

Fig. S1. Slit1 expression during GnRH neuron migration and GnRH neuron number in mice lacking Slit1 and Slit2. (A-I) Expression of Slit1 in the nasal compartment and in the forebrain of embryonic mouse heads. Contiguous sagittal sections taken from E14.5 *Slit1*^{+/-} animals were colabelled with anti-GFP and TuJ1/GnRH antibodies to visualise co-expression of Slit2 and TuJ1 in nasal axons and forebrain neurons (A-C) and migrating GnRH neurons (D-I), respectively. Arrows in B,E,H indicate the areas shown at higher magnification in C,F,I. NC, nasal compartment; OB, olfactory bulb; FB, forebrain; MPOA, medial preoptic area. Scale bar: 150 μ m in A,B,D,E,G,H; 50 μ m in C,F,I.

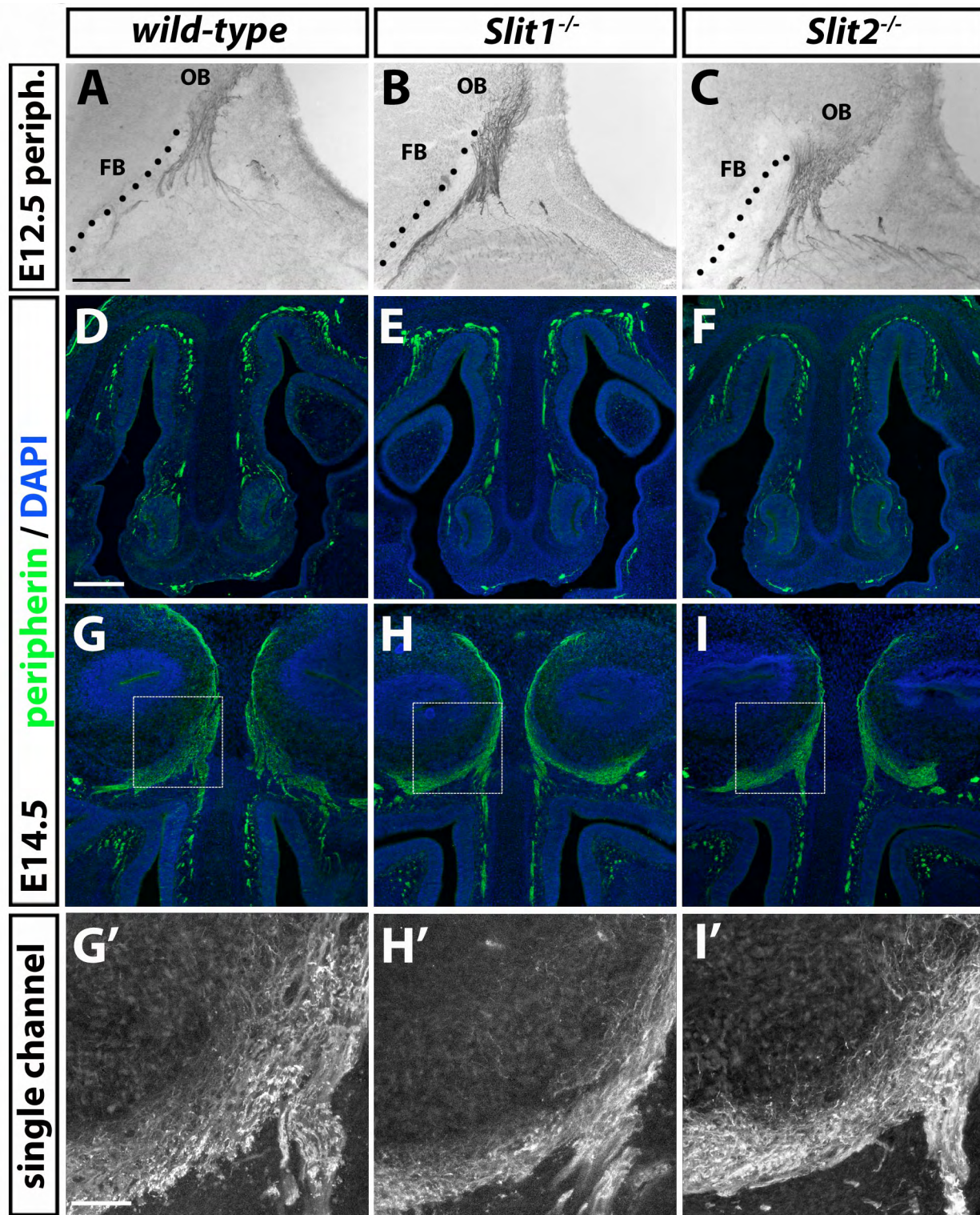


Fig. S2. Lack of *Slit1* or *Slit2* does not impair fasciculation/targeting of OLF/VN axons. (A-I') Normal olfactory/vomer nasal axon organisation in *Slit1* null and *Slit2* null mutants. Sagittal sections of E12.5 (A-C) and E14.5 (D-I) mouse heads from wild-type (A,D), *Slit1* null (B,E) and *Slit2* null (C,F) mutant littermates were immunolabelled for peripherin to reveal the intermingled olfactory/vomer nasal axons in the nasal compartment. No obvious differences were observed in the fasciculation and olfactory bulb (OB) targeting in the genotypes analysed at either embryonic stage. Scale bars: 75 μ m in A-C; 100 μ m in D-I; 50 μ m in G'-I'.

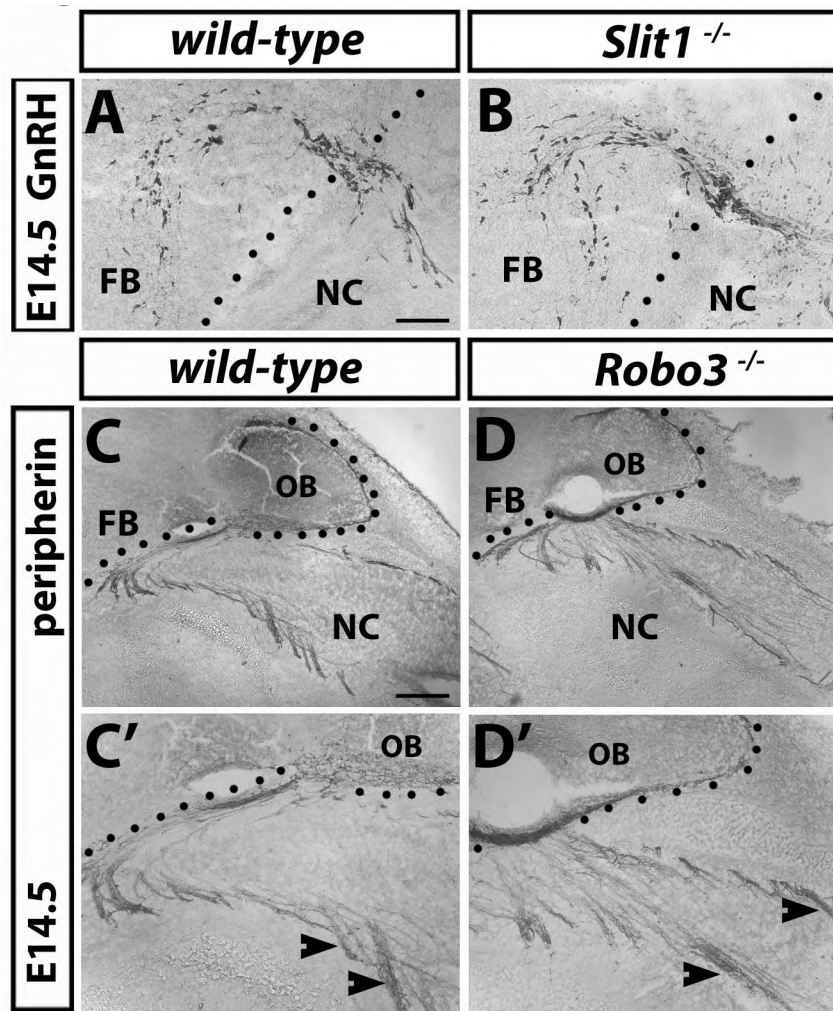


Fig. S3. *Slit1* does not affect migration of GnrRH neurons, and fasciculation and targeting of OLF axons are unaffected in *Robo3* null mice. (A,B) Sagittal sections of E14.5 mouse heads of the indicated genotypes were immunolabelled for GnrRH to reveal neurons migrating in the NC and FB. The FB boundary is delineated with a dotted line. No differences in GnrRH neuron distribution between *Slit1*^{-/-} and wild type were observed. (C-D') The nasal OLF/VN axonal pattern appears normal in sagittal sections of E14.5 *Robo3* null and wild-type mice immunolabelled for peripherin. Arrowheads in high-magnification images (C',D') point to normally fasciculated OLF axons. NC, nasal compartment; OB, olfactory bulb; FB, forebrain; OLF/VN, olfactory/vomerolateral axons. Scale bars: 75 μ m in A,B,C',D'; 100 μ m in C,D.

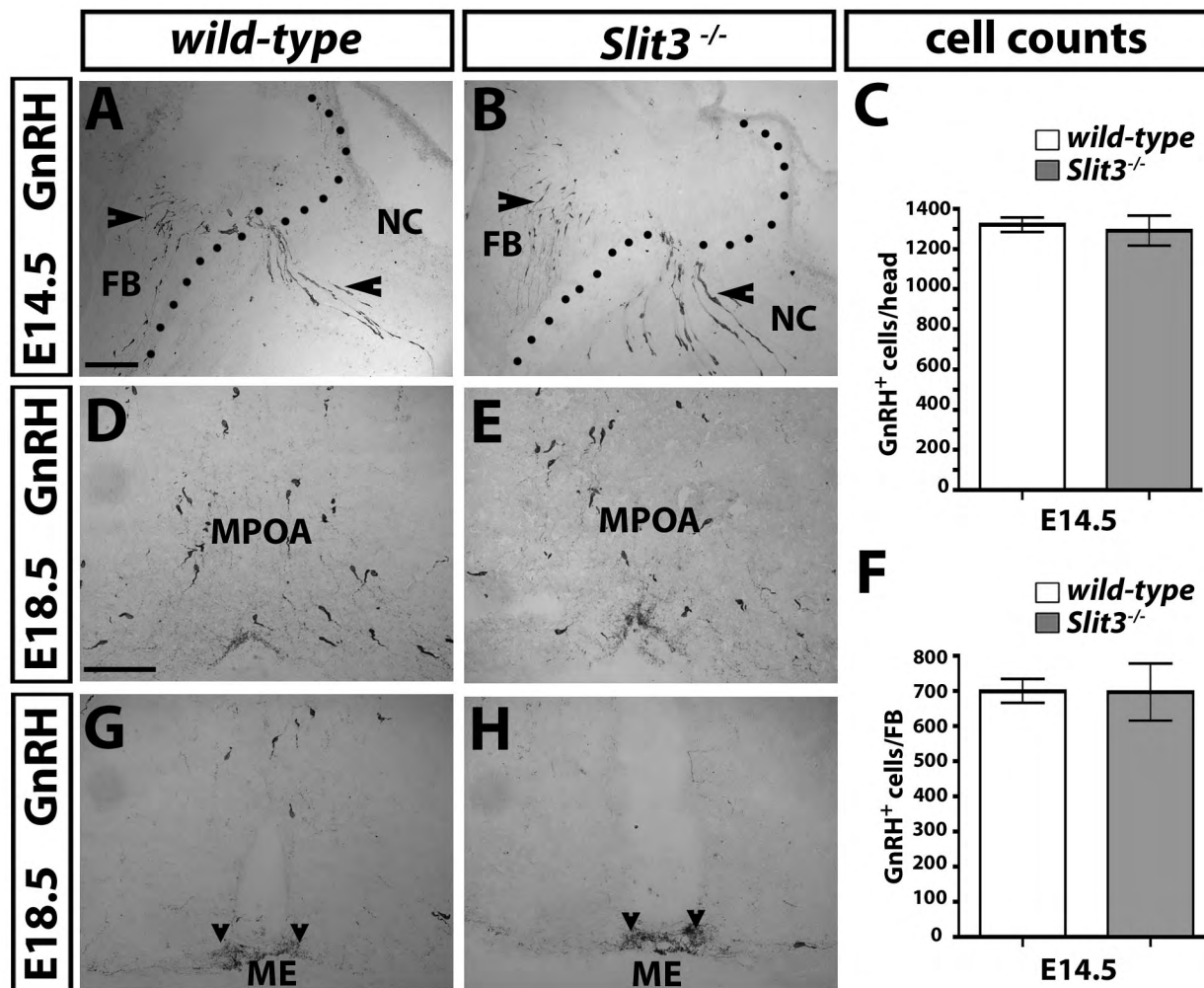


Fig. S4. Normal GnRH neuron migration and olfactory axon organisation in *Slit3* null mutants. (A,B) Sagittal sections of E14.5 mouse heads from wild-type (A) and *Slit3* null mutant (B) littermates were immunolabelled for GnRH to reveal migrating neurons in the nose and forebrain (examples are indicated with arrowheads). Note that GnRH neurons follow a normal path of migration in the mutants at E14.5 (the boundary of the nasal compartment and forebrain is indicated with a dotted line) and are present at normal numbers in the entire head and in the forebrain. (C,F) Quantitation of GnRH⁺ cells per head/FB in wild-type and *Slit3* null mutant. (D,E,G,H) Coronal sections of E18.5 mouse brains of the indicated genotypes were immunolabelled for GnRH to visualise migrating neurons in the MPOA (D,E) and their axons projecting to the ME (arrowheads in G,H). No differences were observed in the migration and axon targeting of GnRH neurons in the *Slit3* null mice compared with wild-type littermates. NC, nasal compartment; FB, forebrain; MPOA, medial preoptic area; ME, median eminence. Scale bars: 100 μ m in A,B; 75 μ m in D,E,G,H.

