

Fig. S1. The length of Tam treatment determines the number of the tagged BECs.
(A) Timeline depicting the cell tagging and imaging scheme. Green box indicates imaged body regions. Tam, Tamoxifen. dpf, days post fertilization.
(B) Confocal images of tagged cells in an 8 dpf basebow ( $\mathrm{n}=10$ animals in each treatment group). Scale bar, $100 \mu \mathrm{~m}$.
(C) Images of optical sections and illustrations depicting the positon of a tagged BEC and SEC. White dashed lines mark basement membrane. SECs, Superficial epithelial cells. BECs, Basal epithelial cells. Scale bar, $100 \mu \mathrm{~m}$.
(D) Quantification of total tagged BECs and SECs ( $465 \mu \mathrm{~m} \times 465 \mu \mathrm{~m} ; \mathrm{n}=6,8,10$ animals, respectively; mean $\pm$ SD).


Fig. S2. Basebow zebrafish has no leaky Cre activity during long-term monitoring.
(A) Whole-mount confocal image of an 8 dpf basebow (stitched; $\mathrm{n}=18$ animals). Scale bar, $200 \mu \mathrm{~m}$.
(B) Whole-mount epifluorescence image of a 53 dpf basebow (stitched; $\mathrm{n}=24$ animals). Scale bar, 1 mm .


Fig. S3. Clonal behaviors on both sides of the fish body surface are symmetrical.
(A) Illustration depicting the left and the right side of the fish body.
(B) Representative whole-mount epifluorescence images of a 35 dpf basebow (stitched). Right side, top. Left side, bottom. Yellow circles mark examined BEC clones. Scale bar, 1 mm
(C) Quantification of clone numbers ( $\mathrm{n}=33$ animals; mean $\pm$ SD; two-tailed Student's $t$-test).
(D) Quantification of clone number ratios. Clone numbers on both sides of the animal are symmetrical as the average ratio is close to one ( $n=33$ animals; mean $\pm$ SD).
(E) Quantification of clone size ( $\mathrm{n}=33$ animals; mean $\pm$ SD; two-tailed Mann-Whitney test).


Fig. S4. $\operatorname{Tg}$ (krt19:H2A-mCherry) line labels only the BEC stem cell population.
(A) The Tg(krt19:H2A-mCherry) transgenic construct.
(B) Illustration depicting the development of zebrafish skin epidermis and the cell populations that are labeled in the Tg(krt19:H2A-mCherry)as54 line. SECs, Superficial epithelial cells. BECs, Basal epithelial cells.
(C) Cross-sections of the Tg(krt19:H2A-mCherry)as54 line at 8 and 35 dpf. P63 Ab staining marks the BEC layer (green). DAPI stains all cells (white). 35 dpf images are stitched. White box indicates magnified area shown in the right-most images. White dashed lines mark basement membrane. dpf, days post fertilization. Scale bar, $100 \mu \mathrm{~m}$.


Fig. S5. Macroscopic imaging platform detects BEC clones larger than $0.002 \mathbf{~ m m}^{\mathbf{2}}$ in size.
(A) Quantification of absolute clone size from all examined clones. BEC clones smaller than $0.002 \mathrm{~mm}^{2}$ are below the detection limit of the imaging platform. The sizes of animals examined here ranged from SL10 to SL16 mm ( $\mathrm{n}=742$ clones; mean $\pm$ SD).
(B) Quantification of relative clone size as percentage of body surface area. BEC clones smaller than $0.007 \%$ of the body surface area are below the detection limit of the imaging platform ( $n=742$ clones; mean $\pm$ SD).
(C) Illustration depicting how 2D maximum intensity projection clone images may underestimate actual clone size.


Fig. S6. Lamb1a expression is spatially regulated in developing and homeostatic skin tissues.
(A) Illustration depicting scale-plucking from either AS or PS region of the animal body surface. Plucked scales containing skin BECs from distinct body regions were subjected to RT-qPCR analyses.
(B-G) RT-qPCR analyses of lef1, fgf20a, pola2, ddx52, lamb1a, and lamb1b expression in skin tissues collected from either the AS or PS region of the animal body surface. ( $\mathrm{n}=4$ biological replicates in each data point; mean $\pm$ SEM; two-tailed Student's $t$ test).


Fig. S7. Temporal inactivation of Lamb1a activity has no significant influence on BEC proliferation in developing skin tissues.
(A) Illustration depicting the temperature shift (TS) and EdU incubation scheme. Green box indicates imaged body region.
(B) Whole-mount EdU staining of sde1 and mem mutant larvae. Proliferating BECs were visualized by double staining of P63 (red) and EdU (green). Yellow circles mark double-positive cells. Scale bar, $100 \mu \mathrm{~m}$.
(C) Quantification of EdU + P63 ${ }^{+}$double positive cells in sde1 and mem mutant larvae upon heat treatment. The mem mutant, which carries a ts allele of the gene DNA polymerase alpha subunit 2 (pola2), was included as a positive control for the heat treatment ( $\mathrm{n}=6$ sde1/+, 6 sde1; $5 \mathrm{mem} /+, 7$ mem; mean $\pm$ SD; two-tailed Student's $t$-test).

Table S1. List of primers used for RT-qPCR.

| RT-qPCR |  |  |
| :---: | :---: | :---: |
| Gene | Direction | Primer sequence |
| lef1 | Forward | CTG GTC AAC GAG ACA GAA ATC A |
|  | Reverse | CCT GTG CTT CTC ATG GTA AGA G |
| fgf20a | Forward | TGG ATA GCG GAT TGT ATC TGG |
|  | Reverse | CAA ACT GCT CCC TGA ACA CA |
| pola2 | Forward | GTT CGG ACC TTT TGT TGA TTC A |
|  | Reverse | TCC ACT ATG CTG TCC ATA CAT C |
| $d d x 52$ | Forward | GGT CTG GAG CAA AGT TTG ATT T |
|  | Reverse | ATC ACT TGC TTC TCC TTC TTC A |
| lamb1a | Forward | CCC CTG TCG ATG GAA CTG |
|  | Reverse | TAC ACA TAC AAT GAC CAT GAA CCA |
| lamb1b | Forward | CTC CCA ACC GCC TTA AAA C |
|  | Reverse | AAT CCA GCT GAA TGG TCA CA |
| actb | Forward | TAC ACA GCC ATG GAT GAG GAA AT |
|  | Reverse | TCC CTG ATG TCT GGG TCG TC |

