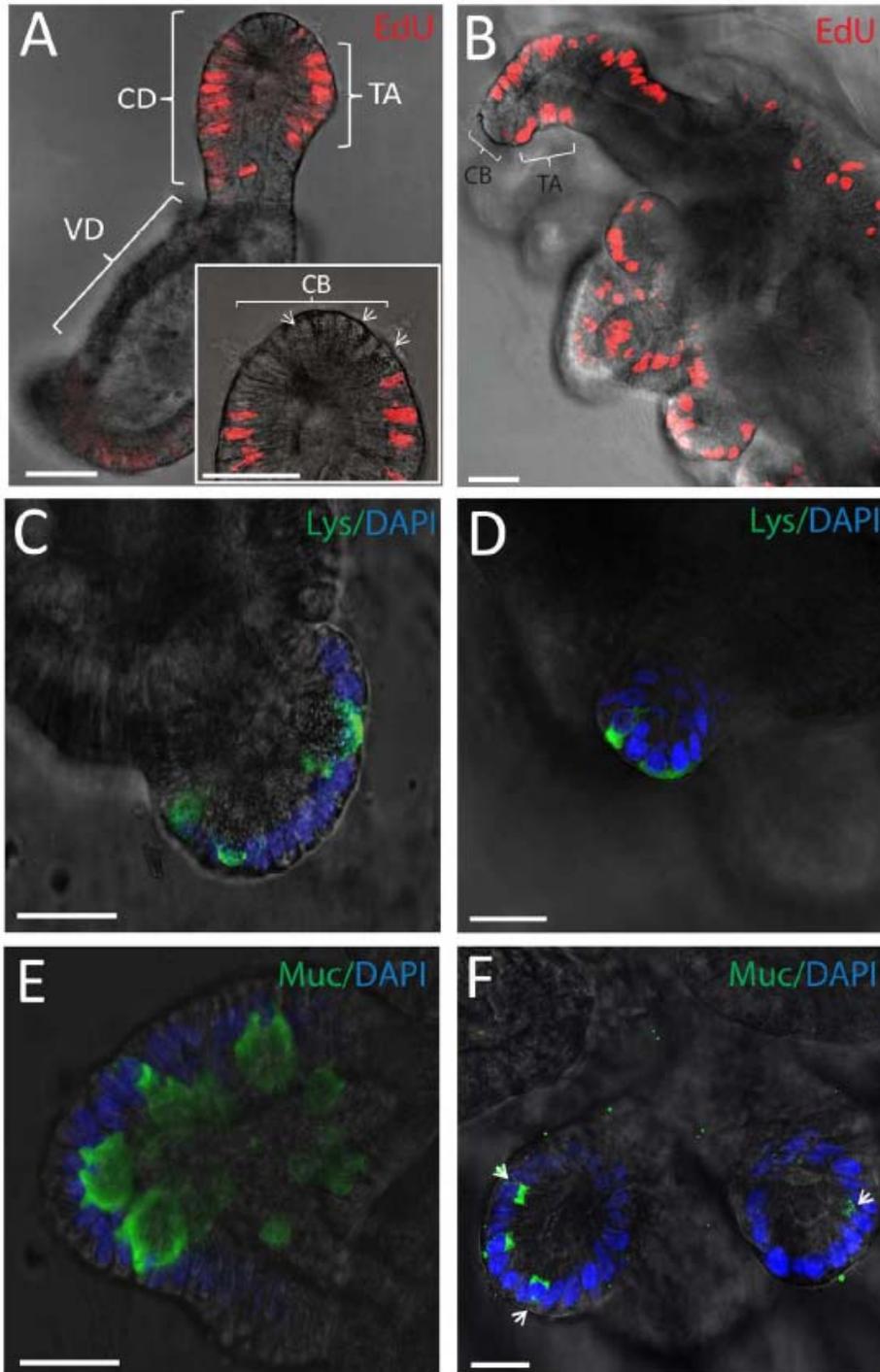


SUPPLEMENTARY DATA

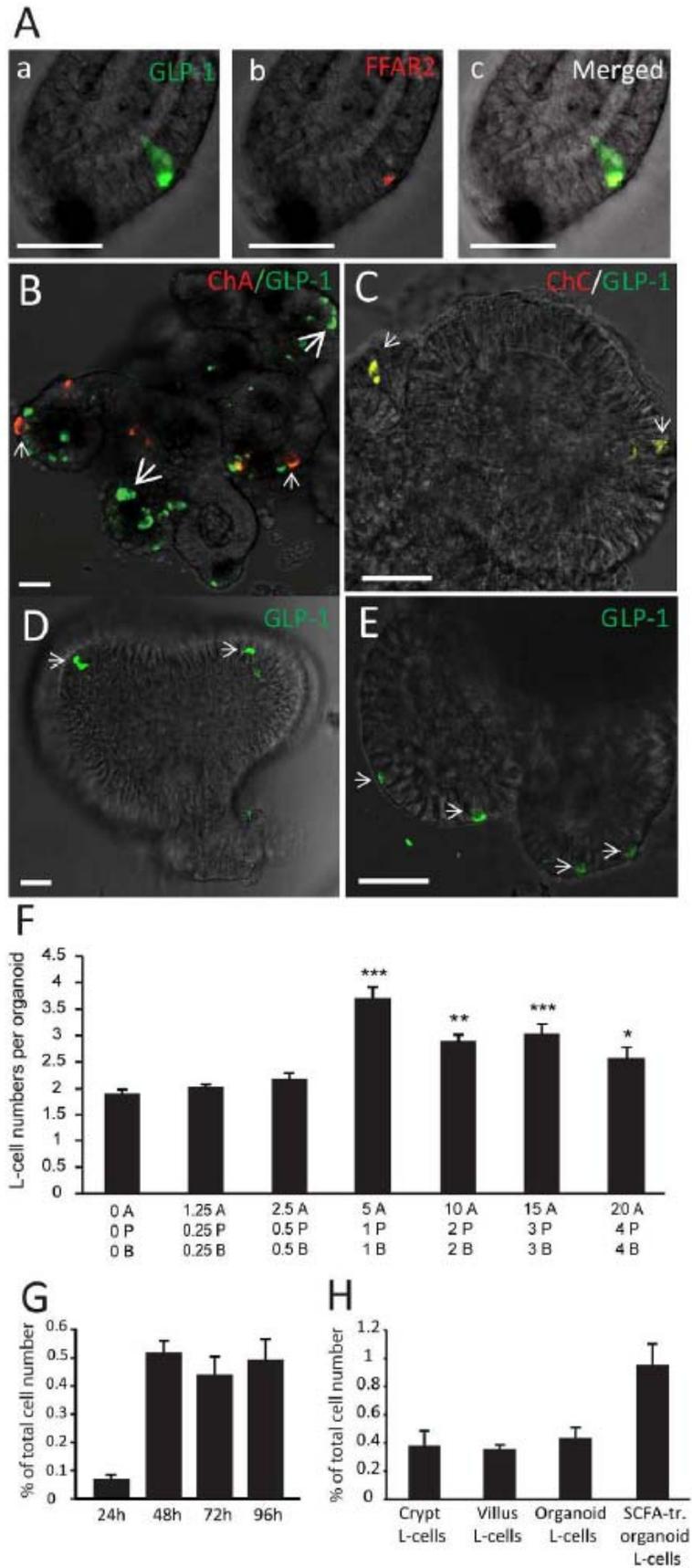
Supplementary Figure 1. Epithelial cell types in small intestine organoids. A: EdU labeling in mouse organoids shows the zone of transit-amplifying cells (TA). Crypt domain; CD and villus domain; VD. The crypt base (CB), containing intestinal stem cells, is identified by the position of a group of Paneth cells (arrowheads). B: Human organoids. Transit-amplifying cell zones, TA, are shown by EdU labeling (red) and un-stained crypt base, CB. C: Paneth cells in mouse organoids. D: Paneth cells in human organoids shown by lysozyme staining. E: Goblet cells in mouse organoids. F: Goblet cells in human organoids shown by mucin staining (arrows). All confocal images, optical slice thickness is 3 μm . Scale bars: 20 μm .



SUPPLEMENTARY DATA

Supplementary Figure 2. L-cell markers in small intestine organoids. A: GLP-1 positive cells (a) express FFAR2 (b). Merged image is shown in c. B: Presence of chromogranin A in L-cells identified by GLP-1 immunostaining (small arrowheads). Co-localisation results in yellow color. ChgA is also present in GLP-1-negative cells (large arrowheads), consistent with its definition of a pan-endocrine vesicular marker. Confocal image of an organoid, optical slice thickness 3 μm . C: Chromogranin C (red) is characteristic for GLP-1-positive L-cells (green). Co-localization results in a yellow color. All confocal images, optical slice thickness 3 μm . Scale bars: 20 μm . D: L-cells are present in mouse colon organoids, where there is no domain distinction. Scale bars: 20 μm . E: L-cells are present in human colon organoids of early passage 1-2 (green). Scale bars: 20 μm . F: Dose effect of SCFA combination on L-cell numbers in mouse small intestine organoids. Concentrations of acetate (A), propionate (P) and butyrate (B) are shown in x-axis. n= 124-132 for each test group. * p<0.05, ** p<0.01, *** p<0.001, for comparison between control and test group, by one-way ANOVA with post hoc Tukey's test. G: Proportion of L-cells in mouse organoids after passaging. Data is the mean \pm SEM calculated in 100 organoids for each group from 3 independent platings. H: Percentage of L-cells in FAC-sorted single cells in fresh crypts (n=7), fresh villi (n=6), control organoids (n=8) and SCFA-treated organoids (n=2). Data is the mean \pm SEM.

SUPPLEMENTARY DATA



SUPPLEMENTARY DATA

Supplementary Figure 3. Expression of transcription factors associated with L-cell development in whole organoids and sorted L-cells from fresh crypts and organoid-derived L-cells: Ngn3 (A), Nd1 (B), Arx (C), Foxa 1/2 (D). Data is relative gene expression determined by the Δ CT method with beta macroglobulin as endogenous control, and presented as mean \pm SEM. N=3-6 for whole organoids, n=3 for crypt L-cells and n=3-5 for organoid L-cells.

