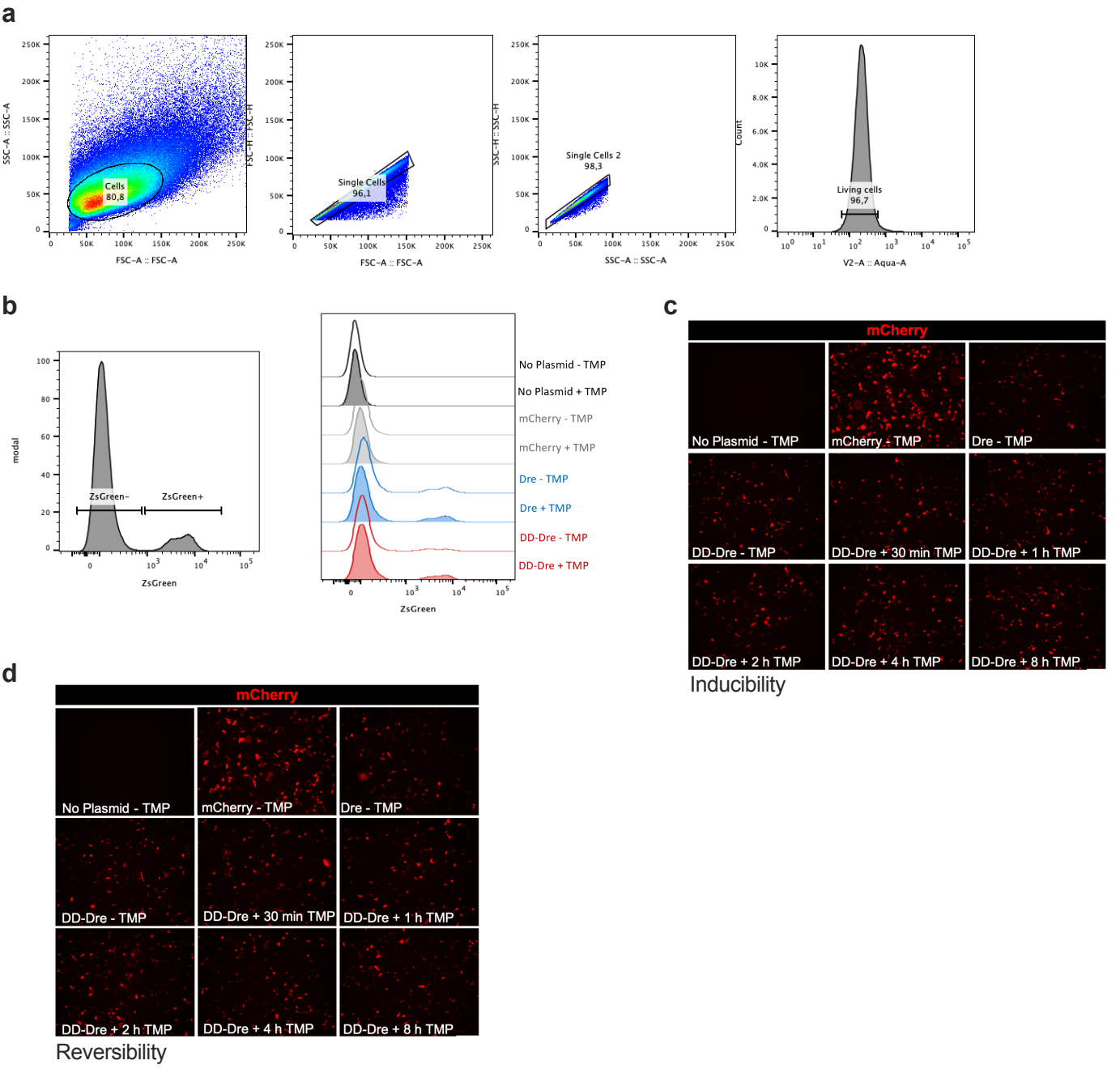


Figure S1: Assessment of a TMP-inducible Dre system *in vitro*

a) Representative FACS gating strategy for Figure 1b. b) Representative plot of the total cell population sorted by ZsGreen fluorescence and split by condition. c) Representative mCherry fluorescence microscopy images as control for a uniform transfection in each sample for Figure 1 C, D. Cells were transfected with pTEDre/pTE-DD-Dre/pCAGGS-mCherry plasmids and treated up to 8 h with TMP. Images were taken 24 h post transfection. Scale bar: 250 µm. d) Representative mCherry fluorescence microscopy images as control for a uniform transfection in each sample for Figure 1 E, F. TMP washout was monitored for 8 h. Images were taken 24 h post transfection. Scale bars: 250 µm.

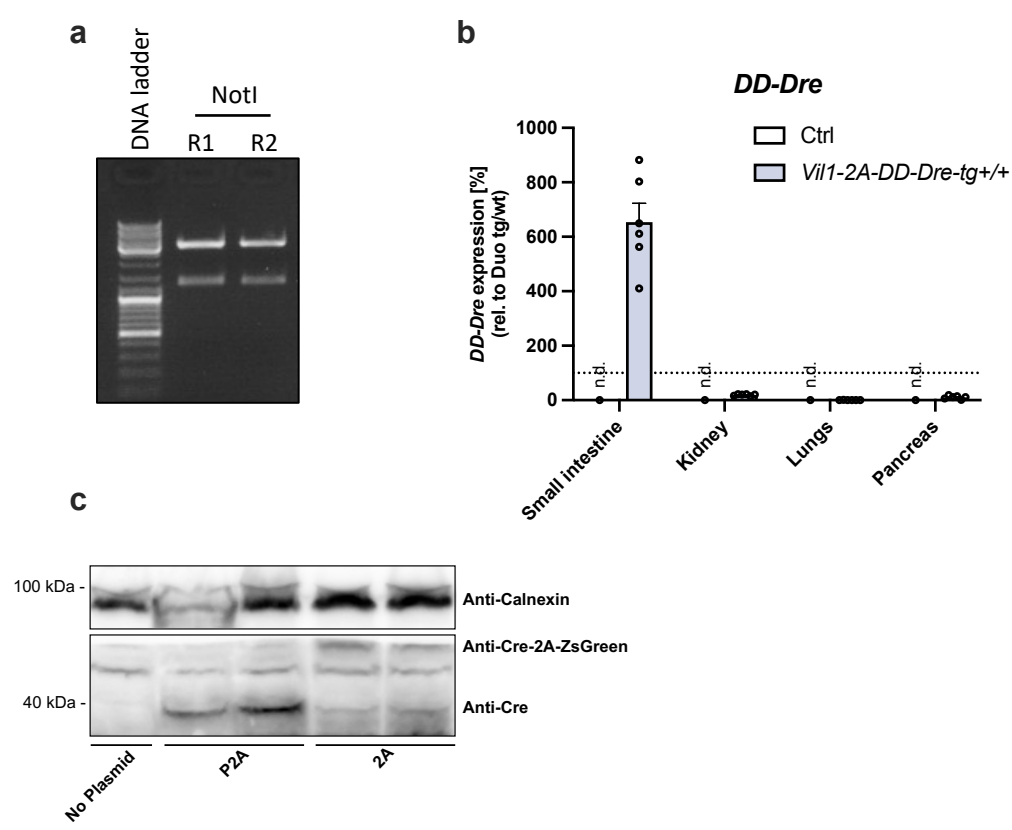
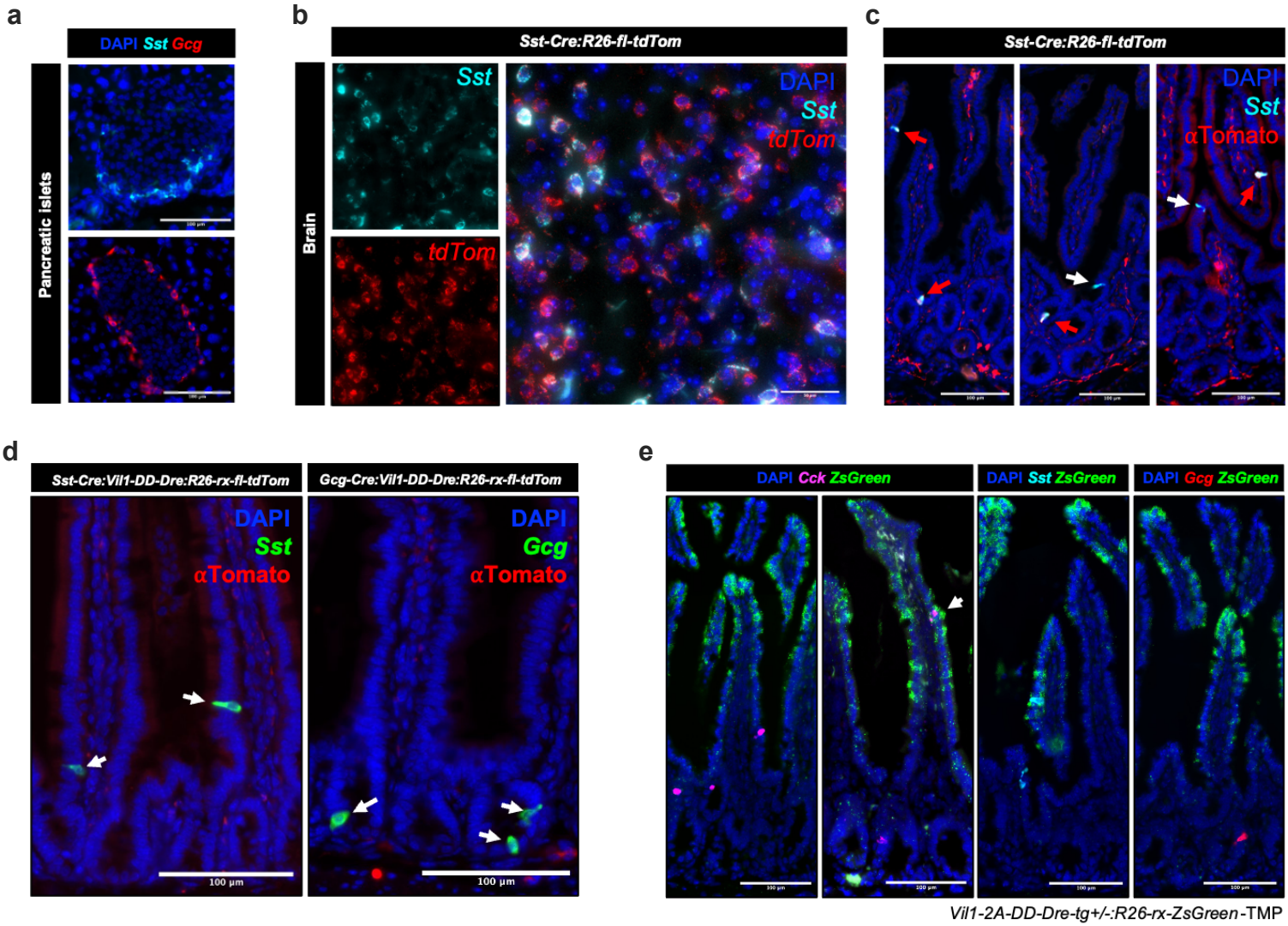
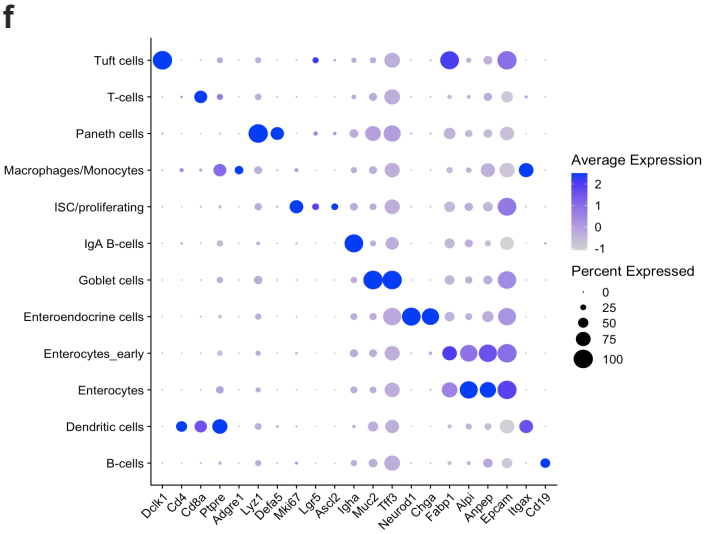
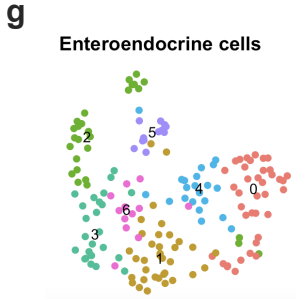
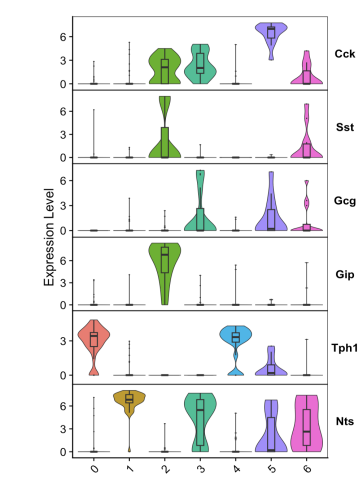


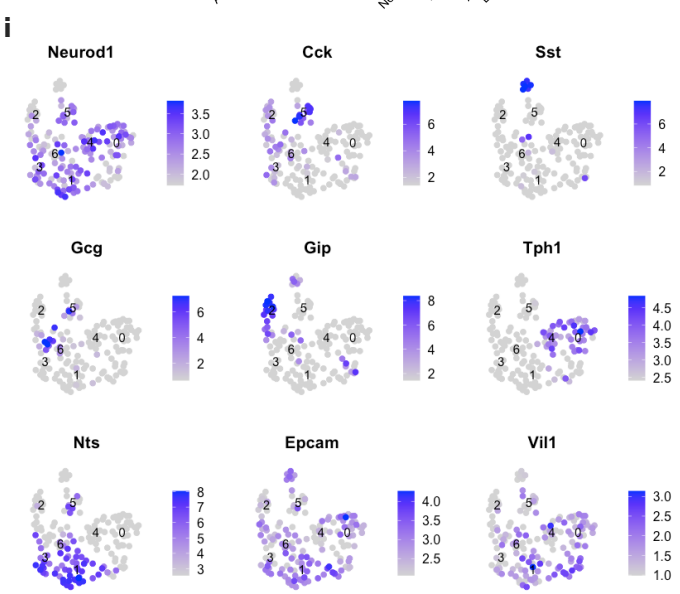
Figure S2: a) Control digest of PCR products of R1 and R2 after successful subcloning into pGEM-T easy using NotI. b) *DD-Dre* expression was measured by qPCR across organs derived from indicated mice. Data were normalized to *DD-Dre* expression in duodenum from *Vil1-2A-DD-Dre-tg+/-* mice shown in Figure 2d. c) Western blot analysis using anti-Cre and anti-Calnexin (loading control) antibodies of MEFs transiently transfected w/o plasmid, with *Cre-2A-ZsGreen* and *Cre-P2A-ZsGreen* encoding plasmids.


a
DAPI Sst Gcg
Pancreatic Islets
b
Sst-Cre:R26-fl-tdTom
Brain
Sst
tdTom
DAPI
Sst
tdTom
c
Sst-Cre:R26-fl-tdTom
DAPI
Sst
αTomato
d
Sst-Cre:Vil1-DD-Dre:R26-rx-fl-tdTom
DAPI
Sst
αTomato
100 μm
Gcg-Cre:Vil1-DD-Dre:R26-rx-fl-tdTom
DAPI
Gcg
αTomato
100 μm
e
DAPI Cck ZsGreen
DAPI Sst ZsGreen
DAPI Gcg ZsGreen
Vil1-2A-DD-Dre-tg+/-:R26-rx-ZsGreen-TMP


f
Tuft cells
T-cells
Paneth cells
Macrophages/Monocytes
ISC/proliferating
IgA B-cells
Goblet cells
Enteroendocrine cells
Enterocytes_early
Enterocytes
Dendritic cells
B-cells
Dclk1 Cd4 Cd8a Ptprc Adgre1 Lyz1 Defa5 Mki67 Lgr5 Ascl2 Igha Muc2 Tff3 Neurod1 Chga Fabp1 Alpi Anpep Epcam Itgax Cd19
Average Expression
Percent Expressed


g
Enteroendocrine cells


h
Cck
Sst
Gcg
Gip
Tph1
Nts
Expression Level


i
Neurod1
Cck
Sst
Gcg
Gip
Tph1
Nts
Epcam
Vil1

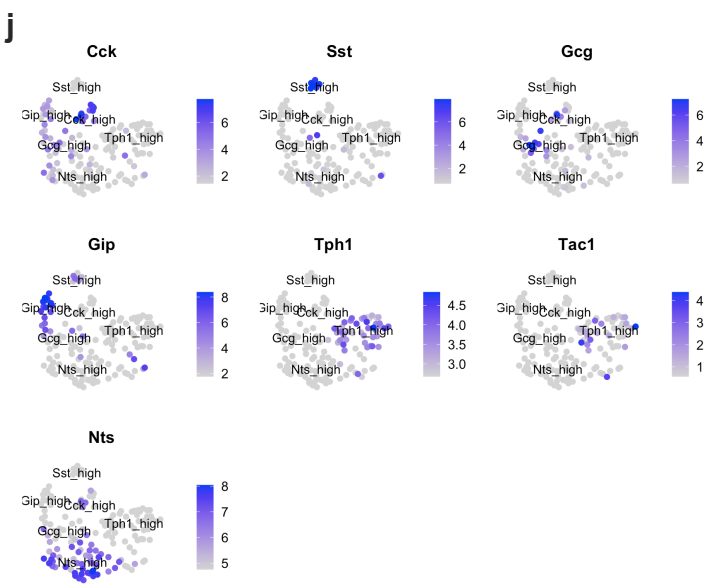

j
Cck
Sst
Gcg
Gip
Tph1
Tac1
Nts
Sst_high
Gip_high
Cck_high
Tph1_high
Gcg_high
Nts_high

Figure S3: An intersectional approach to target mature EEC populations

a) RNAScope ISH of pancreatic islets against *Sst* or *Gcg* counterstained with DAPI. b) RNAScope ISH of brain sections against *Sst* and *tdtomato* counterstained with DAPI of indicated mice. Scale bar: 50 µm. c) Anti-tdTomato IHC combined with RNAScope ISH against *Sst* of small intestines from indicated mice. Red arrows indicate *Sst*/tdTom double-positive cells. White arrows indicate *Sst*-only positive cells. d) Anti-tdTomato IHC combined with RNAScope ISH against *Sst* or *Gcg* counterstained with DAPI of small intestines from indicated mice. White arrows indicate Sst^{+}/Gcg^{+} and $tdTomato^{-}$ EECs. e) *Cck/Sst/Gcg* and *ZsGreen* multiplex RNAScope of *Vil1-2A-DD-Dre-tg+/-:R26-rx-ZsGreen* mice without TMP. White arrow indicates co-expression of *Cck* and *ZsGreen.* Scale bars in a, c, d and e represent 100 µm. f) Gene expression markers that were used for manual annotation of UMAP clusters in Figure 4e across the annotated celltypes. g) Isolated and re-clustered EEC population. 7 clusters were found. However, due to low cell numbers unsupervised clustering was not sufficient to separate EEC subpopulations. h) EEC subtype marker expression across the 7 EEC clusters. i) EEC subtype and IEC marker expression across the 7 EEC clusters. *Vil1* is mostly expressed by cluster 0 and 1. j) EECs subtype marker expression across the annotated EEC subpopulations. Cells were annotated based on threshold expression of the indicated genes.