

Fig. S1. *C. elegans* Wnt signaling pathways. (A) Model for the function of the Wnt/ β -catenin canonical pathway (WBC). In the absence of ligand (left), β -catenin BAR-1 is phosphorylated by the destruction complex and targeted for degradation by the proteasome, consequently target genes are not expressed. In the presence of Wnt ligand (right), BAR-1 is not degraded and translocates into the nucleus where it interacts with the TCF homolog POP-1 and together they activate target gene expression. (B) Model for the function of the Wnt/ β -catenin asymmetry pathway (WBA). Shown are two daughters of a Wnt-regulated asymmetric cell division along the anterior-posterior axis. In the cell that does not receive the Wnt signal (anterior cell, left), there are low levels of the β -catenin SYS-1 in the nucleus, and target genes are repressed by high levels of nuclear POP-1 interacting with transcriptional repressor proteins. In the cell that receives the Wnt signal (posterior, right), a series of events involving MAP kinases and β -catenin WRM-1 cause export of POP-1, decreasing its levels in the nucleus. The level of β -catenin SYS-1 also increases in this cell; SYS-1 binds to POP-1 and together they activate target gene expression. Certain Wnt pathway components such as APR-1, WRM-1, the receptor and POP-1 show differences in localization (cortical versus nuclear) and amount between the daughters of the asymmetric division.

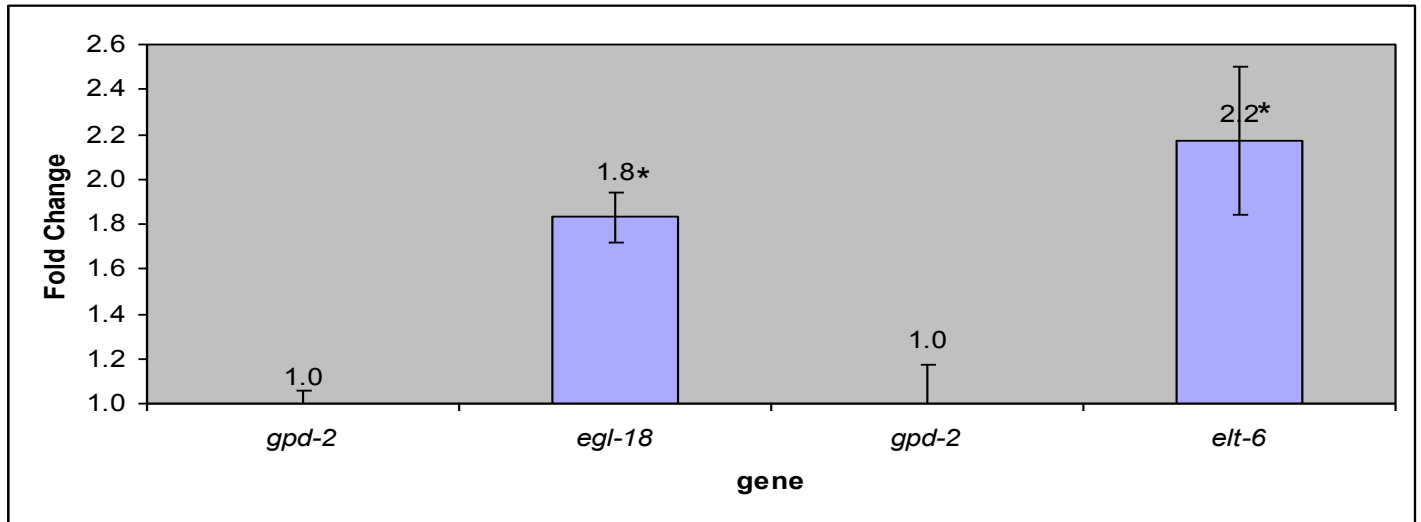


Fig. S2. qRT-PCR analysis shows endogenous *egl-18* and *elt-6* levels increase in response to Wnt pathway activation. Graph shows average fold change of *gpd-2*, *egl-18* and *elt-6* in Wnt pathway activated animals (*hulIs7; egl-18::mCherry*) relative to heat-shocked control animals (*egl-18::mCherry*) 1 hour after heat shock. * $P < 0.05$ average fold change of *gpd-2* compared with *egl-18* and *elt-6* (unpaired t-test).

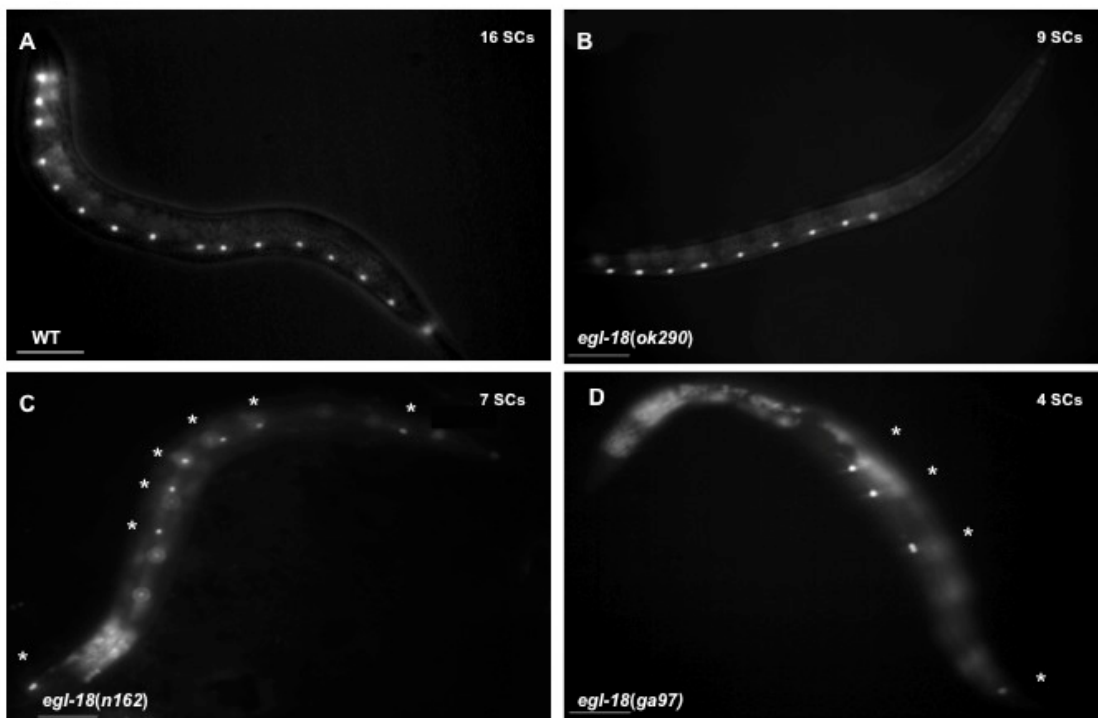


Fig. S3. *egl-18* mutants have fewer seam cells as adults. (A-D) Fluorescent images of young adult worms expressing *scm::GFP*. (A) Wild type, (B) *egl-18(ok290)*, (C) *egl-18(n162)* and (D) *egl-18(ga97)*. In panels C and D, the seam cells in focus on one side are indicated by asterisks and the seam cells on the other side of the worm appear as fainter blurred spots. Anterior is towards the left and ventral is downwards in all panels. Scale bar: 50 μ m.

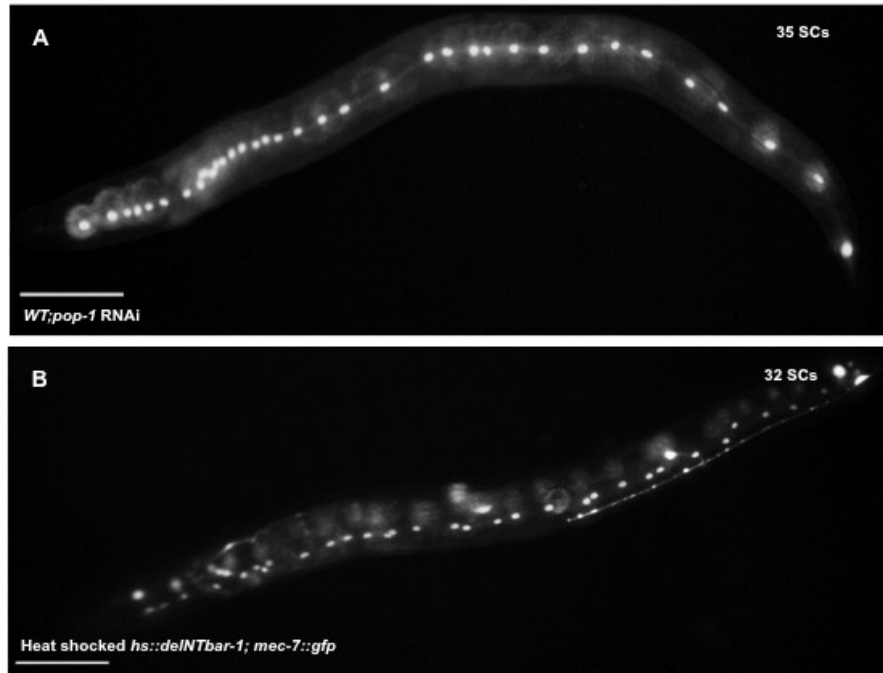


Fig. S4. Overactivation of Wnt pathway results in increased terminal seam cell number. (A,B) Fluorescent images of young adult worms expressing *scm::GFP*. (A) WT; *pop-1*(RNAi). (B) *hs::delINTbar-1*; *mec-7::gfp* heat shocked at L2/L3 molt. The animal in B also contains *mec-7::gfp*. The seam cells on the other side of the animal in B are out of focus and appear as blurred spots. Anterior is towards the left and ventral is downwards in both panels. Scale bar: 50 μ m.

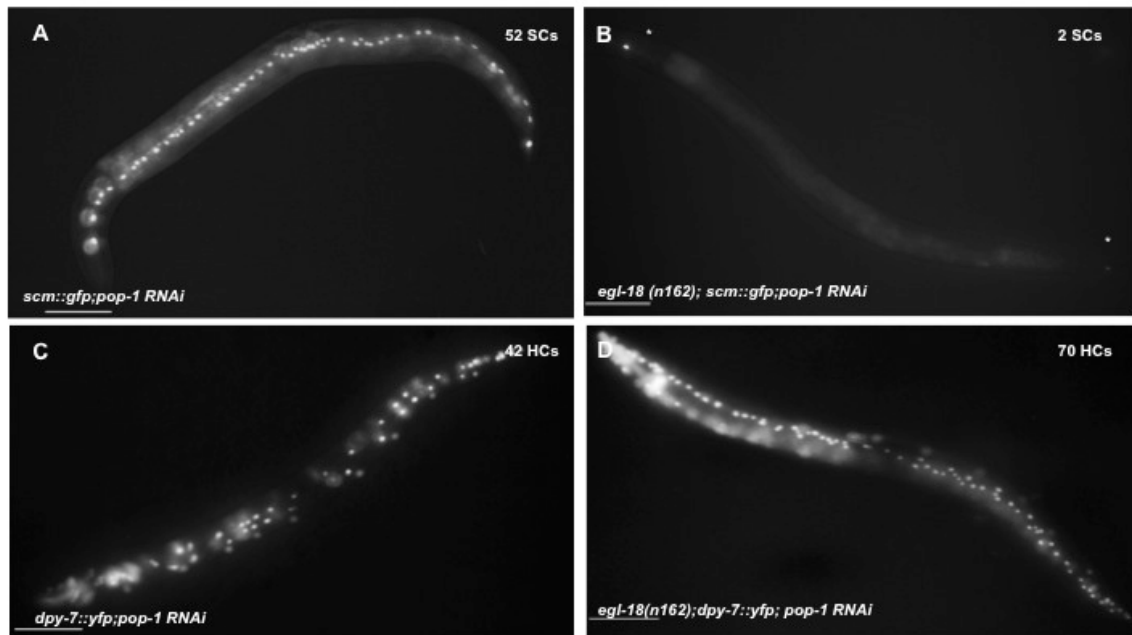


Fig. S5. Increase in seam cell number induced by Wnt pathway overactivation is dependent on *egl-18*. (A-D) Fluorescent images of young adult worms expressing *scm::GFP* (A,B) and *dpy-7::YFP* (C,D). (A) *scm::gfp*; *pop-1*(RNAi), (B) *egl-18*(*n162*); *scm::gfp*; *pop-1*(RNAi), (C) *dpy-7::yfp*; *pop-1*(RNAi), (D) *egl-18*(*n162*); *dpy-7::yfp*; *pop-1*(RNAi). Anterior is towards the left in all panels. Scale bar: 50 μ m.

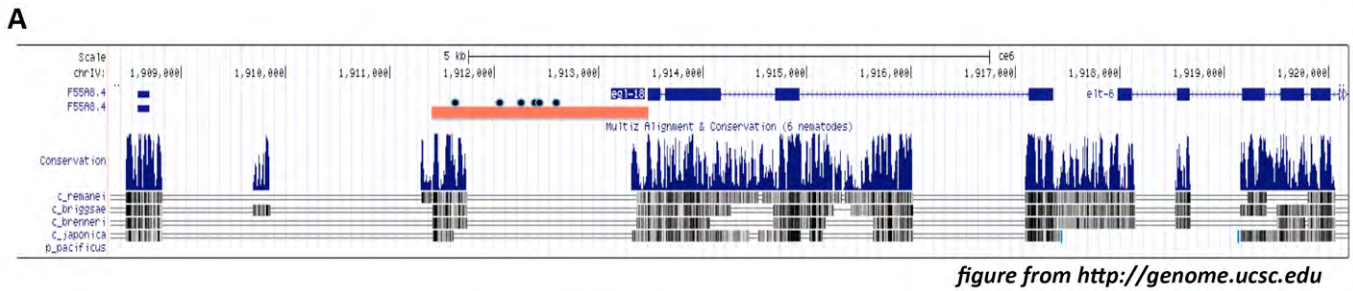


figure from <http://genome.ucsc.edu>

B

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-1038	TTTTGTAA
-1141	TTTCAAAA
-1333	TTTCAAAA
-1806	CTTTGTTC

C

Alignment block 1911416 - 1911735

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C. briggsae tcttttggcgatcgtgcgcaactgattcgggtgtaaacacgcggtgta-----gaggcgaagagg
C. brenneri  tcttctaggcgatcgtgcgcaactgattcgggtgtaaacacgcggtgta-----taatgaatcacg
C. japonica  -----cgatcgcgcaactgattcgggtgtaaacacgcggtgta-----gaaggcgttgag-

C. elegans  gatgttgaggaagaa-----gttgagcgaagagagagacttcg-ttcgaag
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C. japonica  -cgggtgggaaagac-----ggtt-gtcgcca--acggcgagggcgatcgtc-

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C. briggsae gtttt-----tctcccaaacatcttttacatctttgttc
C. brenneri  gtttt-----tcaatctttt-----cttttacatctttgttc
C. japonica  =====

C. elegans  ggactca-----tcacggactcatcaactcattcgtttctgtgttaggacaccactgtttaacggggtc
C. remanei  acagtca-----tca--gcctcatcaact
C. briggsae acagtca-----tca--caatctttcacaactctgatgttaga-----
C. brenneri  atagcaatcccatca-----tcaatcaact-ctcatttctcactatag-----tgtcaaggggac
C. japonica  =====

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D

SC expression	
%	SCs/side
93	14.5
0	0
51	6.1

Fig. S6. An evolutionarily-conserved POP-1 binding site upstream of *egl-18*. (A) Screen shot from the UCSC Genome Browser (<http://genome.ucsc.edu>) (Kent et al., 2002). Coordinates on *C. elegans* chromosome IV are shown at the top, with exon/intron structure of *egl-18* and *elt-6* below. The red bar indicates the 2.0 kb promoter fragment used in YFP reporter constructs. Dots above the red bar indicate the position of sequences matching the POP-1 consensus binding site (YTTTGWWW). The bottom shows an alignment of the *C. elegans egl-18/elt-6* genomic region with regions from *C. brenneri*, *C. briggsae*, *C. remanei* and *C. japonica*. The 2.0 kb fragment contains only one strongly conserved region, at the extreme 5' end. (B) Position and sequence of six sites matching the POP-1 consensus binding site found within 2.0 kb upstream of the *egl-18* start codon. Sites in italics are in reverse orientation from the consensus site. (C) Alignment from five species of the conserved region at 2.08-1.73 kb upstream from the *C. elegans egl-18* start codon [alignment from the UCSC Genome Browser (<http://genome.ucsc.edu>) (Kent et al., 2002), using default parameters]. Single and double lines indicate gaps in the aligned sequences. The red arrow indicates the beginning of the 2.00 kb element used in the YFP reporters. The POP-1 binding site examined for *in vitro* binding and *in vivo* importance is shown in red. Genome assemblies used: *C. elegans* WS190/ce6; *C. brenneri* WUGSC 6.0.1/caePb2; *C. briggsae* WUGSC 1.0/cb3; *C. remanei* WUGSC 15.0.1/caeRem3; *C. japonica* WUGSC 3.0.2/caeJap1. (D) The left shows *egl-18::YFP* reporter constructs created; red box indicates the evolutionarily conserved region (in C), dots indicate sites matching the POP-1 binding site consensus (in B), 'X' indicates mutation of the first POP-1 binding site. Results for expression in seam cells of young adult transgenic animals containing the indicated construct as an extrachromosomal array are shown on the right. Column 1 shows the average percent of animals that showed any expression, and column 2 shows the average number of SCs/side expressing YFP from animals with expression (two to four lines examined for each construct). In addition, expression from the reporter with the mutated POP-1 site was less intense overall than expression from the wild-type construct (Fig. 3 and data not shown).

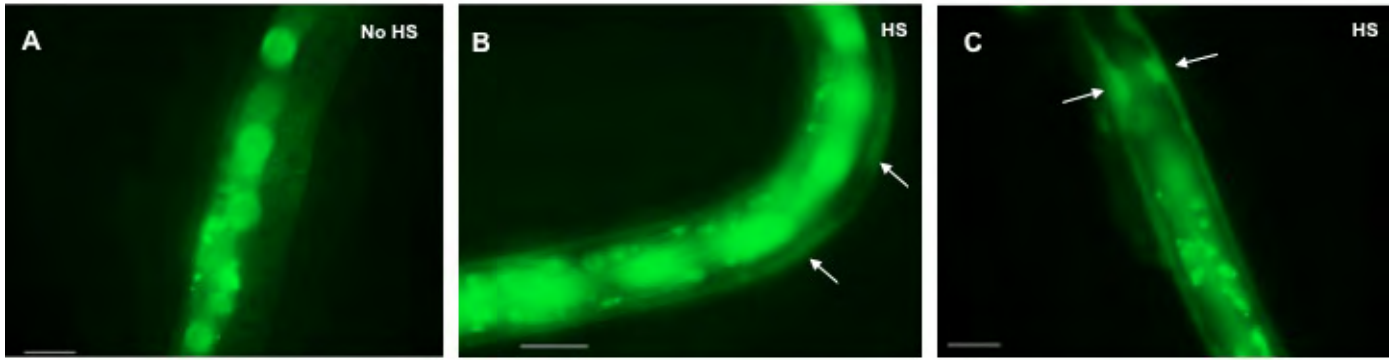


Fig. S7. Ectopic expression of EGL-18 drives expression of seam cell reporter *grd-10::gfp* in head and ventral hypodermal cells. Synchronized, newly hatched *grd-10::gfp; hs::egl-18* L1 larvae were given a single heat shock at the L2/L3 molt and *grd-10::gfp* expression was scored in the same animals as young adults. (A) Non-heat shocked adult showing no ectopic expression in the mid-body. (B) Heat shocked animal showing ectopic seam cell marker expression in the ventral hypodermal cells in the position of non-vulval Pn.p cells (arrows). (C) Heat shocked animal showing ectopic expression in head hypodermal cells in the position of hyp6 cells (arrows). (A,B) Bright expression in the normal seam cells is seen out of focus. (C) GFP intestinal autofluorescence is also seen. HS, heat shock. Scale bar: 50 μ m.