wingless refines its own expression domain on the *Drosophila* wing margin

Eric J. Rulifson, Craig A. Micchelli, Jeffrey D. Axelrod, Norbert Perrimon & Seth S. Blair

Nature **384,** 72–74 (1996)

BECAUSE of an error in the production process all the figures in this Letter were unclear and of poor quality. They are reproduced again below. □

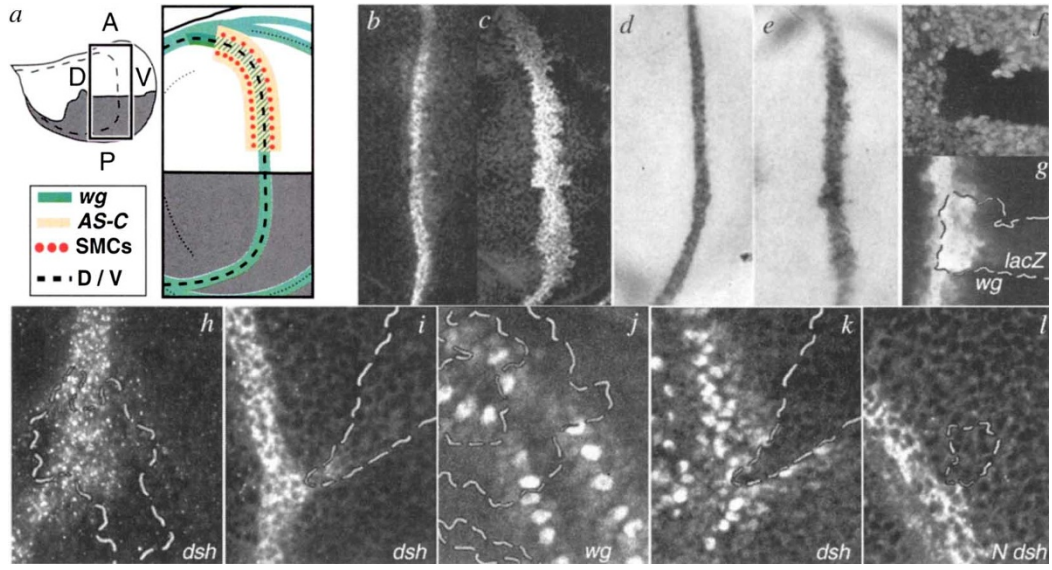

FIG. 1 *a–i*, Margin *wg* expression after loss of *wg* or *dsh* functions. *a*, Diagram of late third instar wing disc. Box outlines margin region shown in the remaining figures, with axes and patterns of gene expression as marked (see text for details). *AS-C*, *acheate–scute complex* expression; D/V, dorsoventral. *b–e*, wg^{ts} (wg^{ts}/wg^{cx4}) margins, stained for Wg protein (*b*, *c*) or messenger RNA (*d*, *e*) expression. *b*, *d*, At permissive temperature, protein and mRNA expression was normal. *c*, *e*, After a 12 h shift to restrictive temperature, protein expression expanded to a region twice the normal width (protein: 55/57 discs; mRNA: 17/19 discs). *f*, *g*, Hypomorphic wg^{LacZ} clone on the margin, shown by the absence of anti-Myc staining (*f*; in *g* and subsequent panels clones are marked by a dotted outline and *). Clones caused expansion of *wg-LacZ* expression (anti-β-gal; 72/97, none showed loss). *h*, *i*, *dsh* clones that intersected (*h*) or sat immediately adjacent to (*i*) the *wg* stripe elevated anti-Wg-staining cell-autonomously (dsh^{75}: 15/17; dsh^{v26}: 12/12). Similar effects were observed in anterior, posterior, dorsal and ventral clones. In some cases, it seemed that the normal *wg* stripe expanded or distorted to meet the ectopic anti-Wg staining within clones away from the margin (see *i*; 7 clones). It is unclear whether ectopic *wg* expression is being induced in wild-type cells between the clone and the margin, or if cells at the margin are distorting or rearranging near the clone. *j*, *k*, Anti-Scute or Achaete staining in wg^- and dsh^- clones. *j*, wg^{cx4} clones that intersected the *wg* stripe showed non-autonomous loss of Sc (18/22 large clones), as did 5/7 wg^{LacZ} clones (not shown). *Sc* was expressed at wild-type levels in *wg* mutant cells that were 1-2 cell diameters from the clone boundary, presumably in response to Wg secreted by wild-type cells. Most small clones, where all cells were close to the boundary, showed normal Sc levels. *k*, dsh^- clones within the margin proneural region showed cell-autonomous loss of sc expression (dsh^{75}: 9/9; dsh^{v26}: 15/15). *dsh* clones that intersected the *wg* stripe, and produced ectopic *wg*, also generated ectopic sc expression outside the normal proneural region in *dsh*/+ cells near the clone boundary (arrow; note *wg* expression in same clone in *i*). *l*, Margin *wg* expression after loss of *Notch* (*N*) and *dsh*. $Notch^-$ dsh^- clones that lay adjacent to (*l*) or intersected the *wg* stripe (not shown) showed cell-autonomous loss of anti-Wg staining without expansion of *wg* expression (29/29 clones). $Notch^-$ dsh^- clones in the anterior also lose anti-Sc staining (E.J.R., C.A.M., M. Halevy and S.S.B., manuscript in preparation).

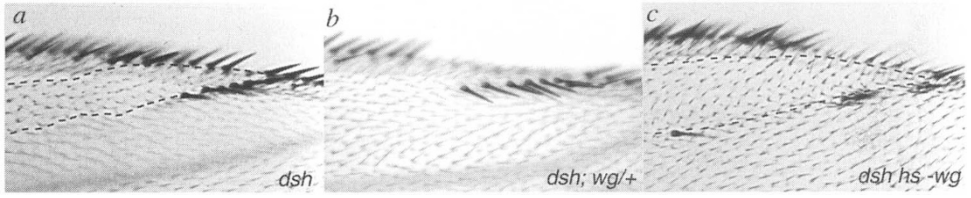

FIG. 2 dsh^- clones induced ectopic bristles off the wing margin in a *wg*-dosage-dependent fashion. *a*, dsh^- clone in adult wing, marked with *y* and f^{36a} (dotted outline), extending from the margin into the interior of the wing blade. The average distance from farthest bristle to margin was 4.5 cells (±1.0, range 3–7, $n = 23$). *b*, *dsh* clone induced in a wg^{IG22} heterozygote. The average distance for all clones observed was 2.71 (±0.73, range 2–4, $n = 14$). *c*, *dsh* mutant clone induced in the presence of heat-shock promoter-*wg*. The average distance for all clones observed was 10 cells (1.6, range 8–12, $n = 5$). *hs-wg* produces insufficient activity to generate ectopic bristles in a wild-type background (not shown). The clones in *b* and *c* are not marked with f^{36a}, but are identified by the cell-autonomous tissue polarity defect caused by dsh^{14} (results not shown). Unmarked dsh^- and $Notch^-$ dsh^- clones were identified in adult wings by the absence of normal margin bristles[6,20]. 22/30 *dsh* clones were accompanied by ectopic bristles distant from the margin, as compared with 5/40 *Notch dsh* clones. The bristles near *Notch* dsh clones were only rarely as contiguously arrayed or as distant from the margin as those near *dsh* clones. Regulation within the margin proneural regions may account for these occasional bristles (data not shown).

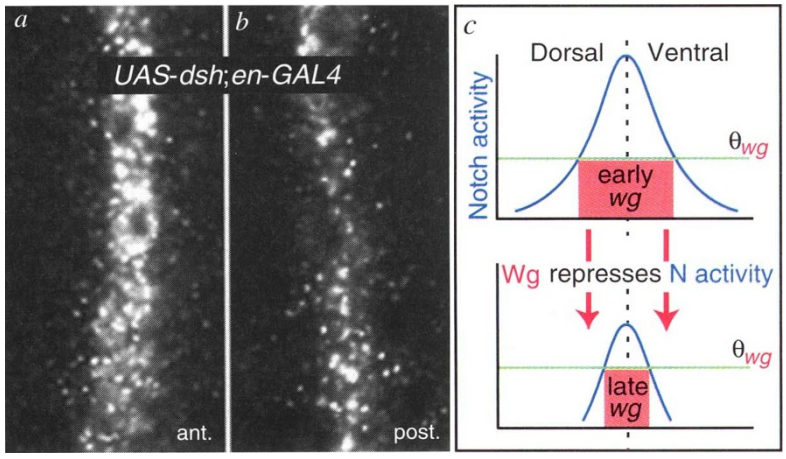

FIG. 3 Interactions between *Notch* and *dsh*. *a*, *b*, *dsh* overexpression in the posterior compartment, using *UAS-dsh* and *en-GAL4*, caused narrowing and occasional loss of margin anti-Wg staining (*b*) when compared with anterior (*a*). Dsh levels, as identified using anti-Dsh, were visibly higher in the posterior compartment (not shown). Both panels are from the same imaginal disc. *c*, Model of Wg self-refinement. Notch signalling activity is highest at the dorsoventral boundary, and levels above the threshold (Θ_{wg}) initially trigger broad *wg* expression (above). Wg represses Notch (N) activity; *wg* expression is maintained only in cells nearest the dorsoventral boundary (below).