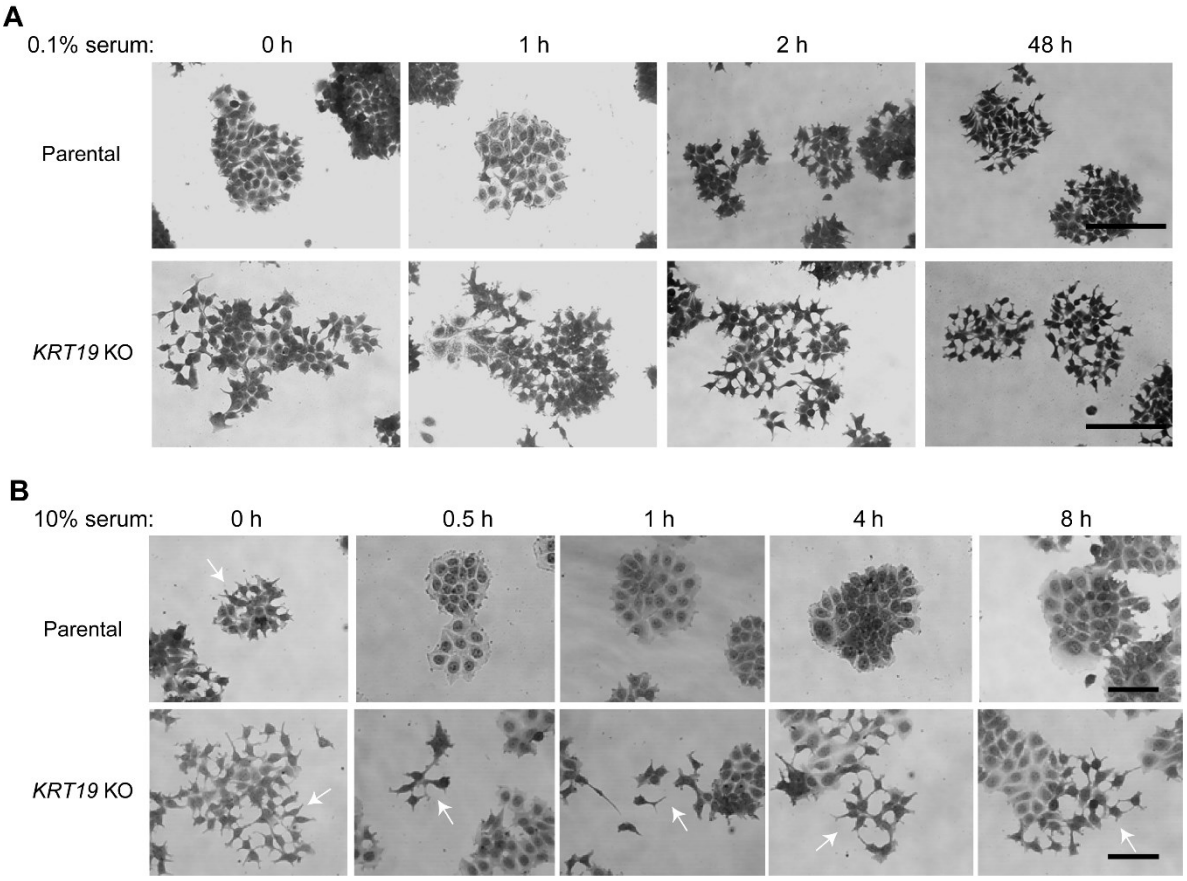


Figure S5. Decreased responsiveness of *KRT19* KO cells for cell-cell adhesion upon serum stimulation. (**A**) Phase contrast images of parental and *KRT19* KO (KO2) cells following serum starvation for indicated time points. Bar, 200 μm. Whereas parental cells show detachments between cells only after 48 h from serum starvation, shape of *KRT19* KO cells remain detached throughout serum starvation. Bar, 200 μm. (**B**) Phase contrast images of parental and *KRT19* KO (KO2) cells serum-starved for 24 h, then stimulated with 10% serum for indicated time points. Parental cells rapidly re-attach from 0.5 h time point but *KRT19* KO cells remain detached throughout serum starvation. Arrows indicate low cell-cell adhesions Bar, 100 μm.