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Reporting Summary

Nature Research wishes to improve the reproducibility of the work that we publish. This form provides structure for consistency and transparency in reporting. For further information on Nature Research policies, see our Editorial Policies and the Editorial Policy Checklist.

Statistics

For all statistical analyses, confirm that the following items are present in the figure legend, table legend, main text, or Methods section.

n/a	Confirmed
☐	☒ The exact sample size (*n*) for each experimental group/condition, given as a discrete number and unit of measurement
☐	☒ A statement on whether measurements were taken from distinct samples or whether the same sample was measured repeatedly
☐	☒ The statistical test(s) used AND whether they are one- or two-sided *Only common tests should be described solely by name; describe more complex techniques in the Methods section.*
☐	☒ A description of all covariates tested
☐	☒ A description of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons
☐	☒ A full description of the statistical parameters including central tendency (e.g. means) or other basic estimates (e.g. regression coefficient) AND variation (e.g. standard deviation) or associated estimates of uncertainty (e.g. confidence intervals)
☐	☒ For null hypothesis testing, the test statistic (e.g. *F*, *t*, *r*) with confidence intervals, effect sizes, degrees of freedom and *P* value noted *Give P values as exact values whenever suitable.*
☒	☐ For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings
☒	☐ For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes
☒	☐ Estimates of effect sizes (e.g. Cohen's *d*, Pearson's *r*), indicating how they were calculated

Our web collection on statistics for biologists contains articles on many of the points above.

Software and code

Policy information about availability of computer code

Data collection	Images were acquired using ZEN Blue (Carl Zeiss, v2.3), iQ3 (Andor, v3.6.2) and Asylum Research v14 (IgorPro 6.2.1) FACS data was collected using FACSDiva (BD Biosciences, v8.0.3) software. Simulations were run using C/C++ based on standard C libraries and GNU Scientific Library (GSL, v2.6).
Data analysis	Images were analysed using Fiji (NIH, 2.0.0-rc-69/1.52p), CellProfiler (v3.1.8) and Imaris (Oxford Instruments, v8.3.1). Sequencing data were analysed using STAR (v2.6.2a) and DEseq2 (v1.24.0). Data were compiled and statistical tests were performed using Excel (Microsoft, v14.5.7), Prism (Graphpad, v7), R (version 3.4.4) and RStudio (Version 1.1.442). Figures were assembled using Adobe Illustrator CS6 (v16.0.0).

For manuscripts utilizing custom algorithms or software that are central to the research but not yet described in published literature, software must be made available to editors and reviewers. We strongly encourage code deposition in a community repository (e.g. GitHub). See the Nature Research guidelines for submitting code & software for further information.

Data

Policy information about availability of data

All manuscripts must include a data availability statement. This statement should provide the following information, where applicable:

- Accession codes, unique identifiers, or web links for publicly available datasets
- A list of figures that have associated raw data
- A description of any restrictions on data availability

All RNA-sequencing data are in the processes of being deposited into the Gene Expression Omnibus and accession codes will be made available before publication. All other data in the manuscript, supplementary materials, and source data are available from the authors upon reasonable request.

Field-specific reporting

Please select the one below that is the best fit for your research. If you are not sure, read the appropriate sections before making your selection.

☒ Life sciences ☐ Behavioural & social sciences ☐ Ecological, evolutionary & environmental sciences

For a reference copy of the document with all sections, see nature.com/documents/nr-reporting-summary-flat.pdf

Life sciences study design

All studies must disclose on these points even when the disclosure is negative.

Sample size	No statistical methods were used to predetermine sample sizes. Sample sizes were determined based on previous publications on similar experiments (Nature volume 501, pages185–190, 2013).
Data exclusions	No data were excluded.
Replication	Every experiment was performed on at least 2 independent litters. All attempts at replication were successful.
Randomization	Samples (mouse embryos) were allocated randomly into experimental groups.
Blinding	Investigators were not blinded to experimental groups/genotypes due to the nature of embryo recovery.

Reporting for specific materials, systems and methods

We require information from authors about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each material, system or method listed is relevant to your study. If you are not sure if a list item applies to your research, read the appropriate section before selecting a response.

Materials & experimental systems

n/a	Involved in the study
☐	☒ Antibodies
☒	☐ Eukaryotic cell lines
☒	☐ Palaeontology and archaeology
☐	☒ Animals and other organisms
☐	☒ Human research participants
☒	☐ Clinical data
☒	☐ Dual use research of concern

Methods

n/a	Involved in the study
☒	☐ ChIP-seq
☐	☒ Flow cytometry
☒	☐ MRI-based neuroimaging

Antibodies

Antibodies used	Antibodies used were as follows: rat anti-RFP (1:1000, Chromotek, 15F8, lot# 60706002AB), rabbit anti-RFP (1:1000, MBL, PM005, lot# 044), chicken anti-GFP (1:2000, Abcam, ab13970, lot# GR3190550-6), goat anti-P-Cadherin (1:500, R&D, AF761), rabbit anti-E-cadherin (1:500, Cell Signaling Technology, 1:500, 9835, lot# GR3190550-6), rat anti-E-cadherin (1:200, M. Takeichi), guinea pig anti-keratin14 (1:500, Fuchs laboratory), rabbit anti-keratin10 (1:1000, Covance, poly19054, lot# D15LF02452), rabbit anti-collagen type IV (1:500, Abcam, ab6586), rat anti-nidogen (1:200, Santa Cruz Biotechnology, ELM1), rat anti-Laminin-β1 (1:100, Abcam, LT3), rabbit anti-Laminin-α5 (1:500, J. Miner, Washington University in St. Louis), rabbit anti-Laminin-332 (1:500, P. Marinkovich, Stanford University), mouse anti-phospho-S22-Myosin light chain 2 (1:100, Cell Signaling Technology, 3675), mouse anti-vimentin (1:200, Dako, 3B4), rat anti-Sca-1 (1:200, Becton Dickinson, D7), rat biotinylated anti-CD45 (1:200, BD Bioscience, 553078, lot# 6294639), rat biotinylated anti-CD140a (1:200, Biolegend, 135910, lot# B236869), rat biotinylated anti-CD31 (1:200, Biolegend, 102504, lot# B208712), rat biotinylated anti-cKit (1:200, Biolegend, cat#105804, lot#B208999), rat PECy7-conjugated-anti alpha6/CD49f (1:1000, Biolegend, 313621, lot# B211003). All secondary antibodies used were raised in a donkey host and were conjugated to one of AlexaFluor488, AlexaFluor546 or AlexaFluor647 (1:500, Life Technologies).
Validation	All antibodies are commercially available and validated by the manufacturer except for rabbit anti-keratin14 (Fuchs lab), rabbit anti-Laminin-α5 (J. Miner) and rabbit anti-Laminin-332 (P. Marinkovich), which were used at the stated concentrations. The subcellular localization of all the proteins analyzed in this study has been previously reported. This was used to validate the specificity of the antibody.

Animals and other organisms

Policy information about studies involving animals; ARRIVE guidelines recommended for reporting animal research

Laboratory animals	Animals were fed regular rodent chow with ad libitum access to food and water. Euthanasia was by CO2 asphyxiation. The following

Laboratory animals	previously generated mouse (Mus musculus) lines were used in this study: Rosa26-SmoM2-YFPfl/fl, FrHRas-G12Vfl/fl, Rosa26-EYFPfl/fl, Rosa26-mTmG, Krt14-rtTA, hIVL-rtTA. C57Bl6J/CD1 mixed background strains were used. Embryos were analyzed from E12.5 to E18.5, and adult mice were collected at P21 and 3-6mo for tumor analysis. Both sexes were used for adult studies, and sex was not determined in embryos.
Wild animals	The study did not involve wild animals.
Field-collected samples	The study did not involve samples collected from the field.
Ethics oversight	All procedures were approved by the Rockefeller University IACUC and performed within an AAALAC-certified animal facility.

Note that full information on the approval of the study protocol must also be provided in the manuscript.

Human research participants

Policy information about studies involving human research participants

Population characteristics	De-identified OCT-embedded fresh, non-fixed tissue blocks of SCCs, BCCs or healthy skin from male and females (age range 49-82) that underwent Mohs micrographic surgery were used.
Recruitment	De-identified tissue blocks were obtained from the Department of Dermatology, Weill-Cornell Medical College (New York, US). This study did not involve the recruitment of new patients, and no self-selection bias is acknowledged.
Ethics oversight	We have complied with all relevant ethical regulations, including obtaining informed consent from patients by Weill-Cornell and the Rockefeller University IRB approved the use of de-identified human samples (EFU-0529).

Note that full information on the approval of the study protocol must also be provided in the manuscript.

Flow Cytometry

Plots

Confirm that:

☒ The axis labels state the marker and fluorochrome used (e.g. CD4-FITC).

☒ The axis scales are clearly visible. Include numbers along axes only for bottom left plot of group (a 'group' is an analysis of identical markers).

☒ All plots are contour plots with outliers or pseudocolor plots.

☒ A numerical value for number of cells or percentage (with statistics) is provided.

Methodology

Sample preparation	Dorsal skins were surgically removed from E15.5 embryos and incubated for 30min at 37C in 1:1 trypsin:versene. Cells were filtered through a 40um cell strainer (VWR) to generate a single cell suspension.
Instrument	BD FACSAriaII, 70um nozzle
Software	BD FACSDIva Software
Cell population abundance	Lin-;LV-Cre+ cell populations were about 5% of the total population at E15.5.
Gating strategy	Cells were gated on DAPI-negative (for live/dead) and FSC/SSC singlets. Lineage-negative cells were identified by staining with biotinylated primary antibodies against CD45 (to exclude immune cells), CD117/cKit (to exclude the melanoblast lineage), CD31 (to exclude endothelial lineage), and CD140a (to exclude fibroblasts). All lin- cells were labelled with a streptavidin-PECy7 conjugated secondary antibody. CD49f/alpha6-high cells (a marker of the basal epidermis) that double labelled with RFP (as a marker of LV-Cre transduction) were isolated to identify WT, SmoM2+ and HRas+ LV-Cre transfected cells (in control, Rosa26-SmoM2-YFPfl/fl and FrHRas-G12Vfl/fl;Rosa26-EYFPfl/+ background genotypes, respectively). Both sexes were used as sex was not determined in embryos). Each sample comprised single cells from at least 3 embryos per genotype.

☒ Tick this box to confirm that a figure exemplifying the gating strategy is provided in the Supplementary Information.