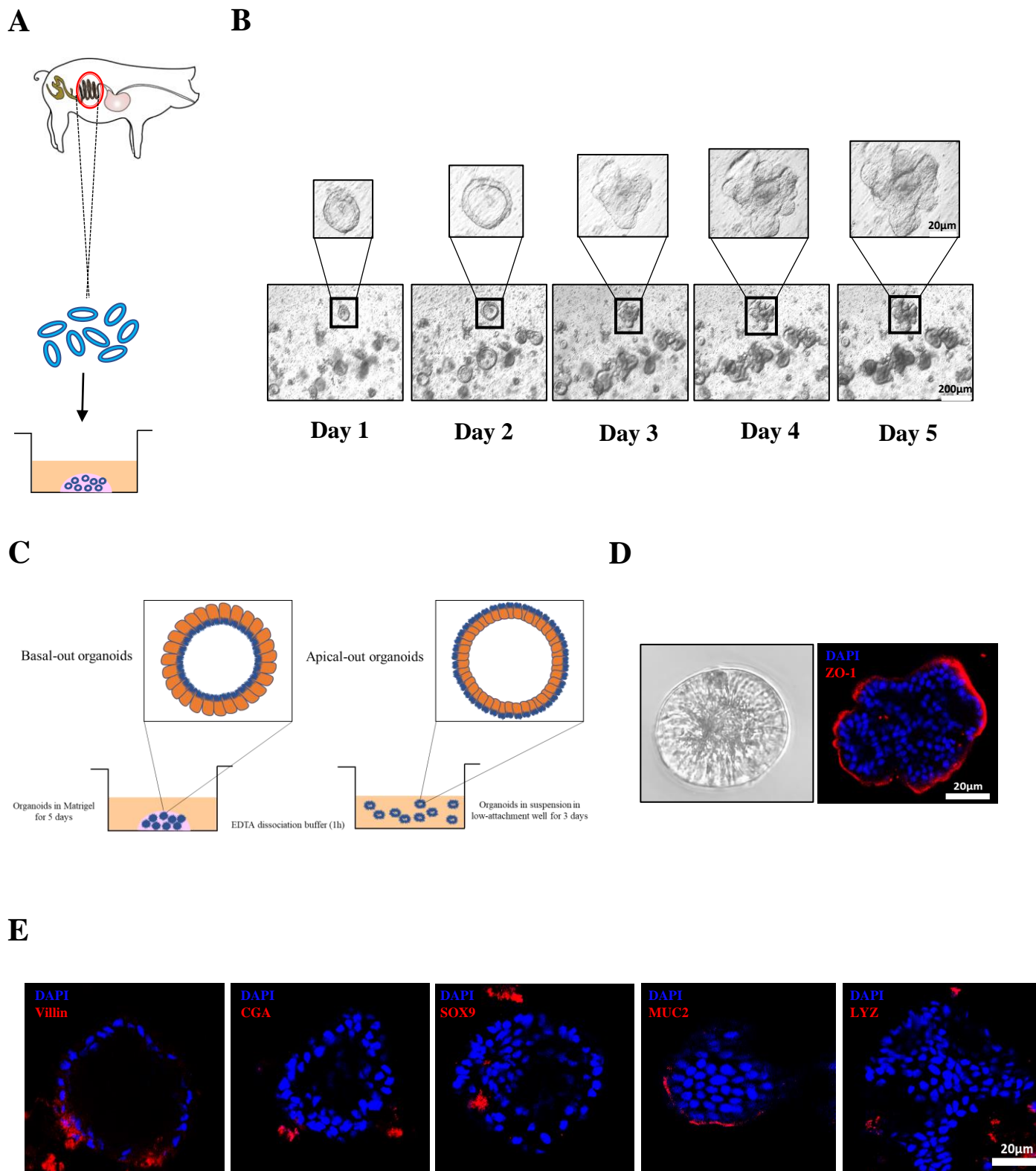
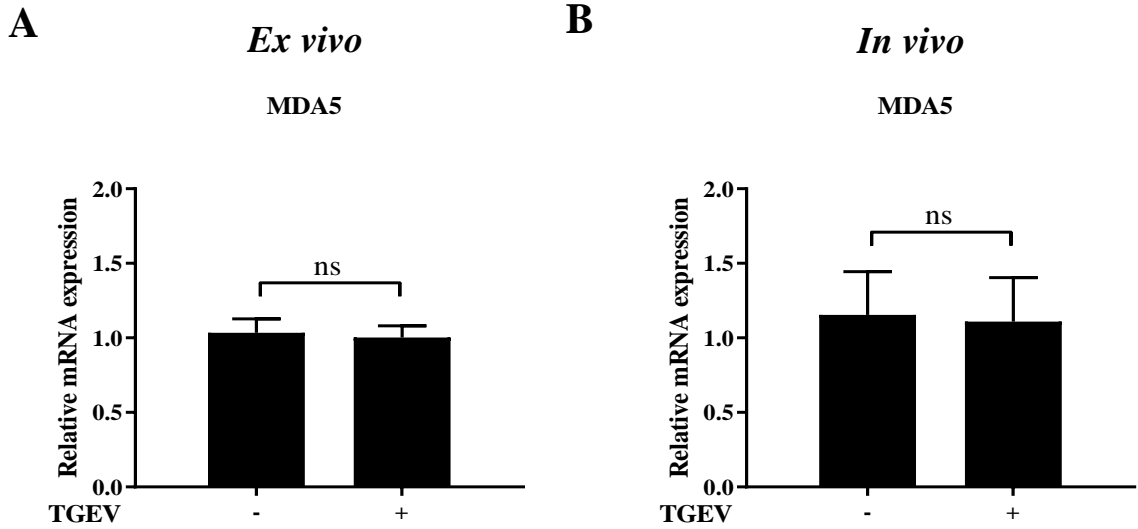


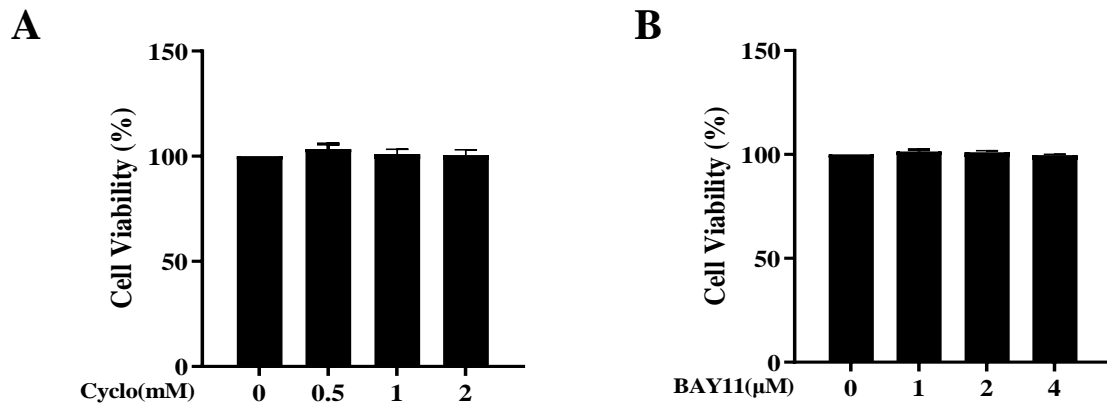
**Fig. S1 Establishment of apical-out porcine intestinal organoids.** (A) Graphical representation for isolation of porcine intestinal crypts. (B) Culture of intestinal organoids from 1 to 5 days; scale bar: 20/200  $\mu\text{m}$  (C) Graphical representation for generation of apical-out organoids (D) Apical-out organoids were stained with ZO-1; scale bar: 20  $\mu\text{m}$  (E) Apical-out organoids was subjected to IFA staining for absorptive enterocytes (Villin), enteroendocrine cells (CGA), stem cells (SOX9), goblet cells (MUC2) and Paneth cells (LYZ) ; scale bar: 20  $\mu\text{m}$ .



**Fig. S2 TGEV infection can not affect MDA5 expression** (A) The transcription level of MDA5 in apical-out organoids at 48 h post TGEV infection was evaluated by RT-qPCR. (B) Transcriptional level of MDA5 in ileum was detected by RT-qPCR. Results are presented as mean  $\pm$  SD of data from three independent experiments ns, no significant determined by two-tailed Student's *t*-test.

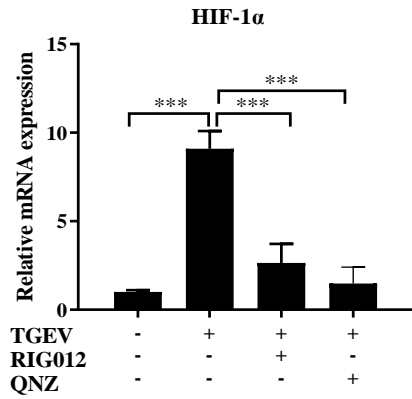


**Fig. S3 Cytotoxicity of Cyclo and BAY11-7082 (A-B)** Intestinal organoids were incubated with different concentrations of Cyclo (A) or BAY11 (B) for 48 h, which was assessed by Cell Counting Kit 8. Results are presented as mean  $\pm$  SD of data from three independent experiments ns, no significant determined by two-tailed Student's *t*-test.

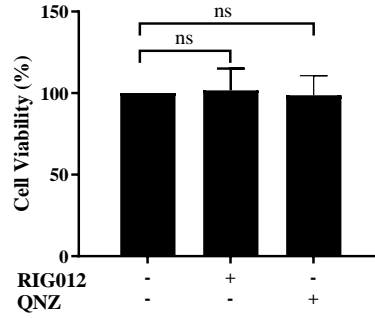


**Fig. S4 RIG-I–NF- $\kappa$ B pathway regulates HIF-1 $\alpha$  expression upon TGEV infection.** (A) Intestinal organoids were infected with TGEV followed by RIG012 (5  $\mu$ M) or QNZ (10 nM) treatment for 48 h. Transcription levels of HIF-1 $\alpha$  in the intestinal organoids post TGEV infection were measured by RT-qPCR. (B) Intestinal organoids were incubated with RIG012 (5  $\mu$ M) or QNZ (10 nM) treatment for 48 h, which was assessed by Cell Counting Kit 8. Results are presented as mean  $\pm$  SD of data from three independent experiments ns, no significant; \*\*\*,  $P \leq 0.001$ , determined by two-tailed Student's  $t$ -test.

**A**

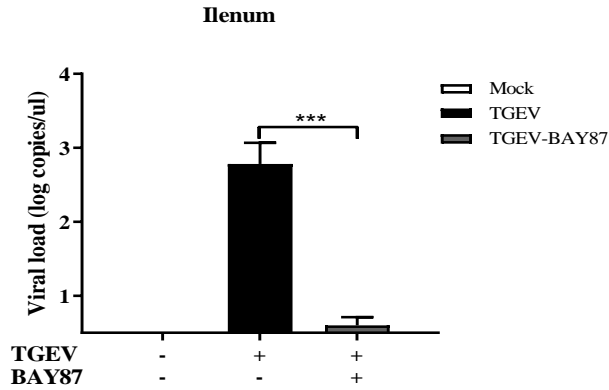


**B**

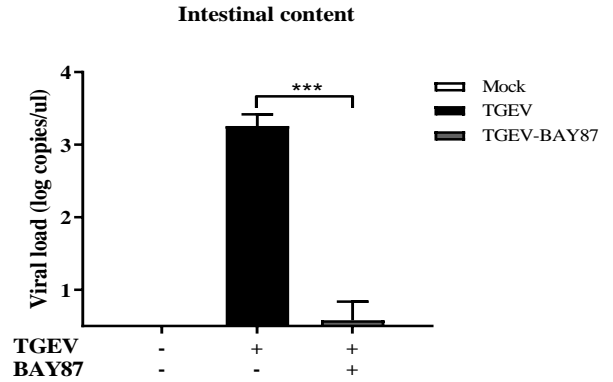


**Fig. S5 Oral administration of BAY87 inhibits TGEV infection in the ileum and intestinal content.** (A-B) TGEV genome copy numbers in the ileum (A) and intestinal content (B) were detected by RT-qPCR. Results are presented as the mean  $\pm$  SD of data from three independent experiments \*\*\*,  $P \leq 0.001$  determined using the two-tailed Student's *t*-test.

**A**



**B**



**Fig. S6 BAY87 cannot change ALT and AST level in serum of pigs.** (A) ALT and AST were detected in serum from piglets sacrificed at 24 hpi. Results are presented as mean  $\pm$  SD of data from three independent experiments ns, no significant determined by two-tailed Student's *t*-test.

**A**

