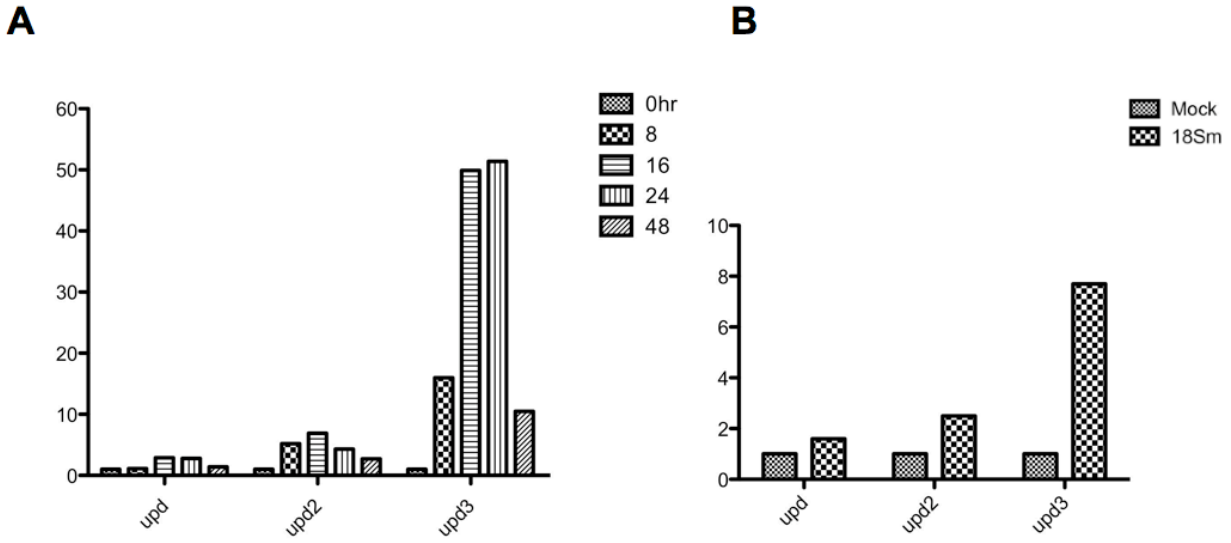
## Supplementary material




| **Normalized ct value** | 0hr | 8hr | 16hr | 24hr | 48hr |
|---|---|---|---|---|---|
| Upd | 34.5 | 34.32 | 32.97 | 33.00 | 34.06 |
| Upd2 | 32.02 | 29.66 | 29.24 | 29.94 | 30.62 |
| Upd3 | 29.73 | 25.75 | 24.11 | 24.07 | 26.36 |

**Fig. S1. Expression levels of the *upd* family genes in response to enteric infection**
**(A)** Time course of *upd* family gene expression in response to *Erwinia carotovora* (Ecc15). The graph indicates the fold induction with respect to the 0hr time point for each gene. The table below the graph indicates the ct values for each gene at each time point after normalization to the corresponding ct values of RP49 expression.
**(B)** *upd* family gene expression in response to 18-hour *Serratia marcescens* infection.

Quantitative RT-PCR results from a representative set of data from three independent experiments. Total gut RNA was extracted from 40-60 fly guts with Trizol reagent (Invitrogen). cDNA was generated by SuperScript II reverse transcriptase (Invitrogen) from the template RNA, and analyzed by quantitative real-time PCR using the LightCycler 480 Master Kit and LightCycler 480 instrument (Roche). Expression levels were normalized to *rp49*. The levels of the *upd* family genes in mock-infected animals were arbitrarily assigned to 1.

Sequence of the primers used for real-time PCR:
updL: gcacactgatttcgatacgg; updR: ctgccgtggtgctgtttt
upd2L: aagttcctgccgaacatga; upd2R: agagtgctcgtcagtgactgtt;
upd3L: cccagccaacgatttttatg. upd3R: tgttaccgctccggctac.
rp49L: cttcatccgccaccagtc; rp49R: cgacgcactctgttgtcg.

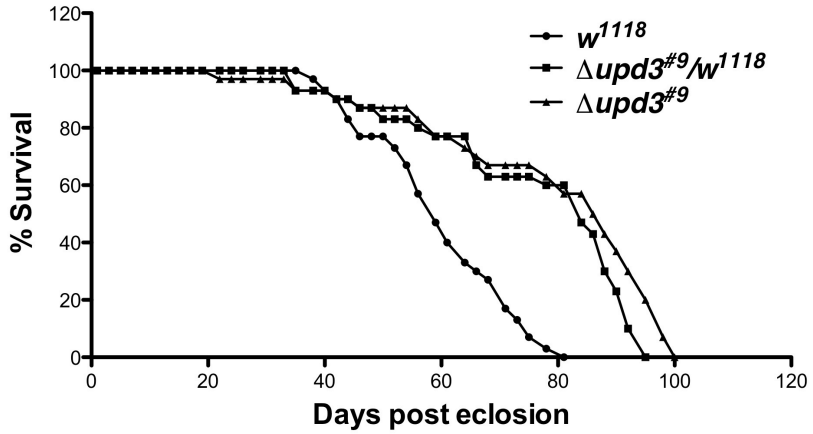



**Fig. S2. Lifespan of the *upd3* mutant**

The percentage of survivors in a cohort of 40 flies is plotted against days post-eclosion. Wild-type ($w^{1118}$) flies displayed a life-span of ~ 80 days after eclosion. The heterozygote ($\Delta upd3^{\#9}/w^{1118}$) and *upd3* ($\Delta upd3^{\#9}$) mutant flies displayed a life-span of ~100 days after eclosion. The number of survivors was scored every 2 days. The food medium was replaced every 4 days for the duration of the experiment.

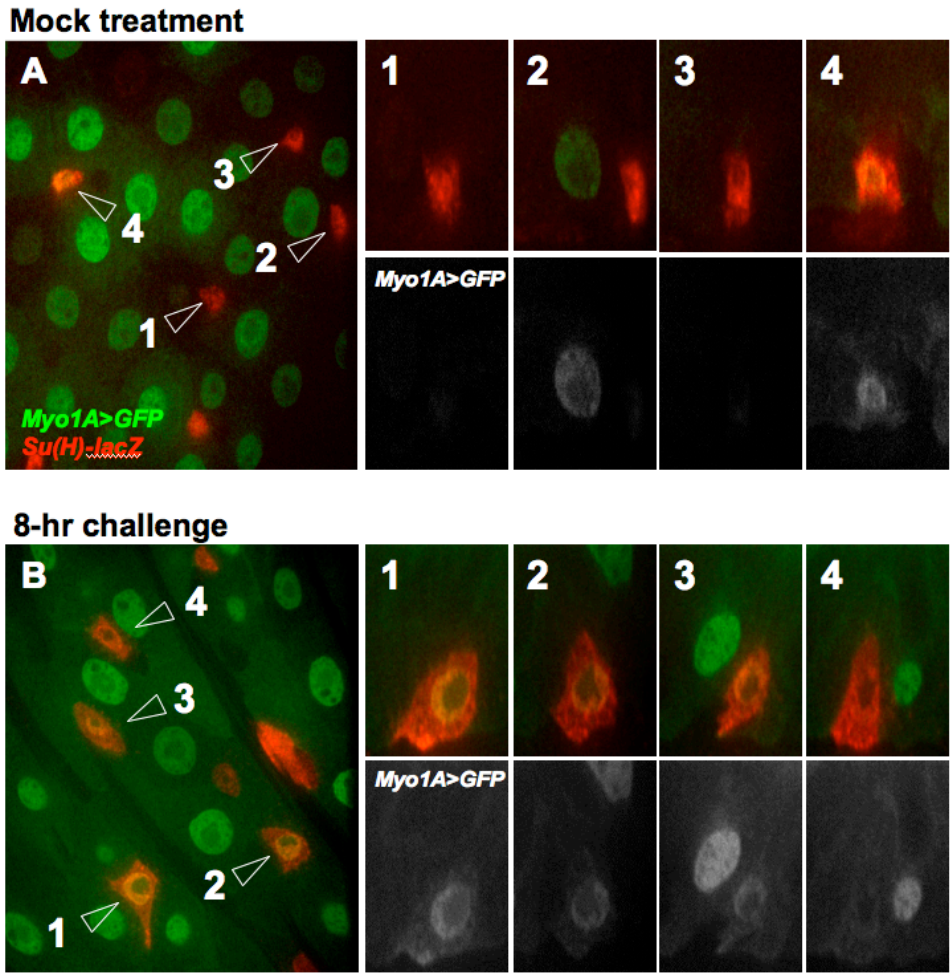



**Fig. S3. EB-to-EC differentiation is characterized by inflated nuclei and expression of the enterocyte marker Myo1A.**

A) and (B), XY images obtained by confocal microscopy of posterior midguts showing enterocytes as large Myo1A-positive cells and enteroblasts as small Su(H)GBE-lacZ-positive cells. The top right panels (1-4) show the XZ reconstruction corresponding to the enteroblasts shown in the XY images (1-4, left panels). The corresponding bottom right panels show the Myo1A marker only.

(A) In mock-treated animals, less than 25% of enteroblasts display large nuclei and express the Myo1A marker.

(B) After 8 hours of *Ecc15* infection, more than 75% of EBs display large nuclei as revealed by nuclear LacZ and express the Myo1A marker as revealed by nuclear and cytoplasmic GFP.

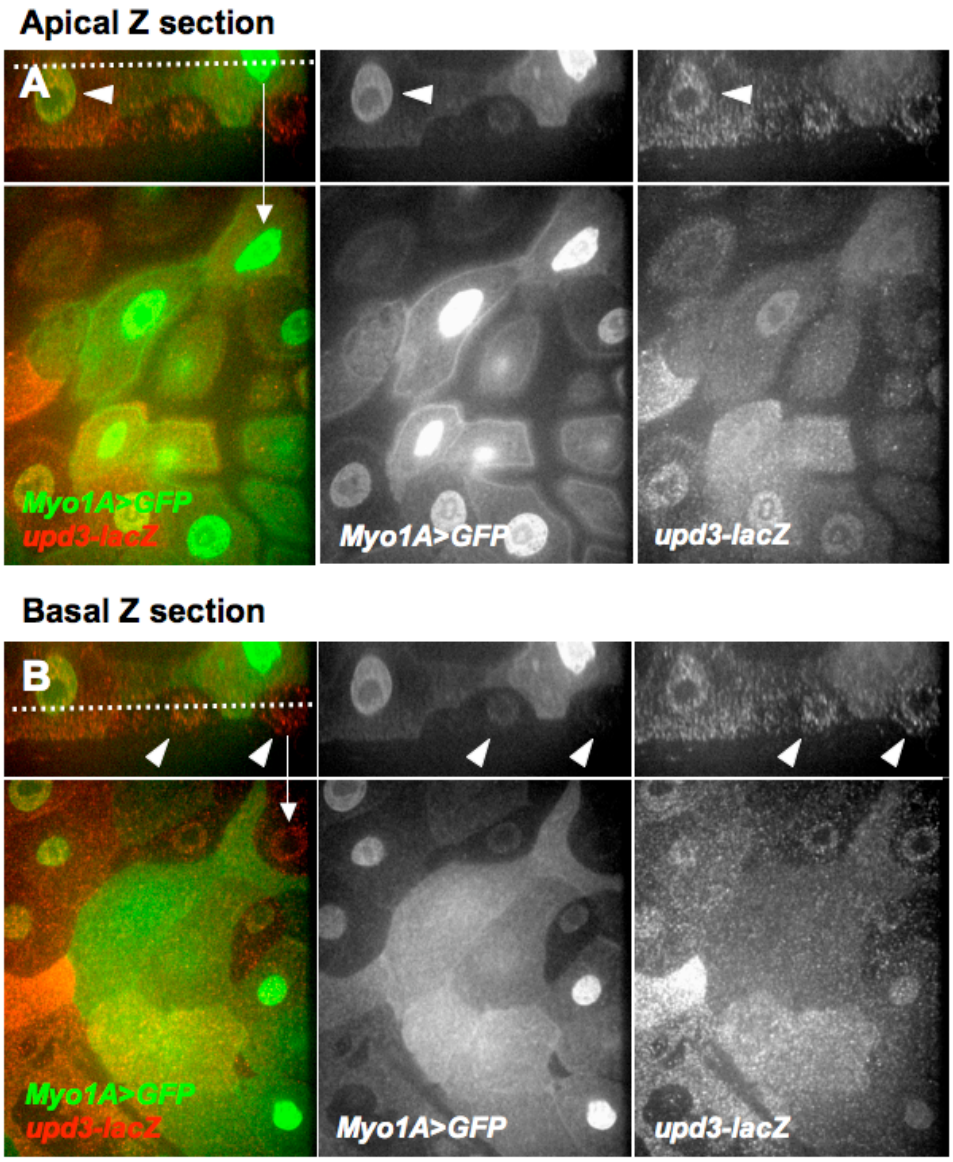



**Fig. S4. Differentiated enterocytes and differentiating EBs express the Myo1A marker and the *upd3-lacZ* reporter in response to challenge.**
(A and B) Confocal microscopy of the same field of view showing apical (A) and basal (B) images of the intestinal epithelium after 8 hours of challenge with *Ecc15*. XY confocal images are shown in the bottom panels for (A) and (B) and XZ reconstructions are shown in corresponding top panels. Apical Z section shows large cells expressing the Myo1A marker and the *upd3-lacZ* reporter, which are mature enterocytes (Apical Z section, top left panel, arrowhead). Underneath the layer of enterocytes, the basal Z section reveals the presence of small cells displaying weak Myo1A expression and expressing the *upd3-lacZ* reporter, which are differentiating enteroblasts (Basal Z section, top and bottom left panel, arrowheads).