



**Supplementary Figure 1.** (A) Loss of mitochondrial pyruvate metabolism in enterocytes results in increased pHH3+ cells in intestines. The EC-specific *myo1A*-GAL4 driver was used to target RNAi for *mCherry* (control) or *dMPC1* in the presence of *Tub-GAL80ts*. Proliferating ISCs are marked by staining for pHH3+ cells (red) and nuclei are stained with DAPI (blue). Images display the R4 region of the intestine. This data is quantified in Figure 1A. Scale bar represents 50  $\mu$ m. (B) Adding *mCherry* RNAi in the background of *dMPC1* RNAi has no effect on ISC proliferation. The *myo1A*-GAL4 driver was used to target RNAi against *mCherry* or *dMPC1*, either alone or in combination. The number of cells staining positive for phosphorylated histone H3 (pHH3+ cells) was quantified per intestine after shifting 4-5 day old animals to 29°C for seven days. Data is plotted as the mean  $\pm$  SEM,  $n \geq 20$  animals for each condition. (C) Feeding N-acetylcysteine (NAC) has no effect on ISC proliferation. The *myo1A*-GAL4 driver was used to target RNAi against *mCherry* (control) or *dMPC1*, in the presence or absence of NAC feeding. The number of cells staining positive for phosphorylated histone H3 (pHH3+ cells) was quantified per intestine after shifting 4-5 day old animals to 29°C for seven days. Data is plotted as the mean  $\pm$  SEM,  $n \geq 20$  animals for each condition. NS=not significant.