Fig.S1

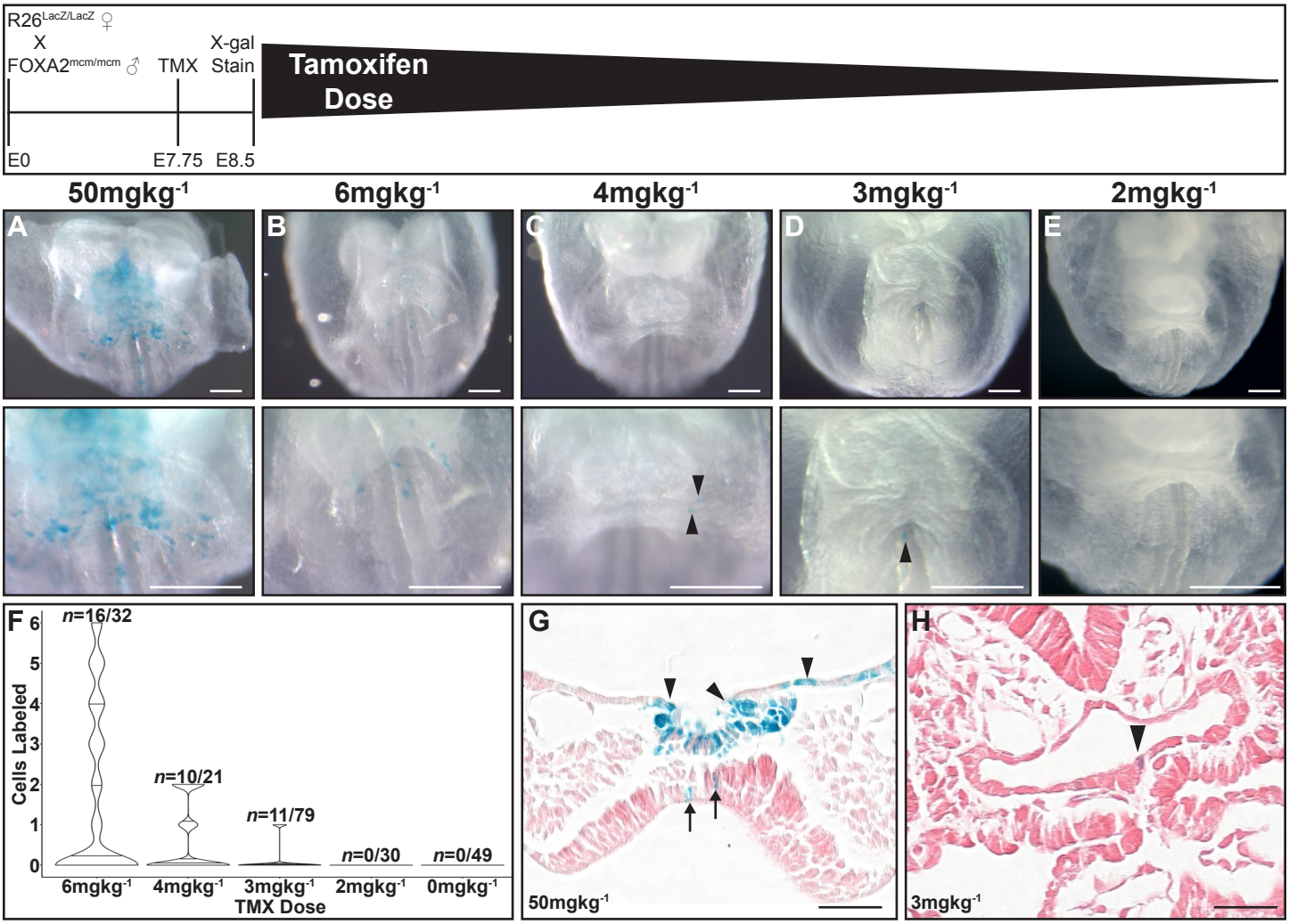
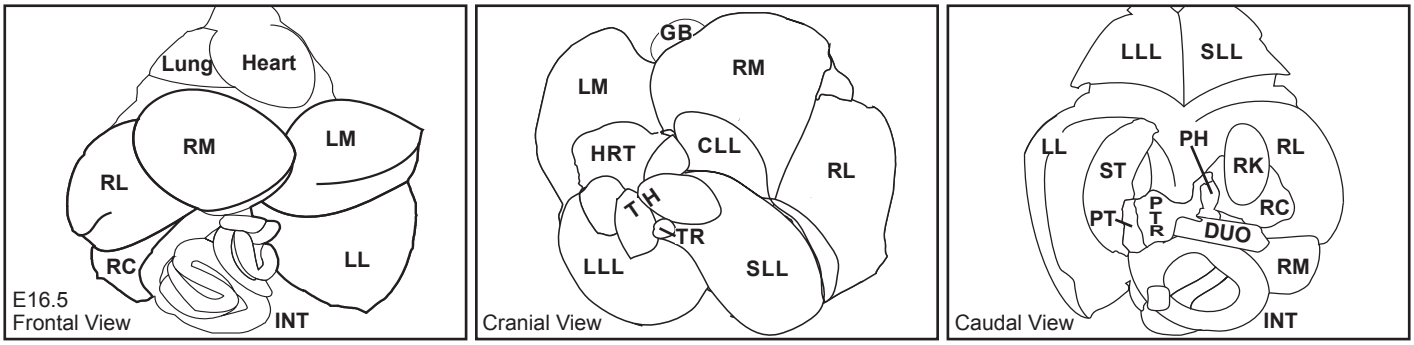


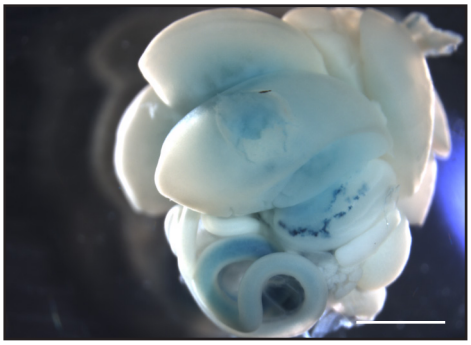
Fig.S1: Defining a TMX dose that reliably promotes single-cell recombination at E8.5. TMX was administered via oral gavage on E7.75 to dams harboring Foxa2$^{mcm/+}$; R26R$^{-/+}$ embryos. Embryos were harvested at E8.5 and subject to X-gal staining to detect the recombined *LacZ*-reporter. (A) 50mgkg^{-1} of TMX induced widespread recombination, producing many blue cells. (B) 6mgkg^{-1} induced more limited recombination. (C) 4mgkg^{-1} induced 1-2 *LacZ*-expressing cells (black arrowheads). (D) 3mgkg^{-1}of TMX infrequently recombines a single *LacZ*-expressing cell in individual embryo (black arrowhead). (E) At 2mgkg^{-1} of TMX, no recombination was observed. (F) A graph depicting the number of labeled cells observed in each embryo at each low TMX dose (ANOVA p = 2.23^{-9}***). The width of each violin plot corresponds to the proportion of samples at that dose and the horizontal lines within each plot indicates the 25%, 50% and 75% quartiles. The numbers provided at the top of each violin plot indicate the number of embryos displaying any *LacZ*-positive cells in the numerator over the total number of embryos receiving the indicated TMX dose. (G-H) Eosin counterstained sections through the foregut of an embryo receiving the indicated TMX dose. (G) A 50mgkg^{-1} dose (n=2) induces recombination in the DE (black arrowheads) as well as the notochord and neural tube (black arrows). (H) A 3mgkg^{-1} dose reveals that all *LacZ*-expressing cells are confined to the DE or the notochord (*n*=9). Scale bars in A-E = 100µm, scale bars in G, H = 50µm.

Fig.S2

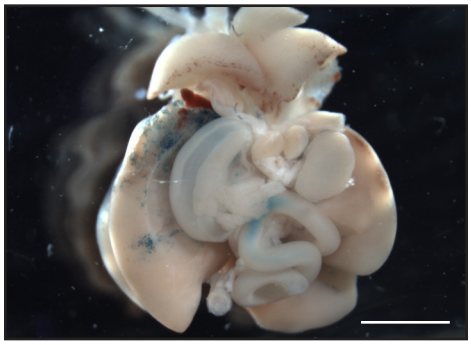


LM: left medial lobe, LL: left lateral lobe, RM: right medial lobe, RL: right lateral lobe, RC: right caudate lobe, LLL: left lung lobe, SLL: superior lung lobe, CLL: cardiac lung lobe, TR: trachea, TH: thymus, ST: stomach, GB: gall bladder, PT: pancreas tail, PTR: pancreas trunk, PH: pancreas head, DUO: duodenum, INT: intestine, RK: right kidney

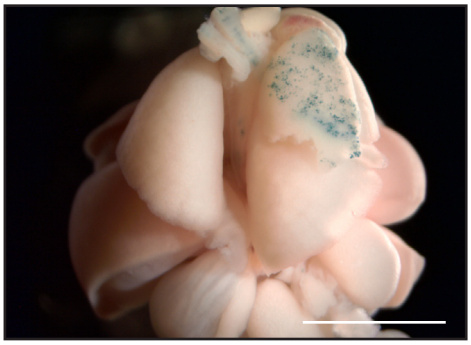
39.1 Left; ST



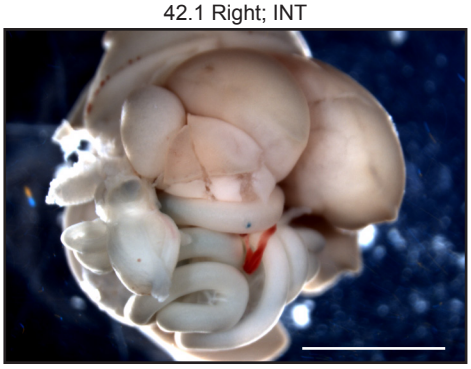
41.1 Caudal; LL, PTR



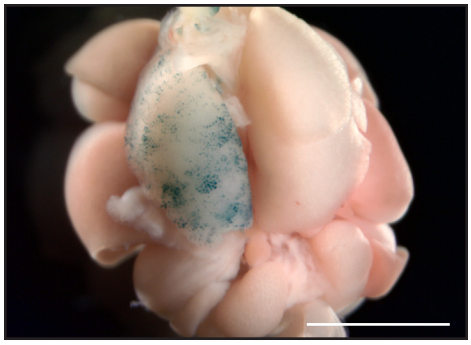
41.2 Dorsal; TR, SLL



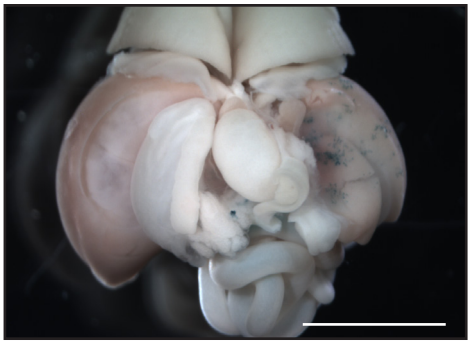
42.1 Right; INT



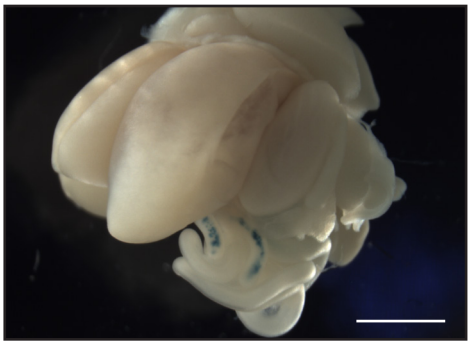
44.1 Dorsal; TH, LLL



46.1 Caudal; LM, RM, RL, RC, PTR

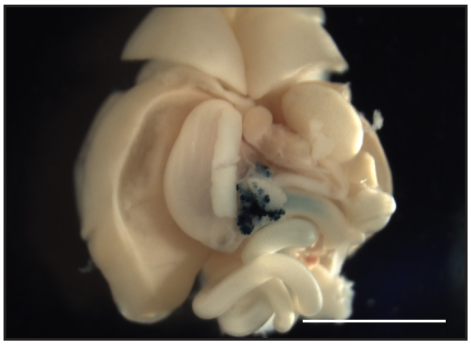


48.1 Left; INT

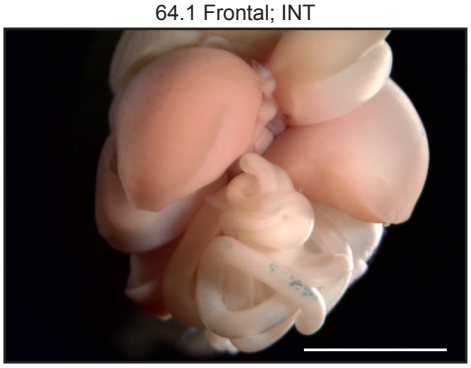


48.2 Frontal; INT

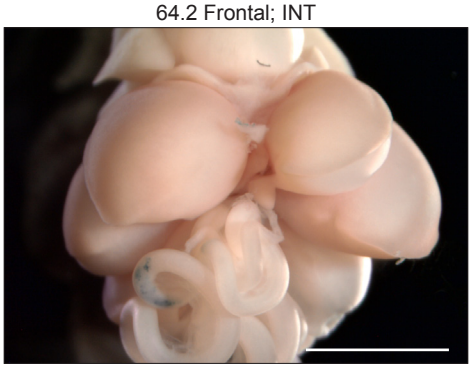
61.1 Caudal; PT, PTR, PH



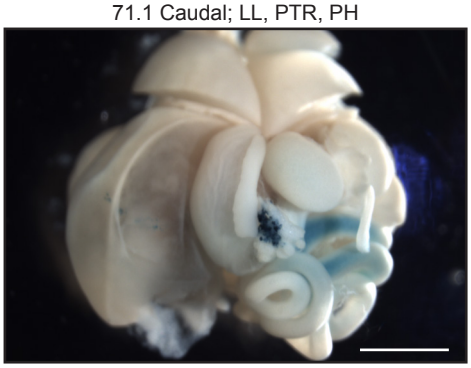
64.1 Frontal; INT



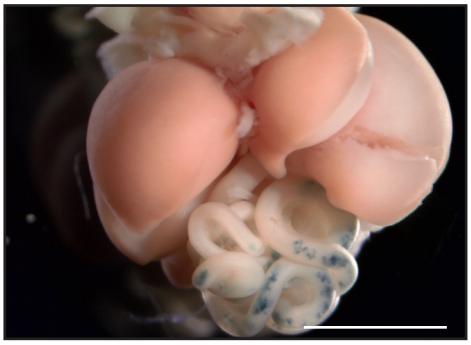
64.2 Frontal; INT



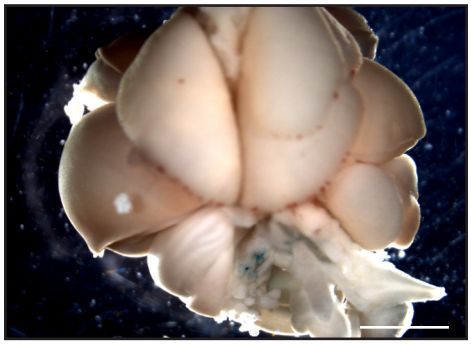
71.1 Caudal; LL, PTR, PH



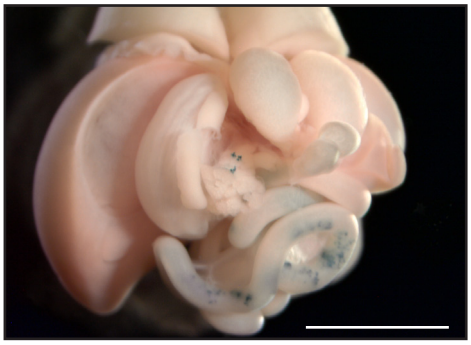
75.1 Frontal; INT



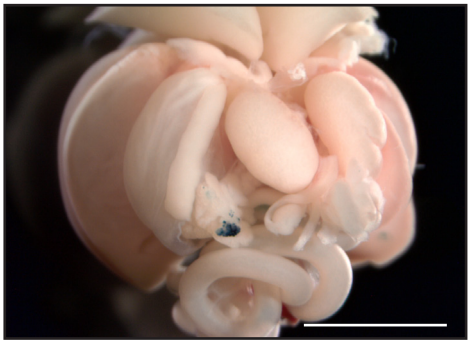
79.1 Dorsal; PTR, PH



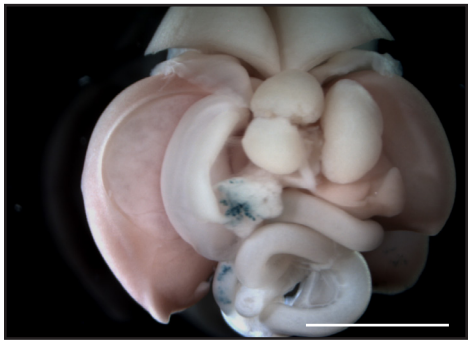
81.1 Caudal; PT, INT



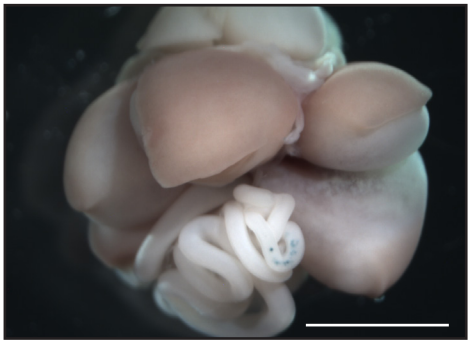
81.2 Caudal; PT



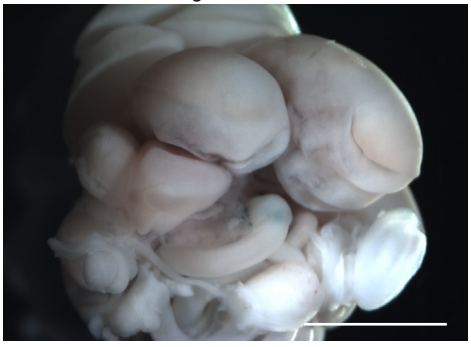
108.1 Caudal; PT, INT



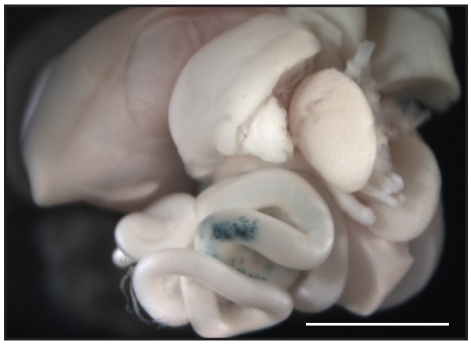
108.2 Frontal; INT



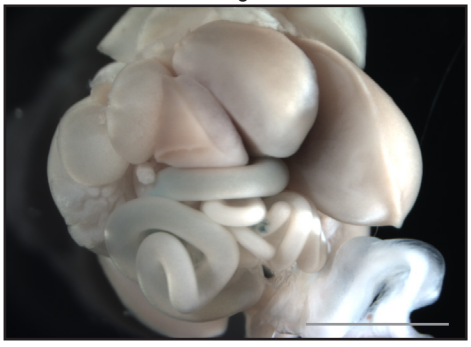
109.1 Right; ST, PH, DUO



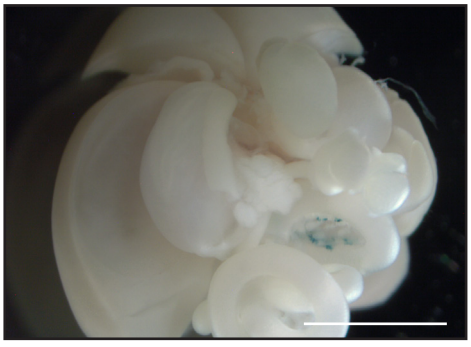
109.2 Caudal; INT



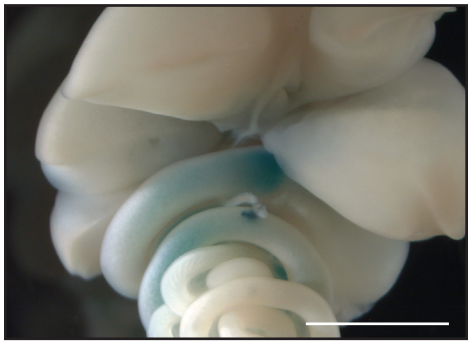
109.3 Right; INT



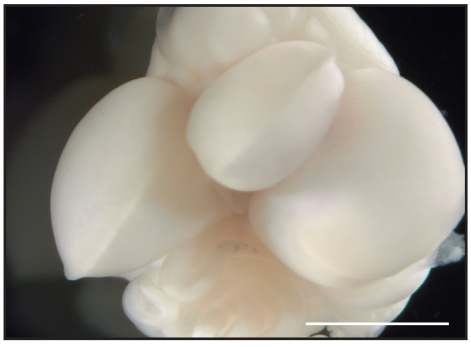
110.1 Caudal; INT



112.1 Frontal; INT



118.2 Frontal; INT



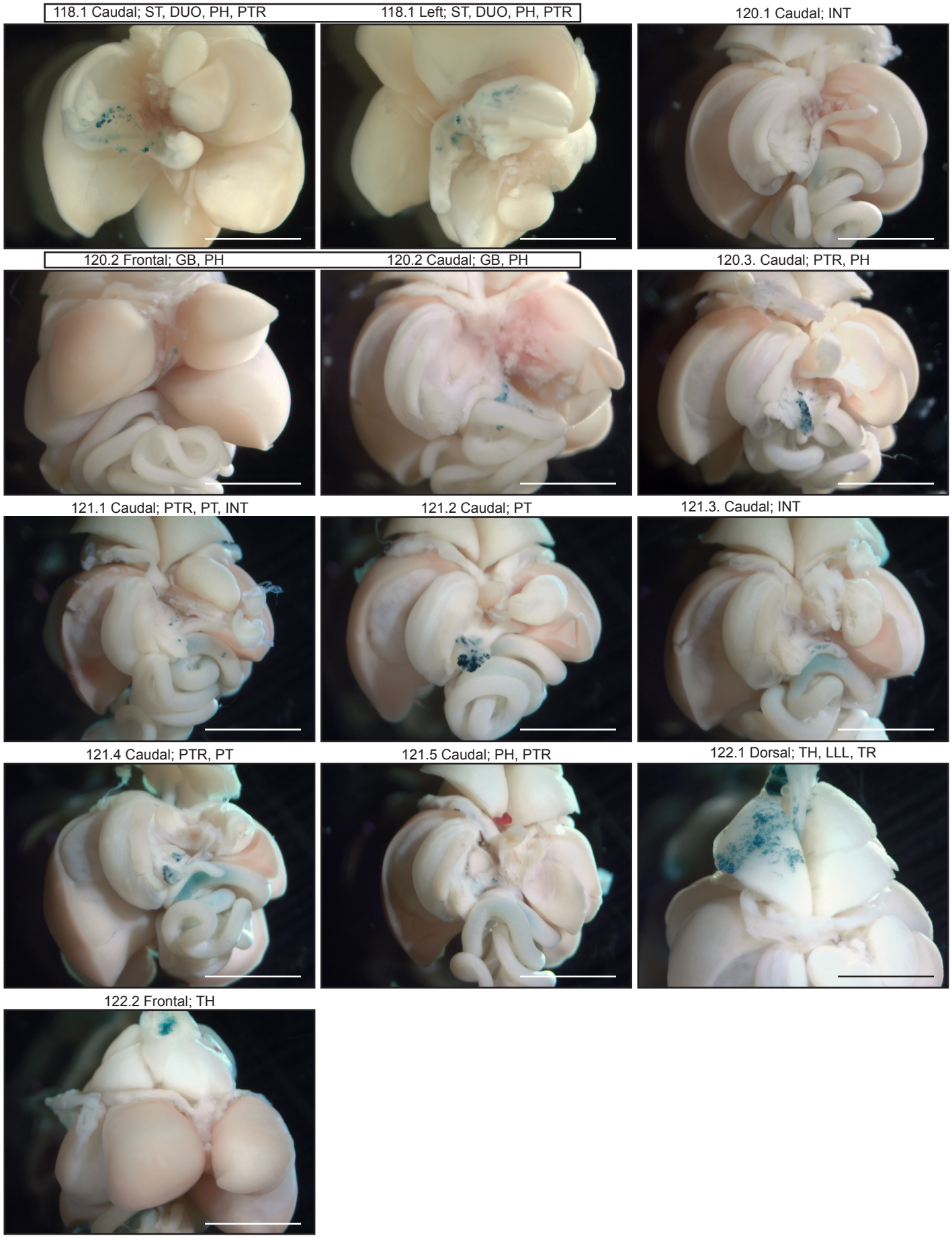
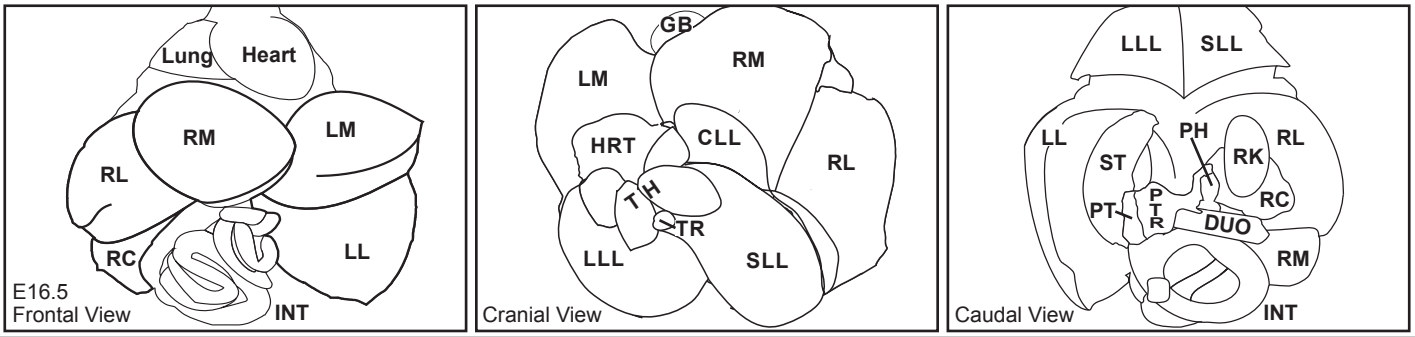


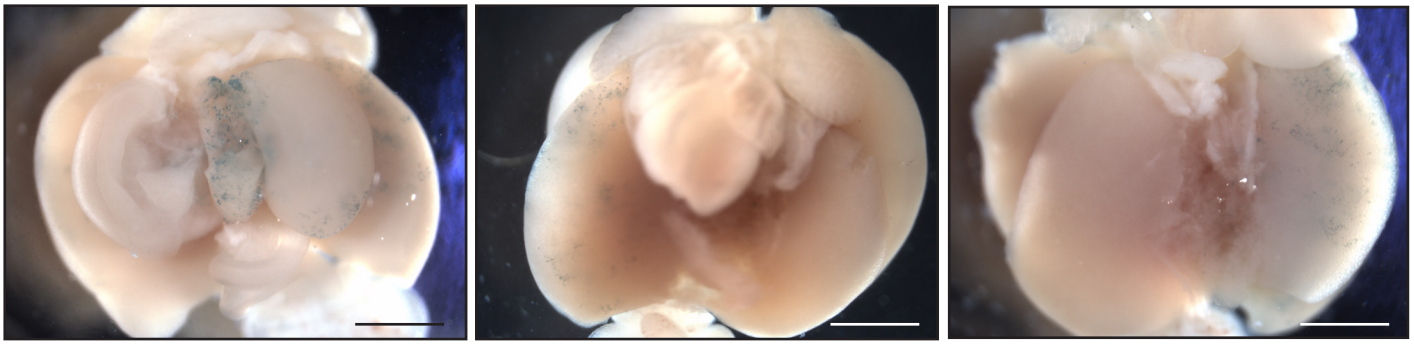
Fig.S2: Images of all extrahepatic clonal descendants in E16.5 intact viscera. Wholemount images of the data summarized in Table 1. Dams each received 3mgkg^{-1} TMX on E7.75 and embryos collected at E16.5. Intact viscera were dissected and X-gal stained to identify embryos with *LacZ*-positive clonal descendants. Illustrated frontal, cranial and caudal views of intact viscera are provided, and the organs seen in each view are annotated. Each embryo harboring extra-hepatic clonal descendants were imaged and listed according to the provided embryo number (*n*=35). This number, the view and the *LacZ*-labeled organs are written at the top of each picture. If a second view was required to visualize a significant portion of the labeled tissue then the headings for those images were boxed. Scale bar = 1mm.

Fig.S3

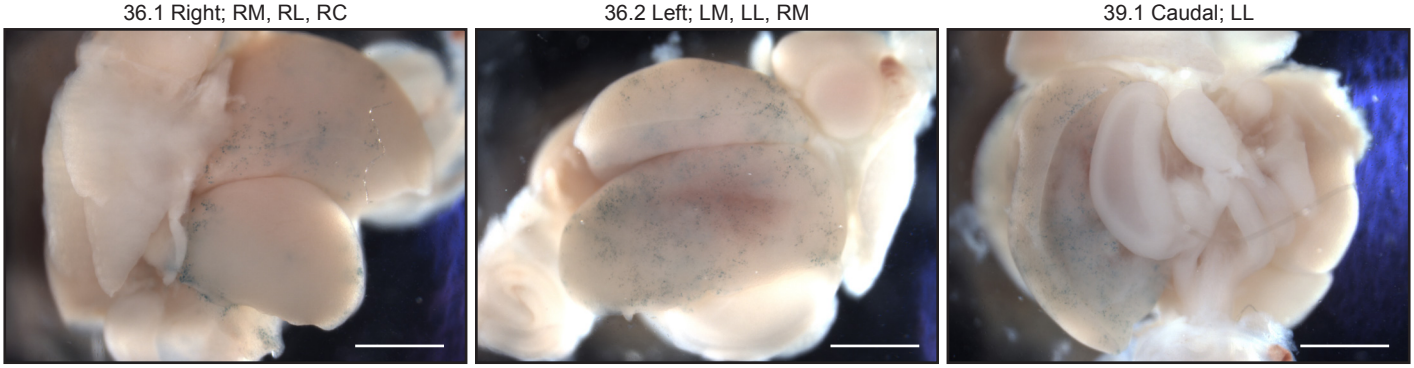


LM: left medial lobe, LL: left lateral lobe, RM: right medial lobe, RL: right lateral lobe, RC: right caudate lobe, LLL: left lung lobe, SLL: superior lung lobe, CLL: cardiac lung lobe, TR: trachea, TH: thymus, ST: stomach, GB: gall bladder, PT: pancreas tail, PTR: pancreas trunk, PH: pancreas head, DUO: duodenum, INT: intestine, RK: right kidney

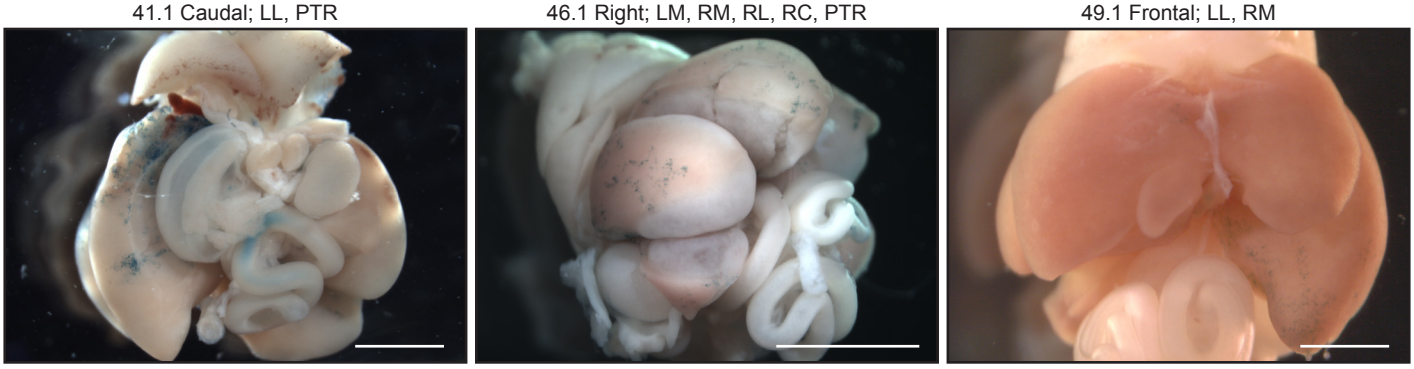
32.1 Caudal; RL, RC | 34.1 Cranial; RM | 34.2 Cranial; LL ,LM



36.1 Right; RM, RL, RC | 36.2 Left; LM, LL, RM | 39.1 Caudal; LL

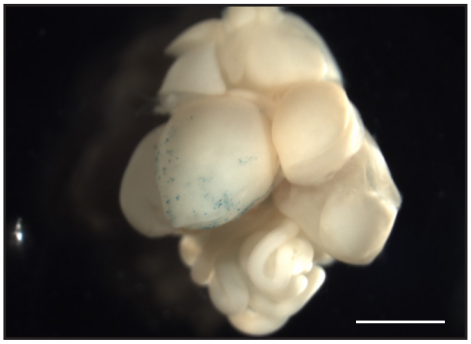


41.1 Caudal; LL, PTR | 46.1 Right; LM, RM, RL, RC, PTR | 49.1 Frontal; LL, RM

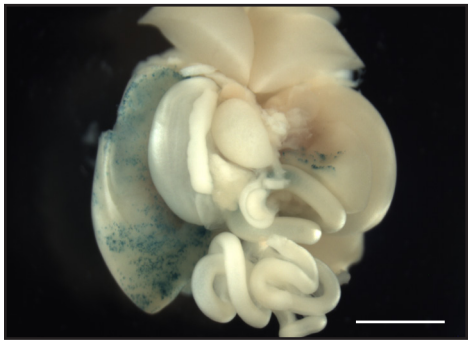


49.2 Left; LL | 52.1 Left; LM, LL | 52.3 Right; RM, RL, RC

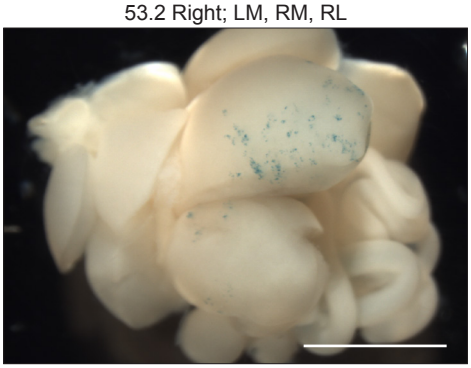
52.4 Frontal; RM



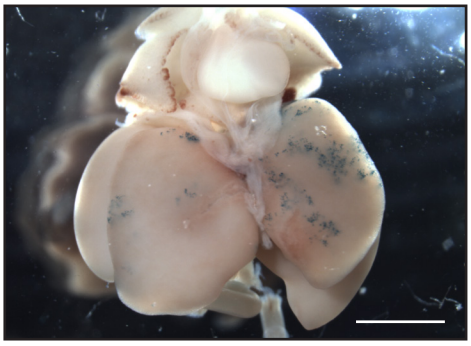
53.1 Caudal; LL, RC



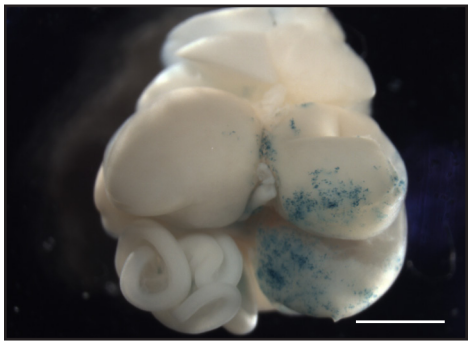
53.2 Right; LM, RM, RL



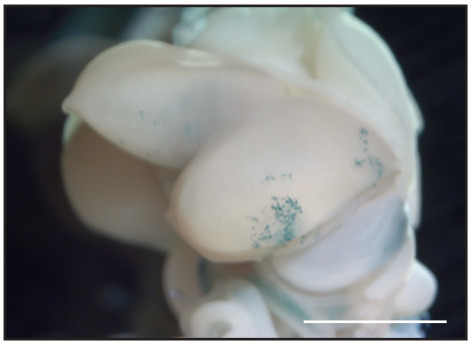
55.1 Cranial; LM, RM



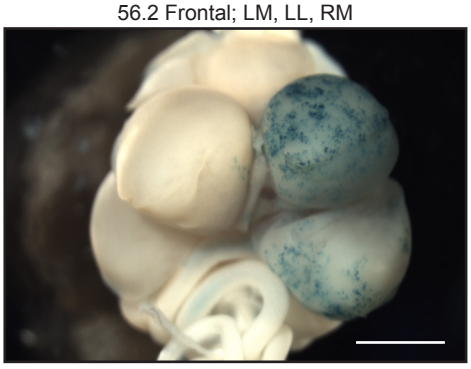
55.2 Frontal; LM, LL, RM



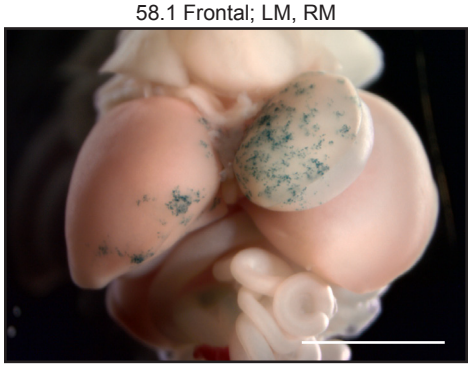
56.1 Left; LL, LM



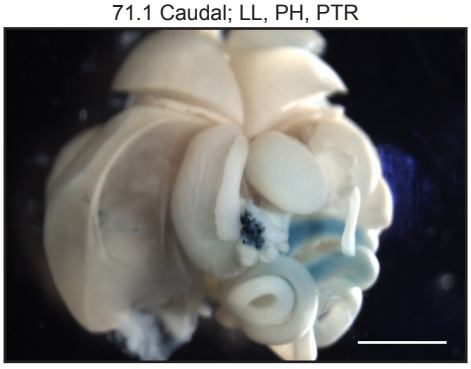
56.2 Frontal; LM, LL, RM



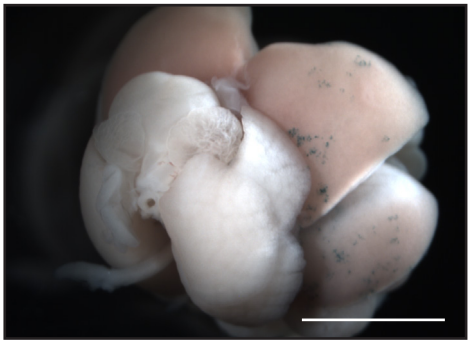
58.1 Frontal; LM, RM



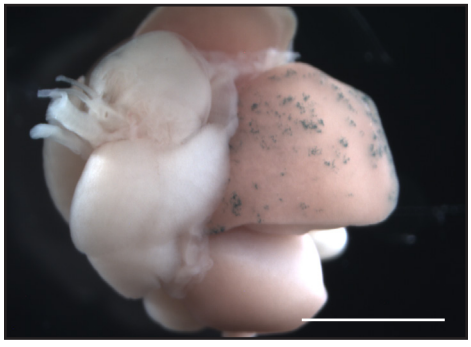
71.1 Caudal; LL, PH, PTR



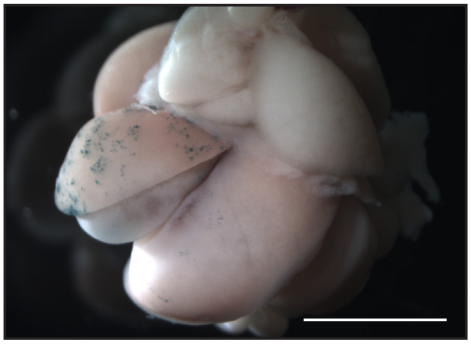
74.1 Cranial; RM, RL



74.2 Cranial; RM



78.1 Left; LM, LL, RM



119.1 Left; LL

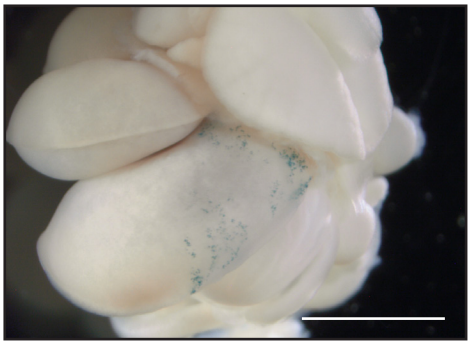
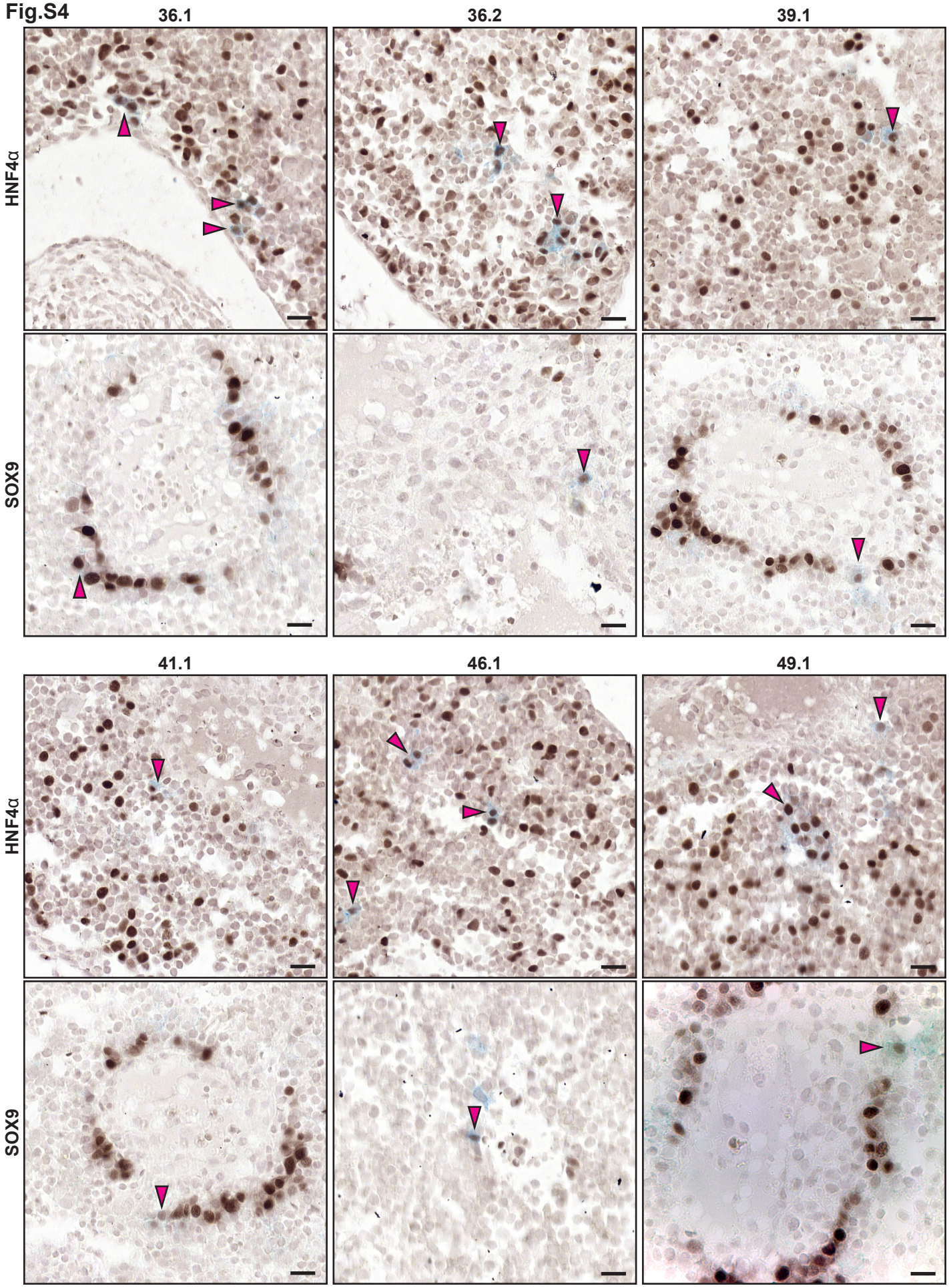
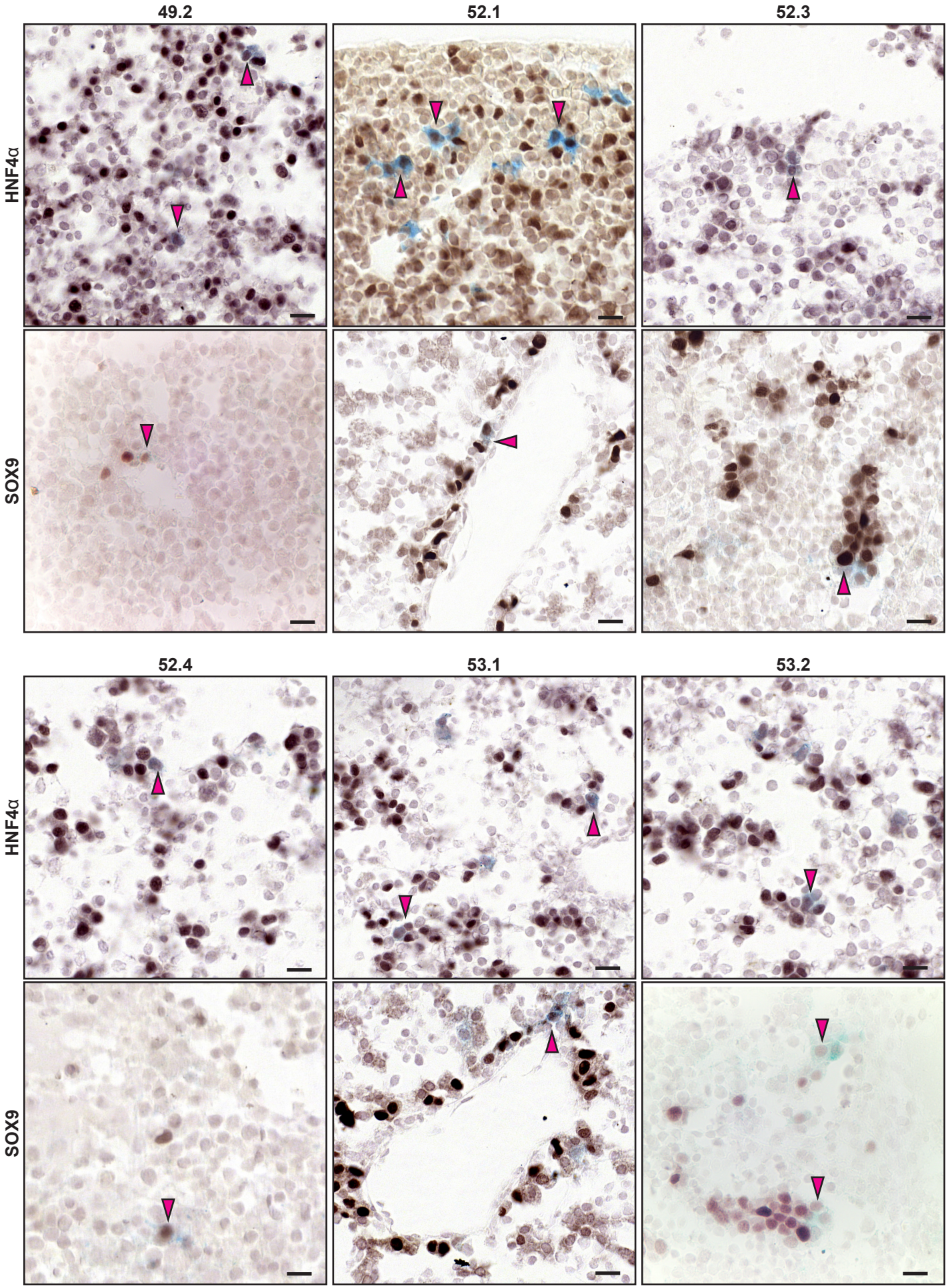
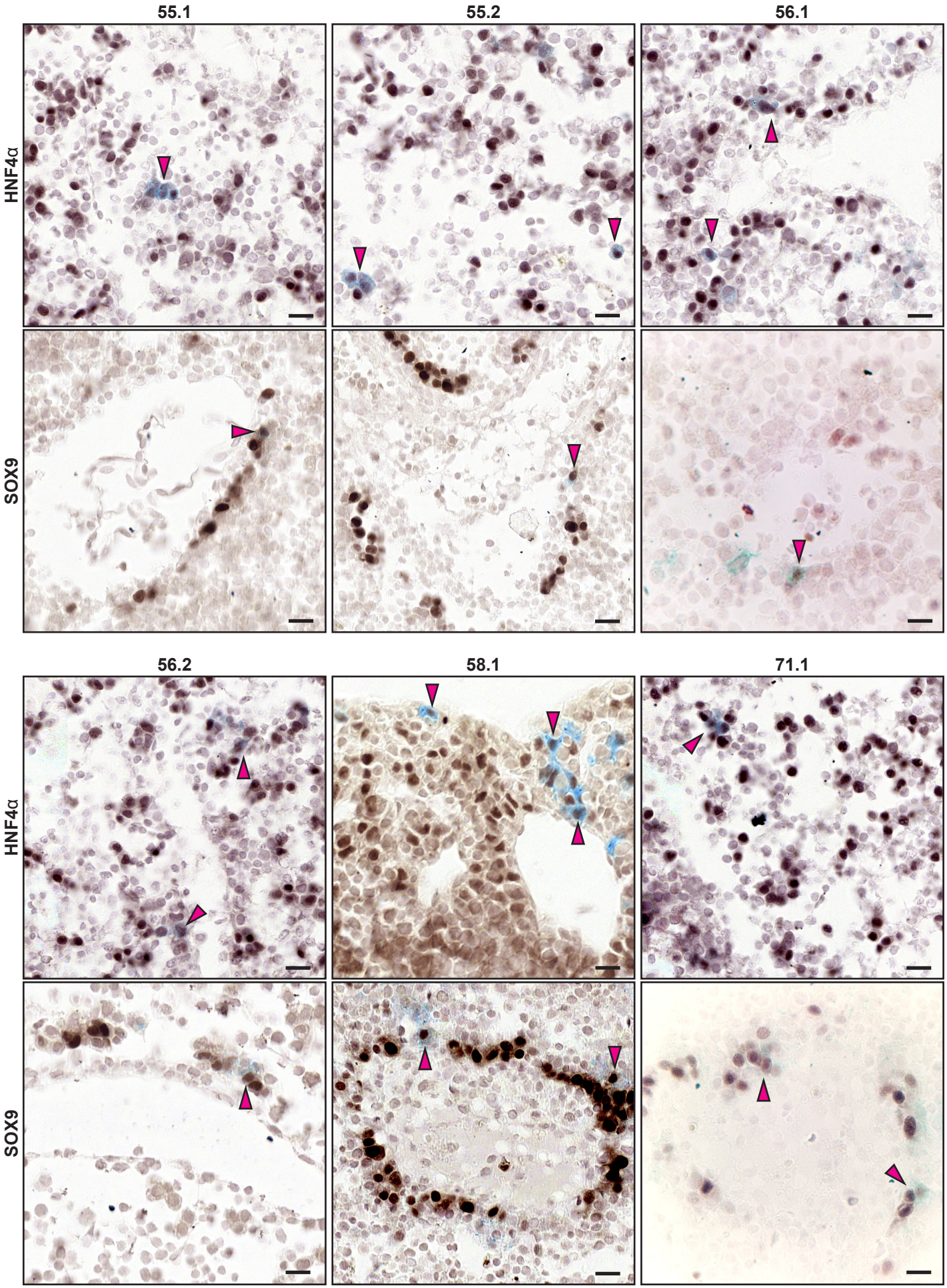


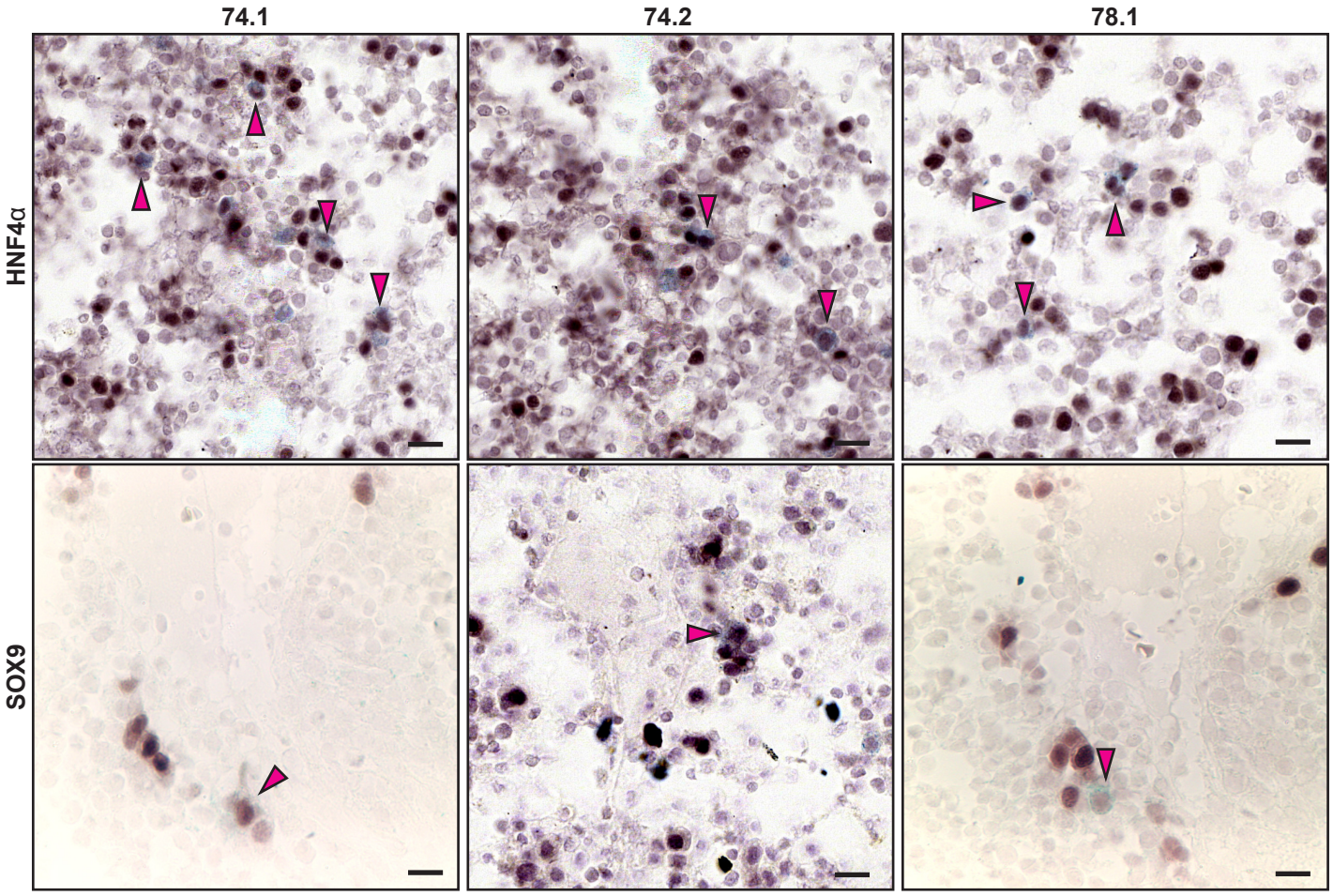
Fig.S3: Images of all clonal descendants present in the liver. Wholemount views of the data summarized in Table 2. Mothers each received 3mgkg^{-1} TMX on E7.75 and embryos collected at E16.5. Intact internal viscera were collected and subject to X-gal staining to identify embryos with *LacZ*-expressing clonal descendants. Whole mount images of all 25 viscera containing *LacZ*-expressing clonal descendants in the liver. The top row represents annotated and illustrated frontal, cranial and caudal views of the intact viscera. The liver is divided into the 5 visible lobes: LM, left medial lobe; LL, left lateral lobe; RM, right medial lobe; RL, right lateral lobe; RC, right caudate lobe. The imaged viscera are organized according to the identification number of each embryo. This number, the view, and the labeled portion of the liver (also listed in Table 2) are provided above each image. The only co-labeled organ includes the pancreas (*n*=3/25). If the pancreas was labeled, then it is also listed next to the labeled portion of the liver. Scale bar = 1mm.

Fig.S4









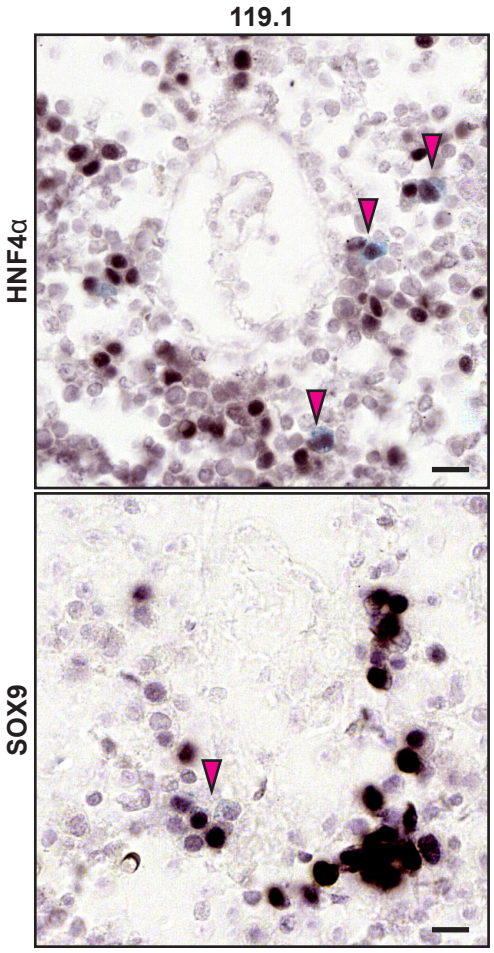


Fig.S4: Immunohistochemical analysis of each E16.5 embryo containing labeled hepatic descendants. Individual sections of the immunohistochemical (IHC) data summarized in Table 2. Mothers each received 3mgkg^{-1} TMX on E7.75 and embryos collected at E16.5. Intact viscera were collected and subject to X-gal staining to identify embryos with *LacZ*-positive clonal descendants. Each *LacZ*-positive liver, identified and organized according to the embryo number at the top of each series (*n*=22), was sectioned onto multiple slides and IHC performed on adjacent or sub-adjacent sections with either the hepatocyte marker, HNF4α (top section in each group) or the biliary epithelial cell marker, SOX9 (bottom section in each group). In each liver analyzed, *LacZ*-positive clonal descendants expressed both HNF4α and SOX9 (**X**^{2}, p = 9.11^{-4}*** < 0.05). In all images, the pink arrowhead denotes *LacZ*-positive cells that are co-labeled with the indicated nuclear-localized antibody. Scale bars = 50µm.