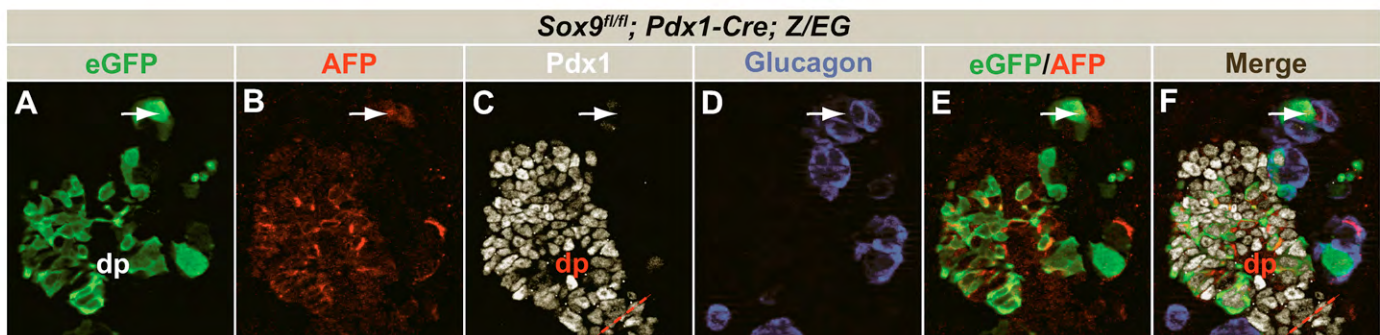
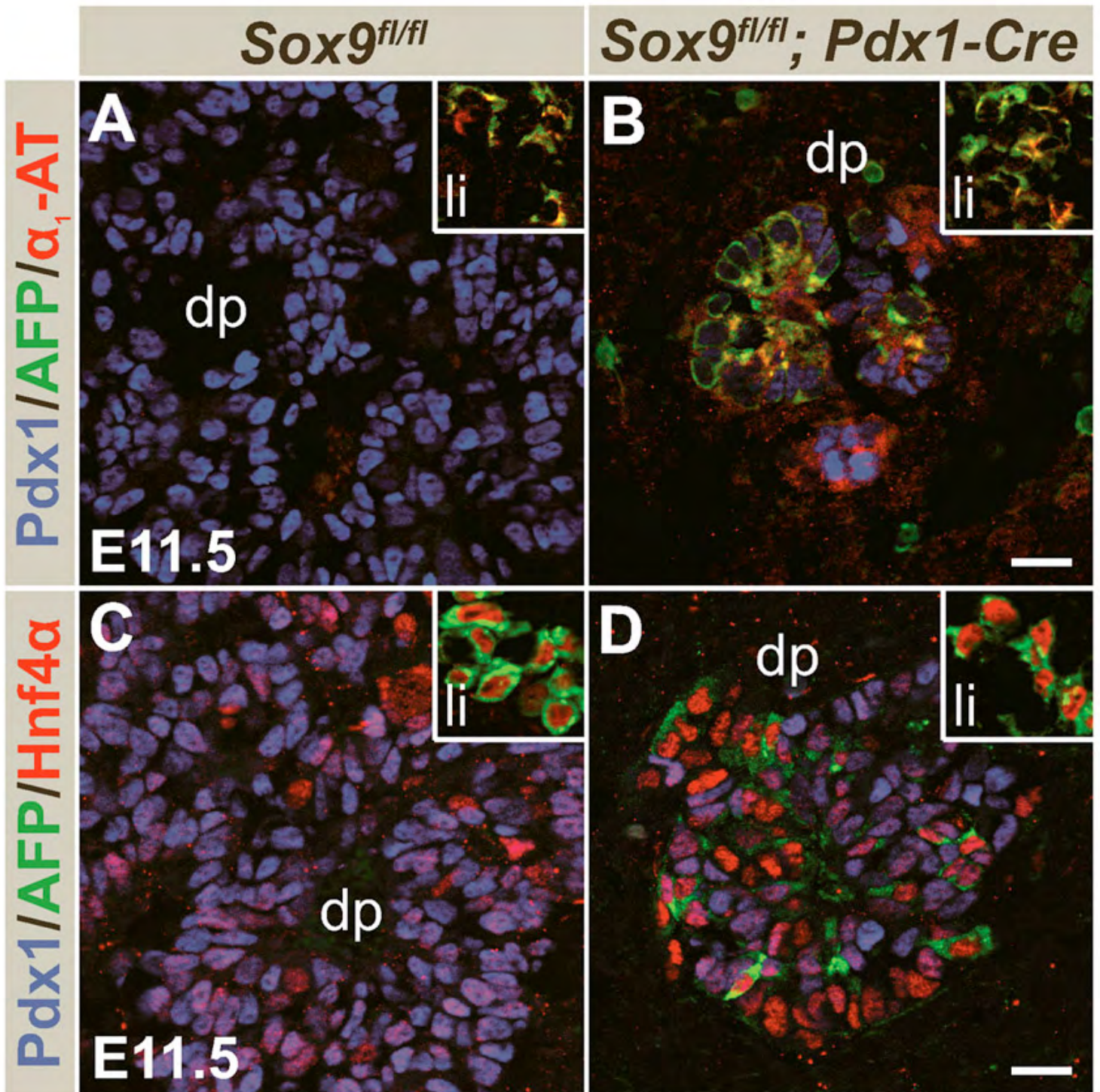


**Fig. S1. *Pdx1-Cre* and *Ptf1a-Cre* are active in the pancreas by E9.5 and E10.5, respectively.** (A-C) X-Gal staining for  $\beta$ -gal activity from the recombined *Rosa26R* reporter allele reveals *Pdx1-Cre* to be active in the dorsal (dp) and ventral (vp) pancreatic buds by E9.5 (A) while *Ptf1a-Cre* is not active until E10.5 (B,C). Scale bars: 250  $\mu$ m.

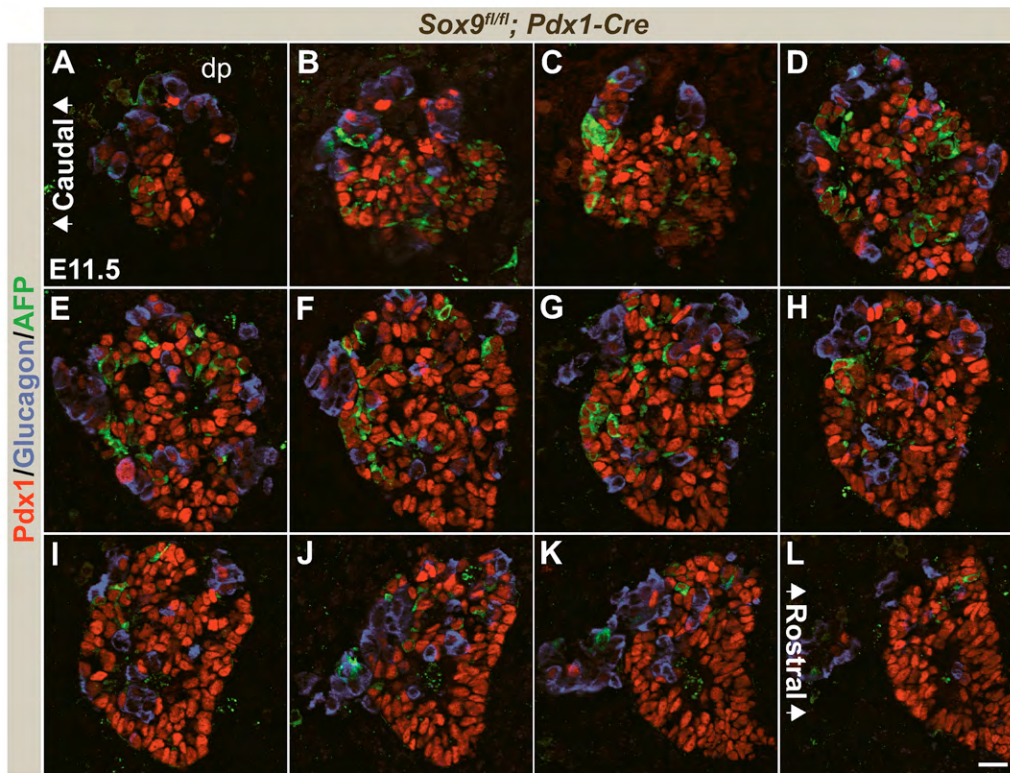


**Fig. S2. AFP<sup>+</sup> cells arise from Pdx1<sup>+</sup> progenitors in *Sox9*-deficient pancreata.** (A-F) eGFP induced by *Pdx1-Cre*-driven recombination of the *Z/EG* reporter allele (A) labels Pdx1<sup>+</sup> cells and their progeny in the dorsal pancreas (dp) (A,F). AFP<sup>+</sup> cells (B), including AFP<sup>+</sup> Pdx1<sup>-</sup> cells (arrows in A-F), are eGFP<sup>+</sup>, showing their derivation from *Pdx1*-expressing pancreatic progenitors in *Sox9*-deleted pancreata (E,F). Most first-wave glucagon<sup>+</sup> endocrine cells (D) are also eGFP<sup>+</sup> and are thus derived from *Sox9*-deleted pancreatic progenitors (F). Broken line indicates the boundary between dorsal pancreas and duodenum (duo). Scale bar: 20  $\mu$ m.

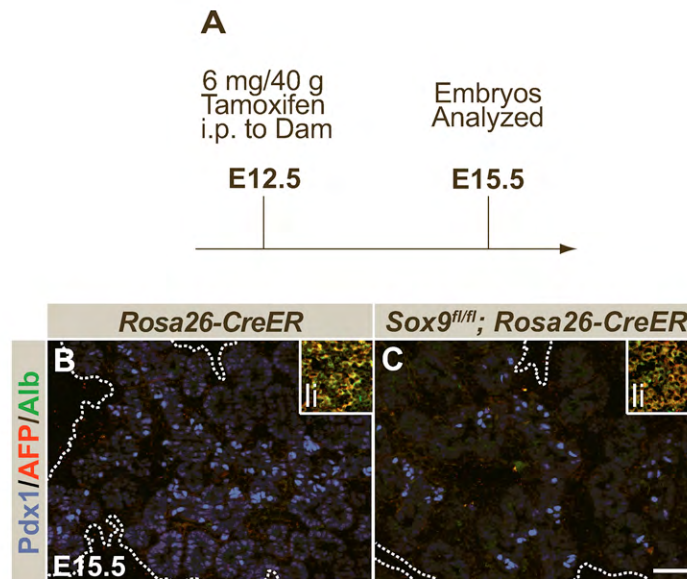


**Fig. S3. *Sox9*-deleted pancreatic progenitors activate a bona fide hepatic program.** (A,B) Immunodetection reveals almost complete absence of AFP or  $\alpha_1$ -antitrypsin ( $\alpha_1$ -AT) in the E11.5 control *Sox9<sup>fl/fl</sup>* dorsal pancreas (dp) (A), but abundant expression in *Sox9<sup>fl/fl</sup>; Pdx1-Cre* pancreas (B). (C,D) Although low-level Hnf4 $\alpha$  nuclear expression is seen in Pdx1<sup>+</sup> pancreatic progenitors in E11.5 control pancreas (C), high-level Hnf4 $\alpha$  expression is evident in AFP<sup>+</sup> cells of *Sox9<sup>fl/fl</sup>; Pdx1-Cre* pancreas (D), similar to that seen in the liver (li) (C,D). Scale bars: 20  $\mu$ m.



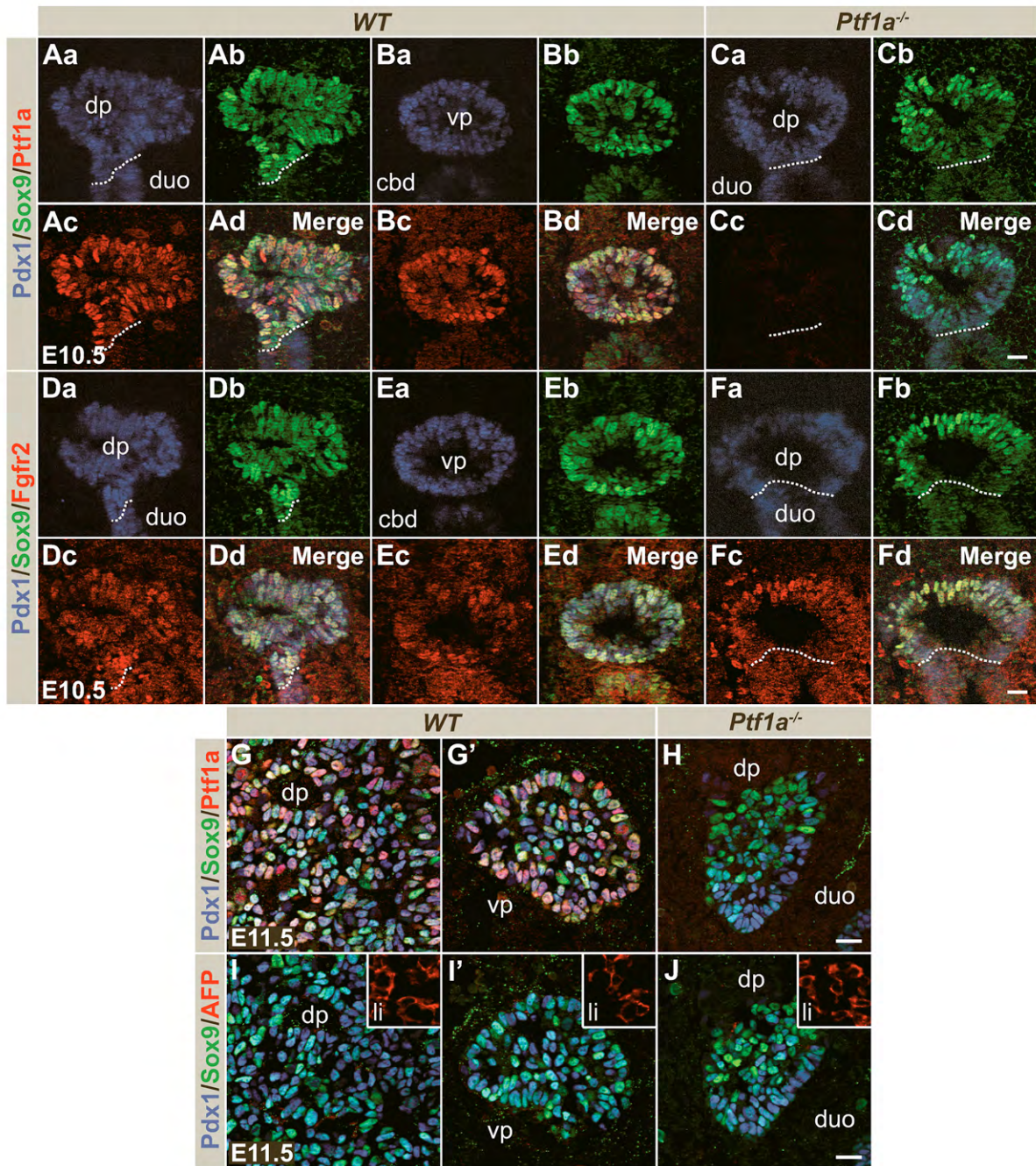


**Fig. S4. Variable ratios of mutually exclusive AFP<sup>+</sup> and glucagon<sup>+</sup> populations.** (A-L) Immunodetection on 12 sequentially ordered (rostral to caudal) serial sections through a single E11.5 *Sox9<sup>fl/fl</sup>; Pdx1-Cre* dorsal pancreas (dp) shows both glucagon<sup>+</sup> cells and AFP<sup>+</sup> cells to occur in clusters. Glucagon and AFP expression are mutually exclusive. The ratio of glucagon<sup>+</sup> cells:AFP<sup>+</sup> cells:Pdx1<sup>+</sup> cells varies greatly between different sections of the same *Sox9<sup>fl/fl</sup>; Pdx1-Cre* pancreas. Scale bar: 20  $\mu$ m.



**Fig. S5. Competence window for *Sox9*-deleted pancreatic progenitors to adopt hepatic fates closes by E12.5.** (A-C) AFP<sup>+</sup> or albumin<sup>+</sup> cells are not detected in the E15.5 *Sox9<sup>fl/fl</sup>; Rosa26-CreER* pancreatic epithelium (indicated by broken lines in B,C) after intraperitoneal (i.p.) tamoxifen-induced *Sox9* deletion at E12.5. li, liver. Scale bar: 50  $\mu$ m.





**Fig. S6. *Ptf1a*-deficient pancreatic progenitors retain pancreatic identity.** (Aa-Cd) Pancreatic progenitors in E10.5 wild-type (*WT*) dorsal (Aa-Ad) and ventral (Ba-Bd) pancreas express Pdx1, Sox9 and *Ptf1a*; both Pdx1 and Sox9 are expressed in dorsal pancreas of *Ptf1a*<sup>-/-</sup> mice (Ca-Cd). (Da-Fd) Like Pdx1 and Sox9, *Fgfr2* expression persists in the *Ptf1a*<sup>-/-</sup> dorsal pancreas (Fa-Fd) as in dorsal (Da-Dd) and ventral (Ea-Ed) wild-type pancreata. (H-J) At E11.5, both Sox9 and Pdx1 are maintained in *Ptf1a*<sup>-/-</sup> dorsal pancreas (H,J), as in wild-type dorsal (G,I) and ventral (G',I') pancreata. AFP is not detected in E11.5 wild-type dorsal (I) and ventral (I') pancreata or dorsal pancreas of *Ptf1a*<sup>-/-</sup> mice (J). *Ptf1a*<sup>-/-</sup> ventral pancreas is not shown as it is histologically undetectable by E11.5. Broken line demarcates the boundary between the dorsal pancreas and duodenum (Aa-Ad,Ca-Cd,Da-Dd,Fa-Fd). dp, dorsal pancreas; vp, ventral pancreas; duo, duodenum; cbd, common bile duct; li, liver. Scale bars: 20 μm.

**Table S1. Sequences of primers used for mRNA quantification by qRT-PCR**

<b>Primer</b>	<b>Forward</b>	<b>Reverse</b>
<i>Sox9</i>	5'-GAGCCGGATCTGAAGAGGGA-3'	5'-GCTTGACGTGTGGCTTG TTC-3'
<i>AFP</i>	5'-CTTCCCTCATCCTCCTGCTAC-3'	5'-ACAAACTGGGTAAAGGTGATGG-3'
Albumin	5'-TGCTTTTTCCAGGGGTGTGTT-3'	5'-TTACTTCCTGCACTAATTTGGCA-3'
Transferrin	5'-TGGGGGTTGGGTGTACGAT-3'	5'-AGCGTAGTAGTAGGTCTGTGG-3'
<i>G-6-P</i>	5'-CGACTCGCTATCTCCAAGTGA-3'	5'-GTTGAACCAGTCTCCGACCA-3'
<i>Fgfr2b</i>	5'-CCCATCCTCCAAGCTGGACTG-3'	5'-CAGAGCCAGCACTTCTGCATTG-3'
<i>Fgfr2c</i>	5'-CCCATCCTCCAAGCTGGACTG-3'	5'-TCTCACAGGCGCTGGCAGAAC-3'
<i>Fgfr4</i>	5'-TTGGCCCTGTTGAGCATCTTT-3'	5'-GCCCTCTTTGTACCAGTGACG-3'