

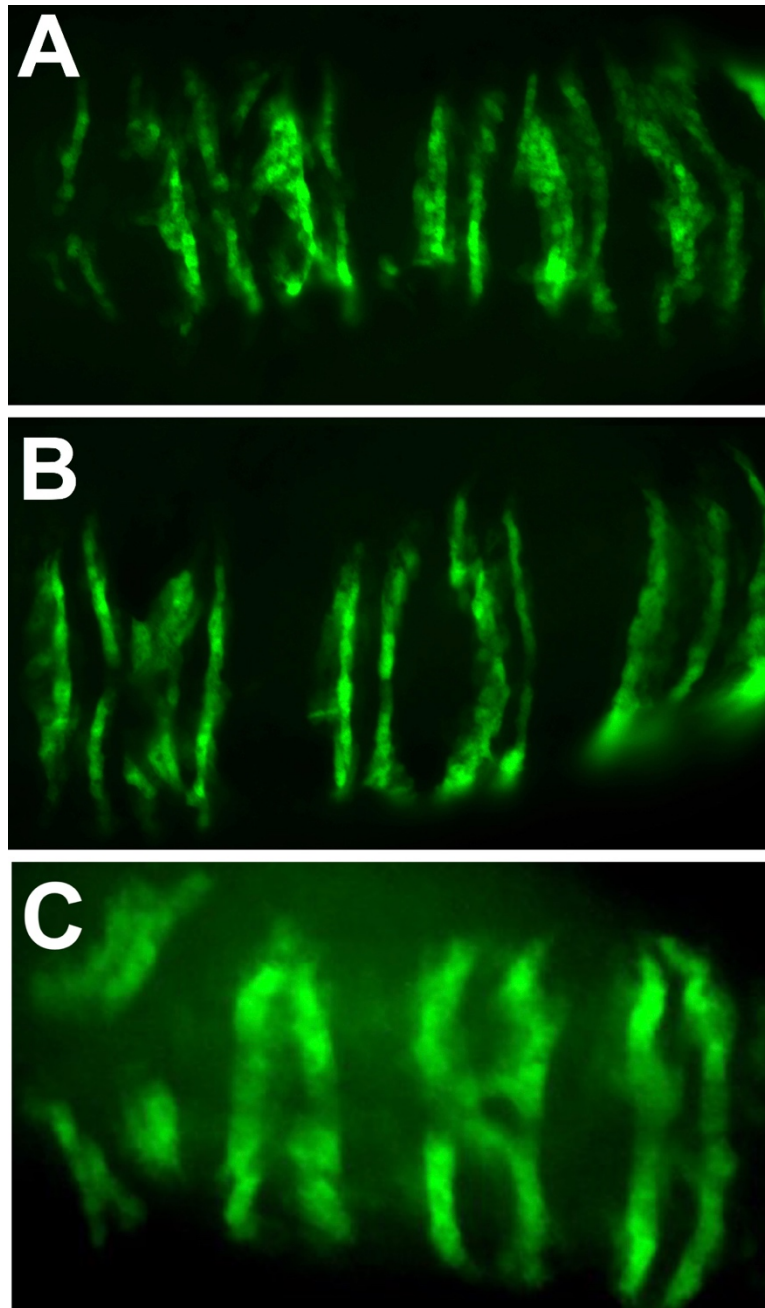
## Supplementary Material

**Table S1. Predicted SoxN binding sites**

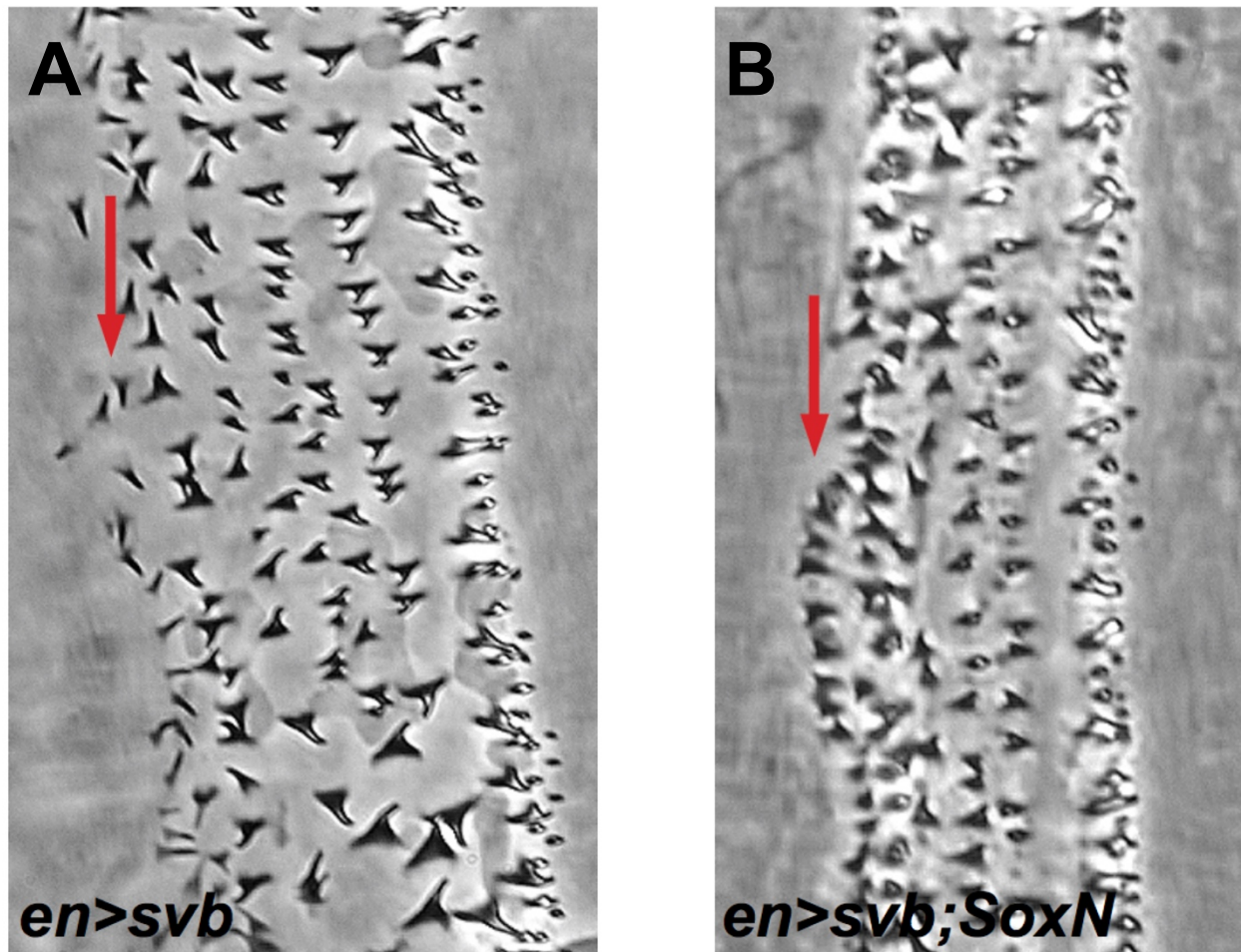
Gene	Binding Site Coordinates	p-value	Coordinate Sequence	Conserved in <i>Drosophila</i> species
CG16885	2L:13932099..13932109	5.13E-05	GCGAACAATTA	
	2L:13932090..13932100	2.70E-04	GCAAACAATGT	
	2L:13934471..13934481	2.95E-04	TGGAACAATGT	<i>sechellia, simulans</i>
	2L:13933769..13933779	2.95E-04	ACGCACATCGC	<i>sechellia, simulans, yakuba, erecta</i>
	2L:13935173..13935183	3.82E-04	CTGTACAATTA	
	2L:13934919..13934929	5.74E-04	ACGGACAGTGT	<i>sechellia, simulans, yakuba</i>
	2L:13933923..13933933	8.40E-04	CCCAACATTGC	<i>sechellia, simulans</i>
	2L:13934972..13934982	8.40E-04	GTGAACACTCG	<i>sechellia, simulans</i>
	2L:13932682..13932692	8.91E-04	CTGTACACTCG	<i>sechellia, simulans, yakuba, erecta</i>
	2L:13935488..13935498	9.06E-04	ACTAACACTTC	
CG30101	2R:17410142..17410152	3.52E-04	GGGAACAATTA	<i>sechellia, simulans, yakuba, erecta, ananassae</i>
	2R:17409197..17409207	4.01E-04	CCGTACACCGT	<i>sechellia, simulans</i>
	2R:17410109..17410119	4.72E-04	CTCAACAATGC	<i>sechellia, simulans, yakuba, erecta, ananassae, pseudoobscura, persimilis, willistoni, mojavenensis, virilis, grimshawi</i>
	2R:17410435..17410445	7.25E-04	GCGTACATTCA	
	2R:17409565..17409575	8.18E-04	CTCAACAATGT	
	2R:17409948..17409958	9.32E-04	CGCCACAATGG	
<i>dusky-like</i>	3L: 4308522..4308532	4.07E-05	ATGAACAATGG	
	3L: 4311650..4311660	9.43E-05	ACGCACAGTTC	<i>simulans</i>
<i>neo</i>	3R:29825746..29825756	5.60E-05	CCGTACAATTA	<i>sechellia, simulans</i>
<i>nyobe</i>	3R:31547625..31547635	5.72E-05	CGGCACAATGG	<i>simulans, yakuba</i>
	3R:31547719..31547729	5.99E-05	GCGAACAATAA	<i>simulans</i>
	3R:31549064..31549074	8.78E-05	ACAAACAATGA	<i>simulans, yakuba, erecta</i>
	3R:31559928..31559938	9.05E-05	ACGCACAGCGC	<i>sechellia, simulans</i>
	3R:31564141..31564151	9.67E-05	GCGAACAACCA	<i>sechellia, simulans</i>

**Table S2. Primers used to generate *in situ* hybridization probes**

Gene	Forward Primer	Reverse Primer	Product Size
CG16885	tgcttagaattcGGTTCCGATCCTTCCAGAAT	taagcaaagcttGACCGTCACCTATCCCGTTA	725bp
CG30101	tgcttagaattcGAAATCGTAGGGTCTAACACGA	taagcaaagcttGGAGAAACCCTACCCAGTCC	725bp
<i>dyl</i>	tgcttagaattcCGCGTTCCTCTTGTAGATCC	taagcaaagcttGTTTCGGGCATTGTCAAGAT	890bp
<i>neo</i>	tgcttagaattcGCACCTCCAAGCTCTTCAAC	taagcaaagcttGGCCATAGGACAACAAGGAC	914bp
<i>nyo</i>	tgcttagaattcCCTATGAACTGTCAGCCTGCTA	taagcaaagcttCGTTACCAAGGCACGAAACT	731bp

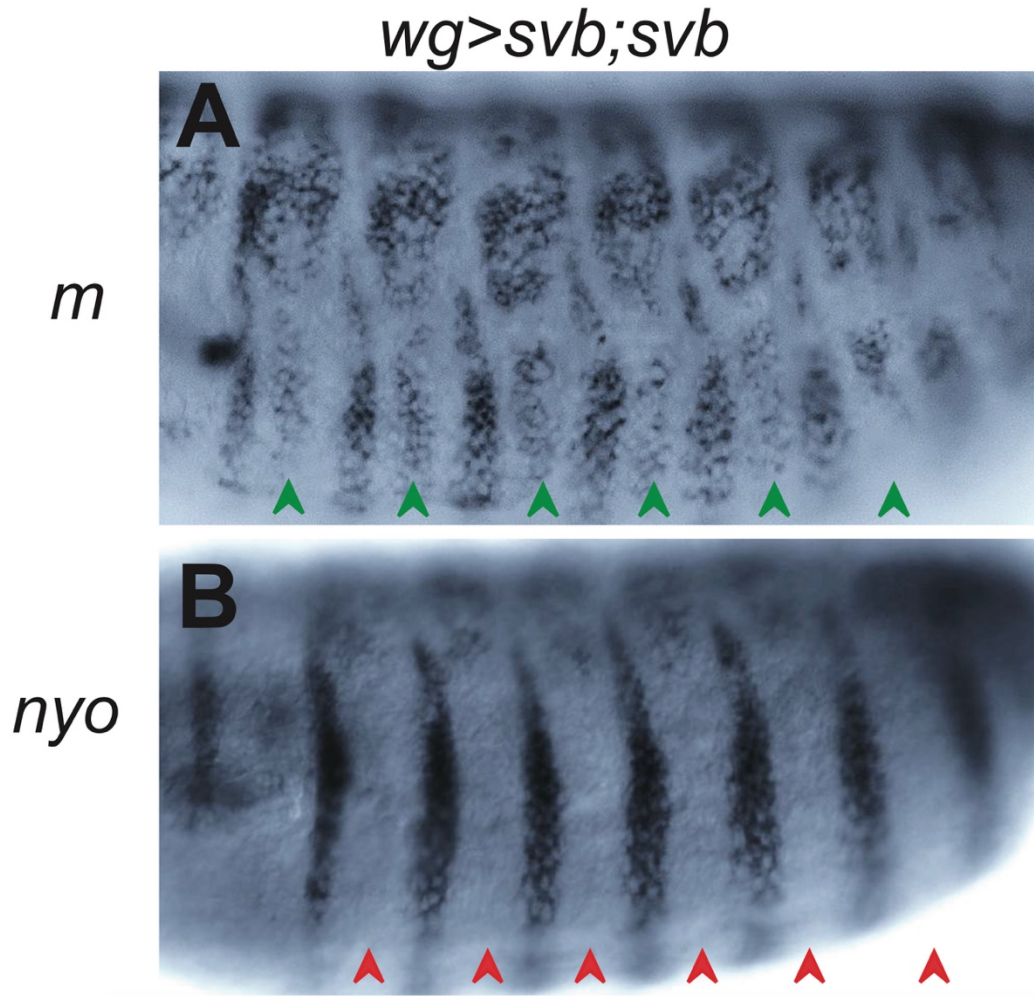


**Fig. S1. *E3-lacZ* promoter element in *wg* mutant embryos.** (A) and (B) Two different *wg<sup>ts</sup>* mutant embryos that were cultured at permissive temperature until 5 hours after egg-laying, to allow generation of denticle diversity, and then shifted to restrictive temperature to remove later Wg function. Both show expansion of the anterior stripe of *E3* expression similar to that observed in the partial-function *wg<sup>PE2</sup>* mutant embryos. Thus the anterior stripe correlates with regions of the denticle belt where diverse denticle types are being specified. (C) *E3* expression in the RNA null mutant, *wg<sup>CX4</sup>*, does not show expansion of the anterior stripe, but instead shows expansion of the posterior stripe so that it matches the width of the anterior stripe. This mutant phenotype is indistinguishable from the *wg<sup>ts</sup>* mutant at restrictive temperature.



**Fig. S2. Ectopic *svb* and *SoxN* coexpression in the *engrailed* expression domain produces denticles of correct morphology.** The *en-Gal4* driver expresses in two rows of cells, one that makes naked cuticle and the other that produces the first row of denticles in each segment. (A) Ectopic expression of *svb* produces thin denticles in the naked cuticle row of cells, similar to those produced by *wg>svb* (red arrow). (B) Ectopic co-expression of *svb* and *SoxN* produces denticles of a more normal morphology (red arrow), similar to those observed in *wg>svb; SoxN* embryos.





**Fig. S3. Doubling the *svb* transgene dose does not increase target gene expression.** (A) *miniature* RNA in situ hybridization in embryos with *wg-Gal4* driving two *UAS-svb* transgenes show same level of ectopic *m* induced (green arrows) as observed with single transgene (Fig. 7A). (B) *nyobe* RNA in situ hybridization in embryos with *wg-Gal4* driving two *UAS-svb* transgenes shows no ectopic expression induced (red arrows), similar to what is observed with single transgene (Fig. 7D).