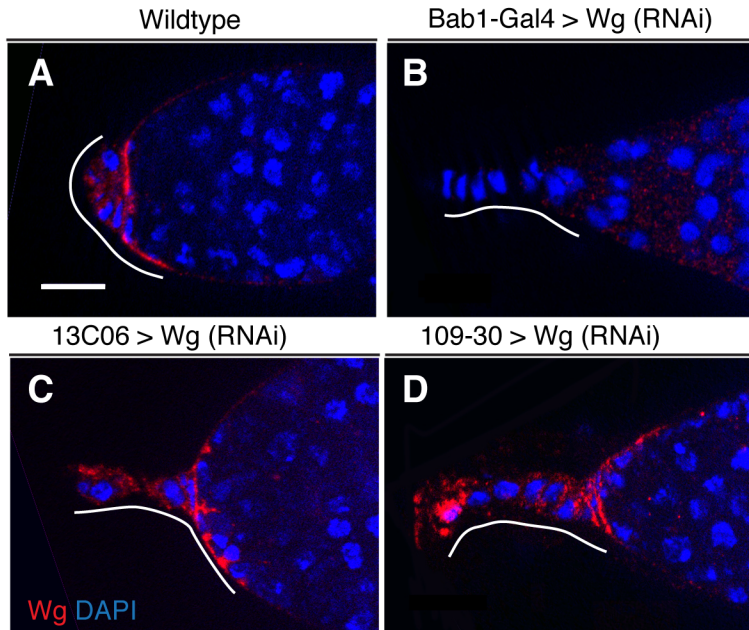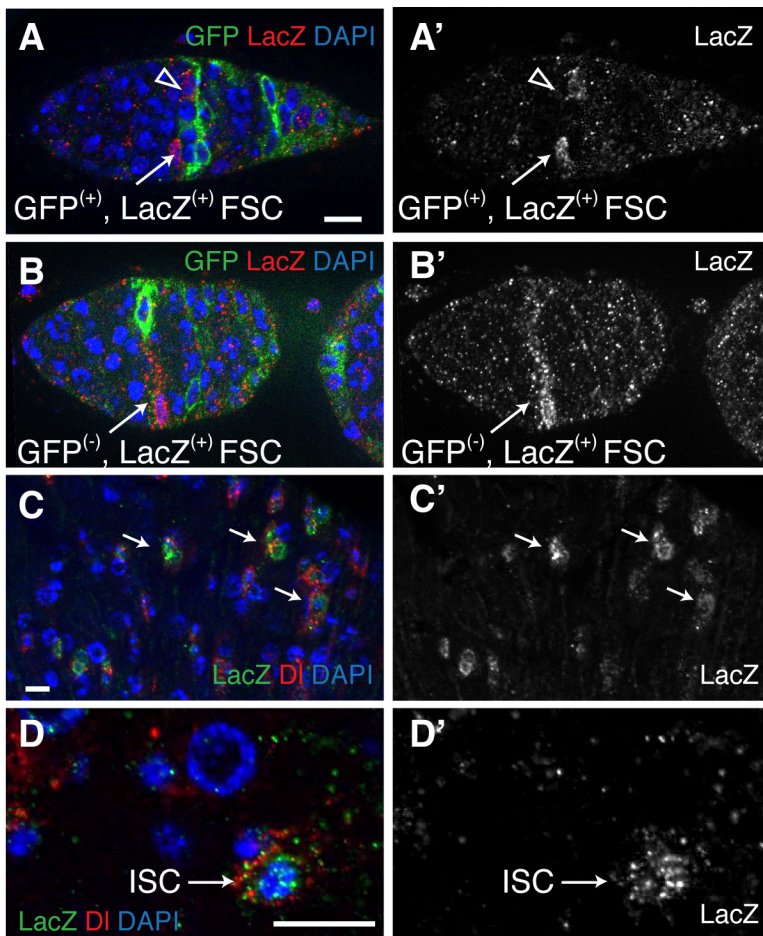# Figure S1

Figure S1. Wg^RNAi eliminates the Wg signal from terminal filament and cap cells

A-D. Germaria from wildtype flies (A) or flies in which Wg^RNAi is driven by Bab1-Gal4 (B), 13C06 (C), or 109-30 (D), at 7 days after flies were shifted to 29°C to repress tub-Gal80^ts and promote Gal4 activity. Tissue is stained for Wg (red) and DAPI (blue). White lines trace the position of the terminal filament cells and cap cells. Note that Wg protein is detectable in the cap and terminal filament cells when Wg^RNAi is driven by 13C06 or 109-30 but not when Wg^RNAi is driven by Bab1-Gal4. Anterior is to the left. Scale bar represents 5 μm.
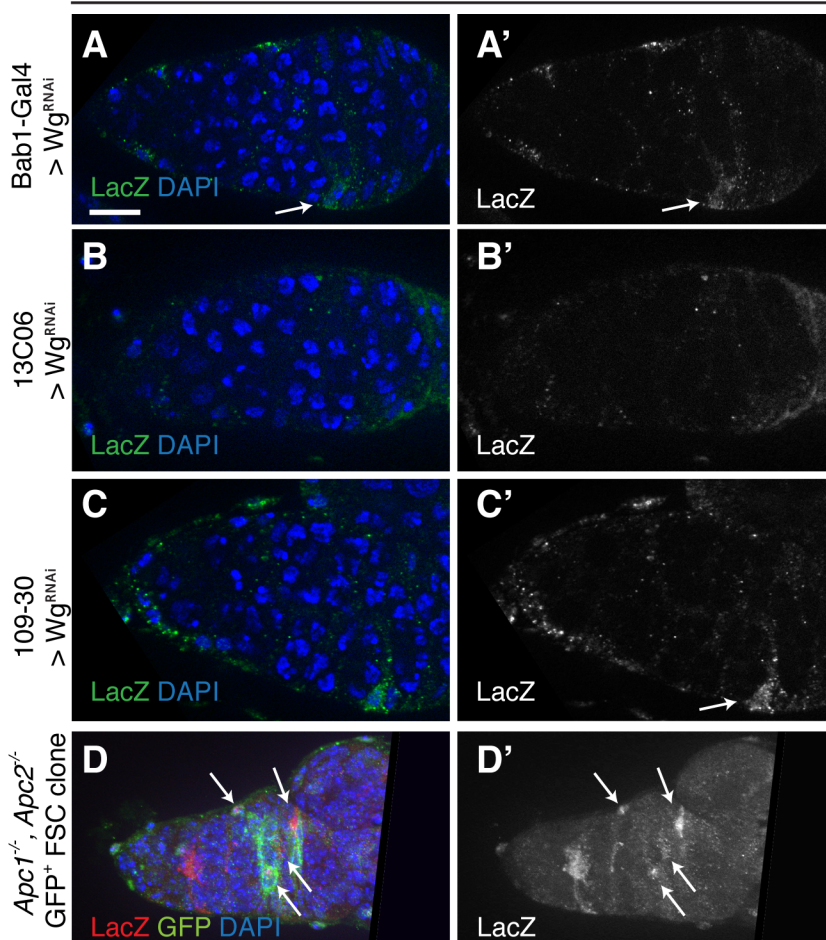
Figure S2

Figure S2. Notum-lacZ labels FSCs and ISCs
A-B. Germaria containing the Wg pathway activity reporter, Notum-lacZ, and a GFP+ FSC clone. In A, both the FSC (labeled) and an early FSC daughter (open triangle) are lacZ+. Because the FSC daughter is GFP+ and positioned near the FSC, we presume this is a recently produced daughter cell that has not yet fully downregulated or turned over the lacZ protein. In B, a lacZ+ FSC is GFP- because it is not part of a GFP+ clone. C-D. Sections of the posterior midgut from flies containing the Notum-lacZ reporter stained with LacZ (green), Dl (red) to mark ISCs, and DAPI (blue). Dl+ cells are also lacZ+, indicating that Notum-lacZ is expressed in ISCs. The lacZ channels in A-D are shown in A'-D'. Anterior is to the left. Scale bar in panel A represents 5 µm and scale bars in B and C represent 10 µm.
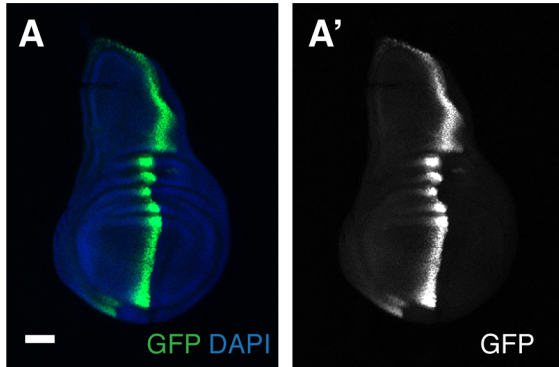
Figure S3

Figure S3. Wg produced by escort cells activates Notum-lacZ in FSCs
A-C. Ovarioles from flies 7 dayes after heatshock that contain the Notum-lacZ reporter, UAS-Wg^{RNAi}, tub-Gal80^{ts}, and either Bab1-Gal4 (A), 13C06 (B), or 109-30 (C). Tissue is stained for lacZ (green) and DAPI (blue). lacZ$^+$ FSCs were present when Wg^{RNAi} is driven by Bab1-Gal4 or 109-30 (white arrows), but not when Wg^{RNA} is driven by 13C06. Anterior is to the left. Scale bar represents 5 μm. D. A maximum image projection of a germarium with a mature GFP$^+$ Apc1$^{-/-}$, Apc2$^{-/-}$ FSC clone. Many mutant FSC daughter cells (white arrows) express Notum-lacZ. The tissue is stained for lacZ (red), GFP (green), and DAPI (blue).

Figure S4

Figure S4. Ptc-pelican-GFP(nls) is a reporter for Hh activity in the wing disc
A. A wing disc that contains Ptc-pelican-GFP(nls). As expected for a reporter of Hh signaling activity, GFP expression is strong along the A/P boundary and tapers off toward the anterior. Anterior is to the left. Scale bar represents 50 μm.

Table S1. The frequency of $lacZ^+$ FSCs in ovarioles with or without a follicle cell defect						
	1 or 2 $lacZ^+$ FSCs		0 $lacZ^+$ FSCs			P-value (Fisher's exact test)
Genotype	Normal (%)	Follicle defect (%)	Normal (%)	Follicle defect (%)	n	
Wild type	71.9	3.1	25.0	0.0	32	1
Bab1-Gal4>Wg^{RNAi}	63.4	7.3	29.3	0.0	41	0.543
13C06>Wg^{RNAi}	30.2	4.7	23.3	41.9	43	0.002
109-30>Wg^{RNAi}	79.5	0.0	17.9	2.6	39	0.205

Table S2

FISH Probe Sequences:

Wingless:
gaagatatagctgatatcca; acagggccatcaggcagatg; gatttctgtttgccctcgac; gtaatgttgttgggttcgcc; tgggtccatgta-
catgatgg; ttctcaacgtagagtggatc; ctgaccaggcgtcgctgttt; ctcccagtacaccgggattg; aagttggcgcccttgaccag;
gtgttggcactcgctaatgg; tccagcggcgatttctgaac; gagaagtttctcgtcgagca; gccgaatagattttttgcccc; ggcagcctc-
gatcaacgatt; attgcgtaaatgaagctcgt; caggccctggcaatcgagtg; aggactctatcgttccttca; gatctcgactggtggctgta;
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gcttgcgtcc; ctccagatagacaaggtcct; ggttcttctcgcagaagctc; catgggttcccaggatgccc; agcgaggtctcattgcactg;
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Hedgehog:
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gaaaaggatgtccctgttgt; cgtttagcttctccttgcag; gatggtagtcctcgtcccag; taggagacccaatcgaatcc; gggaactgatc-
gacgaatct; catgaagaggatcacttcgc; ccaaacgctaaccaggtgag; ttcttctcctcgatgcgatc; gttgatcaccgcatagcaac;
taccaatggatgccattctg; ggattccatctcaatcgtgg; tgctctttgctttatcgctt; tcaaggacatttaacagtgt; ggatgatttag-
gatctgcgc; cagtaacagtcgtctgtgtt