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Supplemental Information

Adult Neurogenesis Is Sustained by Symmetric Self-Renewal and Differentiation

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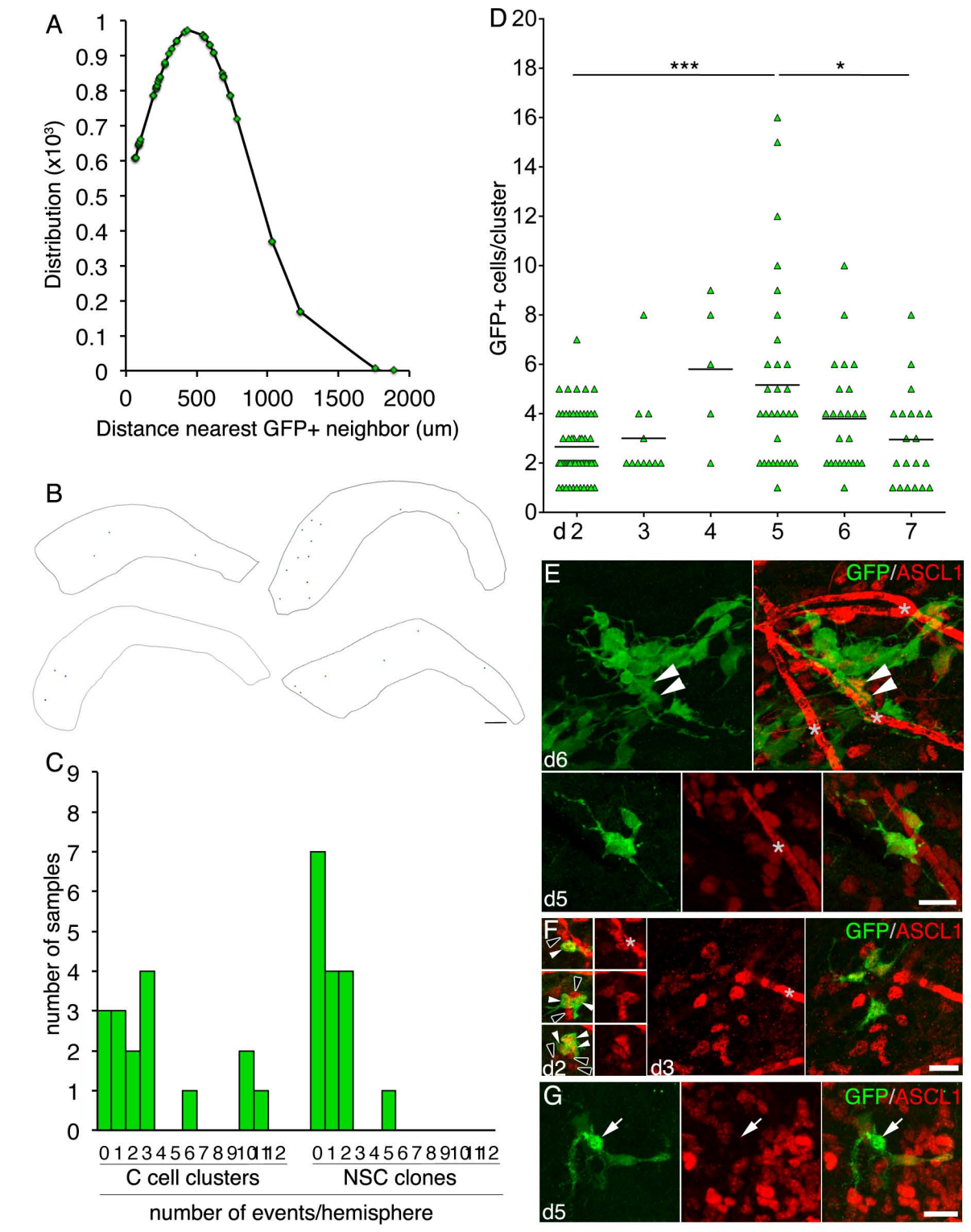
A
Distribution (x10³)
Distance nearest GFP+ neighbor (um)
B
C
number of samples
C cell clusters
NSC clones
number of events/hemisphere
D
GFP+ cells/cluster
***
*
d 2
3
4
5
6
7
E
GFP/ASCL1
d6
d5
F
d2
d3
G
d5

Figure S1: Cluster analysis of GFP+ cells (related to Fig. 1)

(A) Probability distribution of distances between neighboring GFP+ events measured on d2-d3 after RCAS injection. (B) Representative maps of traced GFP+ events used for analysis in (A); green triangles: C cell clusters; blue triangles: NSCs). (C) Number of GFP+ clones (C cell clusters or NSCs) found per whole-mount on d2-d3 after RCAS injection at P21. (D) C cells per GFP+ clone (single cells or clusters). Clones initially increase in size but then decrease (d2 vs. d5 $p < 0.0001$****; d5 vs. d7 $p= 0.0116$*.; one-way ANOVA with Tukey's multiple comparisons). Note that on d2, several C cell clusters consist of one cell only, consistent with a single integration event during M phase. The largest clusters can be found at d5 and one-cell clusters re-appear. (E) At later time-points clusters contain of few Ascl1+ cells and are surrounded by large numbers of GFP+ neuroblasts, possibly their own progeny (upper panel; d6). Other clusters do not contain neuroblasts (lower panel; d5). (F) GFP+ C cells (white arrowheads) in a cluster with comparable number of unlabeled C cells (black arrowheads). (G) A cluster of three GFP+ASCL1+ cells and one NSCl on d5 (arrowhead). Scale bar is 500 μm in (B), all others are 20 μm.

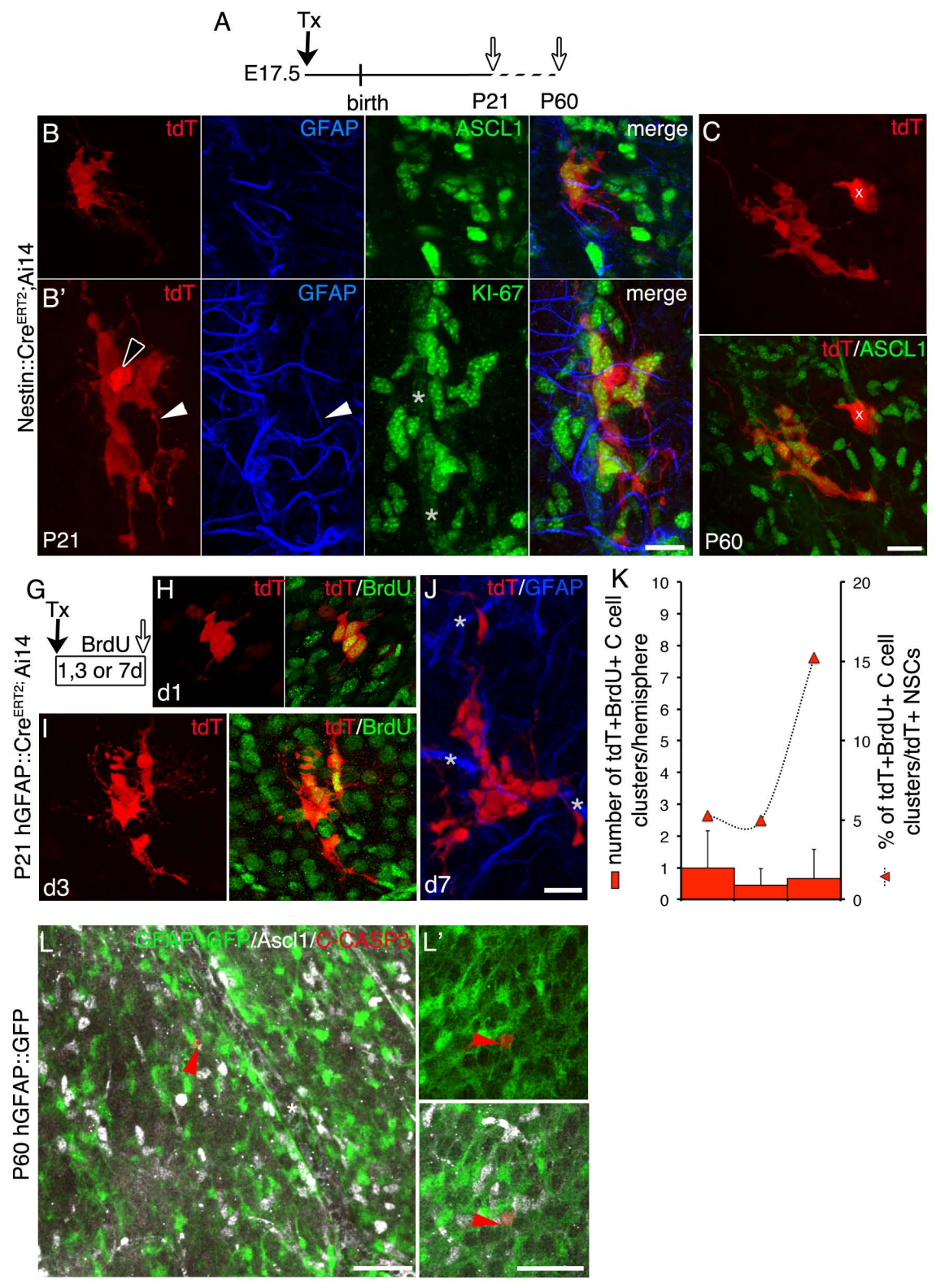
A
Tx
E17.5
birth
P21
P60
B
tdT
GFAP
ASCL1
merge
B'
tdT
GFAP
KI-67
merge
P21
C
tdT
tdT/ASCL1
P60
Nestin::CreERT2;Ai14
G
Tx
BrdU
1,3 or 7d
H
tdT
tdT/BrdU
d1
I
tdT
tdT/BrdU
d3
J
tdT/GFAP
d7
P21 hGFAP::CreERT2; Ai14
K
number of tdT+BrdU+ C cell clusters/hemisphere
% of tdT+BrdU+ C cell clusters/tdT+ NSCs
L
GFAP/GFP/Ascl1/C-CASP3
L'
P60 hGFAP::GFP

Figure S2: NSCs undergo symmetric consuming divisions (related to Fig. 3)
(A) Experimental outline for (B-C): Time-mated pregnant Nestin::Cre^{ERT2};Ai14 mice received one low dose of Tx at E17.5 and V-SVZ whole-mounts of the offspring were prepared at P21 (B) or P60 (C). The majority of tdT+ clusters at P21 were devoid of NSCs (B) whereas a few were mixed (B'; black arrowhead denotes the NSC body; white arrowhead denotes the basal process). (C) A C cell cluster devoid of NSCs at P60. (G) Experimental outline for (H-K): P21 hGFAP::Cre^{ERT2};Ai14 mice received Tx and BrdU (drinking water) for 1, 3 or 7d prior analysis in whole-mount preparations. (H-J) Clusters of tdT+BrdU+ C cells devoid of NSCs, indicating symmetric consuming (differentiation) divisions. (K) Graph shows that the number of tdT+BrdU+ C cell clusters per whole-mount (bars with mean +/- SD; left y-axes) does not change from d1 to d3 to d7 while the percentage of C cell clusters/tdT+ NSCs (percentage of consuming divisions of tdT+ NSCs) increases over time (line plot; right y-axes). (L) CLEAVED_CASPASE-3 staining in whole-mounts of P60 hGFAP::GFP mice revealed low levels of cell death among NSCs (GFAP::GFP+) or C cells (ASCL1+). Scale bars: 20 μm; asterisks indicate blood vessels revealed by antibodies raised in mouse (ASCL1) used on unperfused tissue; x in (C) indicates a tdT+ ependymal cell located at the apical surface.

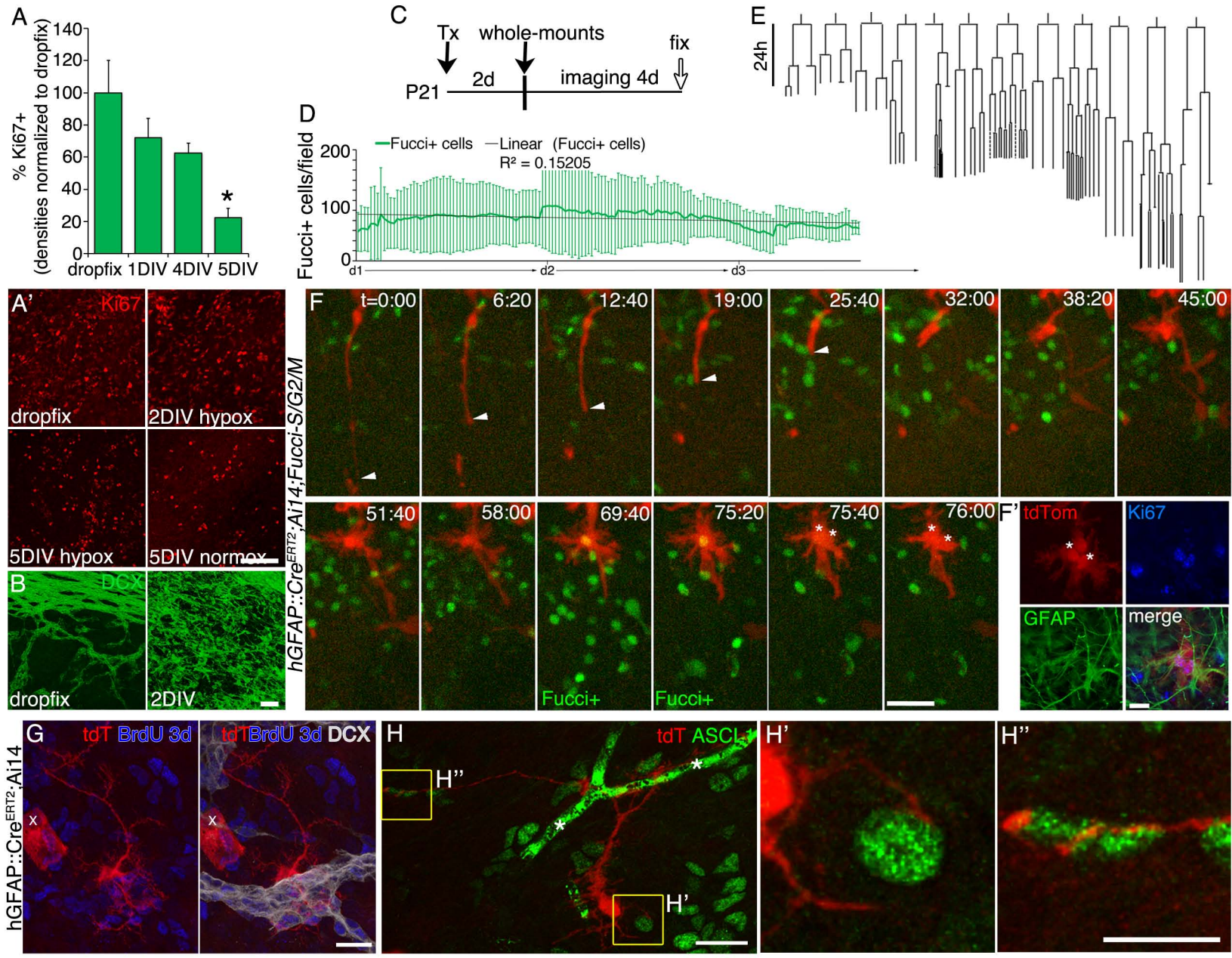
A
% Ki67+
(densities normalized to dropfix)
dropfix 1DIV 4DIV 5DIV
C
Tx
whole-mounts
fix
P21
2d
imaging 4d
D
Fucci+ cells
Linear (Fucci+ cells)
R² = 0.15205
Fucci+ cells/field
d1
d2
d3
E
24h
A'
Ki67
dropfix
2DIV hypox
5DIV hypox
5DIV normox
B
DCX
dropfix
2DIV
F
t=0:00
6:20
12:40
19:00
25:40
32:00
38:20
45:00
51:40
58:00
69:40
75:20
75:40
76:00
Fucci+
Fucci+
hGFAP::Cre^ERT2;Ai14;Fucci-S/G2/M
F'
tdTom
Ki67
GFAP
merge
G
tdT BrdU 3d
tdT BrdU 3d DCX
hGFAP::Cre^ERT2;Ai14
H
tdT ASCL
H'
H''

Figure S3: NSC behavior *in vivo* and *ex vivo* (related to Figure 4)

(A-B) Effects of culture conditions on V-SVZ explants. (A) Density of KI67+ cells (cells/mm^2*10^{-2}) after 1 or 4 days in organotypic explants vitro (DIV) is comparable to that *in vivo* but decreases at 5DIV (p= 0.013 *; two-tailed student's t-test with equal variance). Data are represented as mean +/- SD. (A') Immunostainings for KI67 of explants drop-fixed (*in vivo*), after 2 or 4DIV, or 5DIV under hypoxic or normoxic conditions. Note: normoxic conditions lead to a more severe reduction in the densities of proliferating cells. (B) Typical chain migration of DCX+ neuroblasts seen *in vivo* is disrupted in cultured explants at 2DIV. (C-F) Live-imaging of cells in V-SVZ explants. (C) Experimental outline for (D-F): Postnatal hGFAP::Cre^{ERT2};Ai14 or hGFAP::Cre^{ERT2};Ai14;FucciS/G2/M mice received one injection of Tx; thin slivers of whole-mount preparations were prepared after 2 days, imaged for up to 4 days (also see (A)), and fixed and processed for immunohistochemistry. (D) Levels of proliferation (Fucci) remained constant during live imaging. Data are represented as mean +/- SD. (E) Lineage progression of C cells. C cells divided approximately once a day. Divisions are indicated by horizontal lines, cells followed during imaging by vertical lines (missing vertical lines: cells could not be followed any further). (F) A tdT+ cell with NSC morphology retracts the prominent process (arrowhead) over time, and then divides. Both daughter cells are GFAP+ and KI67+ in post-imaging immunohistochemistry. (G-H) *In vivo*: tdT+ NSCs in whole-mounts from hGFAP::Cre^{ERT2};Ai14 mice (three days after Tx and BrdU). (G) A NSC in contact with BrdU+ cells and neuroblasts. (H) A tdT+ NSC contacts blood vessels

(green) and ASCL1+ cells (green) with its lateral processes (H') and with one of the basal processes (H'') (Note: whereas most NSCs have one basal process, some are bifurcated and harbor two basal processes). Scale bars: 100 μm (A'); 50 μm (B); 20 μm (G-H&F); 10 μm (F'&H"). x in (B) indicates a tdT+ ependymal cell; asterisks indicate blood vessels revealed by antibodies raised in mouse (Ascl1).

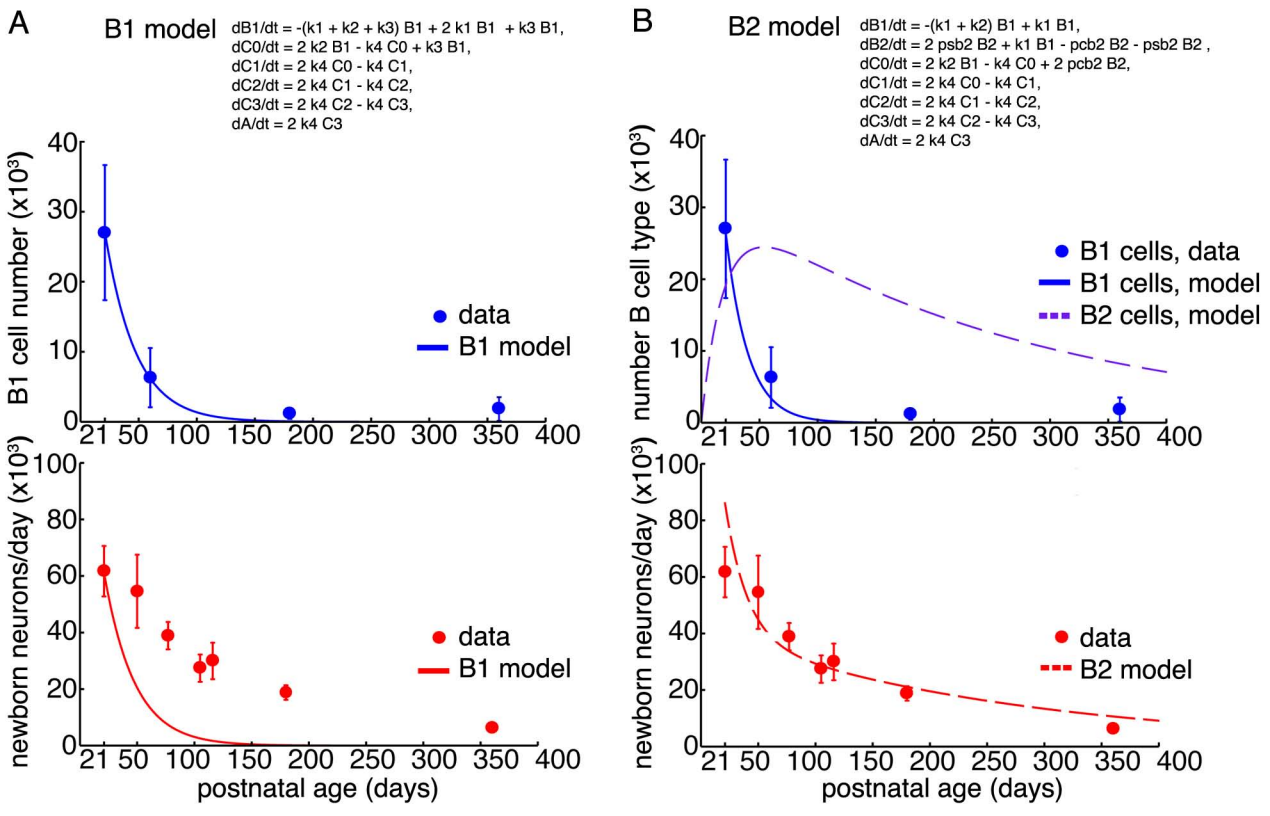
A
B1 model
dB1/dt = -(k1 + k2 + k3) B1 + 2 k1 B1 + k3 B1,
dC0/dt = 2 k2 B1 - k4 C0 + k3 B1,
dC1/dt = 2 k4 C0 - k4 C1,
dC2/dt = 2 k4 C1 - k4 C2,
dC3/dt = 2 k4 C2 - k4 C3,
dA/dt = 2 k4 C3
B1 cell number (x10³)
data
B1 model
newborn neurons/day (x10³)
data
B1 model
postnatal age (days)
B
B2 model
dB1/dt = -(k1 + k2) B1 + k1 B1,
dB2/dt = 2 psb2 B2 + k1 B1 - pcb2 B2 - psb2 B2 ,
dC0/dt = 2 k2 B1 - k4 C0 + 2 pcb2 B2,
dC1/dt = 2 k4 C0 - k4 C1,
dC2/dt = 2 k4 C1 - k4 C2,
dC3/dt = 2 k4 C2 - k4 C3,
dA/dt = 2 k4 C3
number B cell type (x10³)
B1 cells, data
B1 cells, model
B2 cells, model
newborn neurons/day (x10³)
data
B2 model
postnatal age (days)

Figure S4: Mathematical modeling of V-SVZ dynamics and neurogenesis (related to Figure 5)

(A) Population dynamics of B1 cells and neurogenesis rate in base-line (B1) mathematical model. Upper graph: B1 cell number (blue); lower graph: neurogenesis (red). Cell number estimated from experimental data (see Fig. 5D) is indicated as dots, model prediction as lines. (B) Population dynamics of B1 and B2 cell populations (upper graph) in a revised mathematical (B2) model based on the rates of neurogenesis seen *in vivo* (Fig. 5D) (lower graph). Data are represented as mean +/- SD.

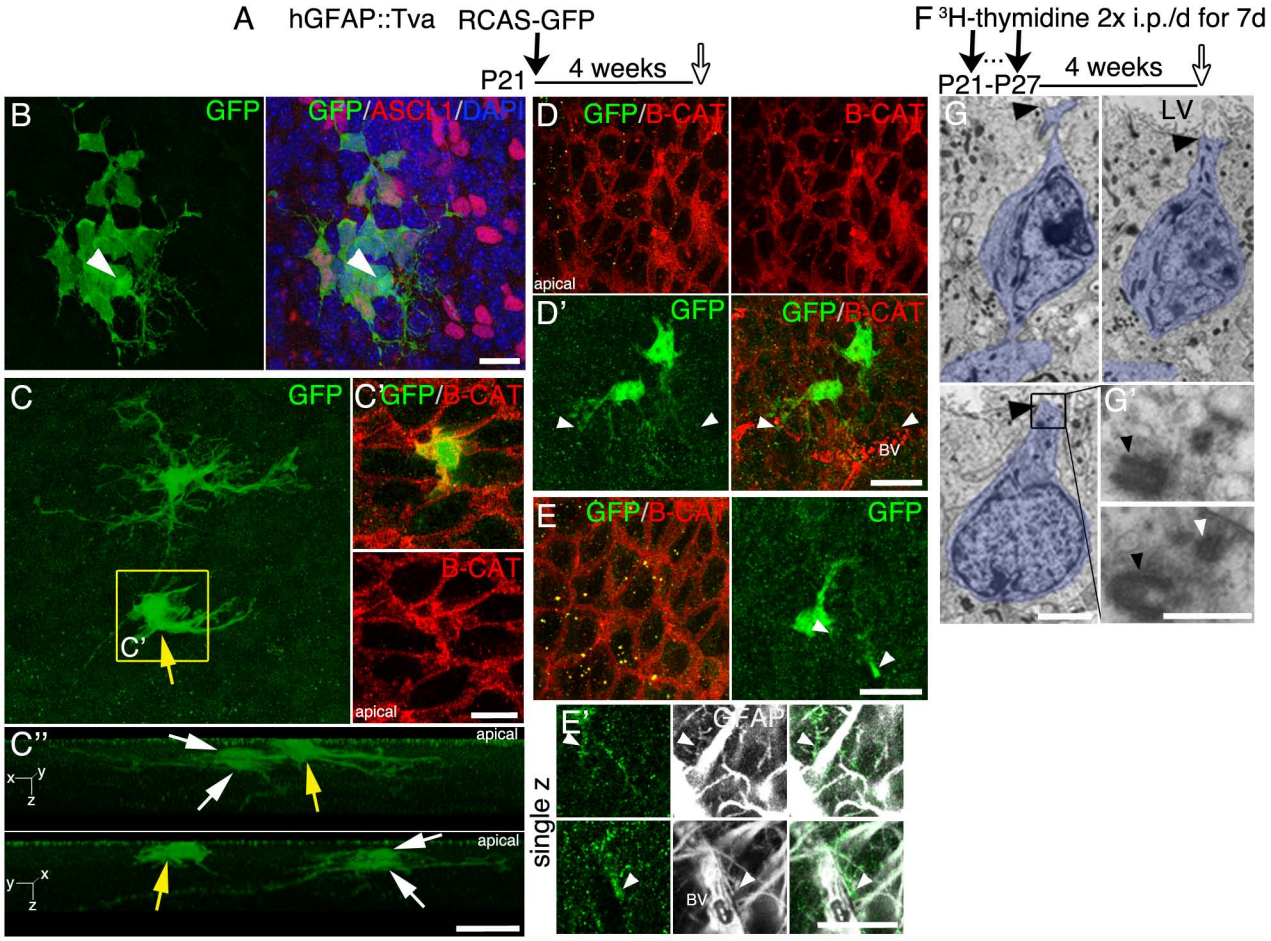
A
hGFAP::Tva
RCAS-GFP
P21
4 weeks
F
3H-thymidine 2x i.p./d for 7d
P21-P27
4 weeks
B
GFP
GFP/ASCL1/DAPI
C
GFP
C'
C'
GFP/B-CAT
B-CAT
apical
C''
apical
apical
D
GFP/B-CAT
B-CAT
apical
D'
GFP
GFP/B-CAT
BV
E
GFP/B-CAT
GFP
E'
GFAP
single z
BV
G
LV
G'

Figure S5: B1 cells give rise to B2 cells (related to Figure 6)

(A) Experimental outline for (B-E): Postnatal hGFAP::Tva mice were injected intraventricularly with RCAS-GFP retrovirus and V-SVZ whole-mounts were prepared after 4 weeks. (B) A mixed GFP+ cluster consisting of ASCL1+ C cells, neuroblasts, and a NSC (arrowhead). (C) A triplet of GFP+ cells: one cell is a B1 cell in contact with the LV (C' and yellow arrow in C") whereas two other cells appear to be without apical contact (white arrows in C"). (C") shows 3D view of cells in (C). Note the lack of a long basal process of the apical B1 cells shown in (C'). (D) A pair of non-apical GFP+ cells contacting blood vessels (BV, arrowheads in D'). (E) A non-apical GFP+ cell contacting blood vessels (BV, arrowhead in E' lower panel) and expressing GFAP (E'). (F) Experimental outline for (G): Postnatal hGFAP::GFP mice received two i.p. injections of ^{3}H-thymidine per day for 7d. Ultrathin (TEM) coronal sections were analyzed after 4 weeks. (G) Pseudo-colored serial TEM micrographs of a ^{3}H-thymidine+ LRC (autoradiography not shown) contacting the LV (black arrowheads). (G') Apical contact boxed in (G) containing the primary cilium (white arrowhead) and its daughter centriole (black arrowheads). Scale bars: 15 μm (C"), 2 μm (G), 500 nm (G'); all others 20 μm.

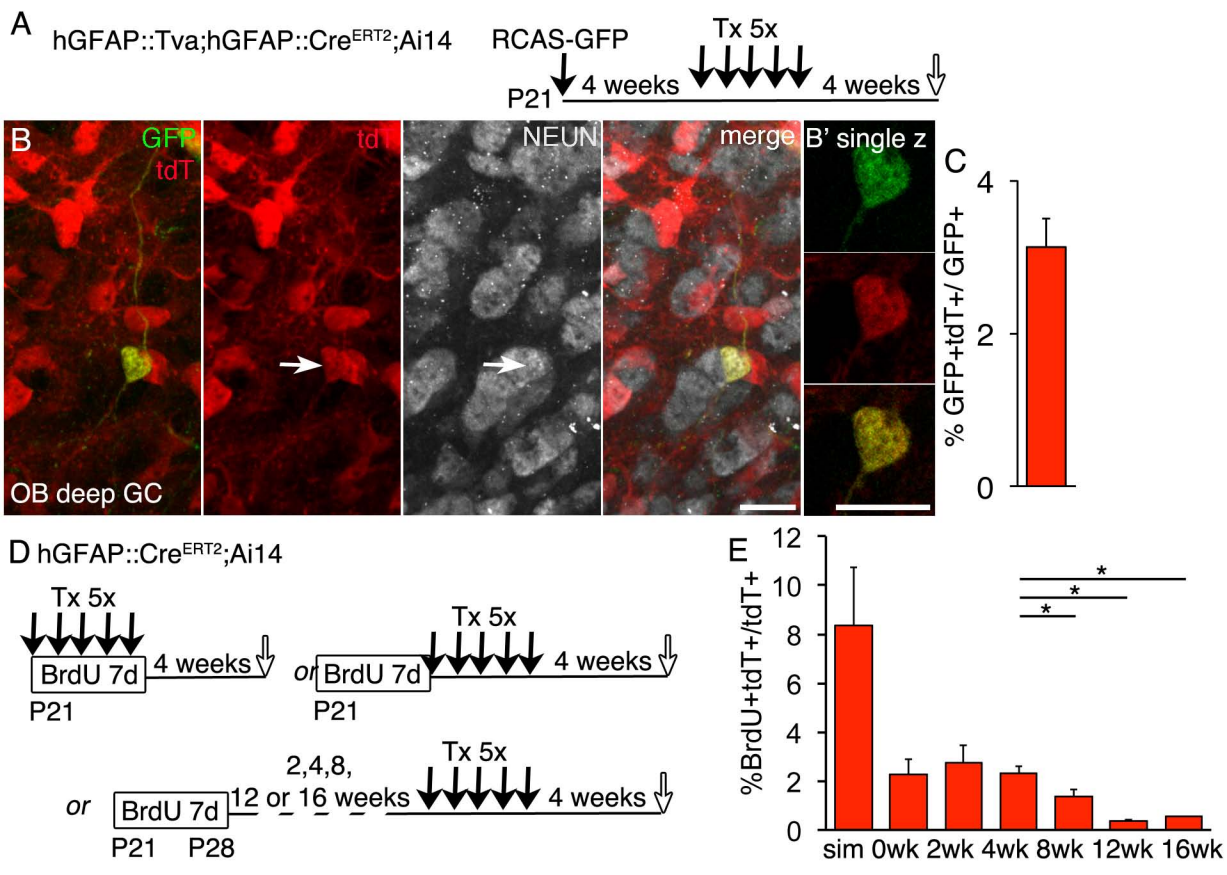
A
hGFAP::Tva;hGFAP::Cre^ERT2;Ai14
RCAS-GFP
Tx 5x
P21
4 weeks
4 weeks
B
GFP
tdT
tdT
NEUN
merge
B' single z
OB deep GC
C
% GFP+tdT+/ GFP+
4
2
0
D hGFAP::Cre^ERT2;Ai14
Tx 5x
BrdU 7d
4 weeks
P21
or
Tx 5x
BrdU 7d
4 weeks
P21
or
BrdU 7d
P21 P28
2,4,8,
12 or 16 weeks
Tx 5x
4 weeks
E
%BrdU+tdT+/tdT+
12
10
8
6
4
2
0
*
*
*
sim 0wk 2wk 4wk 8wk 12wk 16wk

Figure S6: Sporadic reactivation of GFAP+ NSCs (related to Figure 7)

(A) Experimental outline for (B,C): Postnatal hGFAP::Tva mice were injected intraventricularly with RCAS-GFP retrovirus and 4 weeks later received one daily injection of Tx for 5 days. Coronal sections of the OB were prepared 4 weeks later. (B) A GFP+tdT+ granule neuron born from a reactivated GFAP+GFP+ cell. (C) Percentage of GFP+tdT+ OB interneurons born 4 weeks (or later) after viral injection at P21. (D) Experimental outline for (E): Postnatal hGFAP::Cre^{ERT2};Ai14 mice received BrdU (drinking water) for one week (P21-P28) and one daily injection of Tx for five days after different intervals (simultaneously (sim) or immediately after (0wk) BrdU, or 2, 4, 8, 12, or 16 weeks after BrdU). Coronal sections of the OB were prepared 4 weeks after the last Tx injection. (E) Percentages of BrdU+tdT+ interneurons generated from cells recombining the Ai14 reporter when Tx was given after the different intervals indicated on the X-axis (4wk vs. 8wk, p= 0.04*; 4wk vs. 12 wk, p= 0.0004***; 4wk vs 16 wk, p= 0.004**; one-tailed student's t-test with equal variance). Data are represented as mean +/- SD. Scale bar: 15 μm; white arrows in B indicate GFP+tdT+NEUN+ interneurons.

Tables

Table S1: Summary of modes of division (related to Figs. 1-3)

	all events	C clusters	mix. clusters	B1 cell pairs	B1 cells
	short-term lineage tracing				
CRE^{ERT2} d3	(18)	83% (15/18)	0 (0/18)	17% (3/18)	nd
RCAS d2-d3	(75)	75% (56/75)	0 (0/75)	6% (1/18)	25% (18/75)
RCAS d4-d7	(97)	85% (82/97)	*1% / 7% (1/82 C) (1/15 B)*	27% (4/15)	15% (15/97)
	long-term lineage tracing				
RCAS 2 wks	(16)	88% (7/8)	*12% (1/8)*	25% (2/8)	(8)
RCAS 4 wks	(37)	63% (5/8)	*37.5% / 10% (3/8 C) (3/29 B)*	14% (4/29)	(29)
CRE^{ERT2} (E17.5- P21)	(11)	70% (8/11)	*30% (3/11)*	nd	nd
	Mathematical modeling				
	consumption 70%			symmetric self-renewal 30%	

Table S1: Summary of modes of division (related to Figs. 1-3)

Summary of the modes of division observed in postnatal lineage-tracing experiments (RCAS or CRE^{ERT2}) and predicted by mathematical modeling. For short-term experiments (d1-d7), ratios of divisions generating one or two NSCs to those generating clusters of progeny were analyzed. For long-term lineage tracing, cluster composition was analyzed. Data indicated in italics refer to findings whose interpretation is less clear (see text for details). Indicated are percentages of occurrences; total event counts are given in parenthesis.

Table S2: Short-term RCAS-based lineage tracings (related to Figs. 1 & 2)

whole-mount	day	Sample	GFP+ B1 cells (secondary)	GFP+ B1 cell pairs (tertiary)	Clusters	Cluster size (cells/cluster) and composition
1	2	1293	0	0	1	5 C
2	2	1433	1	0	3	1 C, 1 C, 2 C
3	2	1433c	0	0	1	3 C
4	2	1428	0	1	0	0
5	2	no2d2	2	0	10	1 C, 4 C, 2 C, 4 C, 5 C, 3 C, 2 C, 2 C, 5 (3 C+2A), 7 (5C+2A)
6	2	no4d2	2	0	10	2,1,1,1,1,2,2,4,2,5 (all C)
7	2	no3d2	1	0	11	1,4,4,1,2,4,2,1,2,3,5 (all C)
8	3	1298	0	0	3	2 C, 4 C, 2 C
9	3	1427	2	0	1	8 (6 C + 2 A)
10	3	1429	0	0	0	0
11	3	no2d3	1	0	3	2 C, 2 C, 4 C
12	3	no1d3	0	0	2	3 C, 2 C
13	3	no3d3	2	0	2	2 C, 2 C
14	3	5076	5	0	3	3 C, 4 C, 2 C
15	3	5075	1	0	6	2 C, 1 C, 3 C, 4 C, 2 C, 2C
16	3	5081	0	0	0	0
17	4	1350	0	0	3	8 C, 9 C, 4 (2 C + 2 A)
18	4	1352	1	0	1	2 C
19	4	1348	0	0	1	6 C
20	5	no3d5	0	1	6	3 C, 5 (4 C + 1 A), 4 C, 2 C, 8 C, 1 C
21	5	no1d5	3	1	16	4,4,2,2,15,10,2,5,6,10,2,2,2,7,16,5 (all C)
22	5	no1cd5	1	0	5	6 C, 12 C, 4 C, 4 C, 5 (1B+4C; mixed cluster)
23	5	no2d5	1	0	5	9,2,2,6,4 (all C)
24	6	no41d6	1	0	7	4,6,4,2,8,6,5 (all C)

25	6	no41cd6	0	0	1	3 C
26	6	no42d6	0	0	2	2 C, 2 C
27	6	no42cd6	0	0	5	2 C, 2 C, 10 (8C + 2 A), 2 C, 3 C
28	6	no39d6	0	0	10	2,4,2,1,2,6,5,4,4,4 (all C)
29	7	no2d7	2	0	0	0
30	7	no4d7	0	0	1	4 C
31	7	no3d7	1	1	12	8,4,4,4,1,1,1,1,2,2,1,2 (all C)
32	7	no1d7	1	1	7	5,1,4,3,2,6,3 (all C)

Table S2: Short-term RCAS-based lineage tracings (related to Figs. 1 & 2)

RCAS-GFP was injected into hGFAP::Tva mice at P21 and GFP+ cells per V-SVZ whole-mount were quantified after two to seven days (day = days after RCAS injection; Clusters = C cell-only clones or clones containing C cells and A cells clearly associated with the cluster; migrating A cells are excluded from further analysis).

Table S3: Short-term CRE^{ERT2}-based lineage tracings (related to Fig. 3)

whole-mount	Sample	tdT+ B1 cells	tdT+BrdU+ B1 cells	tdT+BrdU+ B1 cell pairs	tdT+BrdU+ clusters	Cluster size (cells/cluster) and composition
1	1578	38	2	1	2	8 C, 15 C
2	1575	26	4	0	2	7 C, 8 C
3	1570	62	4	1	0	0
4	1575	20	0	0	1	4 C
5	1572	58	3	1	3	2 C, 2 C, 2 C
6	1582	106	9	0	2	18 C, 2 C
7	1583	55	1	0	1	4 C
8	1578	100	6	0	4	4 C, 4 C,8 C, 5 C
9	1649	21	0	0	0	0

Table S3: Short-term CRE^{ERT2}-based lineage tracings (related to Fig. 3)

Tamoxifen (6.66 mg/Kg BW) was injected into hGFAP::Cre^{ERT2};Ai14 mice at P21 and mice received BrdU three days before analysis. Cells labeled by tdT or tdT/BrdU were quantified per V-SVZ whole-mount (Clusters = C cell-only clones or clones containing C cells and A cells clearly associated with the cluster; migrating A cells are excluded from further analysis).

Table S4: Short-term CRE^{ERT2}-based lineage tracings (related to Fig. 3)

whole-mount	day	Sample	tdT+ B1 cells	tdT+BrdU+ B1 cells	tdT+BrdU+ B1 cell pairs	tdT+BrdU+ clusters	Cluster size (cells/cluster) and composition	ratio tdT+ B1 cells/C clusters
1	1	1454	20	1	0	0	0	-
2	1	1455	3	0	0	0	0	-
3	1	1449R	21	2	0	2	3 C, 3 C	10.5
4	1	1449L	14	2	0	2	4 C, 1 C	7
5	3	1324R	12	1	0	1	2 C	12
6	3	1324L	17	3	0	1	1 C	17
7	3	1322	19	2	0	1	8 C	19
8	3	1285	16	0	0	0	0	-
9	3	1276	5	0	0	1	6 A cells (excluded)	5
10	3	1274R	5	0	0	0	0	-
11	3	1274L	12	0	0	0	0	-
12	3	1571	14	1	0	0	0	-
13	3	1569	15	2	0	0	0	-
14	3	1647	4	0	0	1	16 C	4
15	3	1652	5	0	0	0	0	-
16	7	773R	12	2	0	0	0	-
17	7	773L	9	1	0	2	26 (18 C + 8 A), 6A (excluded)	4.5
18	7	775R	3	1	0	0	0	-
19	7	775L	1	0	0	1	10 C	1
20	7	777R	1	?	0	1	5 C	1
21	7	777L	1	0	0	0	0	-
22	7	3	1	0	0	0	0	-
23	7	4R	7	3	0	0	0	-
24	7	4L	2	0	0	0	0	-

25	7	5	3	0	0	0	0	-
26	7	6	1	0	0	0	0	-
27	7	8	1	0	0	0	0	-
28	7	757R	13	2	0	0	0	-
29	7	757L	9	nd	-	1	12 (8 C + 4 A)	9
30	7	762	3	nd	-	2	7 C, 6 C	1.5
31	7	763R	3	nd	-	0	0	-
32	7	763L	6	nd	-	1	17 (4 C+13 A)	6
33	7	753	4	nd	-	0	0	-
34	7	758	5	nd	-	3	13 C, 10 A (excluded), 5 A (excluded)	1.7
35	7	3	8	3	0	1	13 C	8

Table S4: Short-term CRE^{ERT2}-based lineage tracings (related to Fig. 3)

Tamoxifen (1.67-3.33 mg/Kg BW) was injected into hGFAP::Cre^{ERT2};Ai14 mice at P21 and mice received BrdU for one, three, or seven days before analysis. Cells labeled by tdT or tdT/BrdU were quantified per V-SVZ whole-mount (Clusters = C cell-only clones or clones containing C cells and A cells clearly associated with the cluster; migrating A cells are excluded from further analysis).

Table S5: Distribution of GFP+ cells in the OB (related to Fig. 6)

region\sample	#732	#767	#765	#768	#731	#766	%GFP+
total GFP+/OB	160	93	203	227	411	64	-
PGL	5	4	3	1	nd	nd	2.34 +/- 0.86
GCL	138	81	168	222	nd	nd	88.48 +/- 3.25
deep GCL	85	69	148	209	nd	nd	73.07 +/- 7.96
superf. GL	53	12	20	13	nd	nd	15.4 +/- 6.09
IPL	3	0	0	0	nd	nd	0.47 +/- 0.47
mitral	6	0	2	0	nd	nd	1.18 +/- 0.89
EPL	1	1	1	0	nd	nd	0.55 +/- 0.22
core	7	2	16	4	nd	nd	4.04 +/- 1.4
core, A cells	0	6	12	0	nd	nd	3.09 +/- 1.79

Table S5: Distribution of GFP+ cells in the OB (related to Fig. 6)

The OBs from hGFAP::Tva mice that had received RCAS-GFP at P21 and BrdU at P49-P55 were analyzed four weeks after BrdU administration for the presence of GFP+ cells. Shown are the numbers of GFP+ cells in serial sections and their layer distribution. The percentages of GFP+ cells found in specific layers are shown in bold. PGL periglomerular layer; GCL granule cell layer; IPL internal plexiform layer; EPL external plexiform layer.

Table S6: Distribution of GFP+BrdU+ cells in the OB (related to Fig. 6)

region\sample	#732	#767	#765	#768	#731	#766	%GFP+ BrdU+
total GFP+BrdU+/OB	8	11	11	11	11	2	-
PGL	0	0	0	0	nd	nd	0
GL	8	10	11	11	nd	nd	97.73 +/- 2.27
deep GCL	4	9	10	10	nd	nd	78.41 +/- 9.71
superf. GCL	4	1	1	1	nd	nd	19.32 +/- 10.23
IPL	0	0	0	0	nd	nd	0
mitral	0	0	0	0	nd	nd	0
EPL	0	0	0	0	nd	nd	0
core	0	1	0	0	nd	nd	2.28 +/- 2.28
core, A cells	0	0	0	0	nd	nd	0

Table S6: Distribution of GFP+BrdU+ cells in the OB (related to Fig. 6)

The OBs from hGFAP::Tva mice that had received RCAS-GFP at P21 and BrdU at P49-P55 were analyzed 4 weeks after BrdU administration for the presence of GFP+BrdU+ cells. Shown are the numbers of GFP+BrdU cells in serial sections and their layer distribution. The percentages of GFP+BrdU+ cells found in specific layers are shown in bold. PGL periglomerular layer; GCL granule cell layer; IPL internal plexiform layer; EPL external plexiform layer.

Supplementary movies

Movie S1: A self-renewing NSC maintains its prominent (basal) process during division (related to Fig. 5B)

A tdT+ NSC divides *ex vivo* and generates a daughter cell with NSC morphology that grows a prominent process in parallel to the process of its mother cell. Images acquired every 35 minutes. Time is in hh:mm:ss.

Movie S2: A self-renewing NSC maintains its prominent (basal) process and outgrowth additional processes during division (related to Fig. 5C)

A tdT+ NSCl divides *ex vivo* and generates a daughter cell with NSC morphology. Images acquired every 35 minutes. Time is in hh:mm:ss.