

# Regeneration can take place across *Drosophila* compartments or segments with different Hox gene expression

RAFAEL ALEJANDRO JUÁREZ-URIBE<sup>1</sup>, PALOMA MARTÍN, LAURA UTIEL, BLANCA L. ARRABAL<sup>2</sup>,  
MARINA BLANCO, ROBERTO YAGÜE-SERRANO<sup>3</sup>, EDUARDO CAZALLA<sup>4,5,6,7</sup>,  
ERNESTO SÁNCHEZ-HERRERO

Centro de Biología Molecular Severo Ochoa (CBM), CSIC-UAM,  
Nicolás Cabrera 1, Universidad Autónoma de Madrid, Cantoblanco, 28049 Madrid, Spain

<sup>1</sup>Present address: Department of Pathology and Laboratory Medicine, University of Rochester Medical Center, Rochester, New York, U.S.A.

<sup>2</sup>Present address: Centro de Neurociencias Cajal (CNC), CSIC, Av. León 1, Alcalá de Henares, 28805, Madrid, Spain

<sup>3</sup>Present address: Health Research Institute La Fe (IIS La Fe), Avenida Fernando Abril Martorell 106, 46026, Valencia, Spain

<sup>4</sup>Present address: -Department of Biochemistry, School of Medicine, Universidad Autónoma de Madrid (UAM), 28049 Madrid, Spain

<sup>5</sup>Present address Instituto de Investigaciones Biomédicas "Sols-Morreale" (CSIC-UAM), 28029 Madrid, Spain

<sup>6</sup>Present address Instituto de Investigación Sanitaria La Paz (IdiPaz), 28046 Madrid, Spain

<sup>7</sup>Present address Centro de Investigación Biomédica en Red de Enfermedades Neurodegenerativas (CIBERNED), 28046 Madrid, Spain

## Supplementary Material

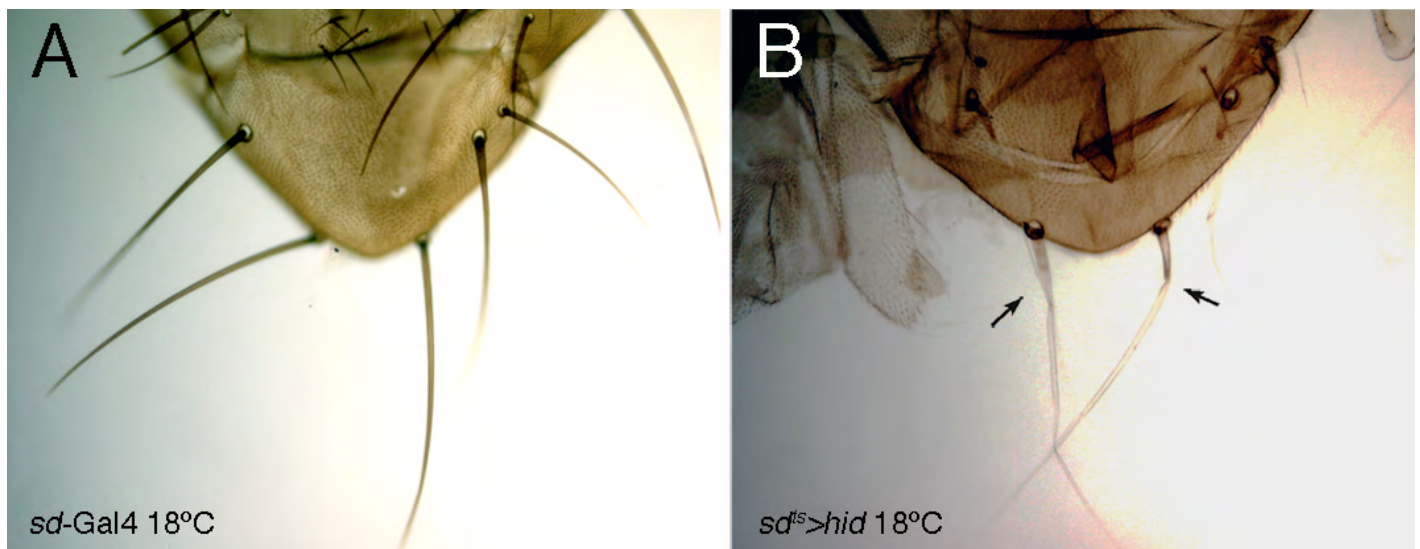


**Fig. S1. Effect of *hid* expression with *tub-Gal80*<sup>ts</sup> on bristle development at 18°C.** (A) Scutellum of a wildtype fly. (B) Scutellum of a *scalloped-Gal4*; *UAS-hid tub-Gal80*<sup>ts</sup>/+ fly grown at 18°C, showing thin and near transparent bristles (*scalloped* is expressed in bristles, Garg et al., 2007).

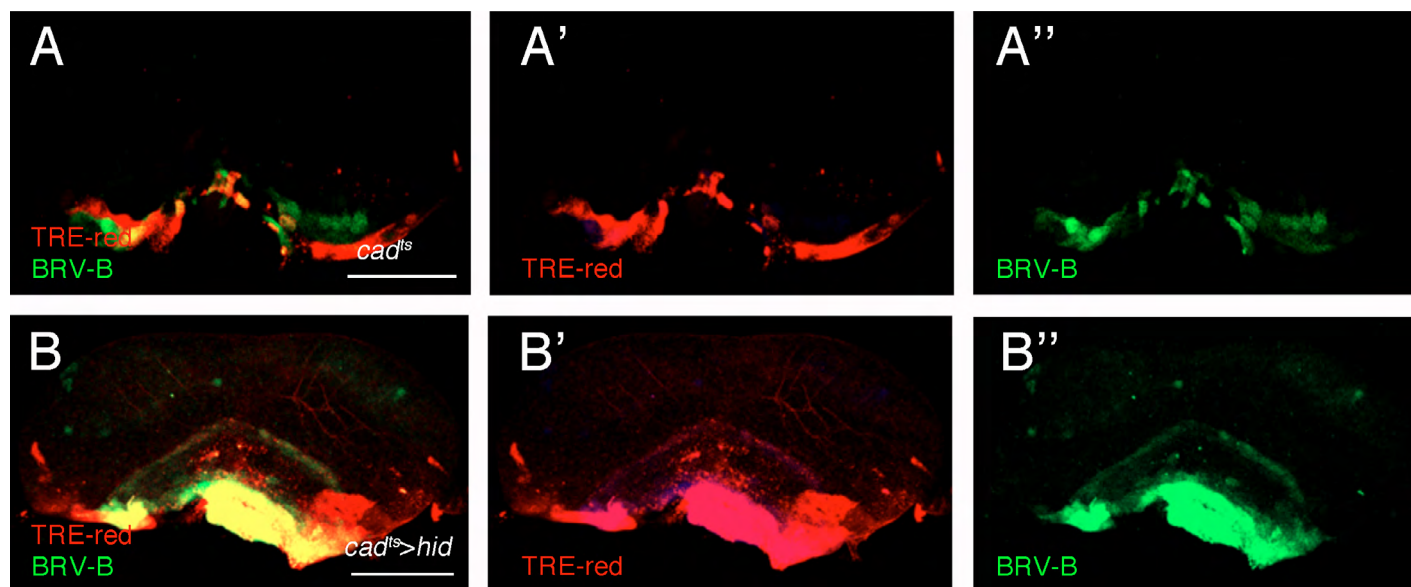


**Fig. S2. JNK pathway and wingless reporter activation after induction of cell death in the genital disc.** (A-A'') *cad-Gal4 tub-Gal80<sup>ts</sup>/+*; BRV-B-GFP TRE-DsRed/+ third instar male genital disc (larvae shifted from 18°C to 30°C), in which JNK activation (TRE-DsRed reporter) or BRV-B-GFP wg-reporter signal (in green) is limited to the stalk region of the disc. (B-B'') In *cad-Gal4 tub-Gal80<sup>ts</sup>/UAS-hid*; BRV-B-GFP TRE-DsRed/+ third instar male genital disc (larvae shifted from 18°C to 30°C) both TRE-DsRed and wg reporter expression is extended in the A10 segment. Scale bars are 50 μm.

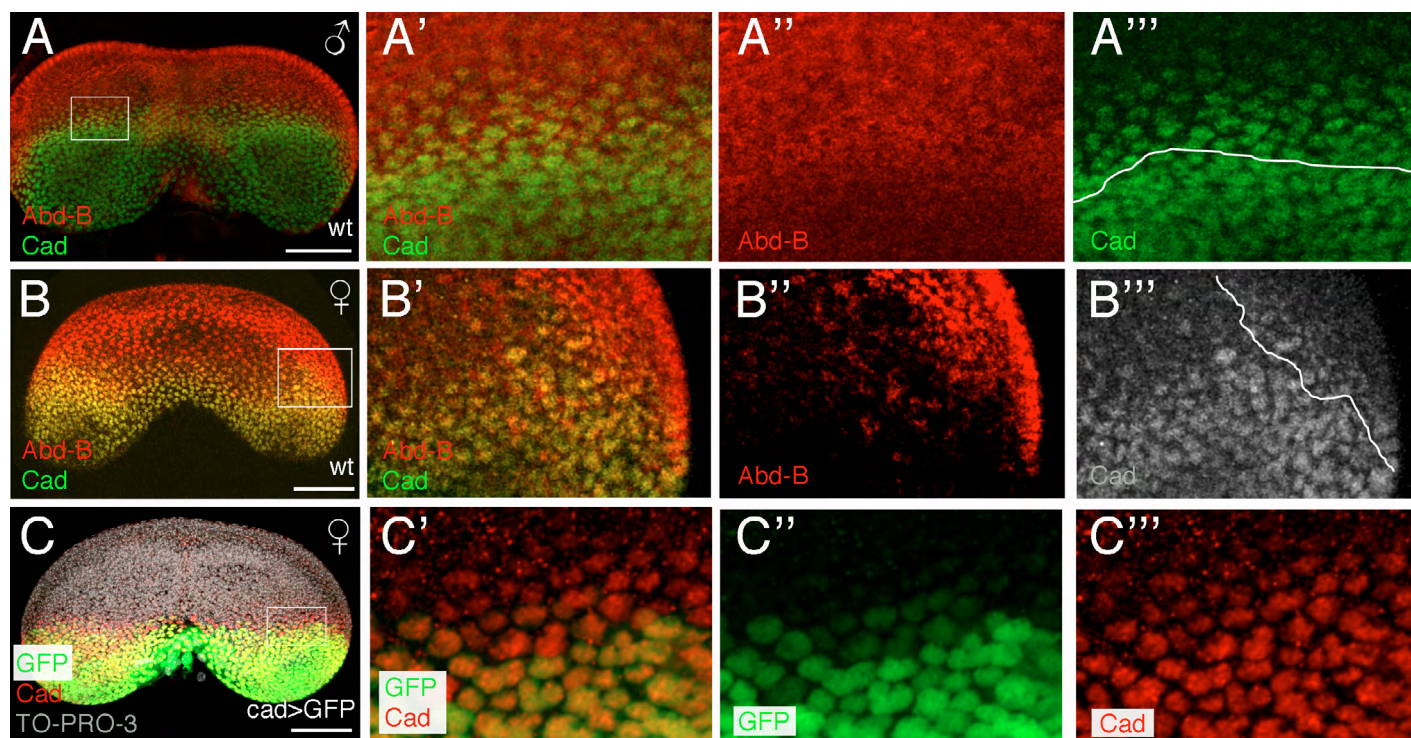


**Fig. S3. Caudal and Abdominal-B expression overlap in the larval genital discs.** (A-B''') Abd-B (red) and Cad (green) antibody signal in the third instar male (A-A''') and female (B-B''') genital discs. The insets in A and B are amplified in A'-A''' and B'-B''', respectively, and show the overlap in the expression of the two proteins. (C-C''') *cad-Gal4 UAS-GFP* third instar female disc stained with anti-Cad, showing *cad* expression anterior to GFP expression (the inset in C is amplified in C'-C'''). Scale bars are 50 μm.

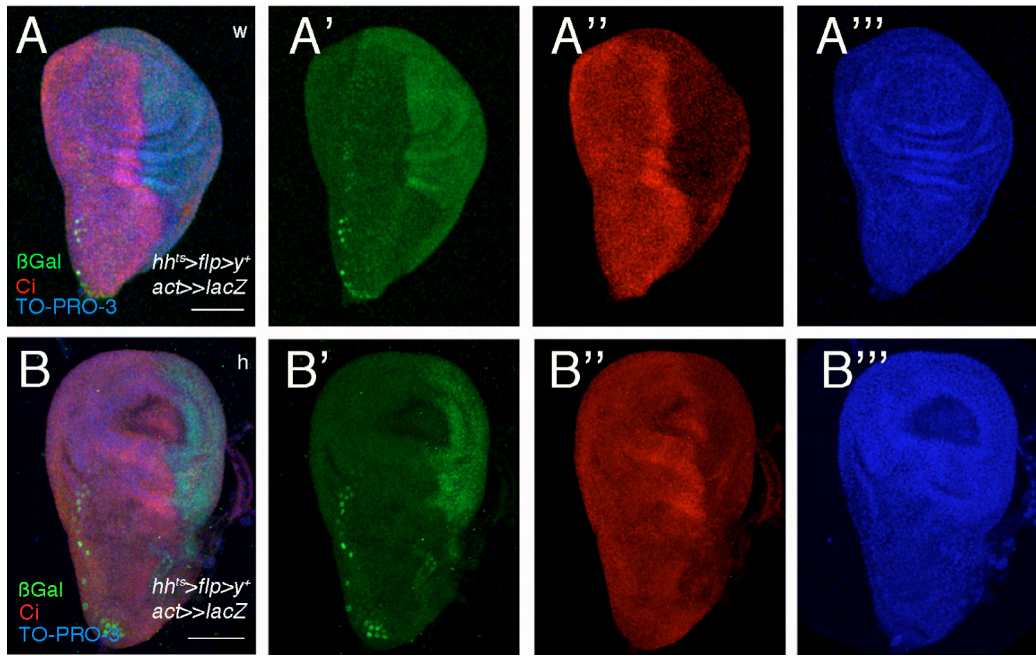


**Fig. S4. Complementary expression of *cubitus interruptus* and posterior compartment cell lineage.** (A-B''') Wing (w) (A-A''') and haltere (h) (B-B''') third instar discs showing Ci expression in the anterior compartment complementary to the cell lineage of the posterior compartment, marked by expressing lacZ through cassette recombination (genotype: UAS-*y*<sup>+</sup>/+; UAS-*flp* *act*> *stop*>lacZ/*hh*-Gal4/*tub*-Gal80<sup>ts</sup>). Second instar larvae were shifted from 18°C to 30°C. To-PRO-3 is in blue. Scale bars are 100 µm in A and 50 µm in B.

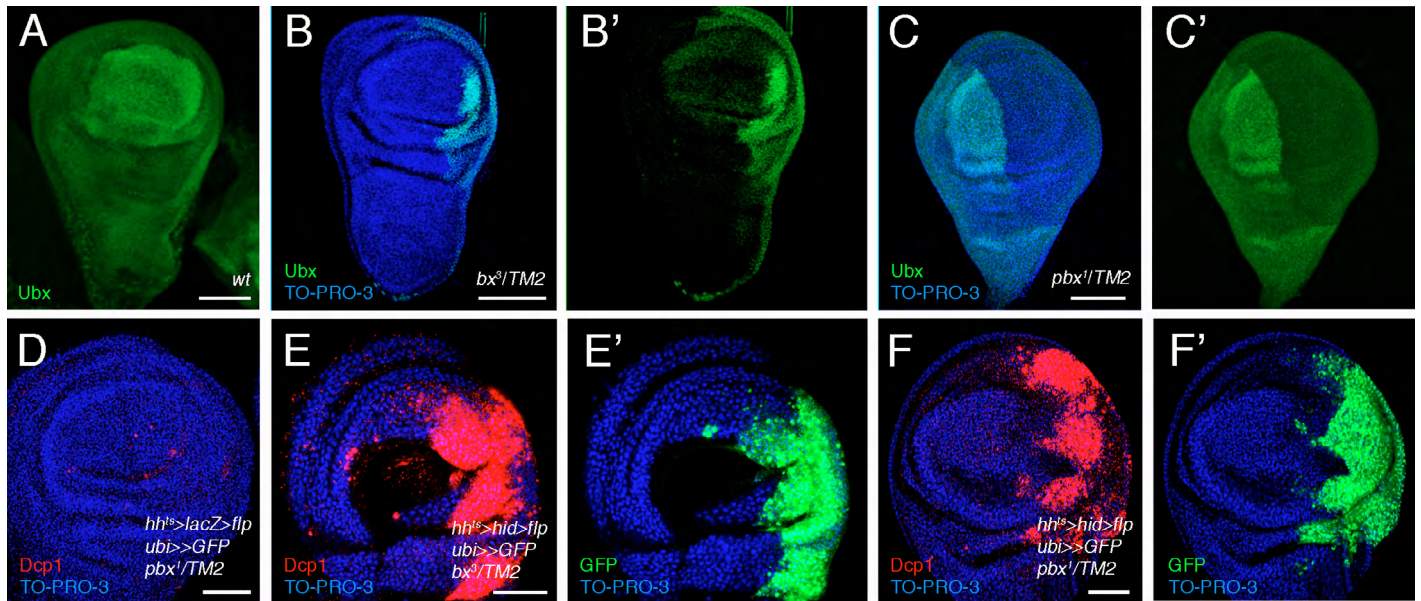


**Fig. S5. *Ubx* expression and cell death induction in *bx* and *pbx* mutants.** (A) *Ubx* expression in a wildtype haltere disc. (B-C'). *Ubx* expression in *bx*<sup>3</sup>/TM2 (B, B') and *pbx*<sup>1</sup>/TM2 (C, C') haltere discs. (D) Dcp1 signal in a *ubi*>*stop*>GFP UAS-*flp*/UAS-*lacZ* *tub*-Gal80<sup>ts</sup>; *pbx*<sup>1</sup> *hh*-Gal4/TM2 haltere disc (similar absence of Dcp1 expression is observed in a *bx* mutant background). E-F') Dcp1 signal (E, F) and cell lineage expression (E', F') after cell death induction with *hid* in the posterior compartment of *bx*<sup>3</sup>/TM2 (E, E') and *pbx*<sup>1</sup>/TM2 (F, F') haltere discs (*ubi*>*stop*>GFP UAS-*flp*/UAS-*hid* *tub*-Gal80<sup>ts</sup>; *bx*<sup>3</sup> or *pbx*<sup>1</sup> *hh*-Gal4/TM2). Treatment of discs in panel C was 48h 30°C, and for D it was 30h 30°C. Scale bars are 100 µm in A, B and C, and 50 µm in D, E and F.

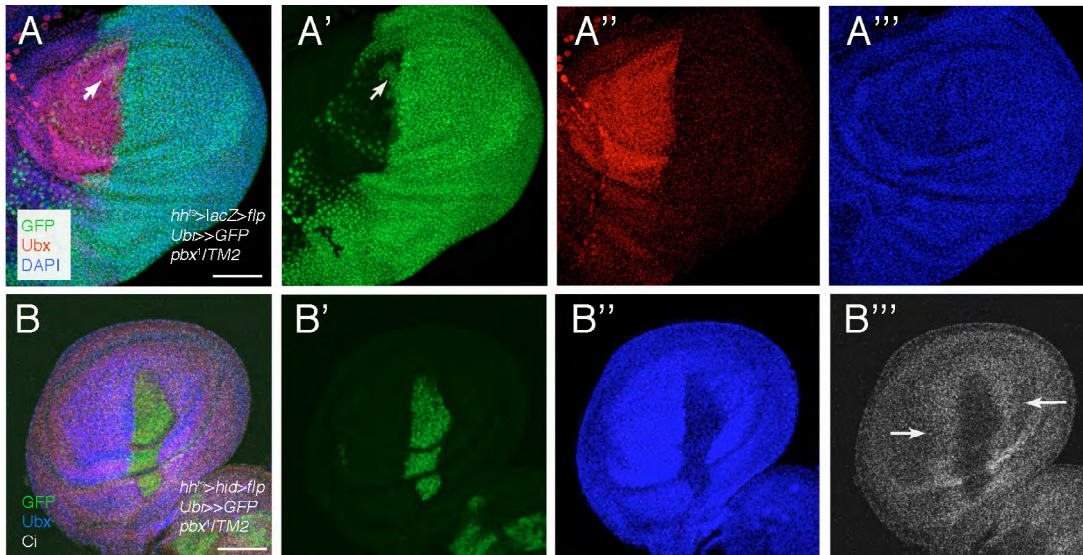


**Fig. S6. Cell lineage and *Ubx* expression in control and experimental (*hid* expression) *pbx* mutant discs.** (A-A''') Cell lineage and *Ubx* antibody signal after expression of *lacZ* in the posterior compartment of *pbx<sup>1</sup>/TM2* haltere discs (*ubi>stop>GFP UAS-flp/UAS-lacZ tub-Gal80<sup>ts</sup>; bx<sup>3</sup> hh-Gal4/TM2*). Note a group of posterior cells overlapping *Ubx* signal (arrow). (B-B''') Cell lineage and *Ubx* and *Ci* antibody signals after induction of cell death in the posterior compartment of *pbx<sup>1</sup>/TM2* haltere discs (*ubi>stop>GFP UAS-flp/UAS-hid tub-Gal80<sup>ts</sup>; pbx<sup>1</sup> hh-Gal4/TM2*). The posterior cells are surrounded by anterior ones. Scale bars are 50  $\mu$ m.

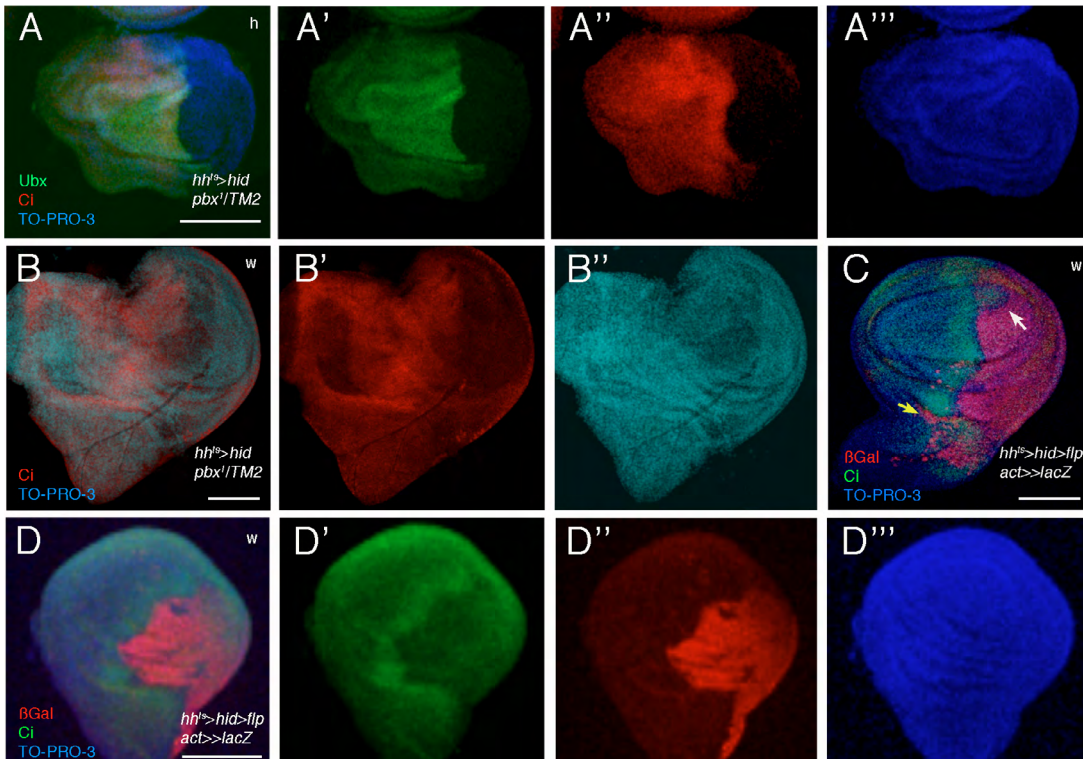


**Fig. S7. Abnormal development of wing and haltere discs after induction of cell death in the posterior compartment.** (A-B') Haltere (h) (A-A''') or wing (w) (B-B''') *pbx<sup>1</sup>/TM2* discs in which *hid* was induced in the posterior compartment, stained with *Ubx* and *Ci* (*UAS-hid tub-Gal80<sup>ts</sup>/+; pbx<sup>1</sup> hh-Gal4 / TM2*). Wing disc (w) in which cell death was induced in the posterior compartment (*UAS-hid tub-Gal80<sup>ts</sup>/+; hh-Gal4/UAS-flp act>stop>lacZ*). The cell lineage of posterior cells is detected by  $\beta$ -galactosidase expression and anterior cells by *Ci* signal. Note anterior cells entering the posterior compartment (white arrow) and posterior cells entering the anterior one (yellow arrow). (D-D''') Wing disc in which cell death was induced in the posterior compartment (*UAS-hid tub-Gal80<sup>ts</sup>/+; hh-Gal4/UAS-flp act>stop>lacZ*). The cell lineage of posterior cells is detected by  $\beta$ -galactosidase expression and anterior cells by *Ci* signal. The posterior compartment is highly reduced and the antero-posterior boundary is irregular. Scale bars are 100  $\mu$ m.