Sen et al., http://www.jcb.org/cgi/content/full/jcb.201206064/DC1

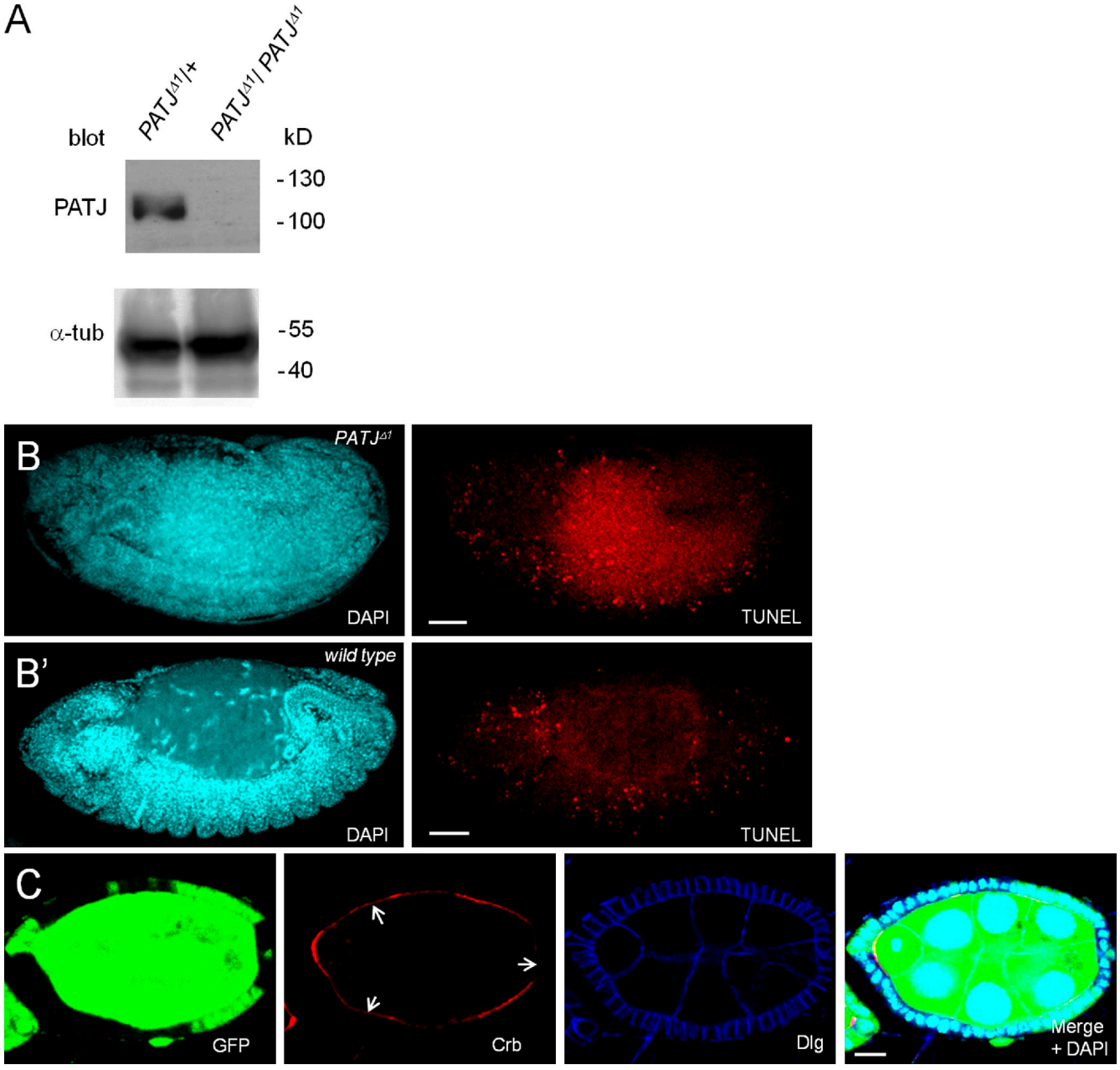


Figure S1. **PATJ mutant embryos and follicle cells.** This figure is related to Fig. 2. (A) Western blot showing that PATJ protein expression is not detectable in embryos, which are maternal and zygotic mutant for *PATJ*$^{\Delta 1}$, but is still present in heterozygous embryos. α-Tubulin (α-tub) was used as a loading control. (B and B′) Embryos homozygously mutant for *PATJ* show only moderate apoptosis in the epidermis of late stage embryos (B), which does not differ from wild-type embryos (B′). TUNEL assay was used to identify apoptotic cells. (C) Follicle cell clones showing decreased protein levels of Crb at the apical junction (arrows). *PATJ* mutant clones are marked by the absence of GFP. Bars: (B and B′) 200 µm; (C) 10 µm.

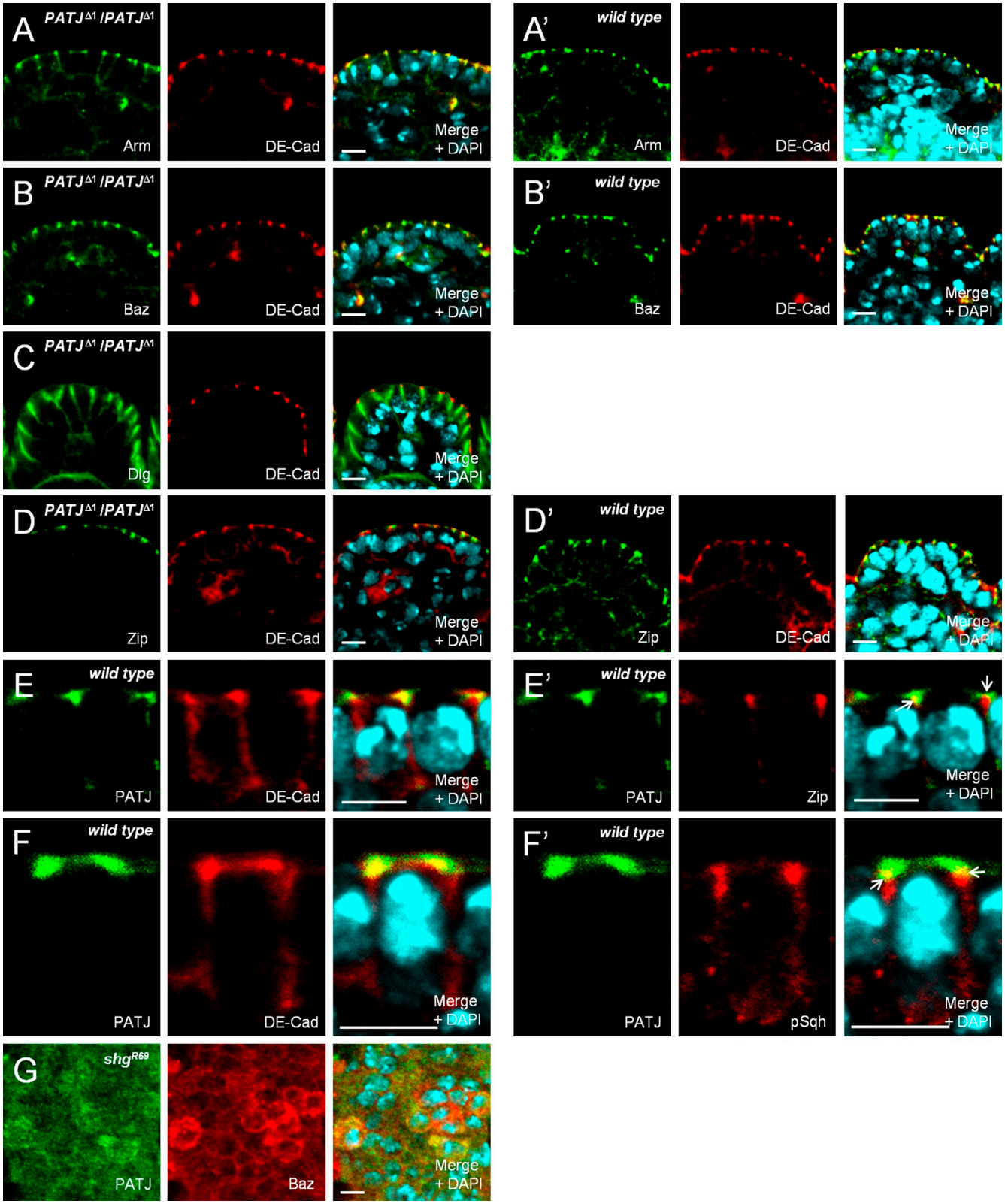


Figure S2. **Localization of Arm, Baz, and Dlg in PATJ mutant embryos.** This figure is related to Fig. 3. (A–D′) Embryos homozygous for *PATJ*$^{\Delta 1}$ (A–D) or wild-type embryos (A′ and B′) were stained against Arm (A and A′), Baz (B and B′), Dlg (C), and Zip (D and D′). Localization of these proteins is indistinguishable from wild-type epithelial cells (A′, B′, D′, and Fig. 2, D and E). (E–F′) High magnifications of epithelial cells showing a partly overlapping localization of PATJ and DE-Cad (E and F) and PATJ and Zip (E′) or pSqh (F′). Cells are the same in E and E′ as well as in F and F′. (G) PATJ and Baz are mislocalized to the cytoplasma in embryos homozygous mutant for *shg*^{R69}. Bars, 5 µm.

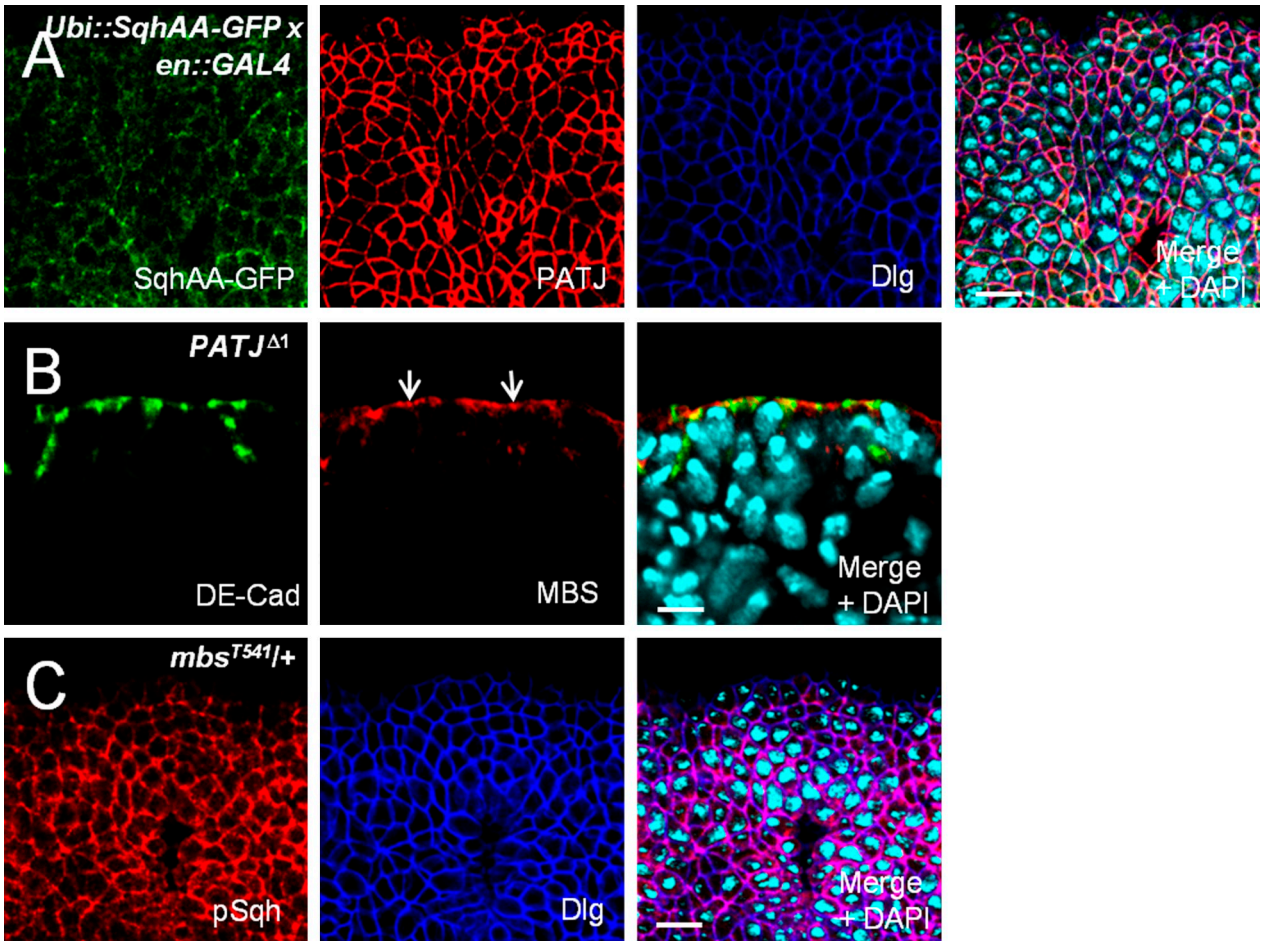


Figure S3. **Localization of nonphosphorylatable GFP-Sqh in wild-type embryonic epidermis.** This figure is related to Fig. 4. (A) Flies expressing SqhAA under a ubiquitous promoter were crossed with the driver line engrailed::GAL4 and stained against GFP, PATJ, and Dlg. Note that there is no accumulation of Sqh-GFP at the junctions in the absence of PATJ overexpression. (B) Localization of MBS in *PATJ* mutant epithelial cells is indistinguishable from wild-type epithelia (junctional MBS is marked by arrows). (C) Phosphorylation of Sqh in embryos heterozygous for mbs^{T541}. Bars: (A and C) 10 µm; (B) 5 µm.

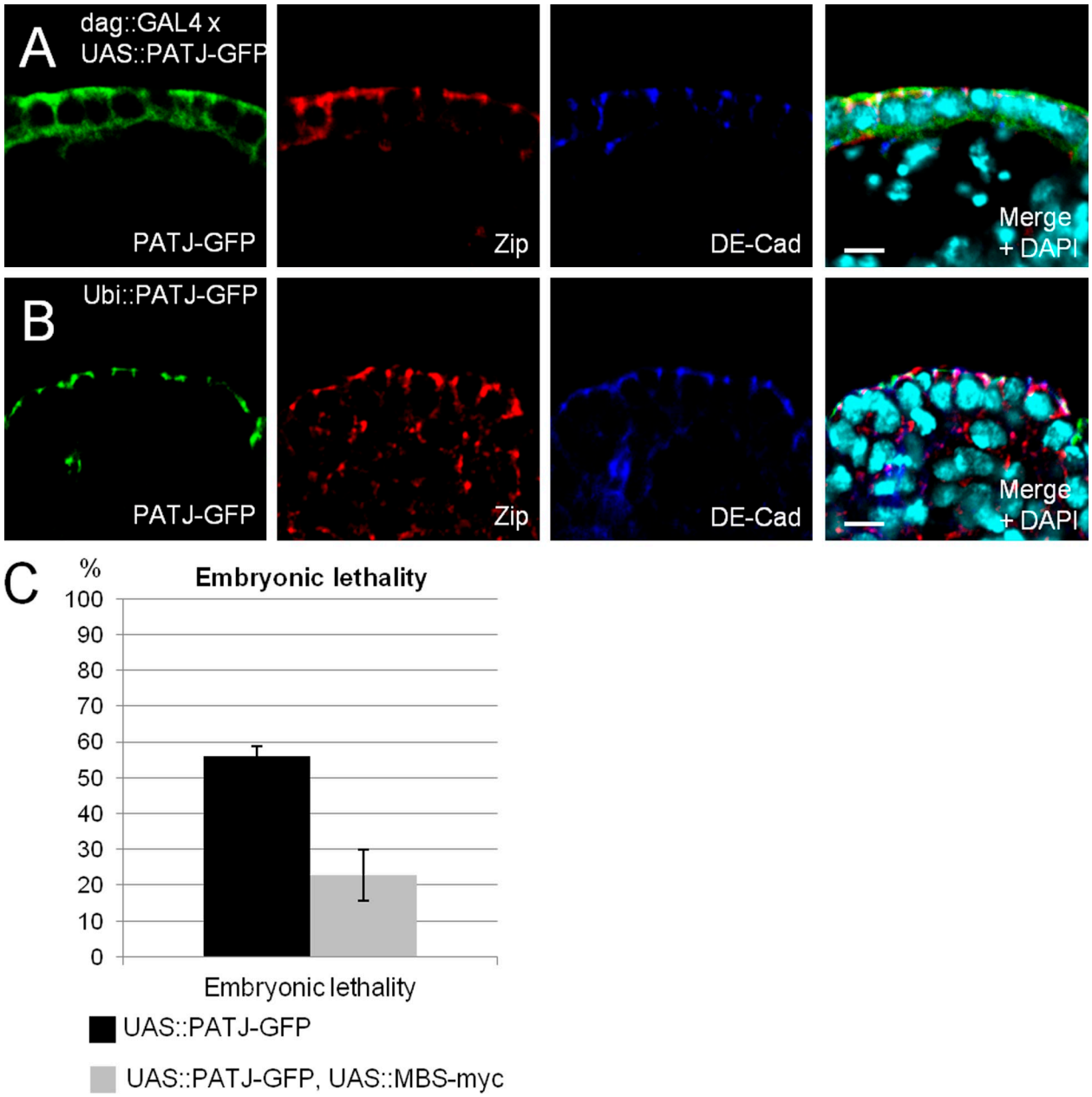
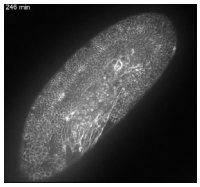


Figure S4. **Overexpression phenotypes of PATJ.** This figure is related to Fig. 5. (A) Overexpression of PATJ-GFP results in a mostly cytosolic protein localization, whereas Myosin and DE-Cad accumulate normally at the apical junctions. Daughterless (dag)::GAL4 was used to overexpress PATJ-GFP. (B) Moderate expression of PATJ-GFP by a ubiquitous promoter (polyubiquitin) results in a physiological protein localization and functional protein (not depicted). (C) Overexpression of PATJ-GFP by daughterless::GAL4 results in an increased embryonic lethality, which can be to some extent rescued by concomitant overexpression of MBS-myc. UAS, upstream activation sequence. Error bars show SDs. Bars, 5 µm.



Video 1. **Life imaging of a wild-type *Drosophila* embryo expressing DE-Cad–GFP.** This video is related to Fig. 2 and is shown at 10 frames/second. Images were recorded every 6 min for 15 h using a spinning-disc laser-scanning microscope (Axio Observer Z1; AxioCam MRm, 20× objective NA 0.8; Carl Zeiss). Each picture is a projection of a stack of four images to prevent out-of-focus slipping of the embryonic epidermis (calculated with ImageJ; National Institutes of Health). Anterior is in the lower left corner. Time in minutes is indicated. DE-Cad–GFP is expressed under a ubiquitous promoter. The video starts shortly after cellularization, showing the normal embryonic development of a representative wild-type embryo with germband elongation, segmentation, germband retraction, and finally, dorsal closure and head development.

Video 2. **Life imaging of a *PATJ* mutant *Drosophila* embryo expressing DE-Cad–GFP.** This video is related to Fig. 2 and recorded as Video 1. Anterior is in the lower left corner. The embryo shown is derived from $PATJ^{\Delta 1}$ germline clones, lacking the maternal and zygotic expression of PATJ. The video starts shortly after cellularization, showing a rather normal germband elongation and segmentation but an incomplete germband retraction with the posterior end of the germband ending at ∼20% embryonic length instead of 0% as in the wild-type embryos (compare with Video 1). Out of 39 *PATJ* mutant embryos investigated, eight exhibited defects in germband retraction and head development, five died early in development, and the rest developed normally.