Expanded View Figures

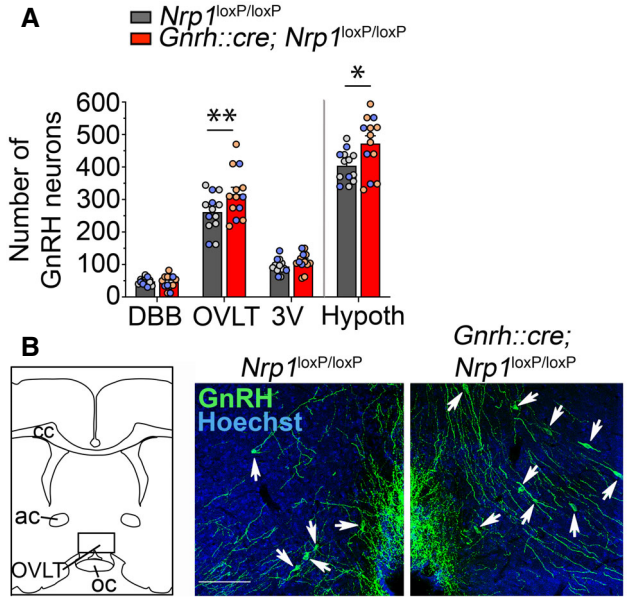

Figure EV1. Migration of supernumerary GnRH neurons in adulthood in the hypothalamus of mice lacking neuropilin-1 expression in GnRH neurons.

A Total number of GnRH neurons in the forebrain and the hypothalamus (Hypoth) and their regional distribution (DBB: diagonal band of Broca; OVLT: organum vasculosum of the lamina terminalis; 3V: periventricular area of the median preoptic) in *Nrp1*$^{loxP/loxP}$ and *Gnrh::cre; Nrp1*$^{loxP/loxP}$ littermates using conventional immunofluorescence. Individual males and females used for this analysis are represented by blue and gray/light-red dots, respectively. Two-way ANOVA, Fisher's LSD multiple-comparison test, n = 13 (nine females and four males).

B Representative immunohistofluorescence for GnRH (green) and Hoechst staining (blue) of sections showing the OVLT region (area framed in the schematic) from control (left) and mutant (right) adult mice. More GnRH cells are observed in OVLT sections of mutant mice (white arrow). Ac, anterior commissure; cc, corpus callosum; Scale bar: 100 μm.

Data information: Bar graphs show individual values and means ± SEM. *P < 0.05, **P < 0.01.

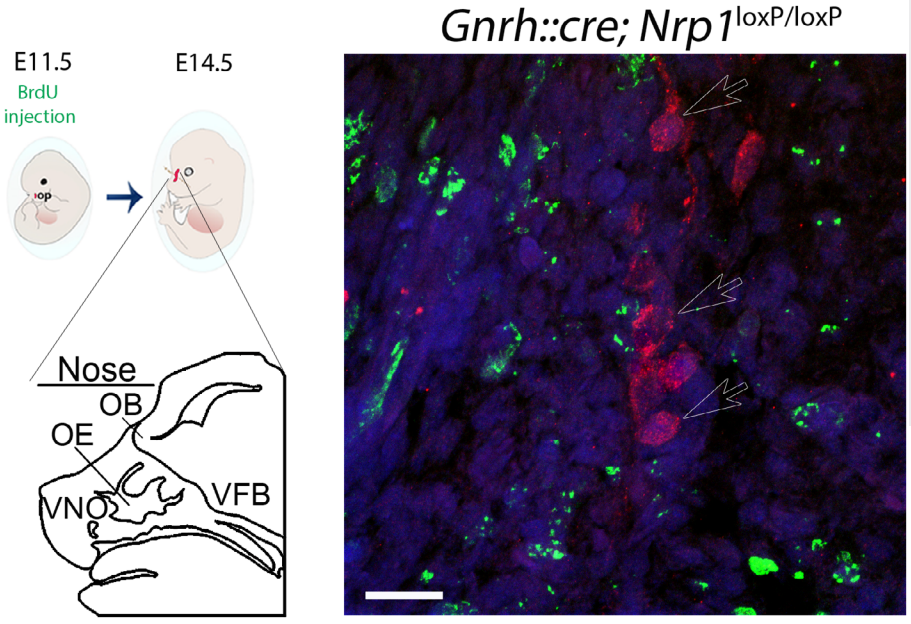

Figure EV2. Knocking out neuropilin-1 in GnRH-expressing cells does not alter cell cycle exit.

Left panels. Schematic diagram illustrating BrdU treatment of dams carrying mutant and control mouse embryos at E11.5 and their collection 3 days later (E14.5), and schematic representation of key anatomical landmarks in the head of an E14.5 embryo (VNO, vomeronasal organ; OE, olfactory epithelium, OB, olfactory bulb; VFB, ventral forebrain). *Right panel.* As in *Nrp1*$^{loxP/loxP}$ control embryos, no BrdU immunofluorescence (green) was detected in GnRH-positive cells (red) in *Gnrh::cre;Nrp1*$^{loxp/loxp}$ mutant embryos at E14.5 (n = 3 per genotype from two independent litters). Empty arrows show GnRH-immunoreactive neuronal cell bodies, which all lack BrdU labeling. Scale bar: 20 μm.

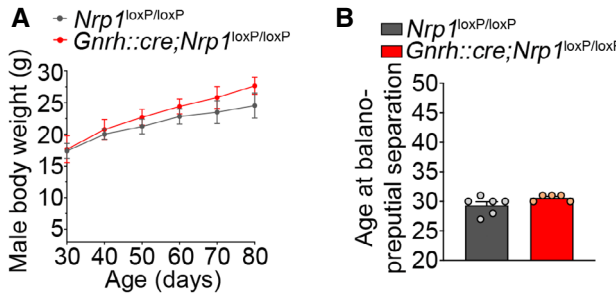

Figure EV3. Male mice lacking neuropilin expression in GnRH neurons show no alteration in body weight evolution and initiation of sexual maturation.

A Evolution of body weight in control and mutant male littermates from 30 to 80 days of age. Two-way ANOVA, n = 3–5 mice.

B Age at balanopreputial separation in males. Mann–Whitney U-test, n = 5–6 mice.

Data information: Bar graphs show individual values and means ± SEM.

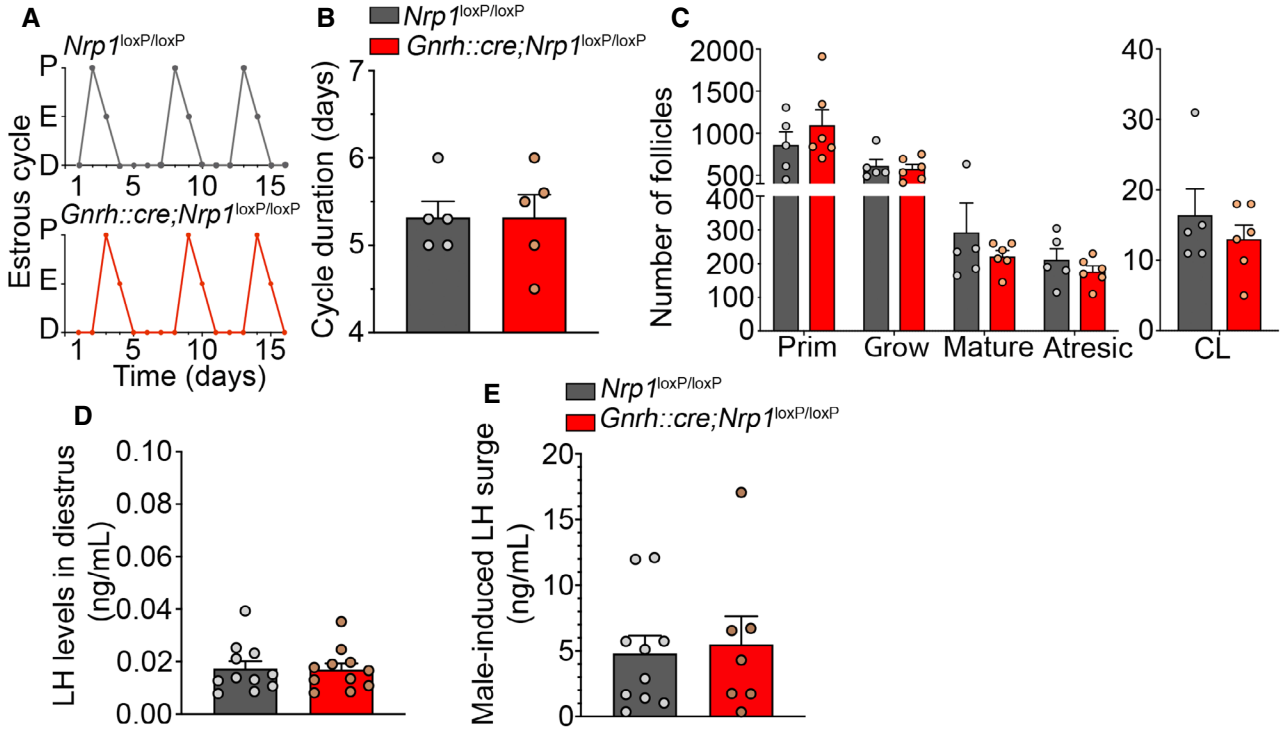

Figure EV4. Female mice lacking neuropilin-1 expression in GnRH neurons show no alteration of adult reproductive physiology.

A Representative estrous cycles from an *Nrp1*$^{loxP/loxP}$ control female (top) and a *Gnrh::cre;Nrp1*$^{loxP/loxP}$ mutant female (bottom). D = diestrus, E = estrus, P = proestrus.

B Estrous cycle duration. Unpaired t-test, n = 5.

C Number of follicles in both ovaries in control and mutant females. Prim, primordial; Grow, growing; CL, corpora lutea. Two-way ANOVA, n = 5–6.

D Nadir LH levels in diestrus. Mann–Whitney U-test, n = 11.

E Surge levels of LH induced by male odors. Unpaired t-test, n = 7–10.

Data information: Bar graphs show individual values and means ± SEM.