pH3 **Sox2** **CldU** **Merge**

Figure S1. Tectal NPC description

Xenopus tectal progenitors reside along the ventricular midline. A) pH3 labeling (red) demonstrates cells in M-phase dividing along the midline. B) Sox2 labeling (green) shows that Sox2+ progenitor cells reside along the midline. C) CldU (blue) is incorporated into dividing cells along the midline. D) Merge demonstrates that ventricular midline cells are Sox2 positive and are the dividing population in the tectum. Single optical sections are shown. Scale, 20um.
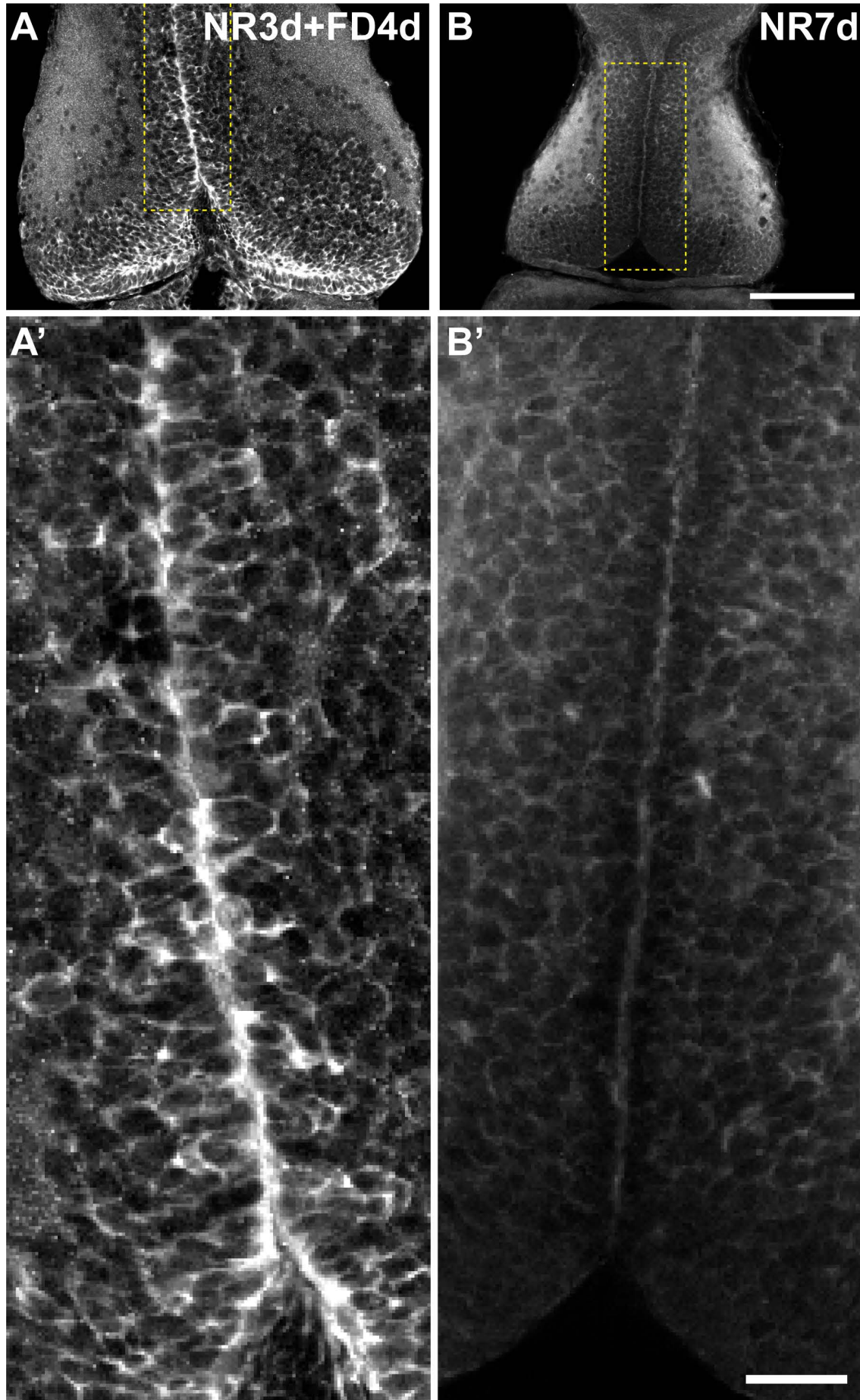
Figure S2. Phospho-ribosomal S6 is decreased in nutrient-restricted progenitors
Whole-mount immunofluorescent images of p-rS6 staining in optic tecta from fed (A, A')
and nutrient restricted (B, B') animals. Confocal images were collected under identical
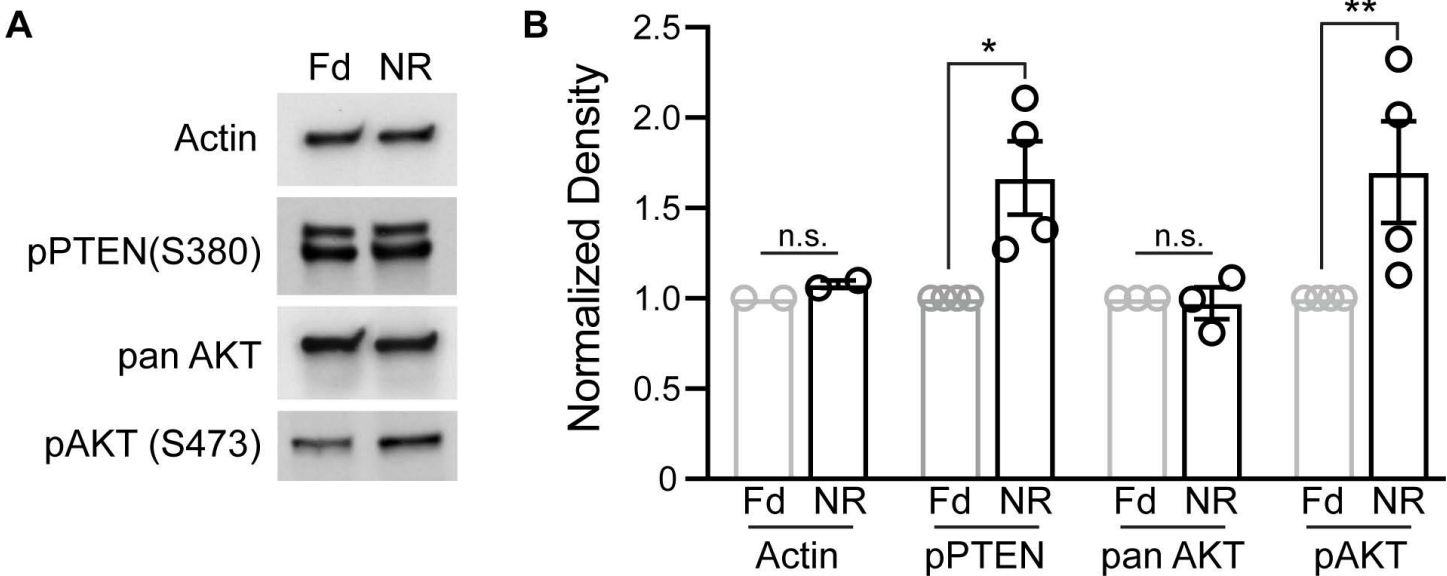imaging parameters, single optical sections shown. Scale, 100um (A,B), 20um (A',B').

Figure S3. Nutrient restriction affects signaling components upstream of mTOR

A) Western blot analysis of NR (3d) and Fed (NR2d +Fd 24h) brains . Nutrient restriction increases p-PTEN (S380) and p-AKT (S473) as compared to Fed animals, but does not change pan AKT levels or Actin. B) Quantification of Western blots shown in A. Samples were corrected for protein loading against PonceauS and then NR (black) were normalized against Fed groups (gray) within each experiment. Results for each condition are shown as individual data points and mean ± SEM are shown as black bars. n=5 brains per treatment per timepoint, a minimum of 3 biological replicates was used for each quantification shown. *p<0.05, **p<0.01, n.s.= not significant, see Table S1.
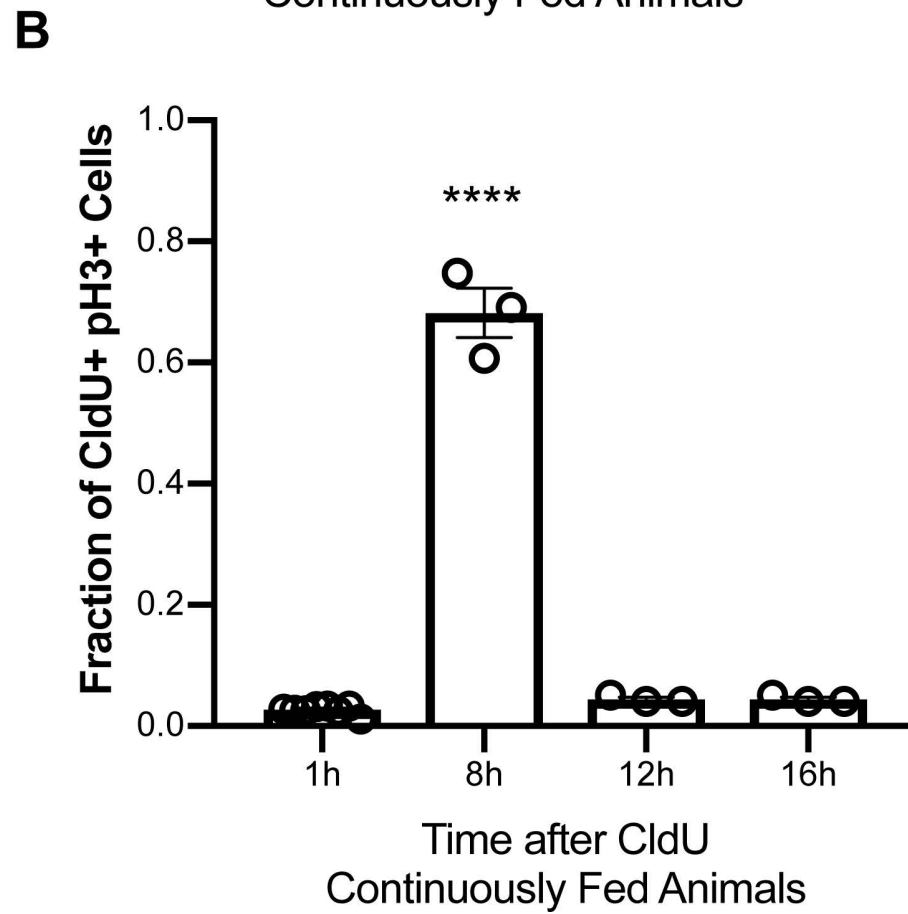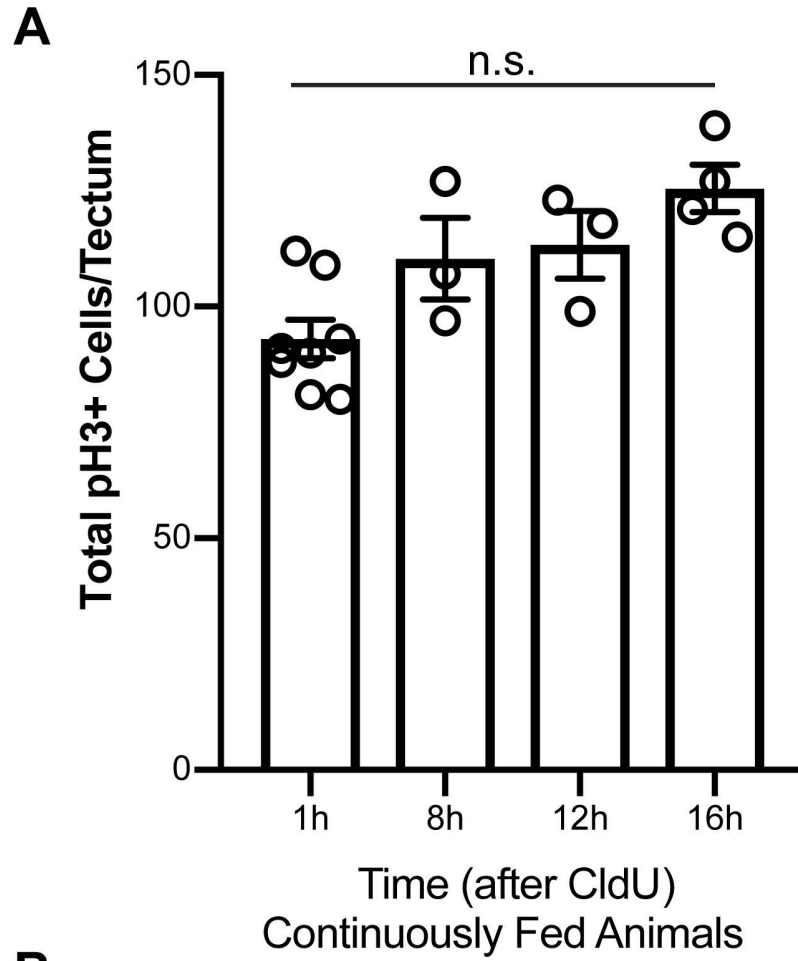
A

B

Figure S4. Cell cycle timing in control fed animals

A) Average number of cells in M-phase in control fed animals. Continuously fed animals were given a 1h pulse of CldU and then fixed at 1, 8, 12, and 16h and processed for immunofluorescence. pH3+ cells were counted throughout the tectum. At any given time, approximately 100 cells are in M-phase in the control fed condition. Note that time is based on CldU administration despite CldU not being measured in this graph. Animals are the same as in B. B) Determination of G2 length in control fed animals. Continuously fed animals were given a 1h pulse of CldU and then fixed at 1, 8, 12, and 16h for CldU and pH3 immunofluorescence. The fraction of double labeled cells (pH3+CldU+/all pH3+) was calculated. The peak of double-labeled cells at 8h shows when most of the previously CldU-labeled cells enter M-phase, indicating the length of G2. ****$p<0.0001$, see Table S1.
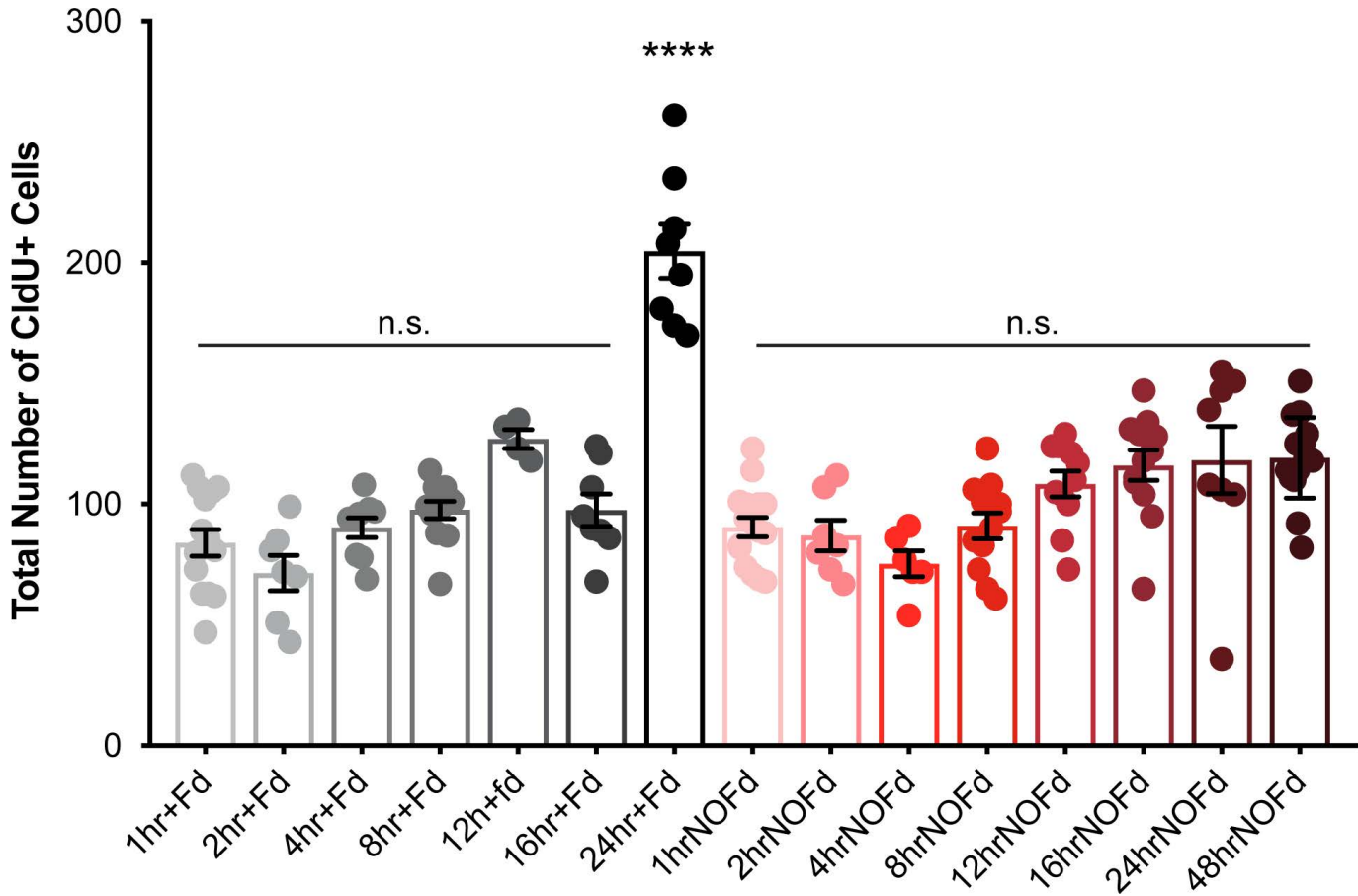
Figure S5. Nutrient restriction causes NPCs to accumulate in G2

CldU+ data from experiment shown in Figure 5. Stage 47 animals were subjected to 2 days of nutrient restriction and then treated with a 1hr pulse of CldU with or without food. Animals were sacrificed at 1, 2, 4, 8, 12, 16, 24h, and 48h after feeding and brains were fixed and processed for CldU immunolabeling. At 24h, the labeled cells in the fed animals have doubled, but even at 48h, the labeled cells in the NR animals have remained unchanged, demonstrating that cells labeled in S-phase are not entering M-phase. ****$p<0.0001$, see Table S1.
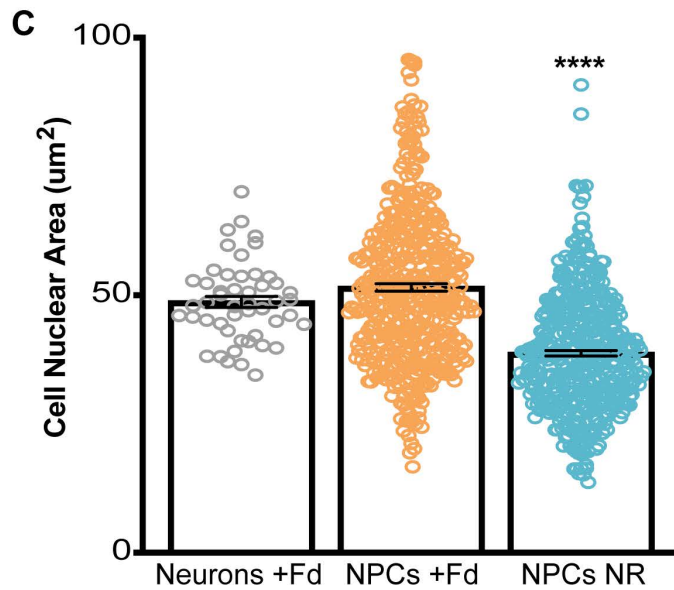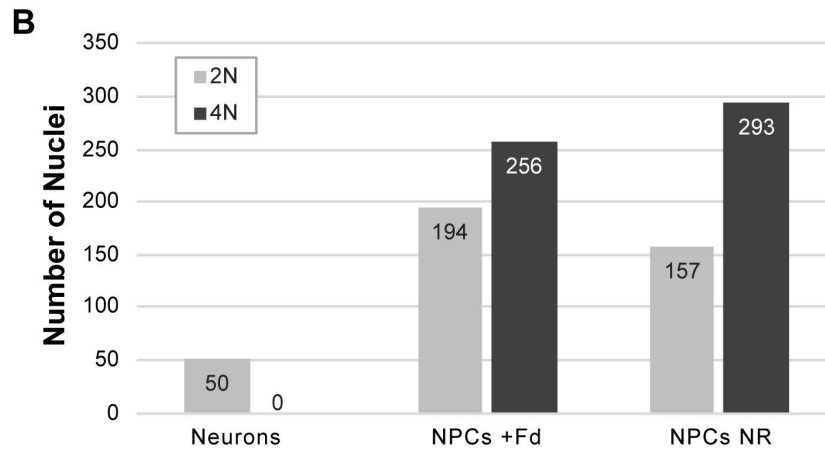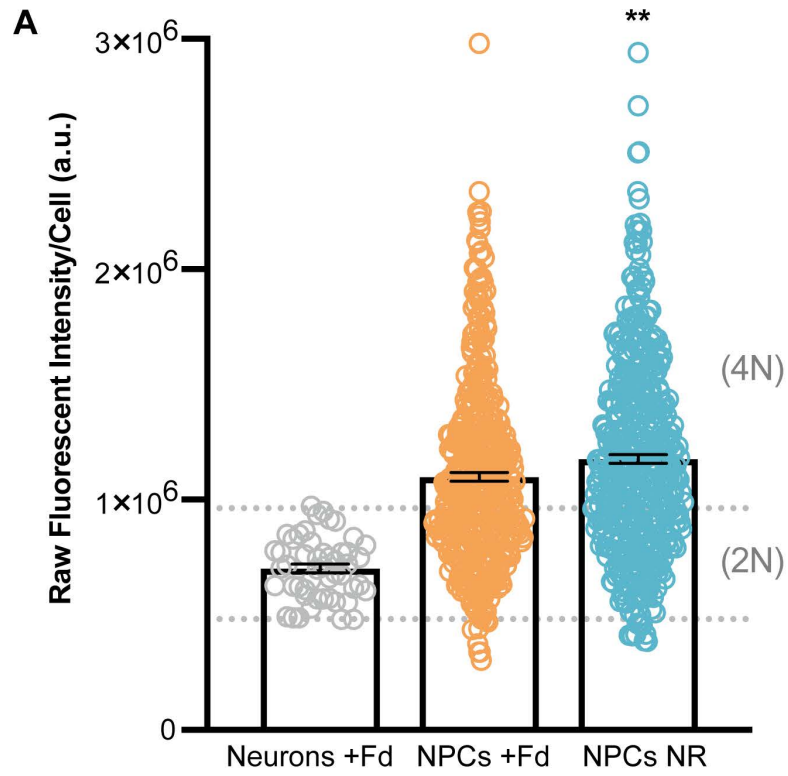
Figure S6. Nutrient-restricted progenitors have increased DNA content and decreased cell size

Data supplement for Figure 7 G-I. A) Raw fluorescent intensity numbers per cell soma in neurons (gray, n=50), fed NPCs (orange, n=450), and NR NPCs (blue, n=450). Gray dotted lines demarcate the range of fluorescence in the neuron population, designated as 2N. Anything above the brightest neuron is categorized as 4N. B) Number of cells from A that fall into the 2N (gray) and 4N (black) bins. C) Area measurements for cell soma in A and Fig 7I. **p<0.01, ****p<0.0001, see Table S1.

Table S1

Figure	Groups	Animals	Animals per group	DFn	DFd	F	ANOVA p-value	Tukey's post hoc	Comments
2	16	304	6-47	15	288	171.4	<0.0001	yes	
3 (B-C)	4	100	25	3	27	60.3	<0.001	yes	
3 (D-E)	12	150	8-17	11	138	56.72	<0.0001	yes	
4	4	60	15	3	56	11.42	<0.0001	yes	
5	15	150	7-16	14	135	21.2	<0.0001	yes	
6	2*	14	7	n/a	n/a	n/a	<0.0001*	no	* *t* test
7 (A-F)	2*	33	16-17	n/a	n/a	n/a	<0.0001*	no	* *t* test
7 (G-I)	2	20	10#	2	947	615.2	<0.0001	yes	# 500 cells/group
8	6	160	11-47	5	154	78.21	<0.0001	yes	
S3	2	130	65	n/a	n/a	n/a	<0.01^	no	^ Wilcoxon S-R
S4	4	17	3-8	3	13	429	<0.0001	yes	
S5	15	150	7-16	14	135	21.2	<0.0001	yes	same as Fig 5
S6	2	20	10#	2	947	33.03	<0.0001	yes	same as Fig 7