



**Figure 5—figure supplement 1. *fz5/8-4*, *wntA*, and *ndk* restrict the brain tissue to the head region.**

(A) Ectopic eyes are shown in 7 dpa trunk fragments of *fz5/8-4* RNAi animals. Black arrows point to ectopic eyes. Total numbers of RNAi animals are indicated. (B) Ectopic eyes (black arrows) are shown in an intact *wntA* RNAi and in a *fz5/8-4; wntA* double RNAi intact animal after 6 RNAi feedings. RNAi experiments were performed three times. Total numbers of RNAi animals are indicated. (C) FISH using the RNA probe *fz5/8-4* shows decreased expression of this gene following its RNAi demonstrating the efficiency of the inhibition (6 RNAi feedings). Graph below (mean  $\pm$  SD) shows quantification of the mRNA levels by qRT-PCR in different RNAi conditions. mRNA was extracted from 6 dpa head fragments. One-way ANOVA, \*\*\*\*  $p < 0.0001$ . (D) Synergistic RNAi effect of several members of the *FGFRL* family on the *fz5/8-4; ndk* RNAi phenotype. RNAi experiments have been performed twice. Total number of RNAi animals is indicated. Black arrows point to ectopic eyes. (E,F) FISH using the neuronal markers: (E) *cintillo*, and (F) *notum* and the photoreceptor marker *opsin* (E). Graph shows the percentage of *cintillo*<sup>+</sup> cells (mean  $\pm$  SD) in intact RNAi animals normalized by the length of the animal (from the tip of the head to the esophagus), ( $n > 8$  animals per RNAi condition, two independent RNAi experiments, One-way ANOVA). Yellow dotted line shows the esophagus location, \* shows pharynx. White arrows show the most posterior cell expressing the neuronal marker analyzed. Anterior, up; dorsal view unless indicated. Scale bar, 100  $\mu$ m.