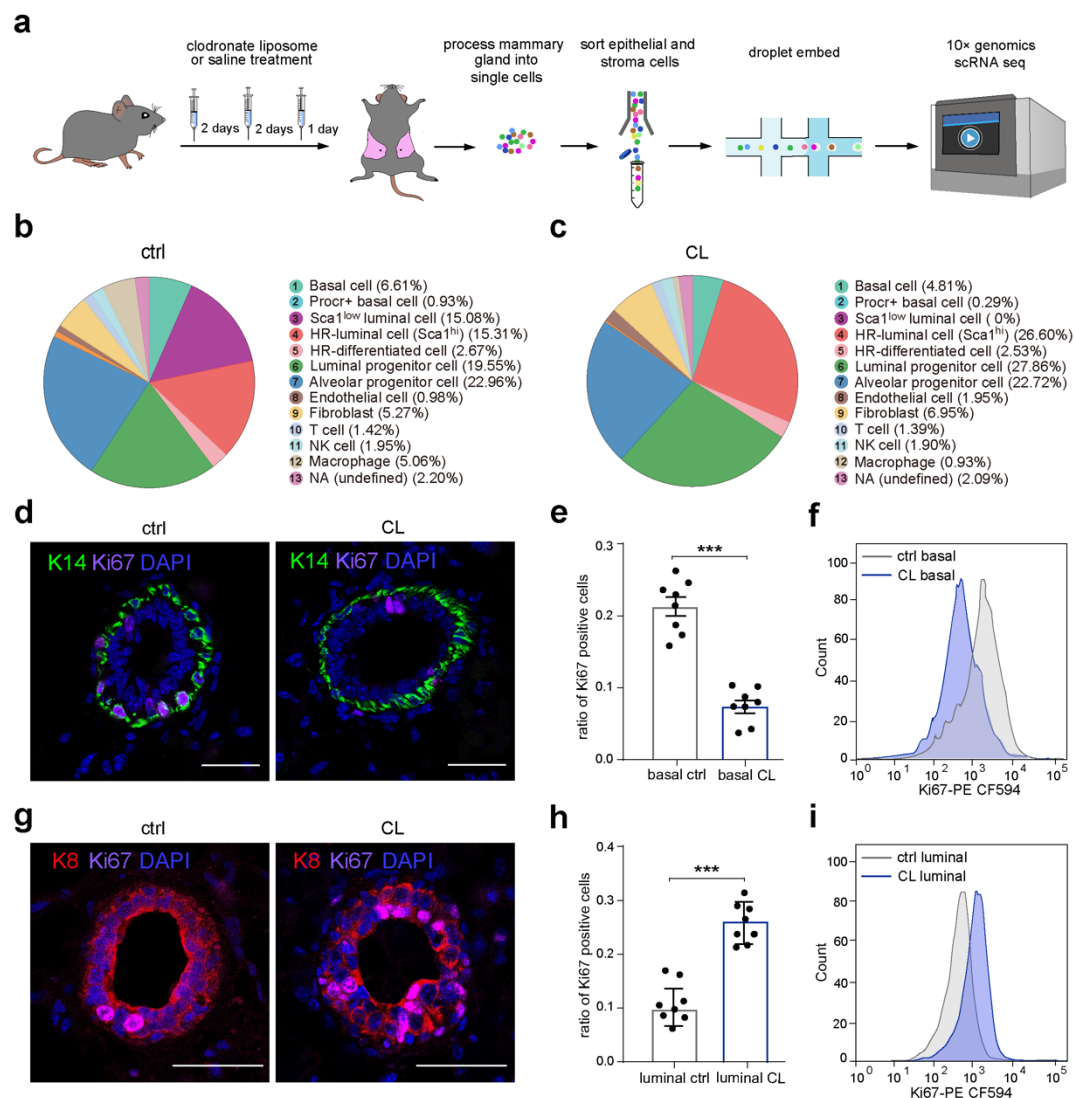


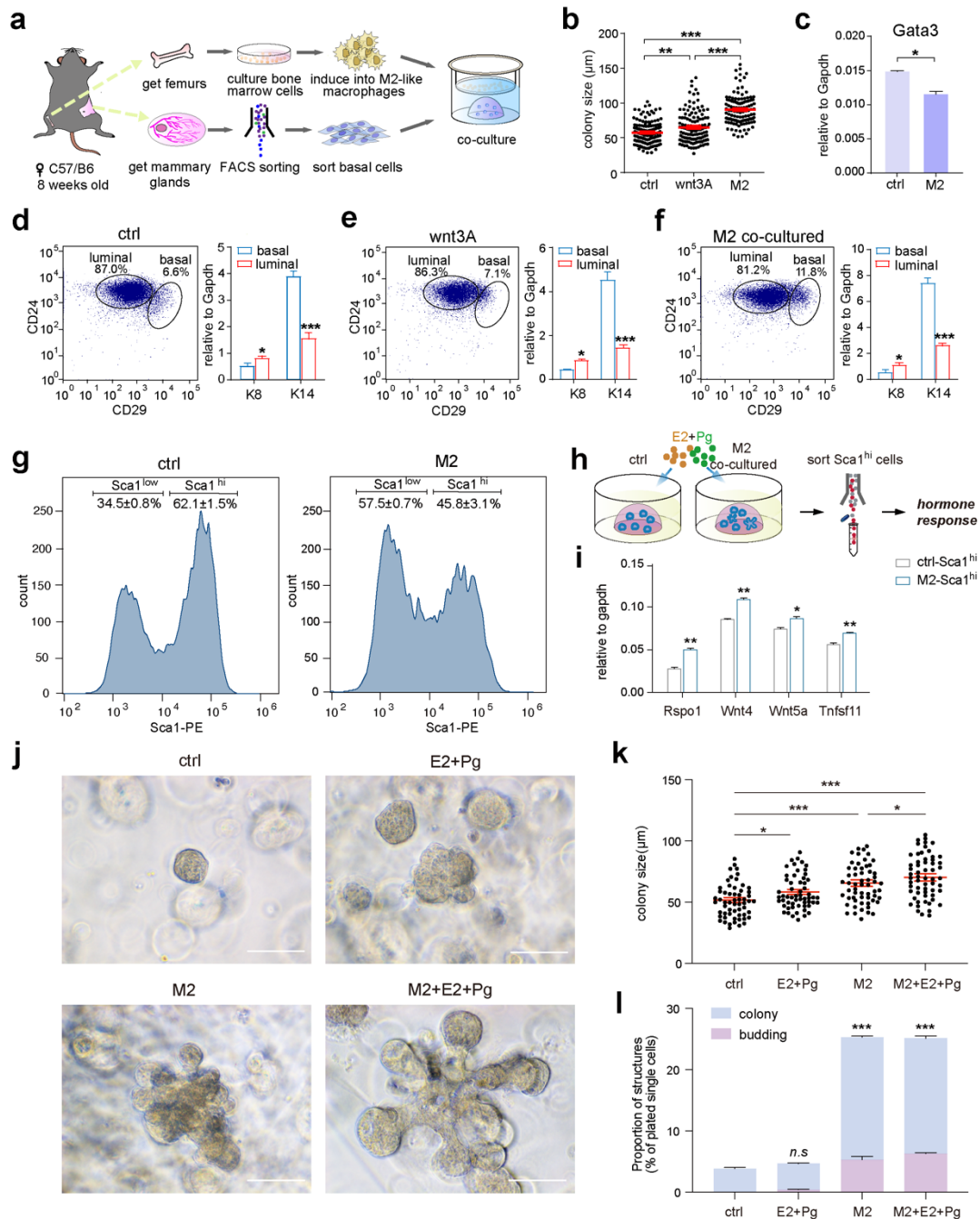
Supplementary Information

Supplementary Figures and Legends



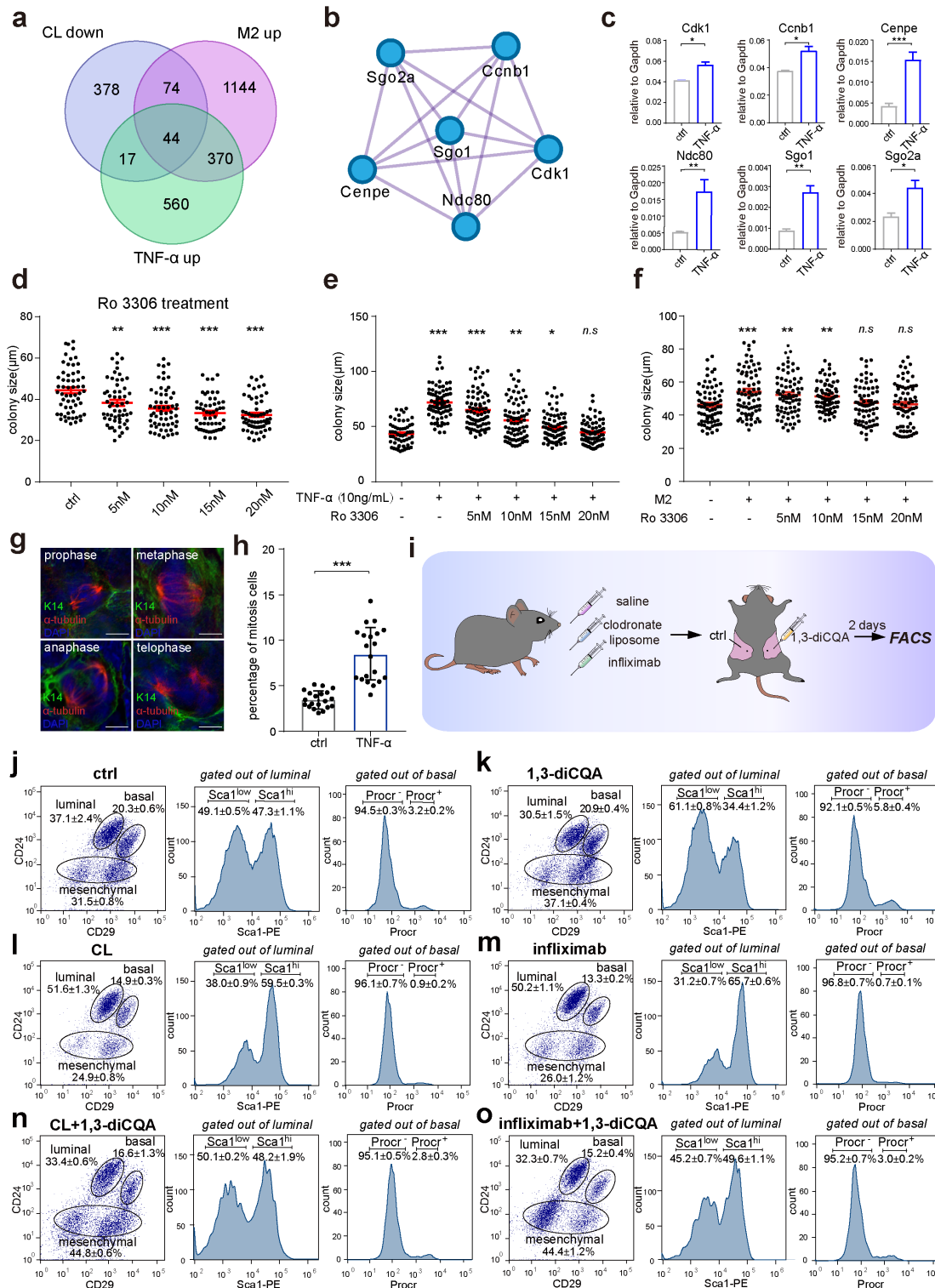
Supplementary Figure 1. Clodronate liposomes (CL) administration inhibits the proliferation of basal cells while promotes the proliferation of luminal cells. **a** Schematic illustration of intraperitoneal injection of clodronate liposomes and single-cell RNA sequencing. **b** The pie chart shows the proportion of various cell clusters of the mammary glands from control mice. **c** The pie chart shows the proportion of various cell clusters of the mammary glands from CL treated mice. **d** Immunostaining of Ki67 and K14 in mouse mammary gland. Ctrl: control mice; CL: CL treated mice, Scale bar: 20µm. **e**

Statistics of the proportion of Ki67 positive cells in basal cells in **(d)**, $n = 3$ mice and 8 images. Data are presented as mean $\pm$ SEM. $***P < 0.001$, unpaired t-test. **(f)** FACS analysis of Ki67 expression in basal cells. The results are representative of 3 independent experiments. **g** Immunostaining of Ki67 and K8 in mouse mammary gland. Ctrl: control mice; CL: CL treated mice, Scale bar: 50 μ m. **h** Statistics of the proportion of Ki67 positive cells in luminal cells in **(g)**. $n = 3$ mice and 8 images. Data are presented as mean $\pm$ SEM. $***P < 0.001$, unpaired t-test. **i** FACS analysis of Ki67 expression in luminal cells. The results are representative of 3 independent experiments.



Supplementary Figure 2. Cell population analysis and hormone response detection of cultured basal cells. **a** Schematic illustration of the co-culture system of basal cells with bone marrow-derived macrophages (M2). **b** Quantification of the colony size of the basal derived colonies treated with Wnt3A or co-cultured with M2, compared with that of control. n = 3 replications, **P < 0.01, ***P < 0.001, unpaired t-test. **c** qPCR analysis of *Gata3* expression

in M2 co-cultured organoids compared with that of control, $n = 3$ replications, $*P < 0.05$, unpaired t-test. **d-f** FACS analysis shows cell populations of digested colonies and organoids. qPCR analysis shows K14 and K5 expression of the colonies and organoids. Control basal cells (**d**); Wnt3A treated basal cells (**e**); M2 co-cultured basal cells (**f**). The results are representative of 3 independent experiments. $n = 3$ replications, $*P < 0.05$, $***P < 0.001$, unpaired t-test. **g** FACS analysis shows Sca1⁻ and Sca1⁺ cell proportion in control colonies and M2 co-cultured organoids. Data are presented as mean $\pm$ SD, the results are representative of 3 independent experiments. **h** Schematic illustration of E2 and Pg treatment with control colonies and M2 co-cultured organoids for hormone response detection. **i** qPCR analysis of *Rspo1*, *Wnt4*, *Wnt5a* and *Tnfrsf11* in sorted Sca1⁺ cells from control colonies and M2 co-cultured organoids. $n=3$ replications, $*P < 0.05$, $**P < 0.01$, unpaired t-test. **j** Representative images of 3D cultured control, E2 and Pg treated, M2 co-cultured and treated with E2 and Pg after M2 co-cultured basal cells, Scale bars: 100 μ m. **k** Quantification of the colony size of the basal derived colonies treated with E2 and Pg, co-cultured with M2, or treated with E2 and Pg after co-cultured with M2, compared with that of control. $n = 3$ replications, $*P < 0.05$, $***P < 0.001$, unpaired t-test. **l** Quantification of colony and budding structures derived from basal cells treated with E2 and Pg, co-cultured with M2, or treated with E2 and Pg after co-cultured with M2, compared with that of control. $n = 3$ replications, $***P < 0.001$, two-way ANOVA.



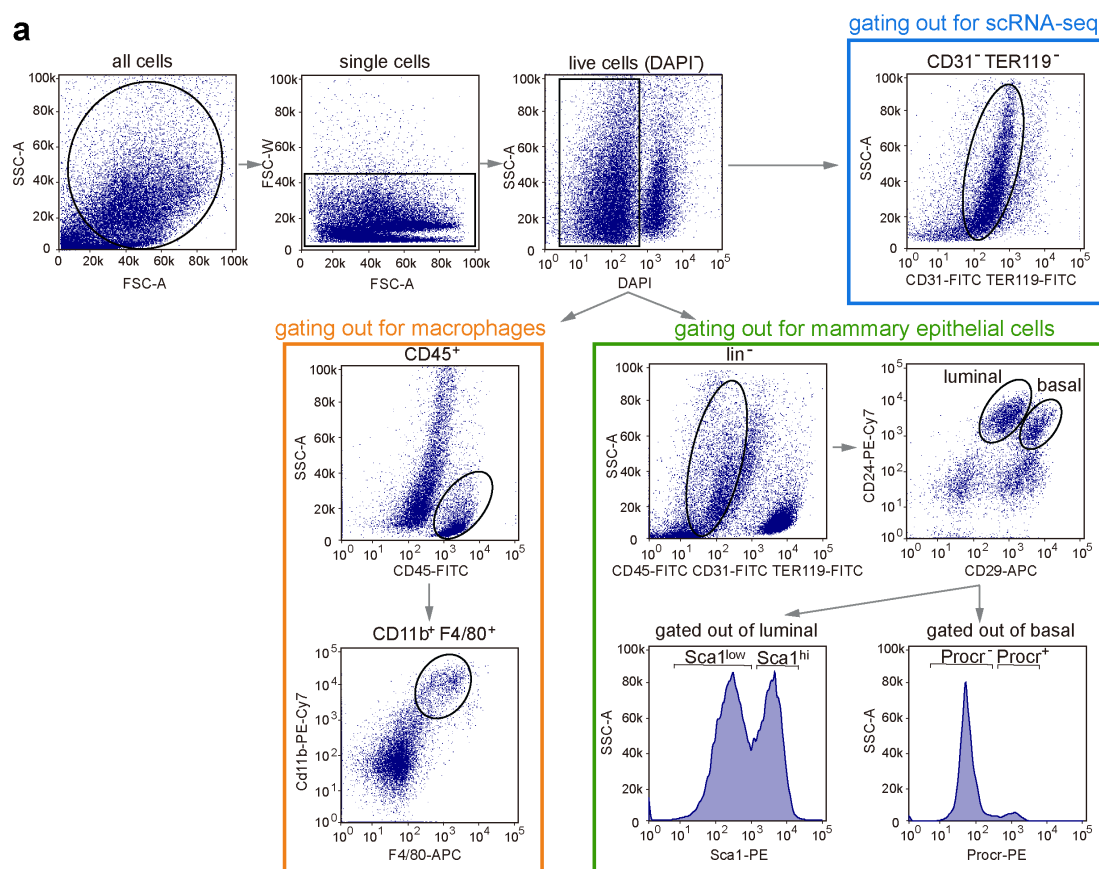
Supplementary Figure 3. Cdk1/Cyclin B1 involved in cell division

regulation in MaSCs and PI3K signaling participated in cell population

shift. **a** Venn diagram displays the overlapping genes of upregulated DEGs in organoids formed by M2 co-culture, upregulated DEGs in organoids induced

by TNF- α , and downregulated DEGs in mammary basal cells from clodronate liposomes treated mice. **b** MCODE out of Protein-Protein-Interaction (PPI) showing the certain functional module of the overlapping genes in (a). **c** qRT-PCR analysis for the expression of *Cdk1*, *Ccnb1*, *Cenpe*, *Ndc80*, *Sgo1* and *Sgo2a* genes in organoids induced by TNF- α . n = 3 replicates, * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$, *n.s* represents no significance, unpaired t-test. **d** Colony size statistics of basal cells treated with Ro 3306 at gradient concentration. n = 3 replicates, ** $P < 0.01$, *** $P < 0.001$, unpaired t-test. **e** Colony size statistics of basal cells treated with Ro 3306 at gradient concentration and TNF- α (10ng/mL) was added for compensation. n = 3 replications, * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$, *n.s* represents no significance, unpaired t-test. **f** Colony size statistics of basal cells treated with Ro 3306 at gradient concentration and M2 macrophages were co-cultured for compensation. n = 3 replications, * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$, *n.s* represents no significance, unpaired t-test. **g** Representative images of the spindles (marked by α -tubulin) and nucleus (marked by DAPI) in different stages of mitosis in basal colonies, Scale bar: 5 μ m. **h** Statistics of the percentage of mitosis cells in TNF- α induced basal cells compared with control. n = 20 colonies, *** $P < 0.001$, unpaired t-test. **i** Schematic illustration of 1,3-diCQA intra fat pad administration after saline/clodronate liposomes/infliximab treatment. **j-o** FACS analysis show mammary epithelial cell population labeled by CD24 and CD29, Sca1 labeled luminal cells, Procr labeled basal cells under separate and dual treatment with 1,3-diCQA, clodronate liposomes, infliximab. Control, treated with saline (**j**); 1,3-diCQA treatment (**k**); clodronate liposomes treatment(**l**); infliximab treatment (**m**); 1,3-diCQA intra fat pad administration

after clodronate liposome treatment (**n**); 1,3-diCQA intra fat pad administration
after infliximab treatment (**o**).



Supplementary Figure 4. FACS sequential gating strategies. **a** The cells sorted for scRNA-seq were CD31⁻TER119⁻ gated out of live cells correspond to supplementary Fig.1a, the gating strategy for macrophages was CD11b⁺F4/80⁺ gated out of CD45⁺ cells correspond to Figure 1b, the gating strategies for mammary epithelial cells were as follows: luminal cells (CD24⁺CD29^{low}) and basal cells (CD24⁺CD29^{hi}) were gated out of lin⁻ cells correspond to Figure 1e and Figure 6e; according to the expression of Sca1, the luminal cells could be divided into Sca1^{low} and Sca1^{hi} correspond to supplementary Fig.3j-o, according to the expression of Procr, the basal cells could be divided into Procr⁻ and Procr⁺ correspond to Figure 1f, Figure 6f and supplementary Fig.3j-o.

Supplementary Tables

Supplementary Table 1. Primers for qRT-PCR

<i>Bcl11b</i>	F: CCCGACCCTGATCTACTCAC	<i>Cenpe</i>	F: GAGCACCAAGAACTATTGACCG
	R: GGAGGTGGACTGCTCTTGT		R: CACGTTGACGCTCACTTGTCT
<i>Gata3</i>	F: CTCGGCCATTCGTACATGGAA	<i>Ndc80</i>	F: GCCTCTCTATGCAGGAGTTAAGG
	R: GGATACCTCTGCACCGTAGC		R: CGGTTTGTGTGTACTCAGCTT
<i>K14</i>	F: AGCGGCAAGAGTGAGATTCT	<i>Sgo1</i>	F: ACGCGTAACGCAGAATTTCTG
	R: CCTCCAGGTTATTCTCCAGGG		R: AATTCATCTCGGCCACTCAGG
<i>K8</i>	F: TCCATCAGGGTGACTCAGAAA	<i>Sgo2a</i>	F: ACCTGAATCCTGGTCTTCGC
	R: CCAGCTTCAAGGGGCTCAA		R: AGGAGTGGCTACCGAGAAGG
<i>Tnfrsf1b</i>	F: ACACCCTACAAACCGGAACC	<i>Rspo1</i>	F: GGGATCAAGGGCAAGAGACAG
	R: AGCCTTCCTGTCATAGTATTCCT		R: CTGGCGGATGTCGTTCCCTC
<i>Mmp3</i>	F: ACATGGAGACTTTGTCCCTTTTG	<i>Wnt4</i>	F: AGACGTGCGAGAACTCAAAG
	R: TTGGCTGAGTGGTAGAGTCCC		R: GGAAGTGGTATTGGCACTCCT
<i>Ccl2</i>	F: TGCCCTAAGGTCTTCAGCAC	<i>Wnt5a</i>	F: CAACTGGCAGGACTTTCTCAA
	R: AAGGCATCACAGTCCGAGTC		R: CATCTCCGATGCCGGAAGT
<i>Ccl7</i>	F: GCTGCTTTCAGCATCCAAGTG	<i>Tnfsf11</i>	F: CAGCATCGCTCTGTTCTCTGA
	R: CCAGGGACACCGACTACTG		R: CTGCGTTTTTCATGGAGTCTCA

Supplementary Table 2. Antibodies for immunochemistry

Antibodies	Source	Identifier	Dilution
Krt14	Wuhan Dia-an Biotechnology Co., Ltd.	Cat#C0740	1:200
Krt8	DSHB	Cat#TROMA-I	1:1000
Ki67	BioLegend	Cat#151202	1:400
α - tubulin	Proteintech	Cat#11224-1-AP	1:200
β -casein	Santa Cruz Biotechnology	Cat#sc-166530	1:50