

Identification of adult midgut precursors in *Drosophila*

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Supplemental Information

Table S1

Summary of pathway components screened. Genotypes and [phenotypes](#) as indicated.

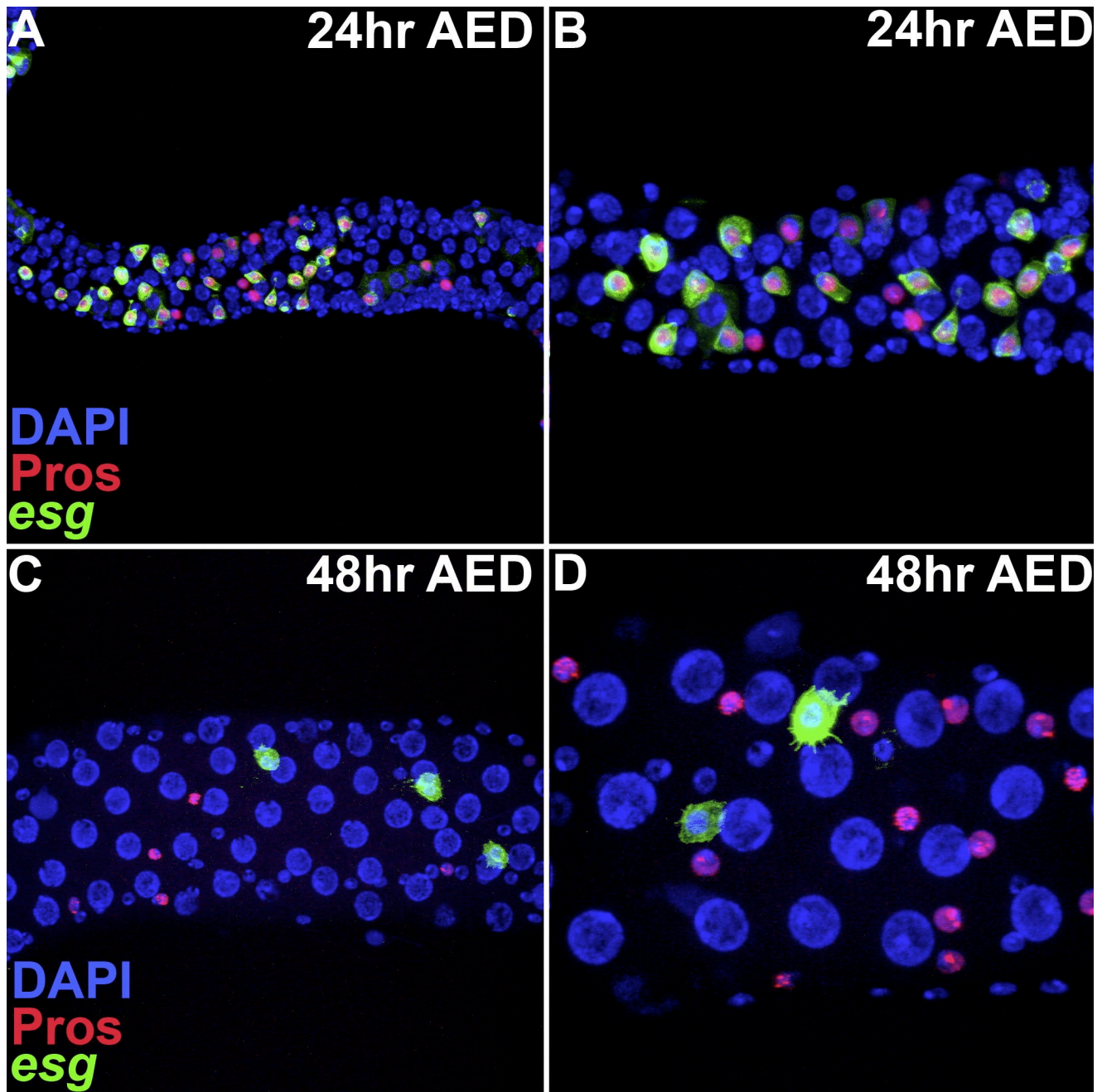
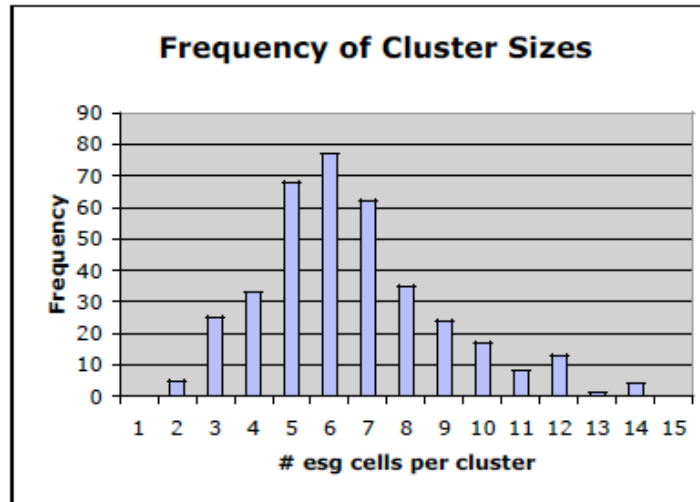


Figure S1

Molecular markers define midgut cell types. (A–D) Distinct populations of small cells can be defined in *esg* > *GFP* midguts (anti-GFP, green; anti-Pros, red; DAPI, blue); *esg*⁺*Pros*[−], *Pros*⁺*esg*[−], *esg*⁺*Pros*⁺. (A and B) First instar. (A) Original magnification, 80×. (B) Original magnification, 160×. (C and D) Second instar. (C) Original magnification, 80×. (D) Original magnification, 160×.

A.



B.

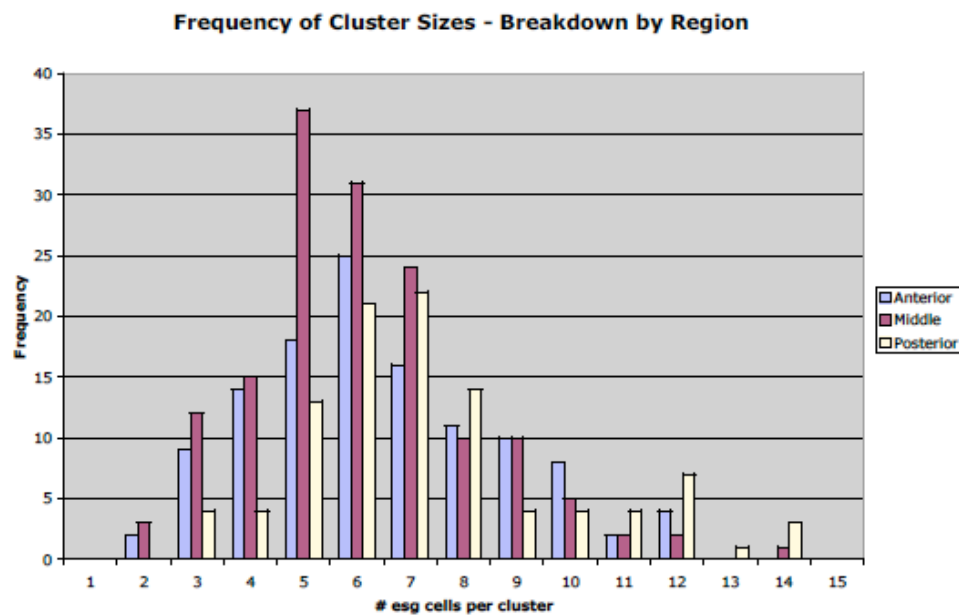


Figure S2

AMP cluster size and distribution in the P3 midgut. (A and B) Histogram displaying the frequency of AMP cluster cell number. The number of DAPI nuclei was counted in *esg*⁺ AMP clusters at WPP. (A) Combined total frequency of AMP cluster cell number from three regions sampled from each gut analyzed; anterior, middle, posterior. (B) Histogram displaying the data in (A) as a function of midgut position. Counts were obtained by scoring single confocal optical sections; thus, these values likely represent a low estimate of actual number of cells.

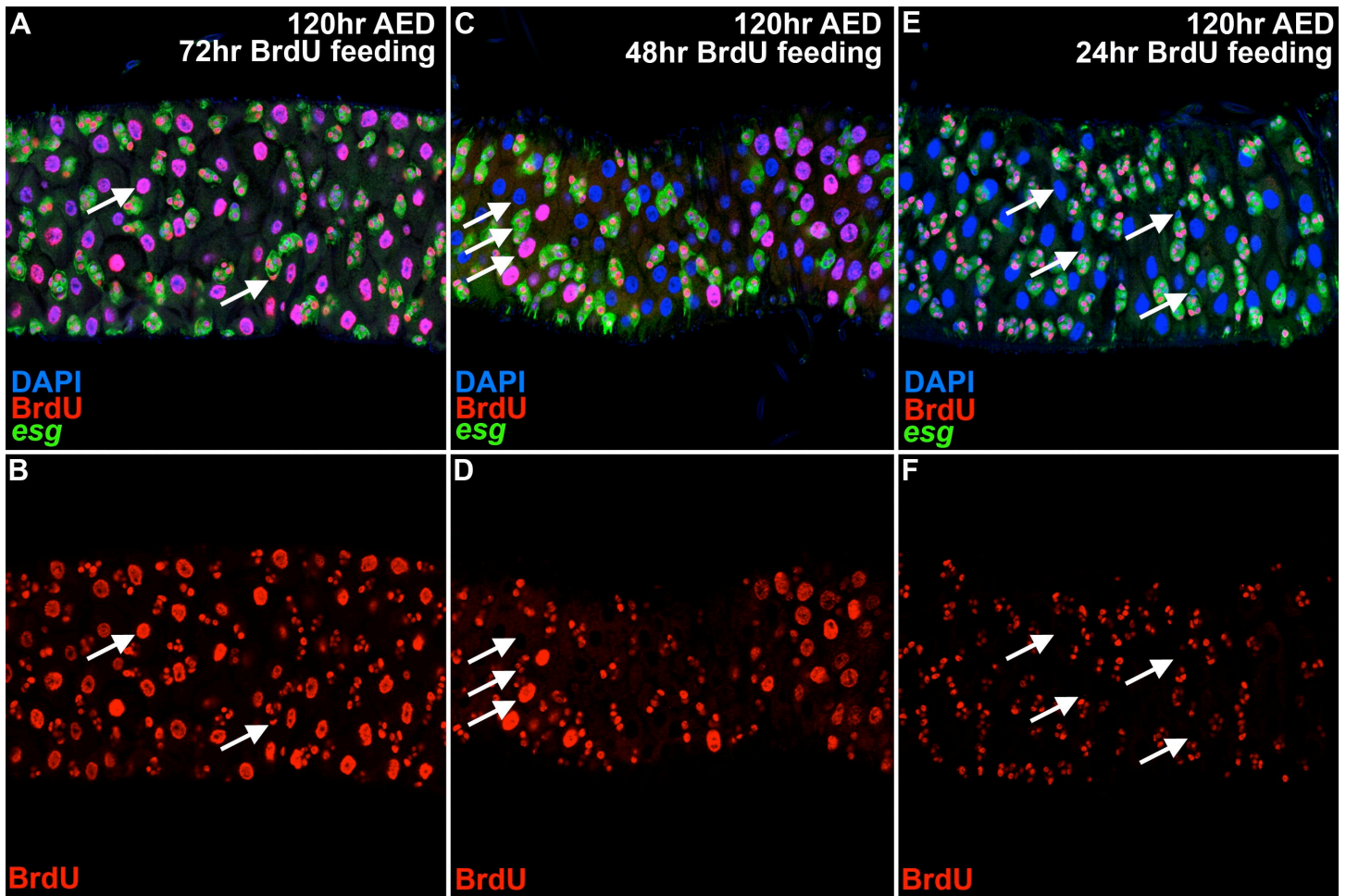


Figure S3

BrdU incorporation in the midgut during larval stages. The extent of BrdU incorporation in the WPP midgut was examined after (A and B) 72 h. (C and D) 48 h and (E and F) 24 h on BrdU-containing media. Arrows indicate different cell types labeled at each time point. Note that cells of the midgut cease [DNA synthesis](#) at different developmental times. 72 h on BrdU labels all cells of the larval midgut and AMPs. 48 h pulse labels many cells of the larval midgut cells and AMP clusters. 24 h pulse does not label larval cells and labels most AMPs. We frequently observed outer cells in the AMP clusters that were unlabeled. Original magnification, 40×.

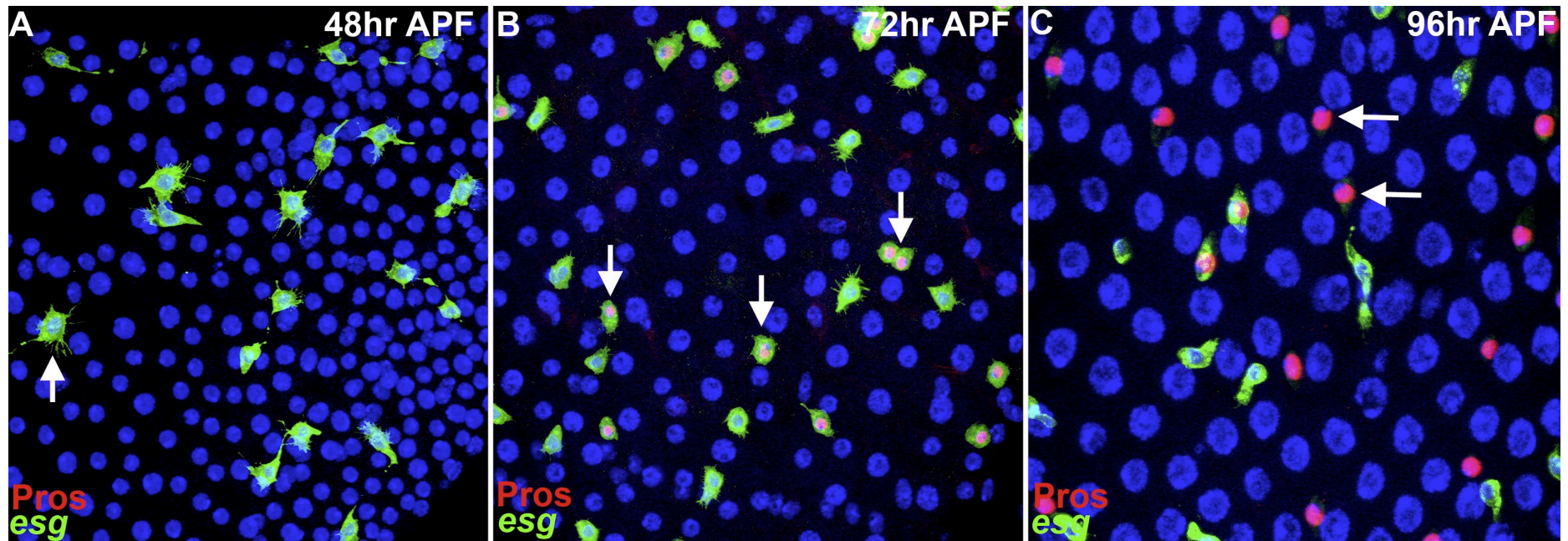


Figure S4

Pros⁺ cells are detectable in late pupal stages. (A–C) Re-emergence of *Pros*⁺ cells in *esg* > *GFP* midguts (anti-Pros, red; anti-GFP, green; *DAPI*, blue). (A) Mid P4. *esg*⁺ cells are expanding; few *Pros*⁺ cells are detected. Original magnification, 80×. (B) Early P5. Many double-positive *esg*⁺*Pros*⁺ cells detected. Original magnification, 80×. (C) Late P5. *Pros*⁺ cells detected. Original magnification, 120×.

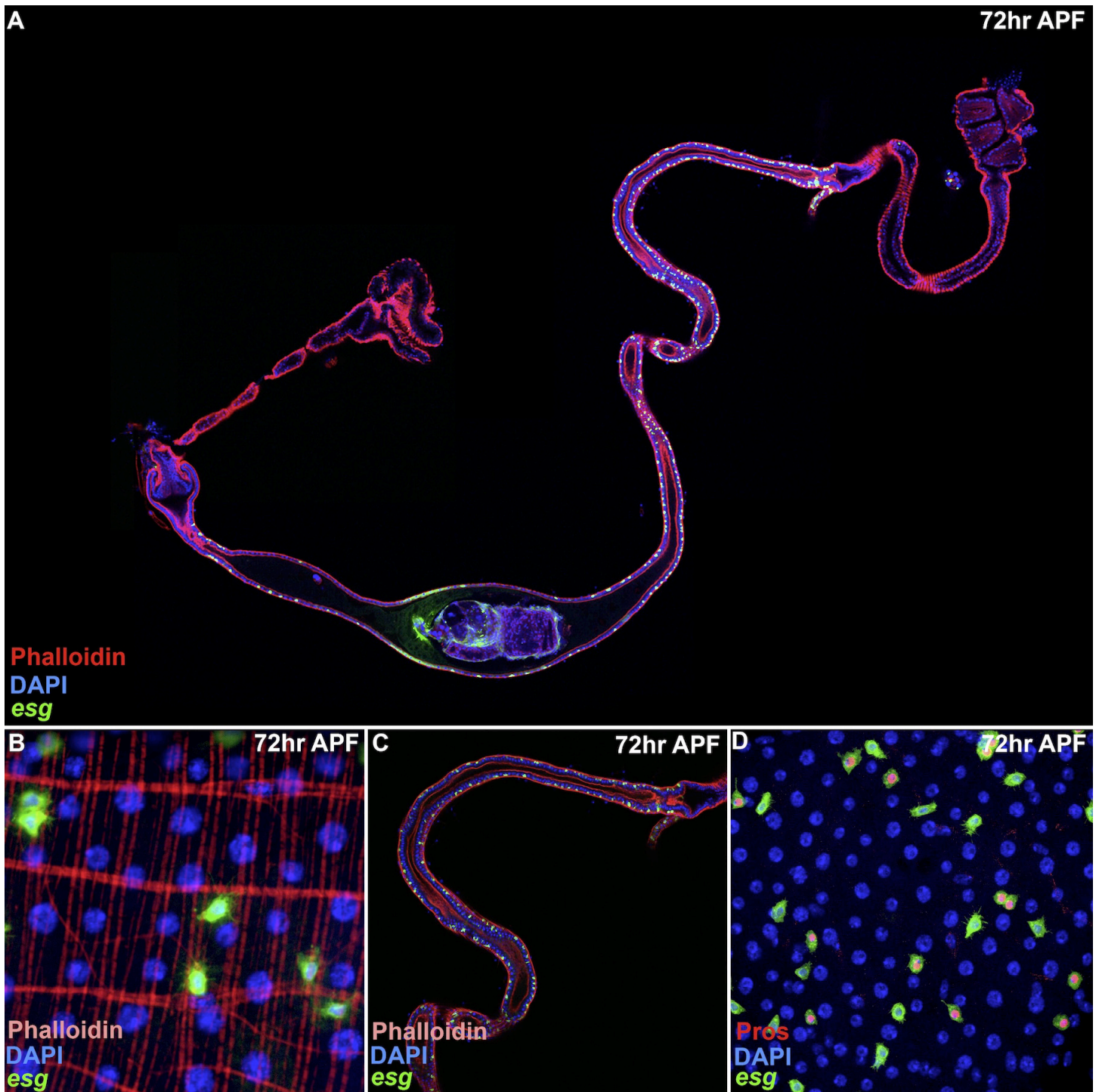


Figure S5

Pupal midgut at 72 h APF. (A–C) *esg* > *GFP* midgut at 72 h APF (phalloidin, red; anti-GFP, green; DAPI, blue). (A) Cross-section. Midgut looping is initiated. Composite micrograph; original magnification, 20×. (B) Superficial section. Original magnification, 160×. (C) Cross-section, original magnification, 20×. (D) Pros⁺ cells can be detected (red). Original magnification, 80×.

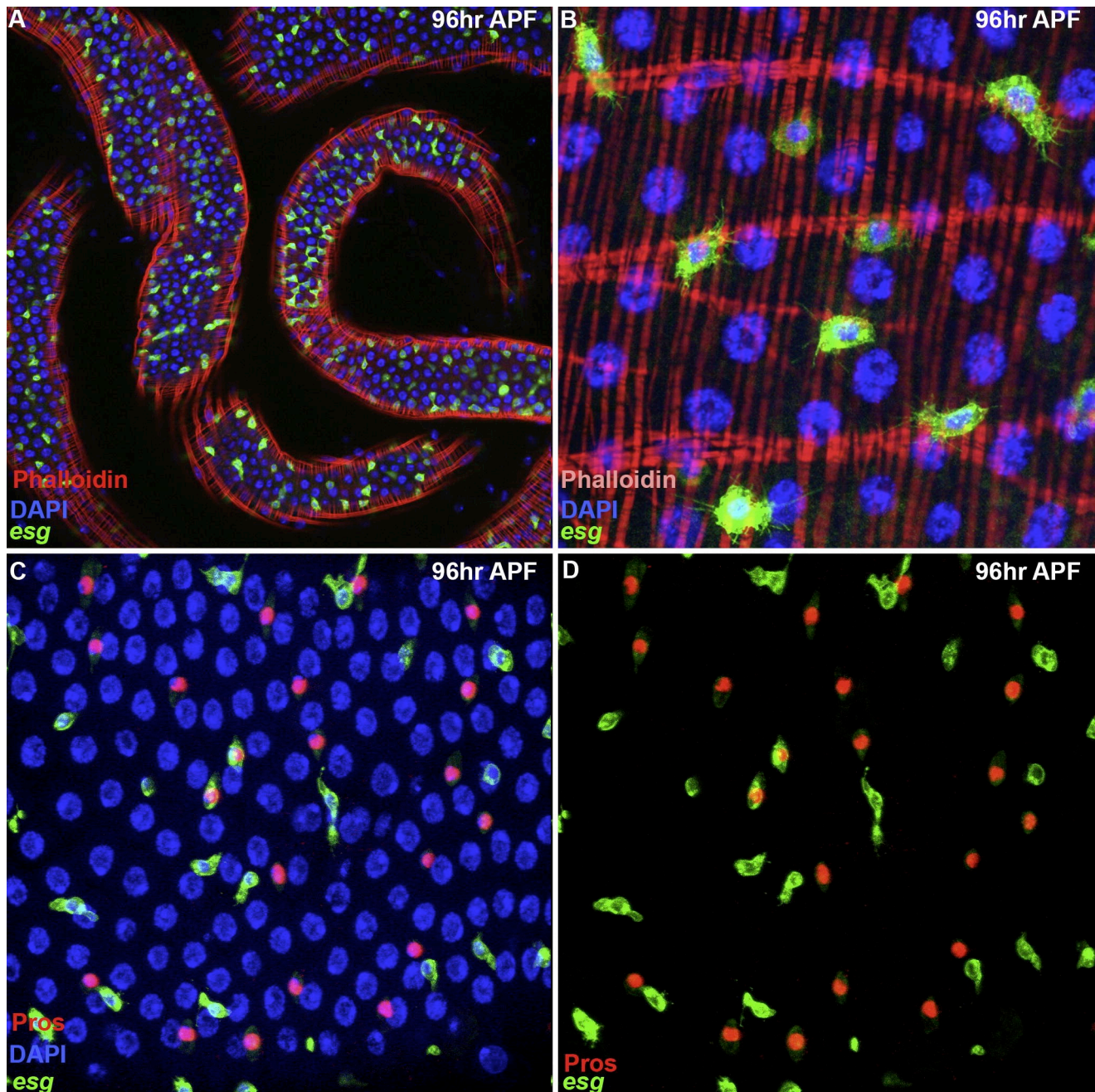


Figure S6

Pupal midgut at 96 h APF. (A–D) *esg* > *GFP* midgut at 96 h APF (anti-GFP, green; *DAPI*, blue). (A) Superficial section, phalloidin, red. Midgut looping is complete. Original magnification, 40×. (B) Superficial section, phalloidin, red. Original magnification, 160×. (C and D) Pros⁺ cells detected in doublets, as in the adult midgut. Original magnification, 120×.