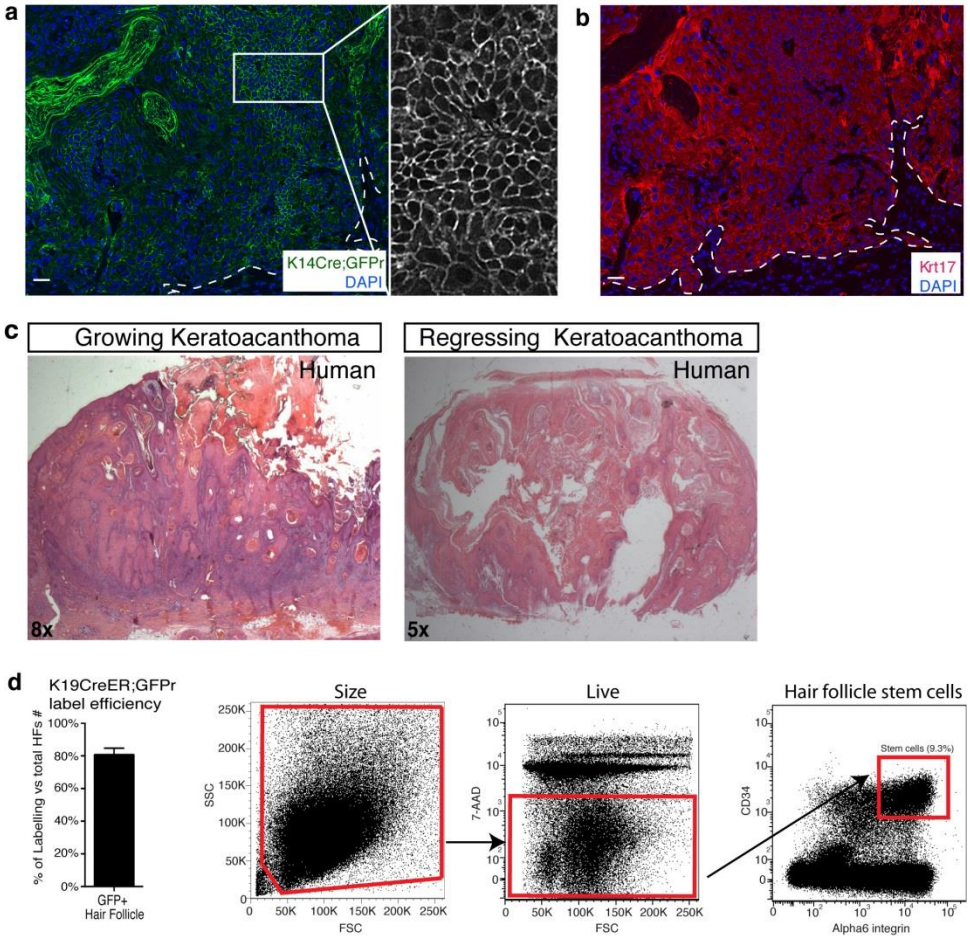
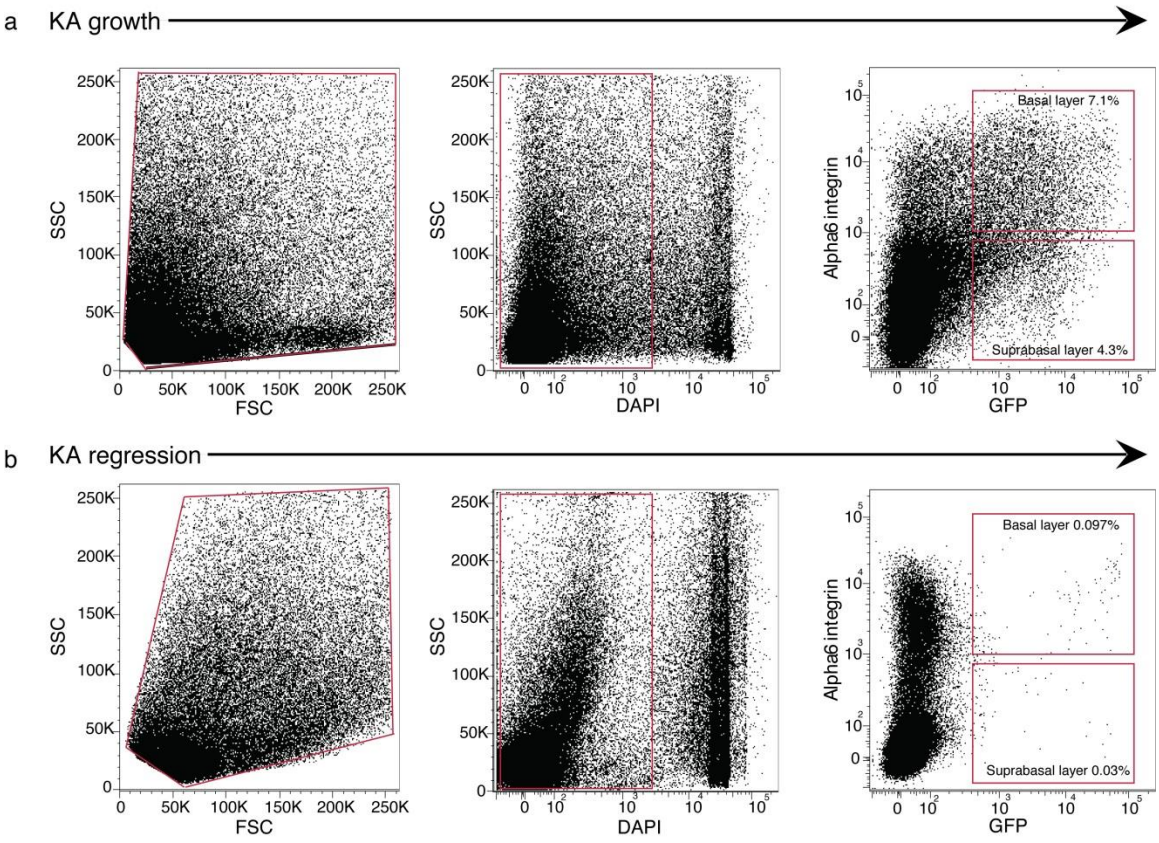
Supplementary Figure1. Characteristics of Keratoacanthoma



(a) *Krt14Cre;Rosa26mTmGFP* mice have been used to address the epithelial origin of KA tumors. Immunostaning for GFP (green) show the epithelial nature of KA tumor. **(b)** Immunofluorescence staining for the epithelial markers Krt17 (scale bar 50µm). Dotted lines delineate the tumor/stroma interface. DAPI (blue) labels all the nuclei.

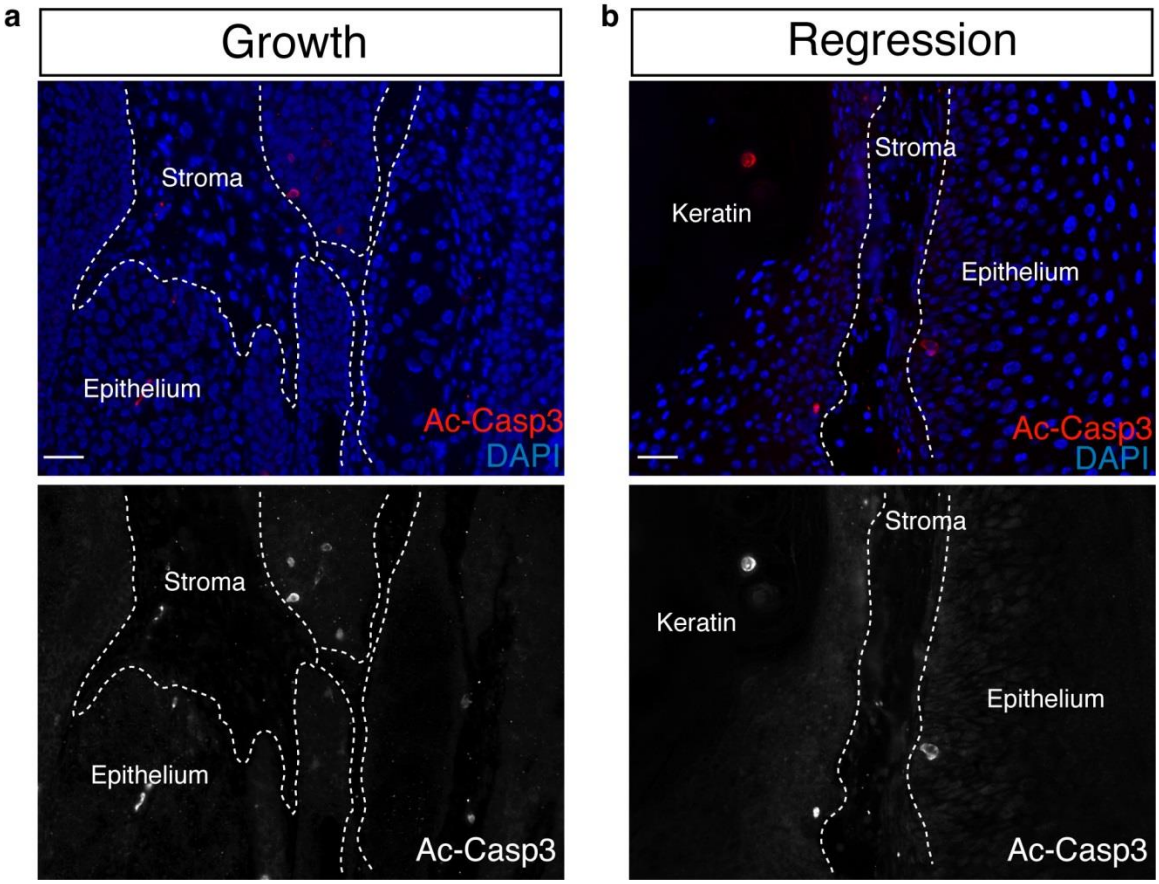
(c) Hematoxylin and eosin stainings show histological characteristics of human KA tumors in growth and regression. **(d-**left panel**)** Quantification of the number of the hair follicles labeled in *Krt19Cre^{ER};Rosa26mTmGFP* mice after 10 days of tamoxifen treatment. Data are represented as mean ± SD (n=3).

Supplementary Figure2. HFSC-derived cells in KAs tumors



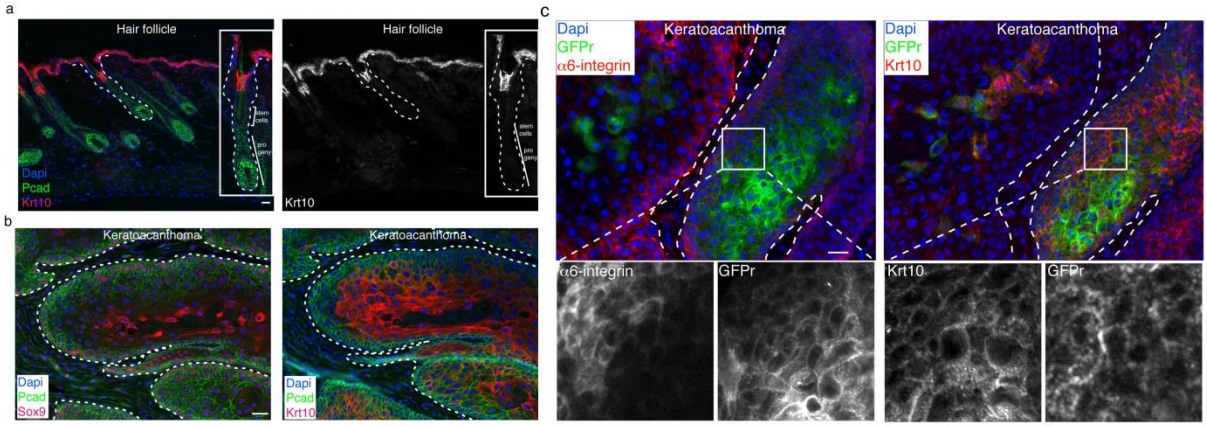
(a) Flow cytometry profile of HFSC- derived cells in growing KAs. **(b)** Flow cytometry profile of HFSC- derived cells in regressing KAs.

Supplementary Figure 3. Apoptosis in KAs tumors



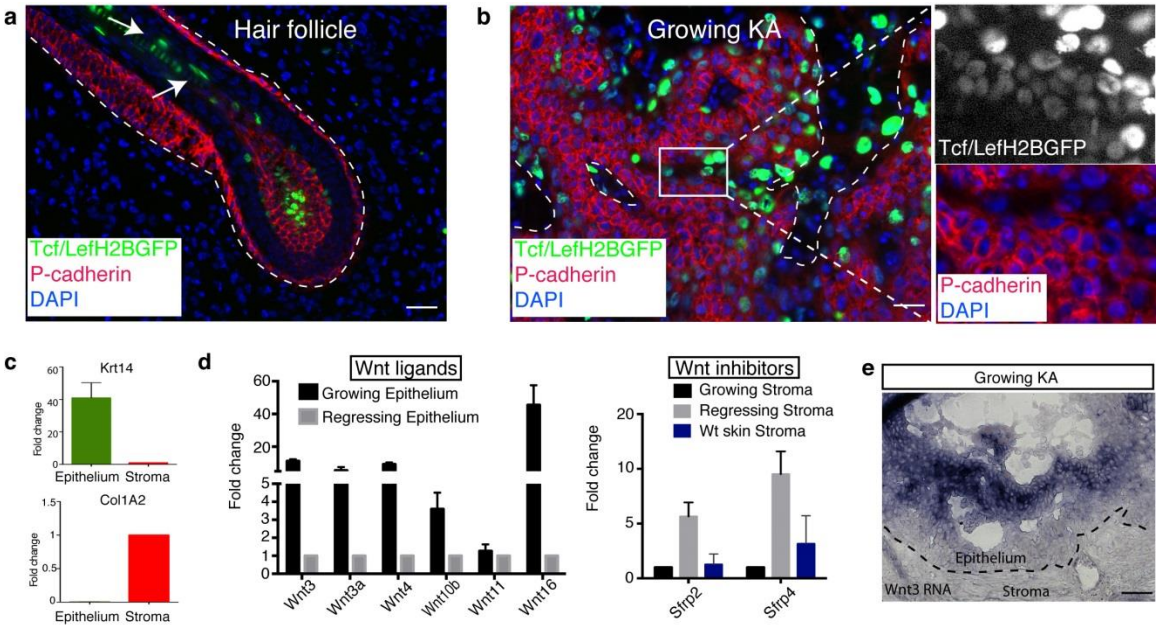
(**a-b**) Immunostaining of KA tumors with Activated caspase3 (red, and grayscale) during tumor growth and regression (scale bar 50μm). DAPI (blue) labels all the nuclei. Dotted lines delineate the tumor/stroma interface.

Supplementary Figure 4. KA tumor expression pattern



(a) Krt10 expression in wt anagen skin. Insets shows Krt10/Pcad co-staining in wt anagen HF. Brackets delineate the stem cells and the expanded progeny. **(b)** Expression pattern of Sox9 and Krt10 (red) in KA tumors. Green cells label the Pcad population (scale bar 50μm). **(c)** Expression pattern of α6-integrin and Krt10 in the HFSC-derived GFP descendants within KA tumors during the growth phase.

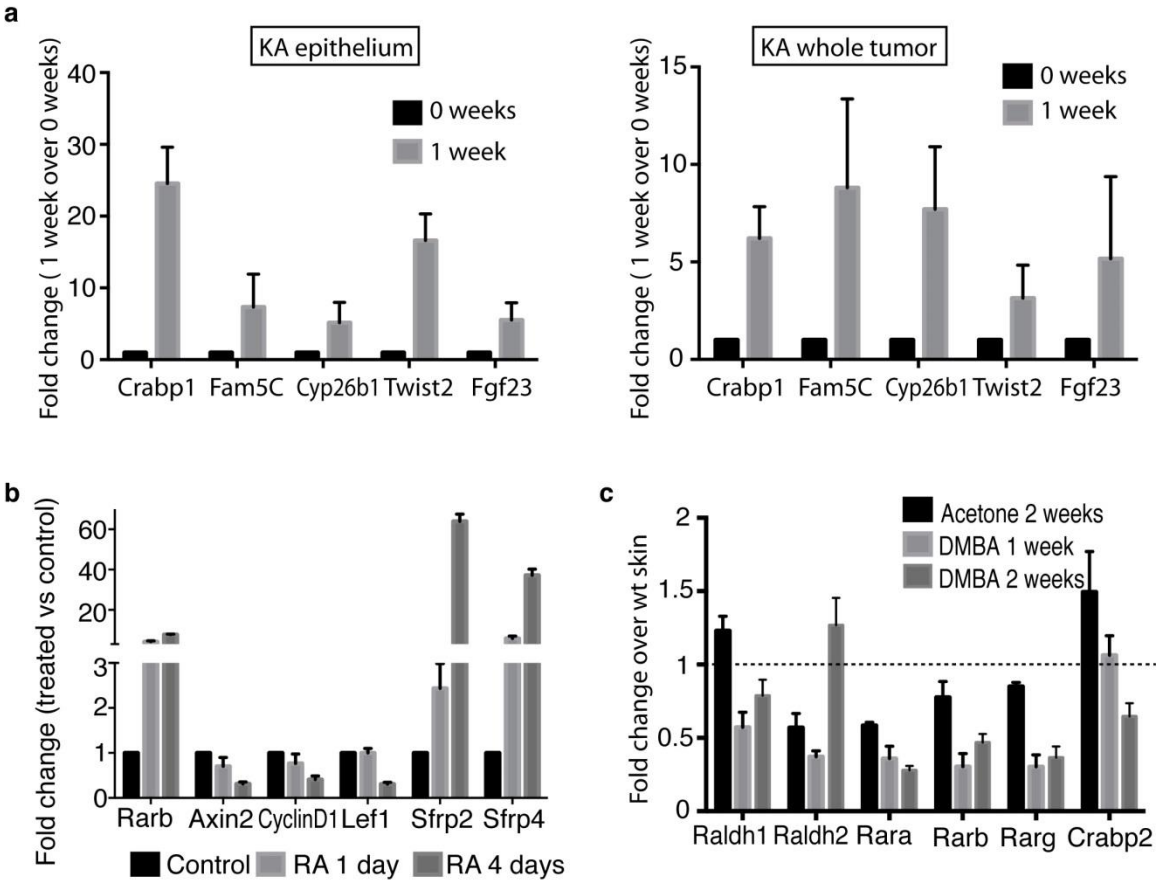
Supplementary Figure 5. Wnt signaling in KA tumors



(a) Hair follicle from *Tcf/LefH2BGFP* reporter mice. Nuclear GFP (green) is localized in the pre-cortex and in the differentiated cells of the hair shaft (white arrows) (scale bar 50μm). Pcadherin (red) outlines the hair follicle. **(b)** Growing KA tumors from *Tcf/LefH2BGFP* reporter mice. Nuclear GFP (green) indicates Wnt activated cells. Pcadherin (red) marks undifferentiated cells (scale bar 50μm).

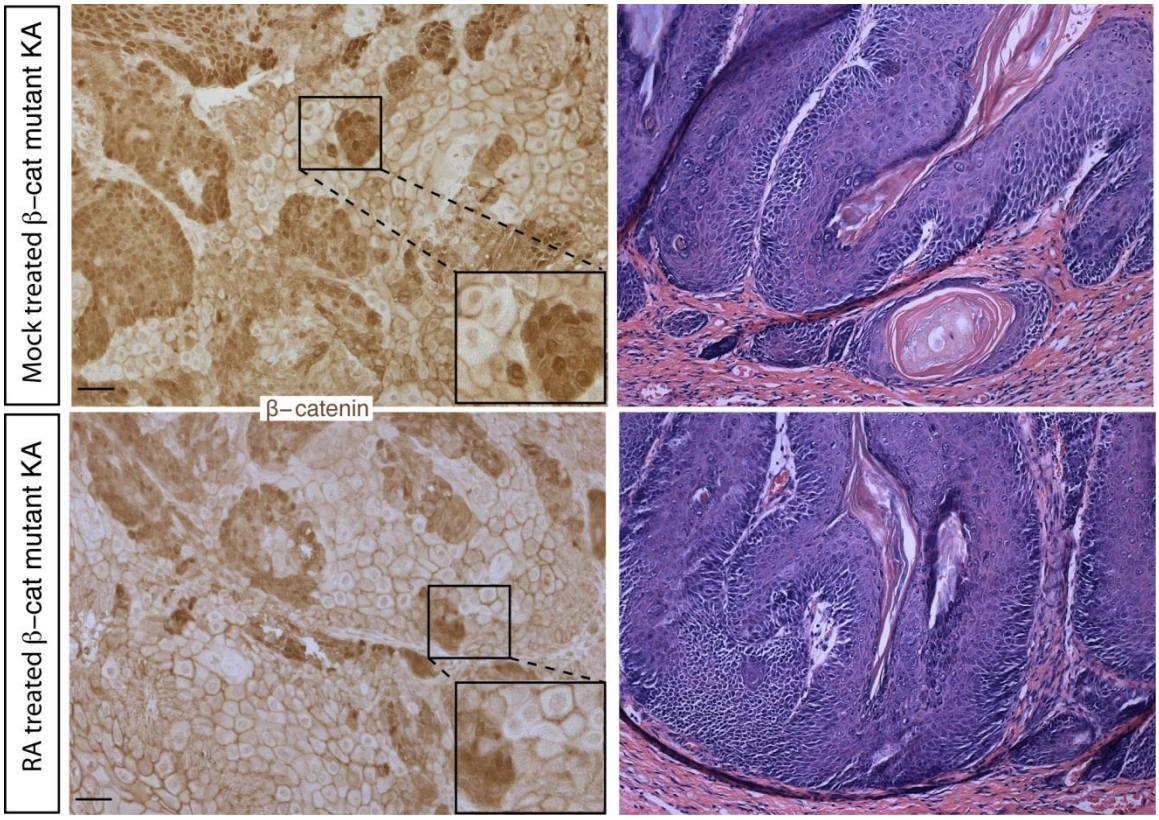
(c) qRT-PCR analysis of epithelial (*Krt14*) and stromal (*Col1A2*) transcripts in FACS sorted cells from growing KA tumors. Data are presented as mean ± SD of three independent experiments (n=3. **(d)** qRT-PCR analysis of Wnt ligands and Wnt inhibitors in epithelial and stromal compartments of KA tumors during growth and regression. Data are presented as mean ± SD of three independent experiments (n=3). **(e)** *In situ* hybridization for Wnt3 in growing KA tumors (n=2) (scale bar 50μm).

Supplementary Figure 6. RNA seq validation and RA treatment



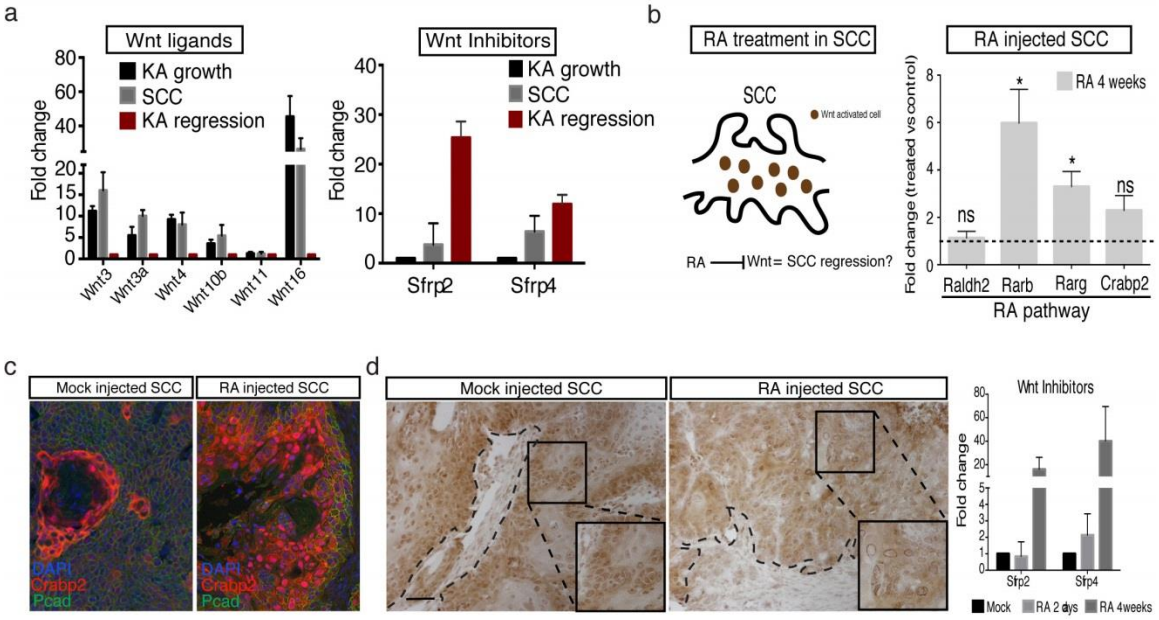
(a) qRT-PCR validation of the genes identified by RNA sequencing in KA epithelial cells (left panel) and whole tumor at 1 week post DMBA treatment. Data are presented as mean ± SD of 3 independent tumors for each timepoint (n=4) **(b)** qRT-PCR of RA treated wild type keratinocytes at 1 and 4 days. Data are presented as mean ± SD of two independent experiments (n=2). **(c)** qRT-PCR analysis of RA signaling pathway in DMBA and acetone treated skin. Data are presented as mean ± SD of two independent experiments (n=2).

Supplementary Figure 7. RA treatment in β-cat activated KAs



Immunohistochemistry for β–catenin and hematoxylin&eosin staining of *K14Cre^{ER}; Rosa26mTmGFP; β-catn$^{flox(Ex3)/+}$* RA or mock treated KA tumors (scale bar 50μm).

Supplementary Figure 8. RA in vivo induces SCC regression



(a) qRT-PCR analysis of Wnt ligands and Wnt inhibitors of growing, regressing KAs and SCC tumors. Data are presented as mean ± SD of three independent experiments (n=6). **(b)** Schematic representation of the potential role of RA to induce SCC regression. qRT-PCR analysis of RA signaling in SCC tumors after RA treatment. Dotted line represents the normalized expression level of each gene analyzed by comparing RA-treated versus mock-treated SCCs. Data are represented as mean ± SD (n=4) (*<0.05, obtained by unpaired t test statistical analysis). **(c)** Crabp2 staining in mock and RA 4 weeks post injection SCCs (scale bar 50μm). **(d)** Immunohistochemistry for β–catenin in RA or mock treated SCC tumors (scale bar 50μm). **(d** right panel**)** qRT-PCR analysis of Wnt inhibitors in RA or mock treated SCC tumors. Data are presented as mean ± SD of three independent experiments (n=6).

Supplementary Table1 Genes differentially by RNA sequencing

Gene Symbol	Log. Fold Change 1 over 0 weeks DMBA	p value
Cox4l2	7.9x	0.000394669
Ccl17	7.3x	0.00448685
Crabp1	6.3x	0.00039136
Vmn2r11	6.1x	0.00460318
Slc19a3	6x	0.00726248
Masp1	5.9x	0.000058299
AU021092	5.9x	0.00069437
Olfr558	5.8x	0.00075273
Alb	5.7x	0.00904593
Wisp2	5.6x	0.000017502
Mmp9	5x	0.000000047
Sfrp2	4.8x	0.000037632
Podnl1	4.7x	0.00012739
Tmem132c	4.6x	0.000065618
Gm600	4.5x	0.00157024
Rspo3	4.5x	0.00065618
Tceal3	4.3x	0.00020289
1300002K09Rik	4.3x	0.00397425
Kcn3	4.2x	0.00649095
Darc	4.1x	0.0007497

Trpc6	4.1x	0.00295936
4833422F24Rik	4.1x	0.00320924
Prl2c5	4.1x	0.00297716
Twist2	4x	0.000088405
Angptl1	4x	0.000027564
Cnn1	4x	0.00177364
Olfr810	4x	0.00539585
Prss35	4x	0.00230389
Mmp10	3.8x	0.0002076
Dbc1	3.8x	0.00028807
Hoxa10	3.8x	0.00153063
F7	3.7x	0.00459826
Sfmbt2	3.7x	0.00021844
BC021767	3.7x	0.00944554
Pdcd1lg2	3.7x	0.00261392
Ppp1r14a	3.7x	0.0068006
Apln	3.6x	0.00704978
Col13a1	3.6x	0.00589896
Kcnip1	3.6x	0.00344606
a	3.5x	0.00784609
Tacr1	3.5x	0.00154033
Prokr2	3.5x	0.00013168
Fam174b	3.5x	0.00083198

Cacna1g	3.5x	0.00023716
Prr16	3.5x	0.00560394
Ddah1	3.5x	0.0002078
Dll4	3.4x	0.00540425
Slc38a11	3.4x	0.00668297
F10	3.4x	0.000271
Fgf23	3.3x	0.0010876
Erg	3.3x	0.00858546
Fam5c	3.3x	0.00403728
Sfrp4	3.2x	0.00413046
Stc1	3.2x	0.00313585
Mirg	3.2x	0.00233529
Ramp3	3.1x	0.00629639
She	3.1x	0.00233529
Pdllm3	3.1x	0.00939842
P4ha3	3.1x	0.00001382
Aqp5	3x	0.00300768
Apcdd1	3x	0.00050177
Slco5a1	3x	0.001208
St6galnac3	3x	0.00591538
Mustn1	3x	0.00555434
Pcdh17	2.9x	0.00689018
Wnt2b	2.9x	0.00154266

Slc6a17	2.9x	0.00563872
Usp13	2.8x	0.00793107
Cap2	2.7x	0.00191436
5430421F17Rik	2.7x	0.00405703
8430408G22Rik	2.7x	0.00253044
Ehd3	2.6x	0.00180346
Adam12	2.6x	0.00030358
Nr5a2	2.6x	0.00595864
Tbx3	2.5x	0.00626062
Rapgef4	2.5x	0.00929844
Fbln2	2.5x	0.0015356
Htr2b	2.5x	0.00359735
Clstn2	2.5x	0.00495792
Il1rn	2.5x	0.00500511
Igfbp3	2.4x	0.00243947
Sema6a	2.4x	0.00379856
Plln4	2.3x	0.00534427
Col12a1	2.3x	0.004257
Bpgm	2.2x	0.00407281
Adamts9	2.1x	0.00447072
9930013L23Rik	2.1x	0.00713945
Sdk1	2x	0.00553886
Ppp1r16b	2x	0.0040297

Cyr61	2x	0.00473784
Fam198b	2x	0.00727547
Nr4a1	2x	0.00742337
Gcnt2	1.9x	0.00768971
Scd1	-1.7x	0.00963149
Serpinb8	-1.9x	0.00555486
Bmp2	-2.1x	0.00421467
Dapk2	-2.2x	0.00848164
Rwdd3	-2.2x	0.00830347
Hddc3	-2.4x	0.0014926
Sertad4	-2.4x	0.00247591
Serpinb6c	-2.7x	0.00249803
Cldn1	-3.7x	0.000021438
Jakmlp2	-4.2x	0.00236162
Gbp1	-5.3x	0.000000014
Rprl3	-8.7x	0.000007224

Supplementary Table 2 List of primers used for qRT-PCR

Primers	Sequence	Primers	Sequence
GAPDH	FW 5'-AACTTTGGCATTGTGGAAGG-3' Rev 5'-ACACATTGGGGGTAGGAACA-3'	**Sfrp4**	FW 5'-ATCCTGGCCATCGAACAGTA-3' Rev 5'-CTCTGGCCAGCTGTGGTTAT-3'
Krt14	FW 5'-TTGAGAGCCTCAAGGAGGAG-3' Rev 5'-GTACTGATCCCGCATCTCGT-3'	**Dkk1**	FW 5'-TGGCCGTGTTTACAATGATG-3' Rev 5'-TACTTGTTCCCGCCCTCATA-3'
Krt10	FW 5'-ACGAAGAGCTGGCCTACCTA-3' Rev 5'-GCGTTCATTTCCACATTCAC-3'	**Dkk2**	FW 5'-CTGGTACCCGCTGCAATAAT-3' Rev 5'-CTTGGAGTGTGGCCTTCCTA-3'
Loricrin	FW 5'-CCTGGTGCTTCAGGGTAAC-3' Rev 5'-TCCTCCACCAGAGGTCTTTC-3'	**Dkk3**	FW 5'-TGTGACAACCAGAGGGATTG-3' Rev 5'-GCTCCAGTTCCCAGGTGAT-3'
Filaggrin	FW 5'-TTCATCAATGGGTACAAGCG-3' Rev 5'-TCAGCAAGTGGGAAGGTGTA-3'	**Wnt3**	FW 5'-CCATCTTTGGGCCTGTCTT-3' Rev 5'-ACTTCCAGCCTTCTCCAGGT-3'
Involucrin	FW 5'-CCTCCTGTGAGTTTGTTTGG-3' Rev 5'-ACCACAGCTGGAACAGTCAC-3'	**Wnt3a**	FW 5'-TTTGGAGGAATGGTCTCTCG-3' Rev 5'-ACCACCAGCAGGTCTTCACT-3'
Ki67	FW 5'-GAGGCTCCATCTGCAGAGAC-3' Rev 5'-CAGGTAGGCCAGAGCAAGTC-3'	**Wnt4**	FW 5'-ACTGGACTCCCTCCCTGTCT-3' Rev 5'-TCACAGCCACACTTCTCCAG-3'
CyclinD1	FW 5'-AAGTGCGTGCAGAAGGAGAT-3' Rev 5'-TTAGAGGCCACGAACATGC-3'	**Wnt10b**	FW 5'-TTCTCTCGGGATTTCTTGGA-3' Rev 5'-CACTTCCGCTTCAGGTTTTC-3'
Sox9	FW 5'-AGAGGCCACGGAACAGACT-3' Rev 5'-CCCTCTCGCTTCAGATCAAC-3'	**Wnt11**	FW 5'-ACCTGCTTGACCTGGAGAGA-3' Rev 5'-AGCCCGTAGCTGAGGTTGT-3'
Axin2	FW 5'- ACTGGGTCGCTTCTCTTGAA-3' Rev 5'-CTCCCCACCTTGAATGAAGA-3'	**Wnt16**	FW 5'-TGATGTCCAGTACGGCATGT-3' Rev 5'-CAGGTTTTCACAGCACAGGA-3'
Lef-1	FW 5'-TGAAGCCTCAACACGAACAG-3' Rev 5'-GCCCAGGATCTGGTTGATAG-3'	**Rara**	FW 5'-AGAGCAGCAGTTCCGAAGAG-3' Rev 5'-CGGAAGAAGCCCTTACAGC-3'
c-Myc	FW 5'-AGCCCCTAGTGCTGCATGA-3' Rev 5'-TCCACAGACACCACATCAATTTC-3'	**Rarb**	FW 5'-ACAAGTCATCGGGCTACCAC-3' Rev 5'-CTCTTTGGACATGCCCACTT-3'
CD44	FW 5'-TCTGCCATCTAGCACTAAGAGC-3' Rev 5'- GTCTGGGTATTGAAAGGTGTAGC-3'	**Raldh1**	FW 5'-CACCATGGATGCTTCAGAGA-3' Rev 5'-ACTTTCCCACCATTGAGTGC-3'
Claud-1	FW 5'-CGAGCCTTGATGGTAATTGG-3' Rev 5- CCACTAATGTCGCCAGACCT-3'	**Raldh2**	FW 5'-AGCCACAGGAGAGCAAGTGT-3' Rev 5'-TGCAAGCTTGTCCAACAGAC-3'
Wif1	FW 5'- AATTCAGGCCGGCGTTCTAAAG-3' Rev 5'-CTGCCGAAATGGAGGTAAATGC-3'	**Raldh3**	FW 5'-ACTGGCACGAATCCAAGAGT-3' Rev 5'-CTTGTCCACATCGGGCTTAT-3'
Sfrp1	FW 5'-CATGCAGTTCTTCGGCTTC-3' Rev 5'- TCGCTTGCACAGAGATGTTC-3'	**p21**	FW 5'-GTACTTCCTCTGCCCTGCTG-3' Rev 5'-TGCGCTTGGAGTGATAGAAA-3'
Sfrp2	Fw5'-AGCCTGCAAAACCAAGAATG-3' Rev5'-CACTGCAGGCTGTCTTTGAG-3'	**p27**	FW 5'-TTGGGTCTCAGGCAAACTCT-3' Rev 5'-TCTGTTCTGTTGGCCCTTTT-3'
Sfrp3	Fw5'-CGAGCCCATTCTCATCAAGT-3' Rev5'-CCCCTCTGCAGTGTCCAG-3'	**Crabp2**	FW 5'-CCAGCAGTCGAGATCAAACA-3' Rev 5'-TTCCACTCTCCCATTTCACC-3'
Wls	Fw5'- TGTTGGAGGGATTCTTCTGG-3' Rev5'- CGCCAGCCATCTTGTTTTAT-3'	**Cyp26b1**	Fw5'- CAAGATCCTACTGGGCGAAC-3' Rev5'- GGGCAGGTAGCTCTCAAGTG-3'
Fam5C	Fw5'- AGCTGGTGCTGAACTGTCCT-3' Rev5'-TGTACTCCTGTGAGCGATGG-3'	**Crabp1**	FW5'- CTTCGAGGAGGAGACAGTGG-3 Rev5'- CAGCTCTCGGGTCCAGTAAG-3'
Twist2	Fw5'- ACCAGTGAGGAAGAGCTGGA-3' Rev5'- CTGCAGCTCCTCGAAAGACT-3'	**Fgf23**	FW5'- TGCTAGGGACCTGCCTTAGA-3' Rev5'- GTACAGGTGGGTCAGGCTTC-3'

Supplementary Table 3: Primary antibodies used for FACS, IF and IHC

Antibody	Company	Catalog number	Species	Dilution
Ki67	Leica	NCL-Ki67p	Rabbit	1:200
Sox9	Millipore	AB 5535	Rabbit	1:500
Krt10	Covance	PRB-159P	Rabbit	1:1000
P-cadherin	R&D	AF761	Goat	1:100
GFP	Abcam	ab13970	Chicken	1:1000
Krt17	Abcam	ab53707	Rabbit	1:1000
β-catenin	BD	610153	Mouse	1:100
α6-integrin	eBiosciences	13-0495-80	Rat	1:1000
Ac-Caspase3	Cell Signaling	9664	Rabbit	1:200
Wntless	Seven Hills	WLAB-177	Rabbit	1:500
Crabp2	Proteintech	10225-1-AP	Rabbit	1:50
CD34-A660	eBiosciences	50-0341-82	Rat	1:75