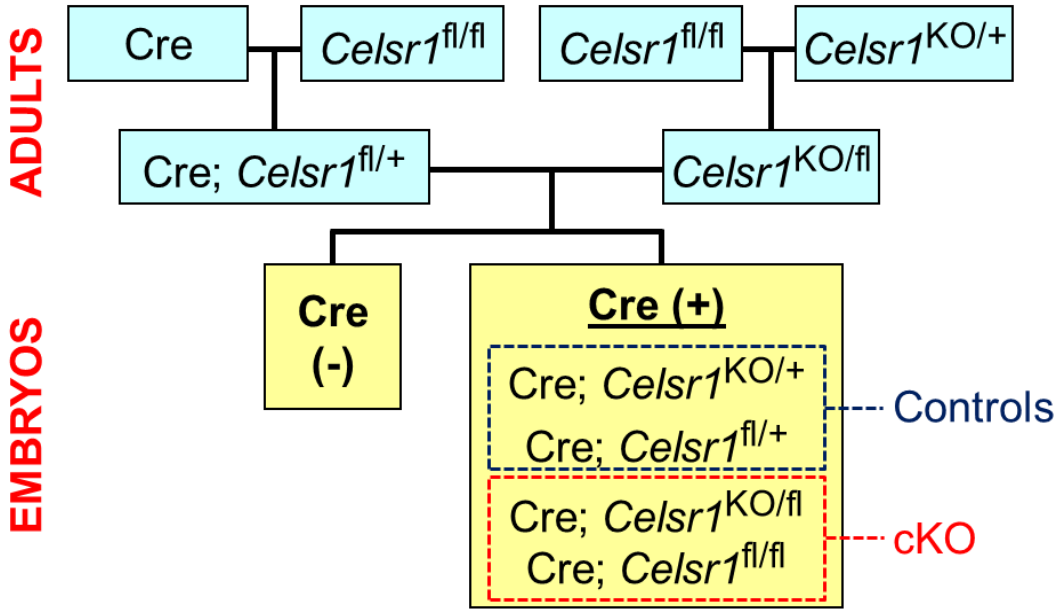
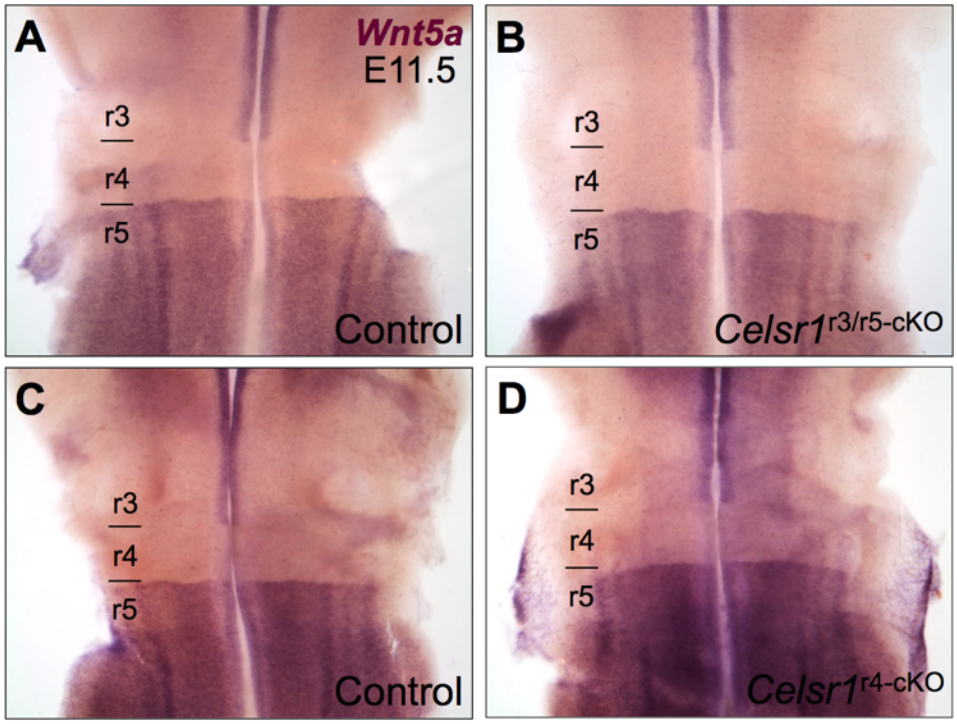
Figure S1



Breeding scheme used to generate *Celsr1* conditional mutant embryos.

To produce male stud mice (Cre; *Celsr1*$^{fl/+}$) from which conditional knockout (cKO) embryos could be generated, mice from each Cre line were crossed with *Celsr1*$^{fl/fl}$ mice. Females for timed matings (*Celsr1*$^{KO/fl}$) were produced by crossing *Celsr1*$^{fl/fl}$ and *Celsr1*$^{KO/+}$ mice. For embryo collection, Cre; *Celsr1*$^{fl/+}$ studs were timed mated with *Celsr1*$^{KO/fl}$ females. Cre-negative embryos were disregarded, while Cre-positive embryos were used for analysis. Cre; *Celsr1*$^{KO/+}$ and Cre; *Celsr1*$^{fl/+}$ embryos served as controls against *Celsr1* cKO embryos, which were either of the *Celsr1*$^{KO/fl}$ or Cre; *Celsr1*$^{fl/fl}$ genotype.

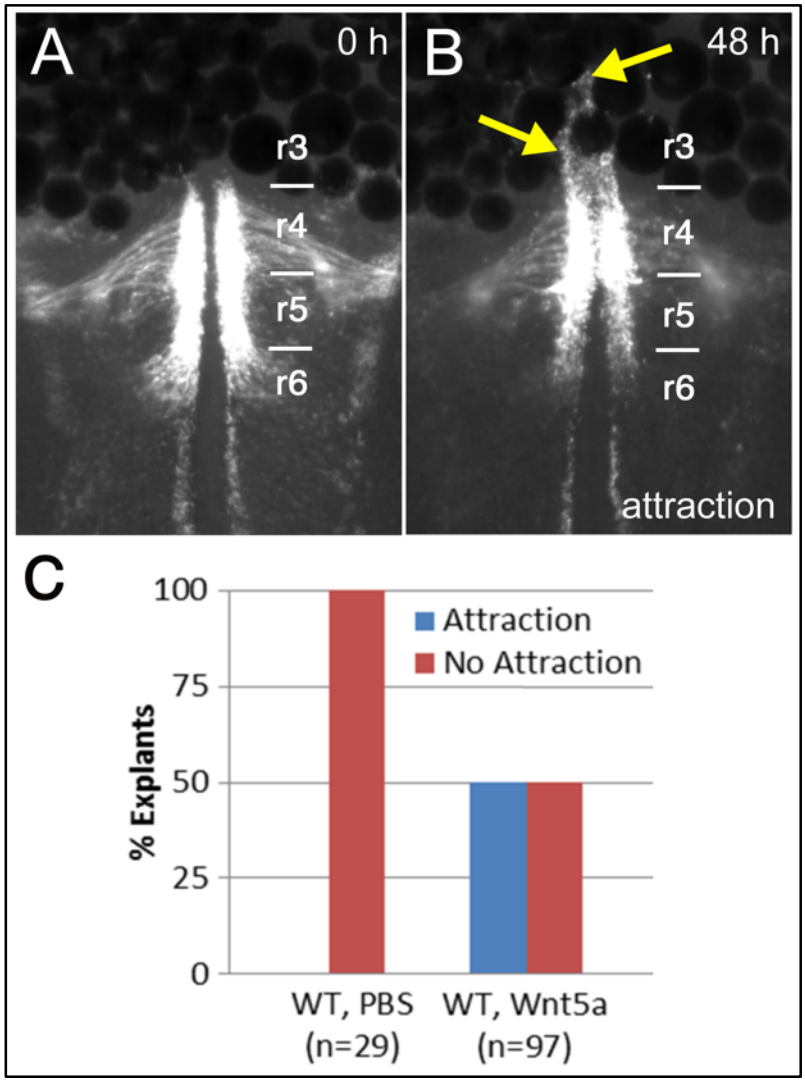
Figure S2



***Wnt5a* is expressed normally in *Celsr1*$^{r3/r5\text{-}cKO}$ and *Celsr1*$^{r4\text{-}cKO}$ embryos.**

A-D, Dorsal views of flat-mounted E11.5 hindbrains processed for *Wnt5a* ISH. In control embryos (A, C) of either Cre; *Celsr1*$^{fl/+}$ or Cre; *Celsr1*$^{KO/+}$ genotypes, *Wnt5a* is expressed along the midline rostral to r4, mostly absent from r4, and expressed in a caudal (high) to rostral (low) gradient caudal to r4. *Wnt5a* is expressed identically in *Krox20*^{Cre}; *Celsr1*$^{KO/fl}$ (B) (n=4) and r4-Cre; *Celsr1*$^{KO/fl}$ (D) embryos (n=4), with sharp boundaries intact at the r3/r4 and r4/r5 borders.

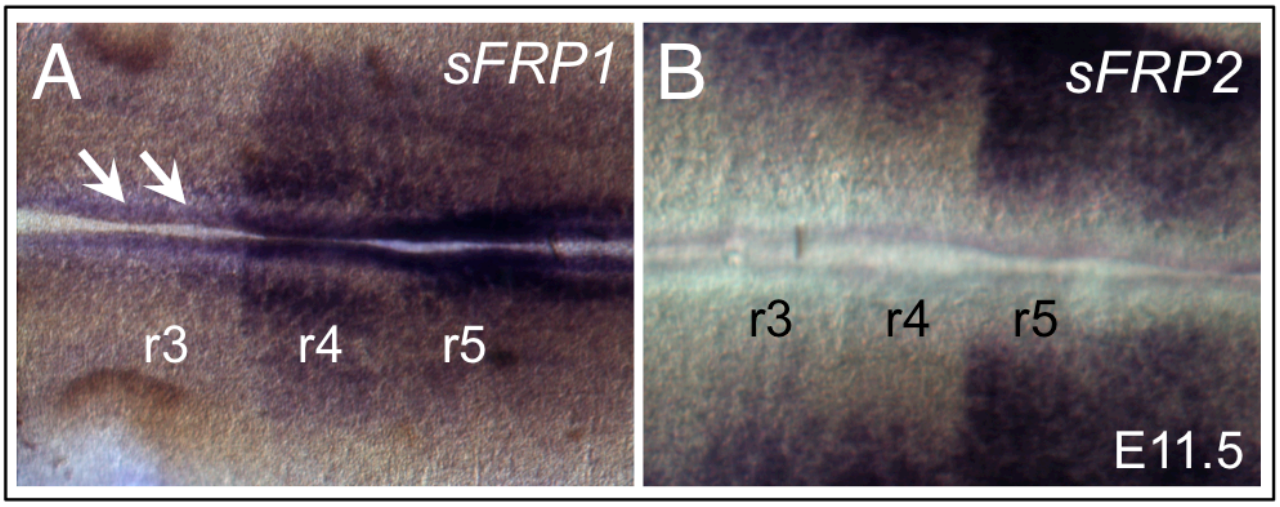
Figure S3



Ectopic Wnt5a can attract FBM neurons to migrate into rhombomere 3.

A, B, Dorsal views of a hindbrain explant from an SE1:GFP wild type mouse (Song et al., 2006). Several (~30) Wnt5a-soaked beads were placed in anterior r3 of E11.5 explant, which was cultured for 48 h. At the start of the experiment (A), all FBM neurons are in r4, respecting the r3-r4 boundary. After 48 hours of incubation (B), many GFP+ve cells have migrated rostrally (arrows) between the beads, which is defined as “attraction”. (C) While attraction was observed in 50% of explants treated with Wnt5a-soaked beads (n=97 explants), it was never seen in explants treated with PBS-soaked beads (n=29 explants).

Figure S4



***sFRP1* and *sFRP2* expression in E11.5 wild type hindbrains.**

A, B, Dorsal views of flat-mounted E11.5 hindbrains processed for *sFRP1* (A) and *sFRP2* (B) ISH. A, *sFRP1* is expressed in the posterior hindbrain neuroepithelium upto the r3/r4 boundary. In r3, *sFRP1* is weakly expressed in the midline tissues (arrows). B, *sFRP2* is expressed in lateral aspects of the anterior hindbrain neuroepithelium up to the r4/r5 boundary. In r5 and posteriorly, its expression expands medially but is still excluded from the midline tissues.