Appendix

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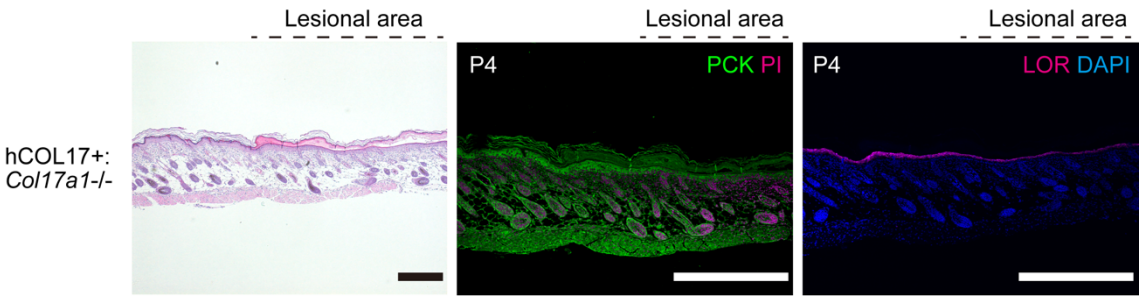
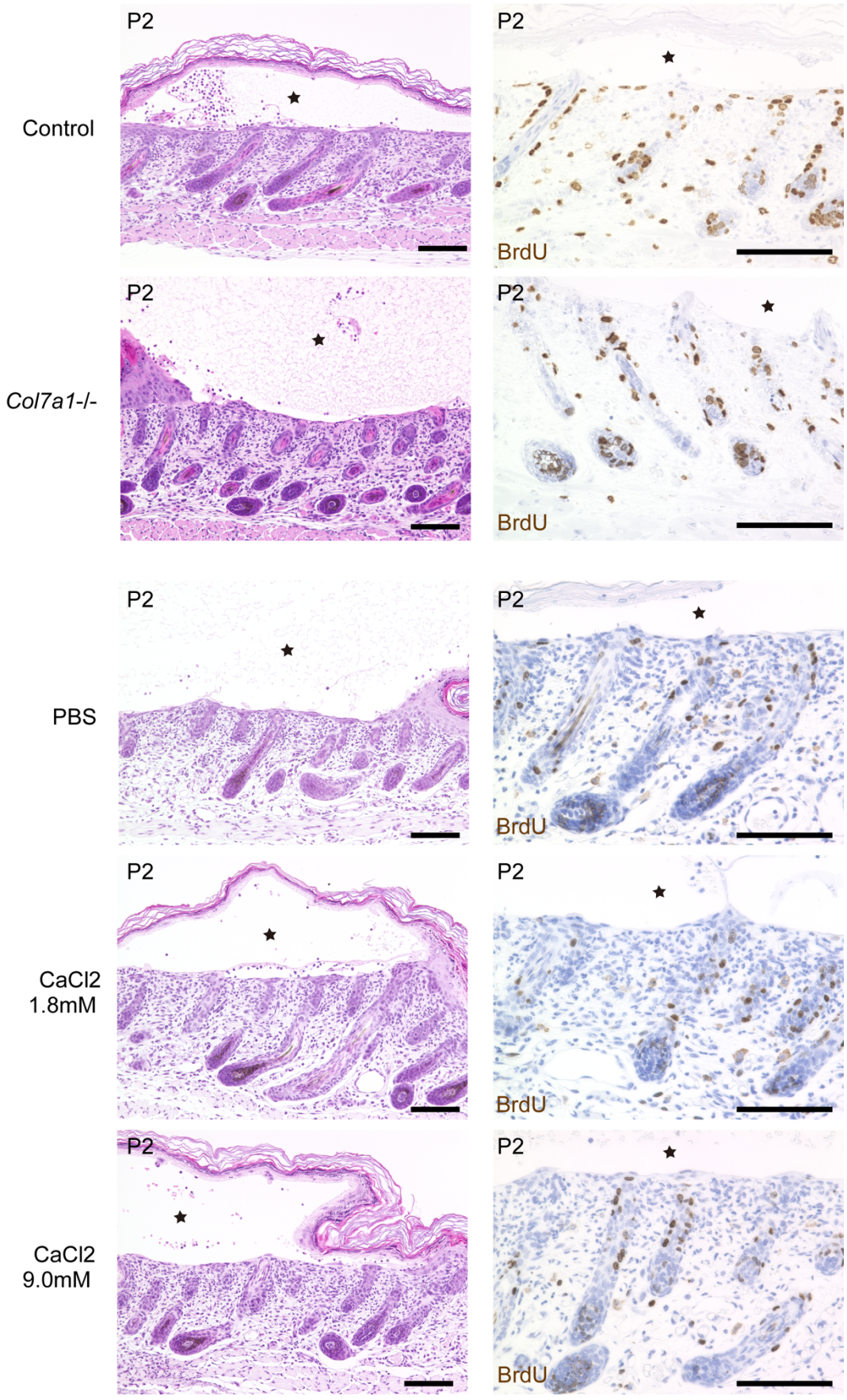


Appendix Figure S1. Transgenic rescue of *Col17a1*-/- mouse blister healing.

Blistered samples of *Col17a1*-/- mice rescued with human COL17 overexpression (hCOL17+;*Col17a1*-/-) (representative images from four mice). H&E, PCK and LOR staining (P4). Scale bar: 500 μm.


Control
P2
P2
BrdU
Col7a1-/-
P2
P2
BrdU
PBS
P2
P2
BrdU
CaCl2
1.8mM
P2
P2
BrdU
CaCl2
9.0mM
P2
P2
BrdU

Appendix Figure S2. H&E and BrdU labeling of *Col7a1*-/- and Ca-treated blisters.

H&E and BrdU labeling of blister samples at P2. *Col7a1*$^{-/-}$ mouse and littermate control (representative images from four and three mice, respectively). WT blistered skin treated with $CaCl_2$ or PBS (representative images from three mice). Scale bar: 100 μm. Blisters are indicated by stars.

Appendix Supplementary Methods

The mathematical model

The mathematical model used for the simulations is based on our previous works [45,46], which has been adapted for the present purpose. We consider cell division dynamics on a flat basement membrane. Two kinds of cells are considered: Stem cells produce transit amplifying cells infinite number of times, and transit amplifying cells undergo division finite number of times. While stem cells are anchored to the basement membrane, transit amplifying cells are weakly adhesive to the basement membrane so that they can leave the basal layer. Details are described below.

Geometry of the system

The system is considered in a three-dimensional region $[0, L_x] \times [0, L_y] \times [0, L_z]$, where periodic boundary is considered in x and y axes. The basement membrane is prepared as densely packed, spatially fixed particles with radius R_m. These particles are arranged in a triangular lattice configuration at $z = 0$ with the x and y coordinates $d_m i + \frac{1}{2} d_m j$ (modulo L_x) and $(\sqrt{3}/2) d_m j$, respectively $(i = 0, \ldots, N_x - 1,\ j = 0, \ldots, N_y - 1)$. The vertical size was set to $L_z = 300$ μm. For the horizontal size, we set $d_m = 2.5$ μm, N_x =240, and $N_y = 277$, so that $L_x =$

$d_m N_x = 600\ \mu\text{m}$ and $L_y = (\sqrt{3}/2) d_m N_y = 599.7\ \mu\text{m}$. Overlapping of membrane particles are allowed, so that their radii matter only when they interact with cells.

Epidermal cell dynamics

Epidermal cells are modeled as oblate spheroids. The position of cell i is specified by the coordinate of the center of mass $\boldsymbol{r}_i(t) = (x_i(t), y_i(t), z_i(t))$. The shape of the cell is characterized by the flatness parameter $\rho_i(t)$, with which the long axis and the short axis are given by $\rho_i(t)R$ and $\rho_i(t)^{-2}R$, respectively, where the volume is assumed to remain unchanged by the shape change. The short axis is assumed to be aligned with z axis.

The position of cell i is governed by the equation of motion:

$$\frac{d\boldsymbol{r}_i}{dt} = -\mu \frac{\partial}{\partial \boldsymbol{r}_i}\left[U_{cell} + \chi_i^{div} U_{div} + U_{base}\right].$$

The first term U_{cell} represents the interaction potential between two cells, which consists of two parts:

$$U_{cell} = \sum_{j \in \Omega_i} \left[K_{ex}^{(1)} u_{ex}\left(\frac{|\boldsymbol{r}_i - \boldsymbol{r}_j|}{R_i^{eff} + R_j^{eff}}\right) + K_{ad}^{(1)} u_{ad}\left(\frac{|\boldsymbol{r}_i - \boldsymbol{r}_j|}{R_i^{eff} + R_j^{eff}}\right)\right].$$

Here Ω_i denotes the set of cells that are inside the interaction range of cell i defined by $|r_i - r_j| < l^*$, The cutoff range l^* is chosen to be sufficiently large. When cell i commits division, its dividing partner, although within the interaction range, is excluded from Ω_i and treated separately, as explained below.

The function $u_{ex}(x)$ represents the exclusion volume effect, which works only when the two cells overlap:

$$u_{ex}(x) = \begin{cases} \frac{1}{12}x^{12} - \frac{1}{6}x^6 + \frac{1}{12}, & x < 1, \\ 0, & x \geq 1, \end{cases}$$

and $u_{ad}(x)$ represents adhesion between cells, which has a cutoff length λ, above which adhesion is lost:

$$u_{ad}(x) = \begin{cases} 0, & x < 1, \\ \frac{(x-1)^2}{2} - \frac{(x-1)^4}{4(\lambda-1)^2}, & 1 \leq x < \lambda, \\ \frac{(\lambda-1)^2}{4}, & \lambda \leq x. \end{cases}$$

The constants R_i^{eff} and R_j^{eff} are the effective radii of i and j, respectively, which reflect the nature of the oblate spheroid, as defined below.

The second term U_{div} is the potential between a pair of cells undergoing division; $\chi_i^{div} = 1$ if i is undergoing division; otherwise $\chi_i^{div} = 0$. On the onset of division at $t = t_0$, the original cell i and the newly created cell i' completely overlap: we set $\boldsymbol{r}_i(t_0) = \boldsymbol{r}_{i'}(t_0)$ and $\rho_i(t_0) = \rho_{i'}(t_0)$. For $t > t_0$, they repulsively interact with each other with the spring force, with the potential given by

$$U_{div} = \frac{K_{div}}{2}\left(|\boldsymbol{r}_i - \boldsymbol{r}_{i'}| - l(t)\right)^2.$$

The natural length $l(t)$ between i and i' grows from zero to $2R$ at a constant rate β as $\frac{dl(t)}{dt} = \beta(2R - l(t))$. Division is complete when the distance between

the pair exceeds $R_{i,i'}^{eff} + R_{i',i}^{eff}$; after that the two cells are treated independently by setting χ_i^{div} to 0).

The third term describes the interaction between cell i and the basement membrane, which is given by

$$U_{base} = \sum_{k \in \Omega_{memb}} K_{ex}^{(2)} u_{ex}\left(\frac{|\boldsymbol{r}_i - \boldsymbol{r}_k^m|}{R_{i,k}^{eff} + R_m}\right) + (1 - \chi_i^{stem}) K_{ad}^{(2)} u_{ad}\left(\frac{|\boldsymbol{r}_i - \boldsymbol{r}_{i^*}^m|}{R_{i,i^*}^{eff} + R_m}\right)$$
$$+ \chi_i^{stem} \frac{K_{bind}}{2}\left(|\boldsymbol{r}_i - \boldsymbol{r}_{i^*}^m| - (R_{i,i^*}^{eff} + R_m)\right)^2.$$

The first term describes the excluded volume effect between the cells and the basement membrane: Ω_{memb} is the set of membrane particles, and $\boldsymbol{r}_k^m$ is the position of membrane particle k, with radius R_m. The second and the third terms represent adhesion to the basement membrane particle nearest to cell i, denoted by i^*. The second term works for a TA cell ($\chi_i^{stem} = 0$), for which i^* changes in time. TA cells can leave the basal layer because the function u_{ad} has a cutoff. The third term works for a stem cell ($\chi_i^{stem} = 1$), whose interaction partner i^* is fixed. Stem cells cannot leave the basal layer because their adhesion is represented by a spring force.

Shape changes of the cell

We assume that stem cells do not change its shape, i.e., $\rho_i(t) = 1$, whereas transit amplifying cells become flattened when they are in the lesional area: ρ_i changes

as $d\rho_i(t)/dt = k_\rho(\rho_{max} - \rho_i(t))$ when cell i is inside of the lesional area, and $d\rho_i(t)/dt = 0$ when outside of the lesional area. The definition of the lesional area is given below. Given a direction of short axis $\boldsymbol{n}_i = (0,0,1)$, the effective interaction radius of cell i interacting with cell j is given by $R_{i,j}^{eff} = \sqrt{\frac{R}{\rho_i^2}\cos\theta_{ij} + \rho_i R \sin\theta_{ij}}$, where $\cos\theta_{ij} = \frac{(\boldsymbol{r}_i - \boldsymbol{r}_j)}{|\boldsymbol{r}_i - \boldsymbol{r}_j|} \cdot \boldsymbol{n}_i$. Note that $R_{i,j}^{eff} \neq R_{j,i}^{eff}$ in general. Although R_i^{eff} depends on $\boldsymbol{r}_i$, we treat them as constant when taking derivative with respect to $\boldsymbol{r}_i$ in the equation of motion.

The flatness approaches the fixed value ρ_{max}, which we choose in the simulations as $1.0, 1.25, 1.5, 1.75,$ and 2.0.

Cell division

The cell division event is governed by the following rule: Each cell that is proliferative has a period T_0 after which it undergoes division stochastically following the Poisson process with the division rate γ, implying that the average division period is $T_0 + 1/\gamma$. The maximum number of cell division for transit amplifying cells is set to 12.

Simulation setup

First, we placed stem cells and ran a simulation until the basement membrane was covered with transit amplifying cells. Then we created the initial condition for a

healing process by replacing transit amplifying cells in the circular region with radius R_{lesion} by "air" particles with the same size, which would not exert forces on cells but get pushed by cells and by themselves due to excluded volume effect. The lesional area was defined by the region occupied these air particles, and the transit amplifying cells that were in contact with at least one air particle was considered as being in the lesional area, and therefore, changes into a flattened shape. The size of the lesional area was set to $R_{lesion} = 0.4\,L$; For simulation efficiency, we created a smaller lesional area than in the experiment.

Model parameters

Parameter values were set as follows: $R_m = 5.0\ [\mu m]$, $R = 5.0\ [\mu m]$, $K_{ex}^{(1)} = 1.0$, $K_{ad}^{(1)} = 0.16$, $l^* = 1.17$, $\lambda = 1.17$, $K_{div} = 5.0$, $\beta = 0.14$, $K_{ex}^{(2)} = 1.0$, $K_{ad}^{(2)} = 21.0$, $K_{bind} = 30.0$, $k_\rho = 0.5$, $T_0 = 36\ [h]$, and $\gamma = 0.0416\ [h^{-1}]$. These parameter values are consistent with those in our previous work, with which epidermal structures were successfully reproduced.