

Figure S1. Moesin knockdown.

(A). Moesin is strongly depleted by an RNAi transgene driven under the *engrailed-Gal4* driver in the posterior half of the wing imaginal disc. The boxed area indicates where Flag-Conu localization was analyzed in this same disc for Figure 2F.

Figure S2. Effects of Rho1 depletion and Rho Gap expression on the wing imaginal epithelium.

(A) RNAi depletion of Rho1 in the dorsal compartment of the disc, under the *apGal4* driver, results in apoptosis, as assayed by Caspase-3 staining, and a smaller dorsal compartment. (B) Expression of a membrane tethered (myristoylated) version of a RhoGAP, *Cv-c*, under *apGal4* results in apoptosis and a smaller dorsal compartment as well. Membrane tethered expression of the C-terminal portion of Conu (residues 313-577) encompassing the GAP domain results in increased apical cell size in expressing cells (to the right of the arrowheads, C) and a furrow at the expression boundary, as can be seen from the bright E-cadherin staining where the two apical surfaces are opposed (D) and the lack of nuclei at the furrow (D'). The boundary of expression is indicated by arrowheads.

Figure S3. Interactions between Conu, Rho1 and Cdc42 in the eye.

(A) Expression of untethered Flag-tagged *conu* has no effect on the adult eye when expressed under *GMR-Gal4*. (B-C) The untethered Conu protein has little if any ability to suppress the rough eye of *GMR>Rho1*. (D) *GMR-Cdc42*, *GMR-Gal4/+* expression has a slight rough eye. (E-G) Co-expression of *myr-conu*, *Myr-conu^{R402A}*, or *myr-conu^{GAP}* under the *GMR-Gal4* driver

along with *GMR-Cdc42* had no effect on the *Cdc42* eye phenotype, appearing similar to *myr-conu*, *myr-conu^{R402A}* or *myr-conu^{GAP}* alone (Fig. 8C, E, J).

Figure S4. Effects of Arf6 expression in the eye.

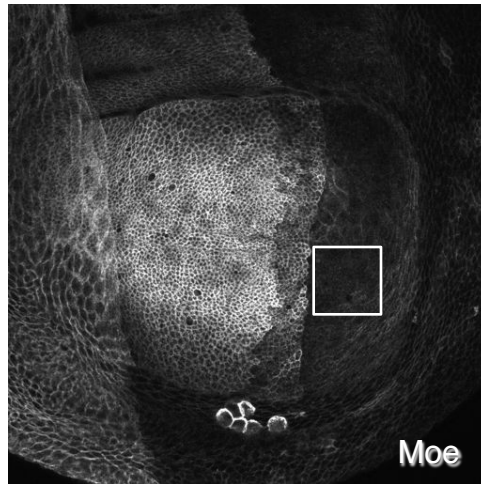
(A) Expression of *Arf6* in the posterior half of the disc under the *enGal4* driver results in cell shape changes compared to wild type cells (posterior is to the right of the arrowheads). (B-C) *Arf6* expression has no effect on Conu or phospho-ERM protein levels. (D-E) Co-expression of *Arf6* in the eye under *GMR-Gal4* alone with *GMR-Cdc42* had little effect on the adult eye, appearing similar to *GMR>Arf6* expression alone (Fig. 9H).

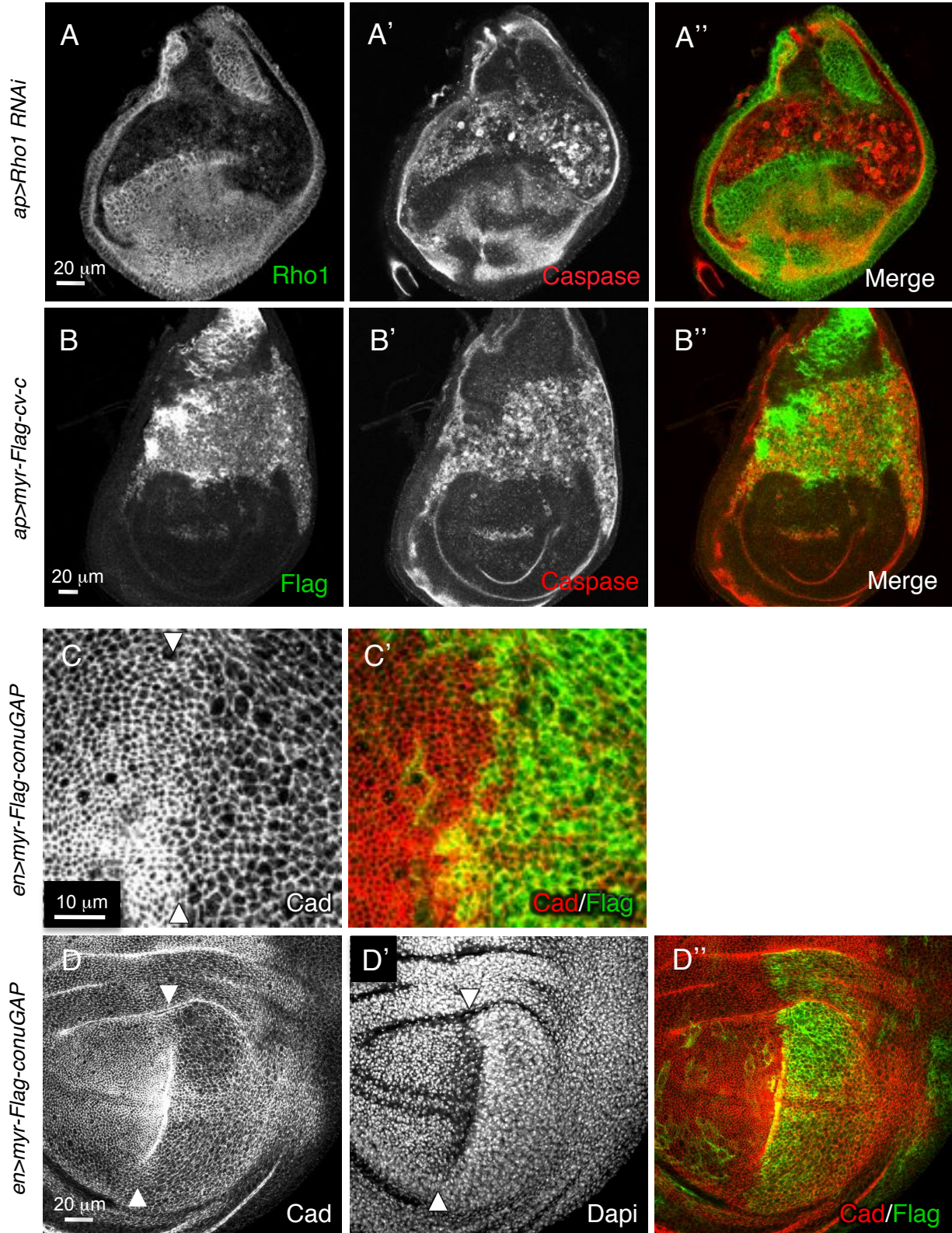
Figure S5. Signaling Pathways and Conu

For all discs, unless otherwise noted, staining of the disc proper is shown. The *enGal4* expressing, posterior half is to the right. A-B) *puc-LacZ*, a readout of JNK signaling is upregulated in cells expressing *myr-conu* compared to control discs. In contrast, readouts for Notch signaling (*E(Spl)Mb-LacZ*; C-D), Wingless signaling (*Dll* and *Sens*; E-H) Hippo signaling (Ex staining; I-J), Hedgehog signaling (*Ci* staining; K-K') are unaffected by *myr-Conu* expression. (L-L') Endogenous pattern of p-Mad staining, a TGFβ signaling readout, in wild-type wing discs. (M-M') p-Mad staining is not elevated in disc proper cells expressing *myr-conu*, but is increased in the overlying peripodial cells. Both layers express *myr-conu*.

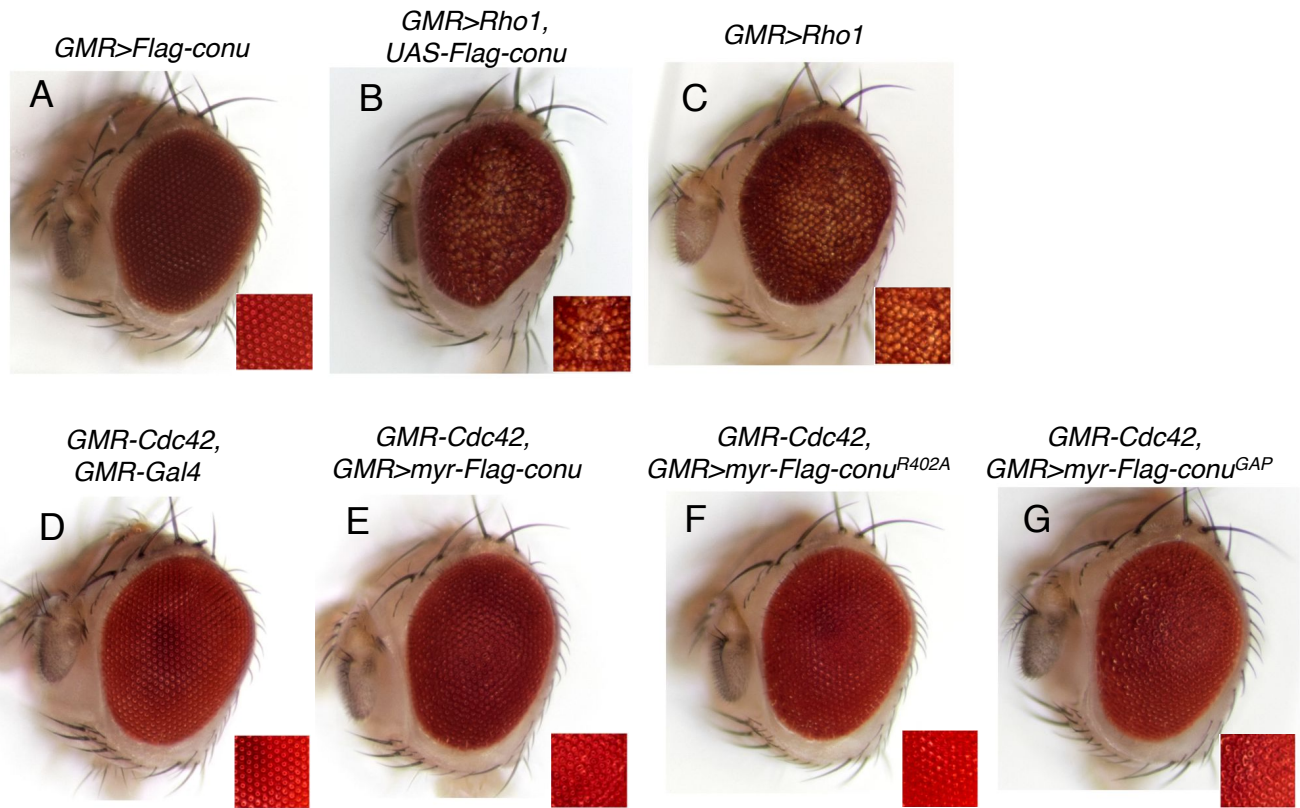
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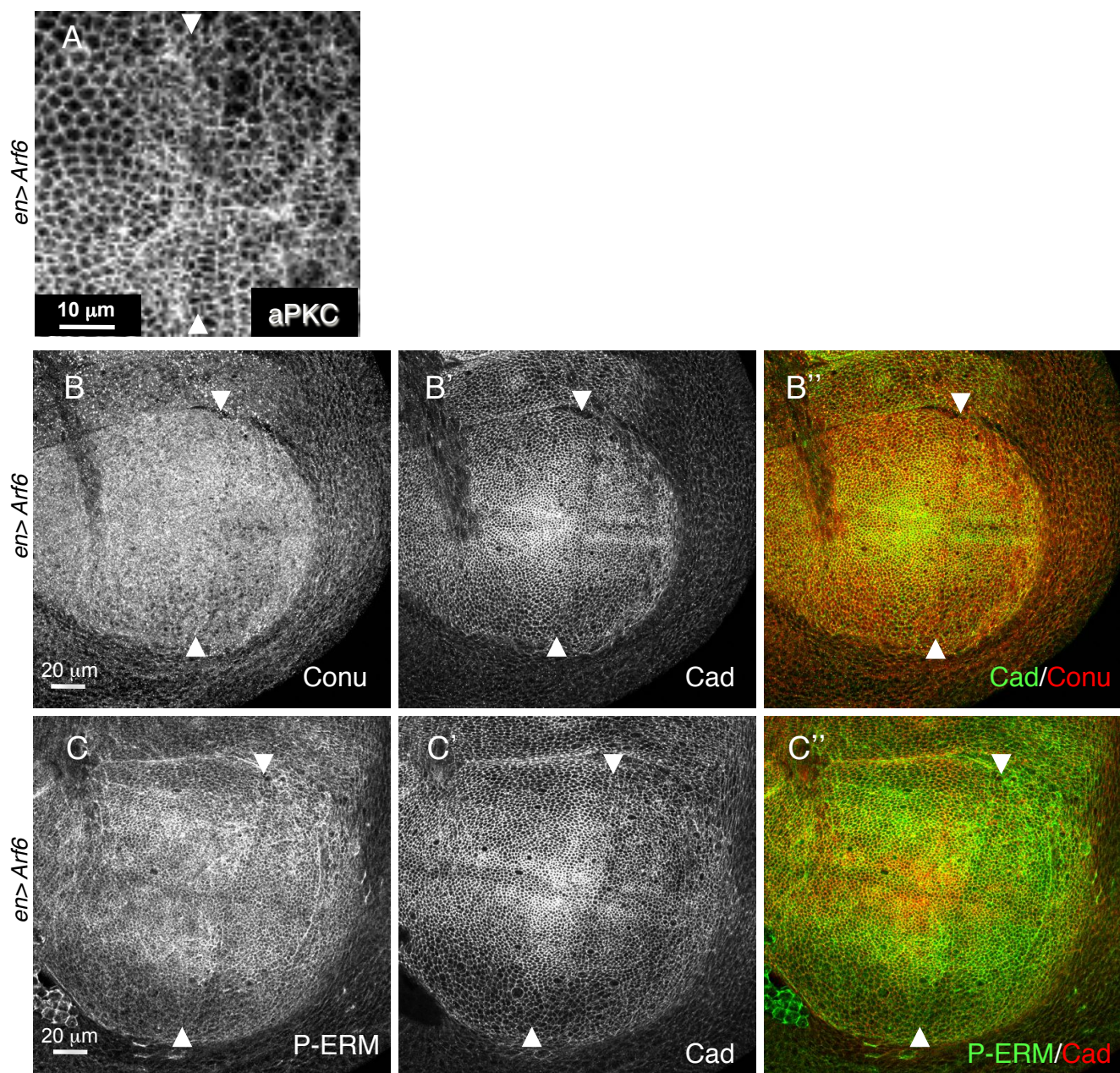
en>Flag-conu, UAS-MoeRNAi





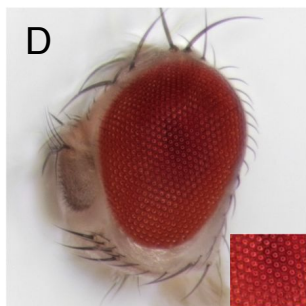
Neisch et al., Figure S2



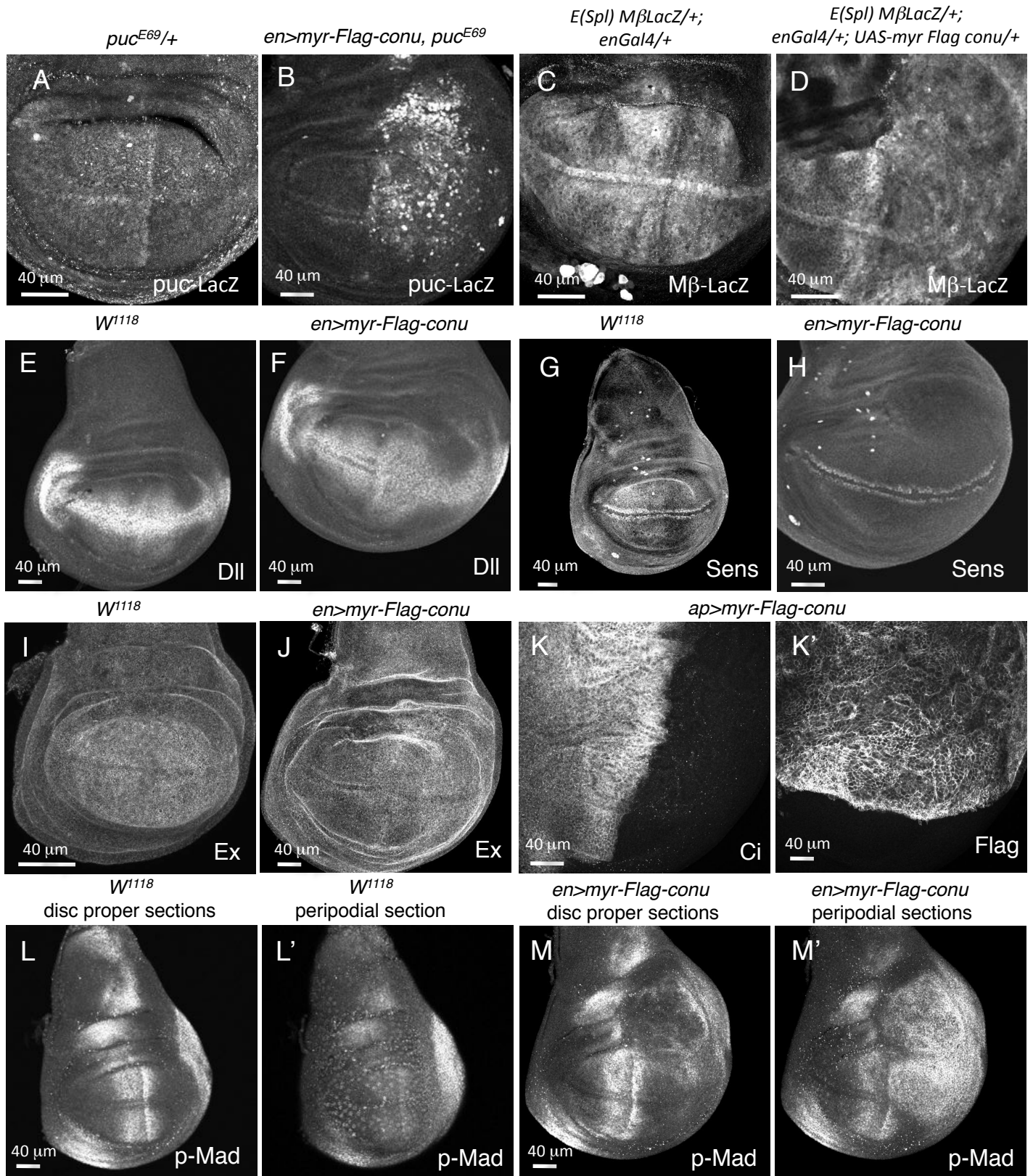


GMR-Cdc42,
GMRGal4

GMR-Cdc42,
GMR>Arf6



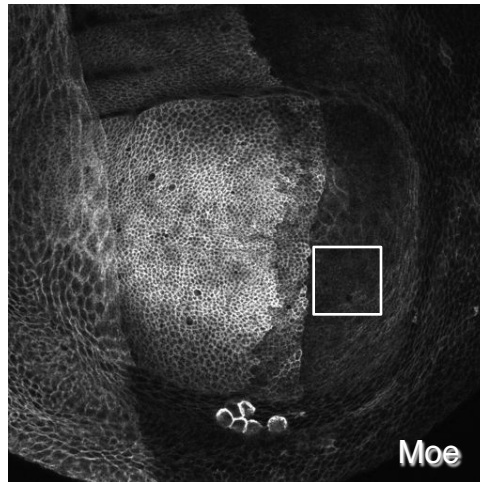
Neisch et al., Figure S4

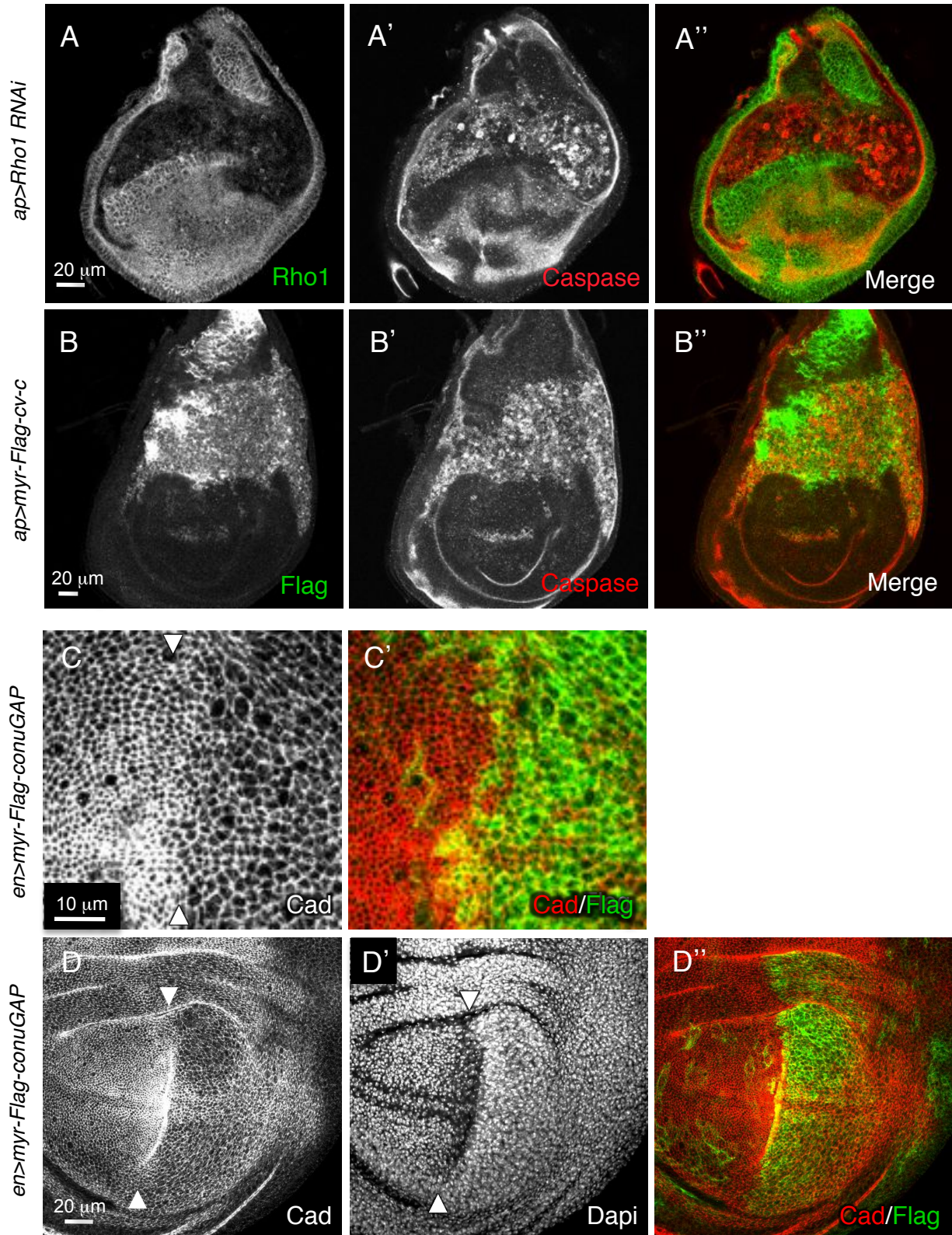


Neisch et al., Figure S5

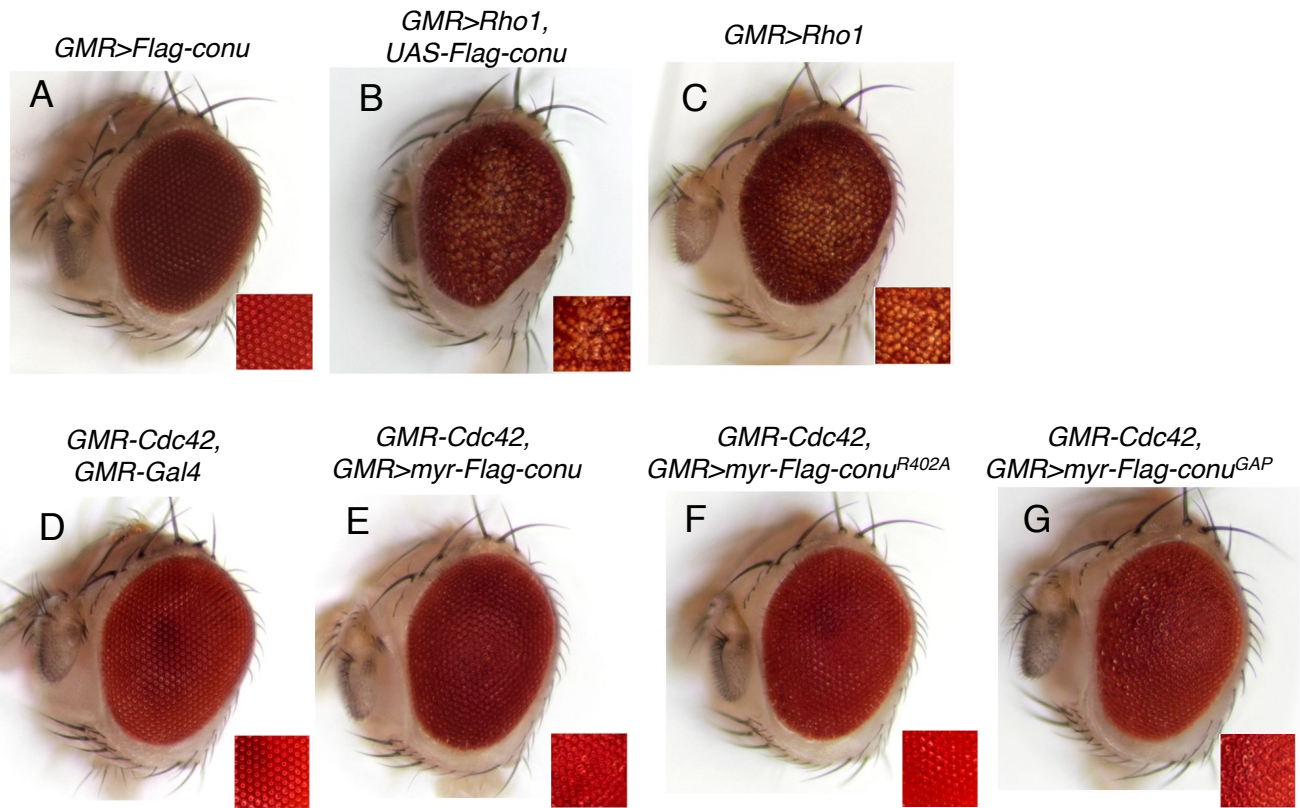
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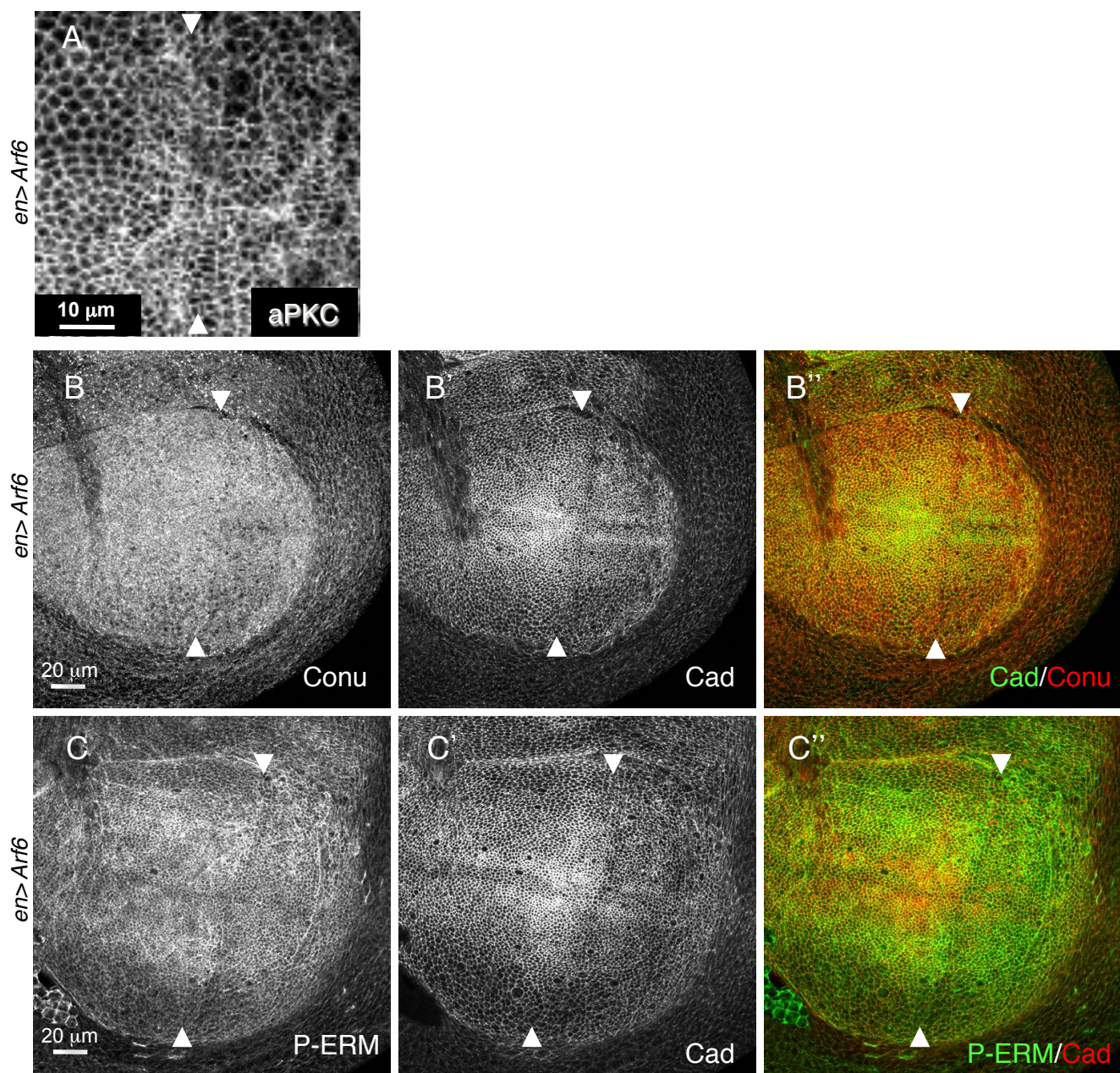
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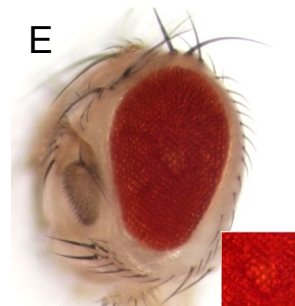
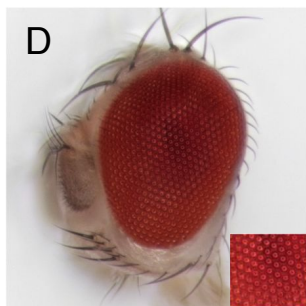
Neisch et al., Figure S2



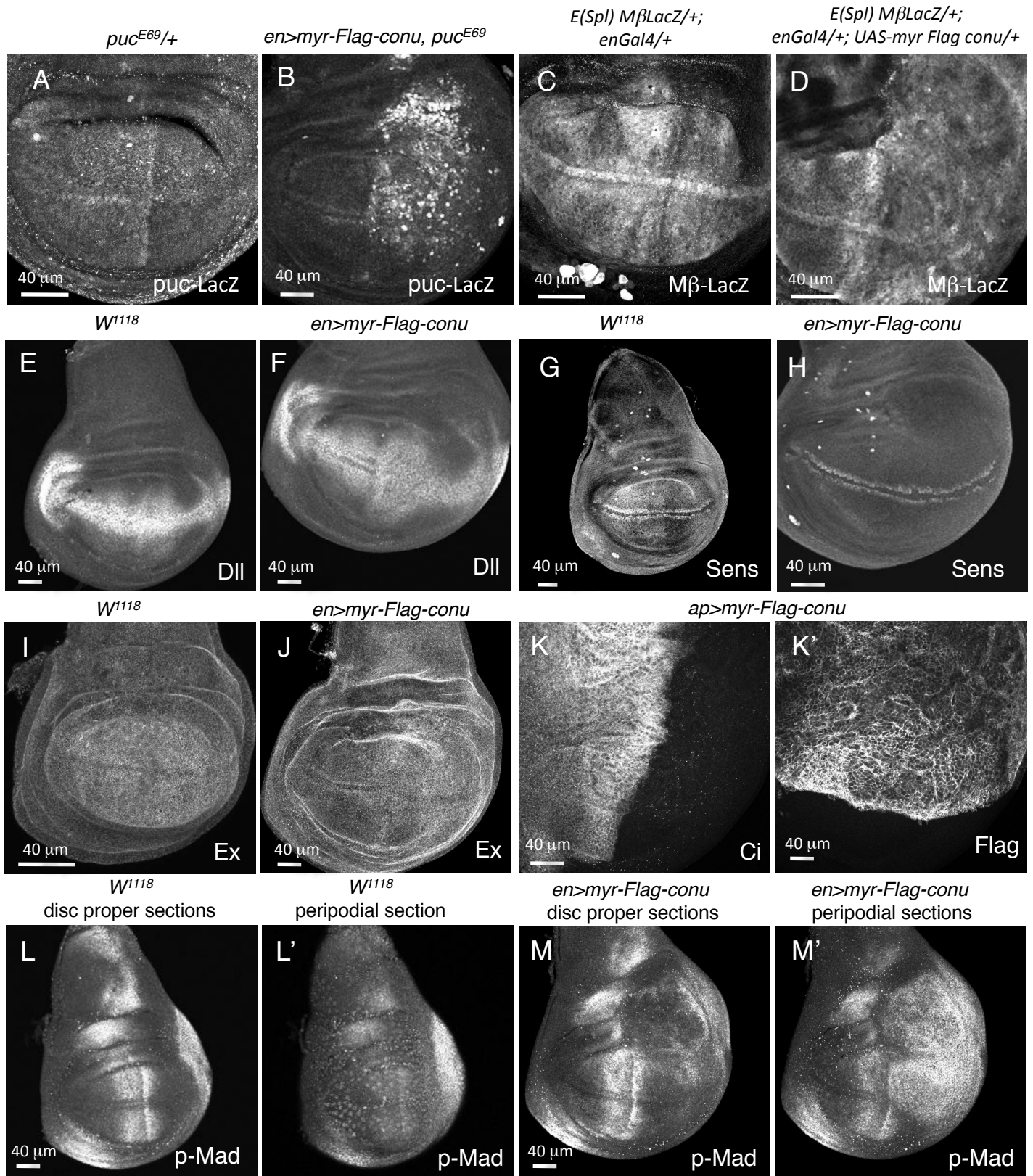


GMR-Cdc42,
GMRGal4

GMR-Cdc42,
GMR>Arf6



Neisch et al., Figure S4



Neisch et al., Figure S5