

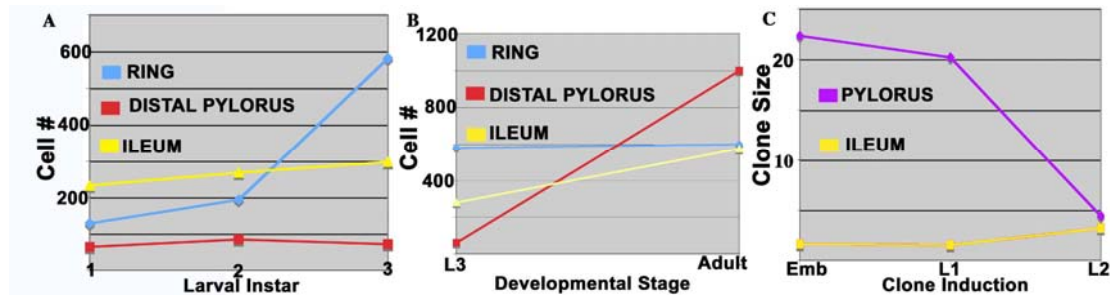
## Supplemental Data

### The *Drosophila* Hindgut Lacks

### Constitutively Active Adult Stem Cells

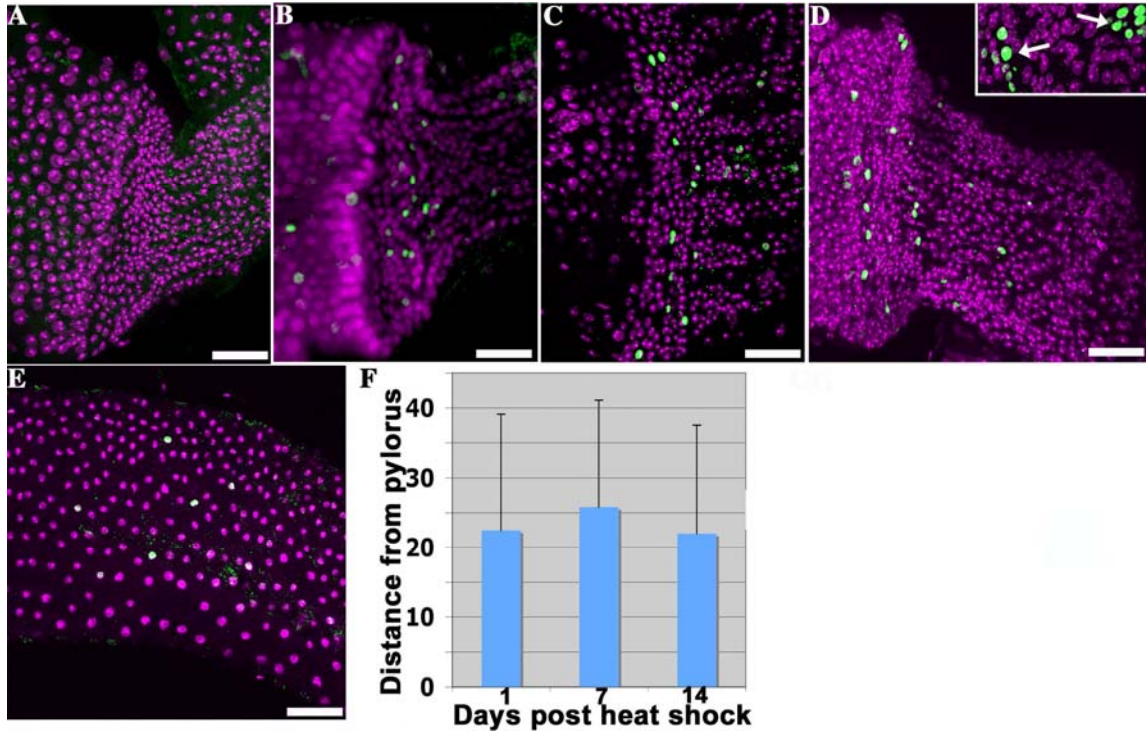
### but Proliferates in Response to Tissue Damage

Donald T. Fox and Allan C. Spradling



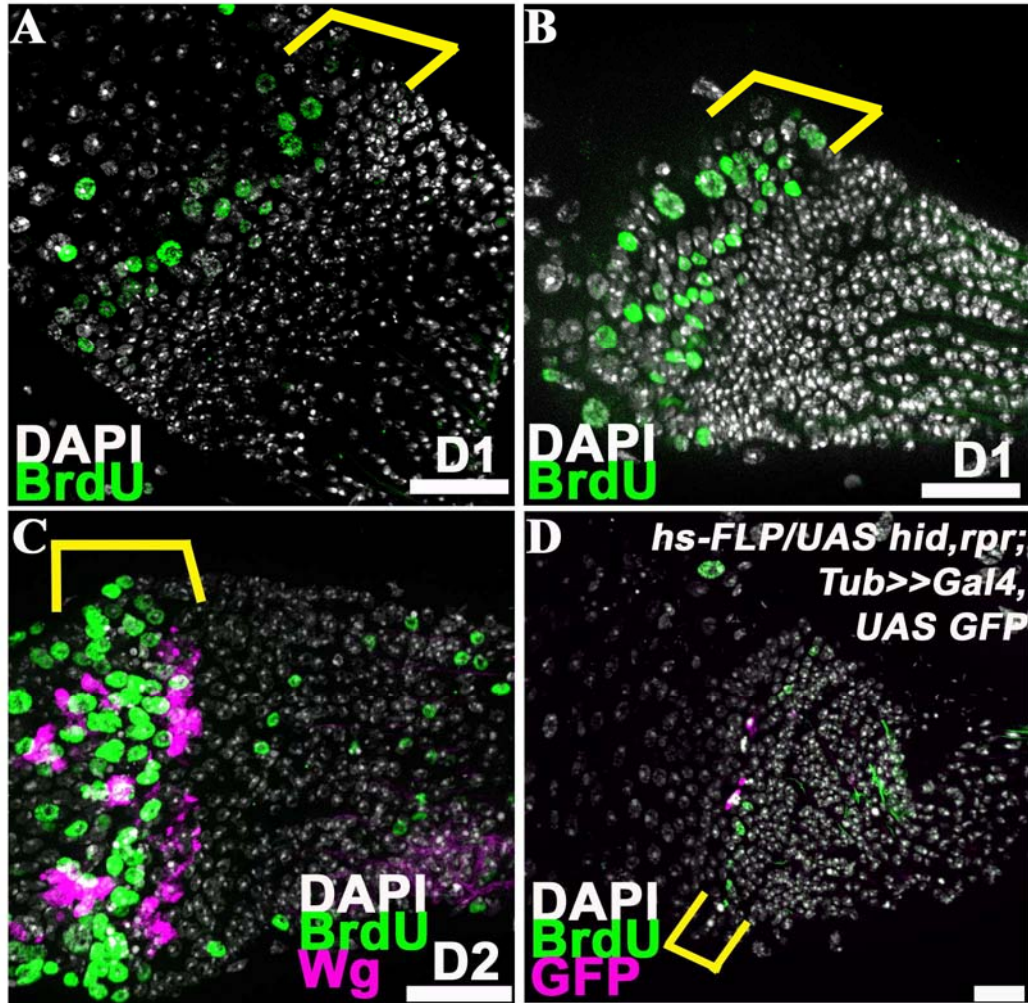
**Figure S1. Cell content and proliferation of the hindgut**

Graph showing the number of cells in the larval ring (blue), the distal pylorus (red) and the ileum (yellow) during the three larval instars. **B)** Graph showing the number of cells in the larval ring (blue), the distal pylorus (red) and the ileum (yellow) before (L3) and after (adult) pupal development. **C)** Graph showing the average size in third instar larvae of cell clones marked at late embryo, 1<sup>st</sup> or 2<sup>nd</sup> instar.



**Figure S2. Clones induced in adult hindguts via the lacZ system do not increase in size**

**A)** Control showing absence of clones in non-heat-shocked animals. **B-D)** Representative images of hindguts at various times following the induction of adult lacZ clones by heat shock. **B)** Day 1 post-heat-shock. **C)** Day 8 post-heat-shock. **D)** Day 15 post-heat-shock. No increase in hindgut clone size is seen, but midgut clones in the same animals have grown to encompass many more cells (inset, arrows). **E)** Representative image of day 8 ileal clones showing no increase in cell number. **F)** Graph showing the average distance of the adult ileal clones from the pylorus at various times after clone induction.



**Figure S3. Adult pyloric cells responds to damage**

**A)** BrdU incorporation (green) is confined to the Wg ring of the adult pylorus during the first 24 hours after initiating UAS-*DTI* expression by shifting the temperature to 30 °C. **B)** BrdU incorporation is confined to the Wg ring of the adult pylorus during the first 24 hours after initiating UAS-*Ricin* expression by shifting the temperature to 30 °C. **C)** BrdU incorporation in the adult pylorus during the second day after temperature shift occurs both in the Wg ring and more posteriorly. **D)** BrdU incorporation during the first 24 hours after inducing FLP-out clones marked with GFP (pink) and expressing *hid* and *rpr*. BrdU incorporation (green) is observed in unrecombined cells adjacent to GFP-expressing cells (see inset **I**). All images- anterior to the left. DAPI (white), BrdU (green). Scale bar=25µm.

**Table S1.** Frequency of pyloric clones

Induction time	Clonal system	Large Clones <sup>1</sup> (>20 cells)	Medium Clones <sup>1</sup> (3-19 cells)	Small Clones <sup>1</sup> (1-2 cells)	Guts analyzed <sup>2</sup>	Clones analyzed <sup>2</sup>
Embryo	lacZ	0.24	0.36	0	25	15
	MARCM	0.97	1.4	0.76	33	107
Pupal Day1	lacZ	0	4.0	.54	38	171
Adult Day 4	lacZ	0	0	40	30	1207
	MARCM	0.014	.20	.15	69	25
Adult control (no $\Delta T$ )	lacZ	0	0	0	52	0
	MARCM	.011	.36	.16	69	37

<sup>1</sup>Clone frequency = number of clones of the indicated size / the number of guts analyzed.

<sup>2</sup>Embryonic, larval and pupal clones were analyzed in 4-7 day-old-adults. Adult clones were analyzed 14-21 days after induction.