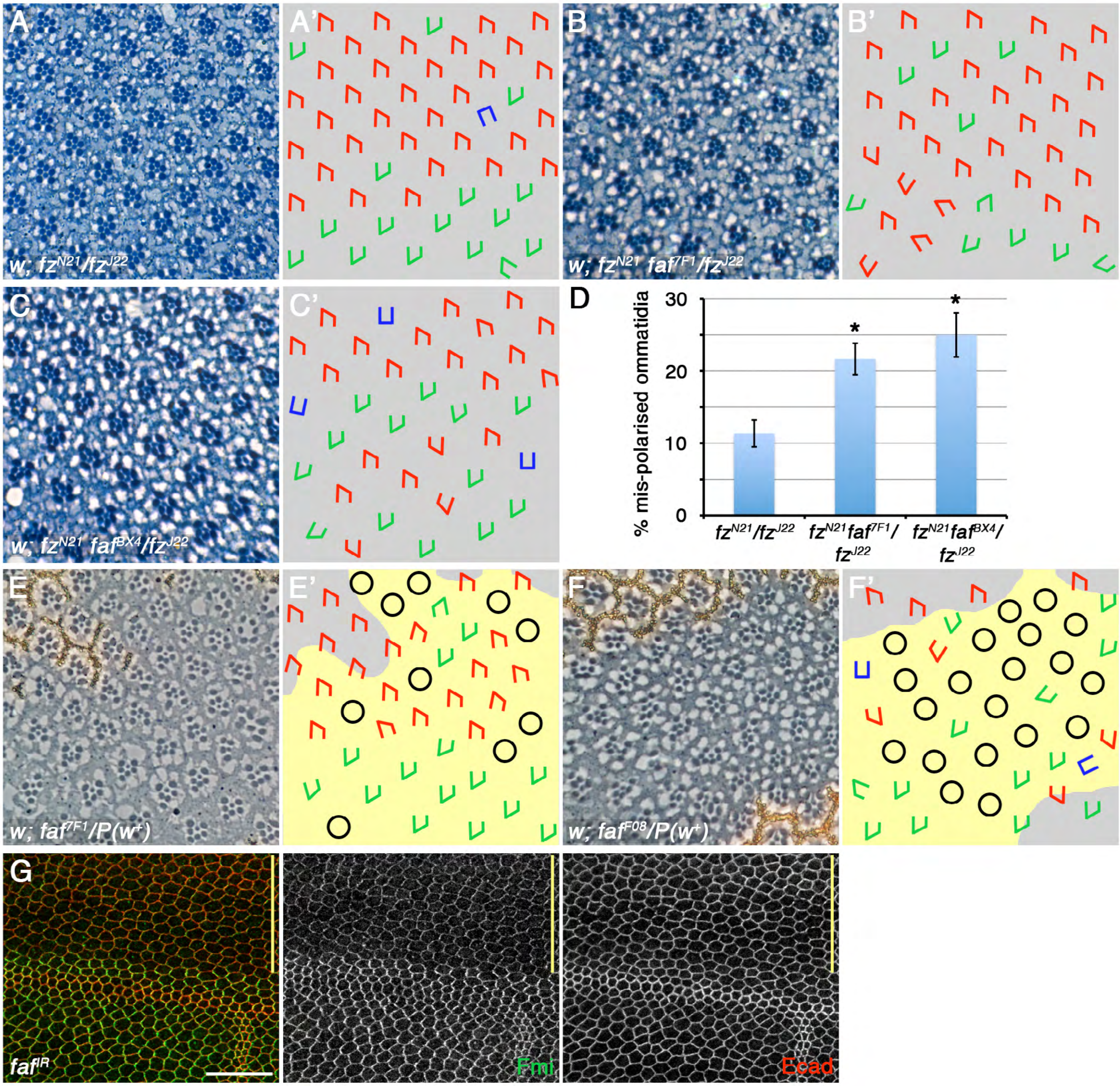

Fig. S1. *faf* mutant phenotypes. (**A-C′**) Adult eye sections and cartoons of fz^{N21}/fz^{J22} (A), fz^{N21} faf^{7F1}/fz^{J22} (B) and fz^{N21} faf^{BX4}/fz^{J22} (C). Red indicates dorsal ommatidia, green indicates ventral ommatidia and blue indicates achiral ommatidia. (**D**) The enhancement of the fz^{N21}/fz^{J22} ommatidial polarity phenotype by *faf*. Error bars are s.e.m.; *P<0.05. (**E-F′**) faf^{7F1} (E) and faf^{F08} (F) eye clones, marked by loss of white pigment, or yellow in the cartoons. Note that in *faf* mutant tissue many ommatidia have photoreceptor recruitment defects (black circles in cartoons), but in ommatidia with normal photoreceptor number there are frequent polarity defects, including chirality defects and misrotations. (**G**) *ptc-GAL4/+; faf*$^{IR\text{-}2956}$/+ pupal wings stained for Fmi (green) or Ecad (red). Yellow bars mark the *ptc-GAL4* domain. Scale bar: 20 µm.

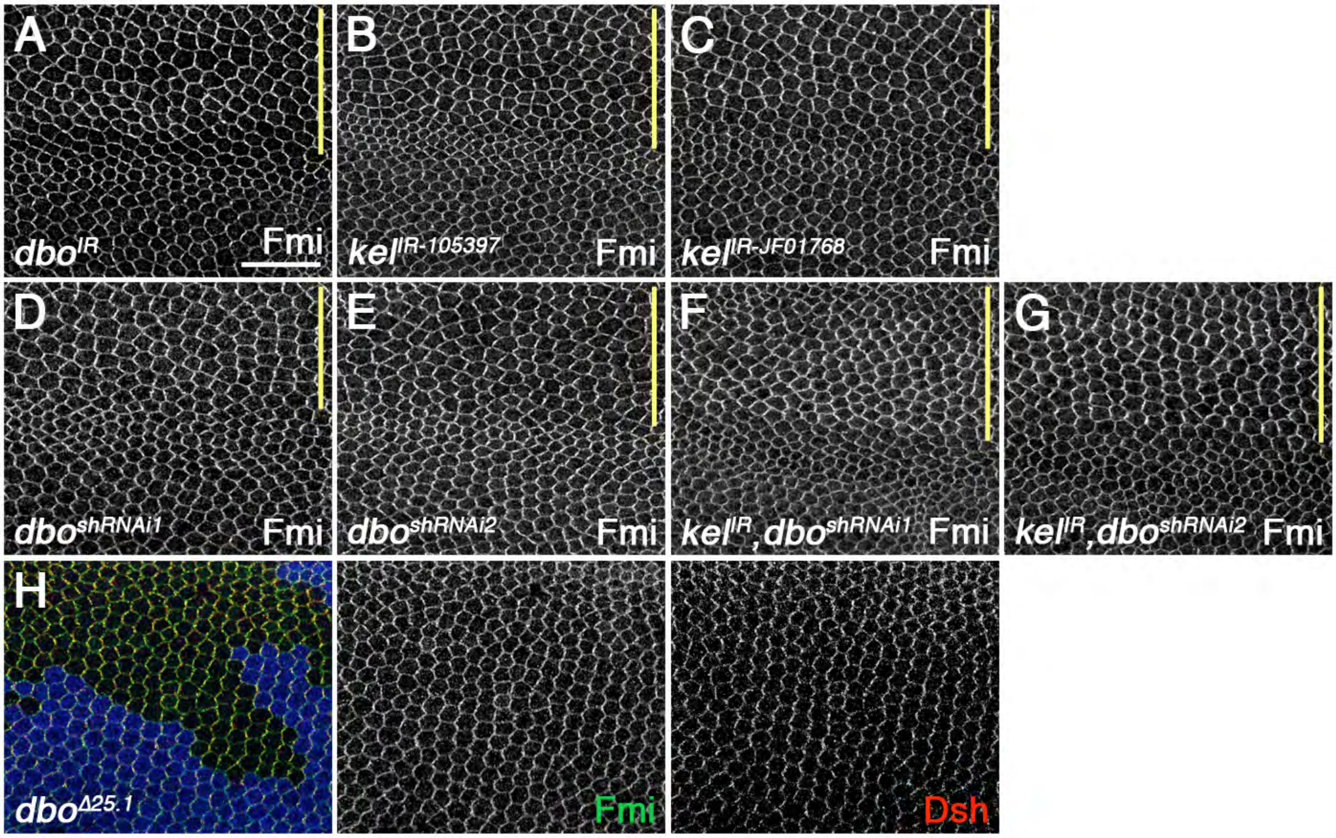

Fig. S2. Effects of RNAi against *dbo* and *kel*. (**A-G**) Pupal wings from *ptc-GAL4/dbo*$^{IR\text{-}105407}$*; UAS-Dcr2/+* (A), *ptc-GAL4/kel*$^{IR\text{-}105397}$*; UAS-Dcr2/+* (B), *ptc-GAL4/+; kel*$^{IR\text{-}JF01768}$*/UAS-Dcr2* (C), *ptc-GAL4/dbo*^{shRNAi1} (D), *ptc-GAL4/dbo*^{shRNAi2} (E), *ptc-GAL4/dbo*^{shRNAi1}*; kel*$^{IR\text{-}JF01768}$*/UAS-Dcr2* (F) and *ptc-GAL4/dbo*^{shRNAi2}*; kel*$^{IR\text{-}JF01768}$*/UAS-Dcr2* (G) stained for Fmi. Note that the increase in core protein localisation caused by RNAi against *dbo* and *kel* (A-C) was not apparent in every wing, probably owing to variations in staining and the flatness of the wings. (**H**) *dbo*$^{\Delta 25.1}$ pupal wing clone stained for Fmi (green) and Dsh (red); clone marked by loss of β-gal (blue). Yellow bars mark the *ptc-GAL4* domain. Scale bar: 20 μm.

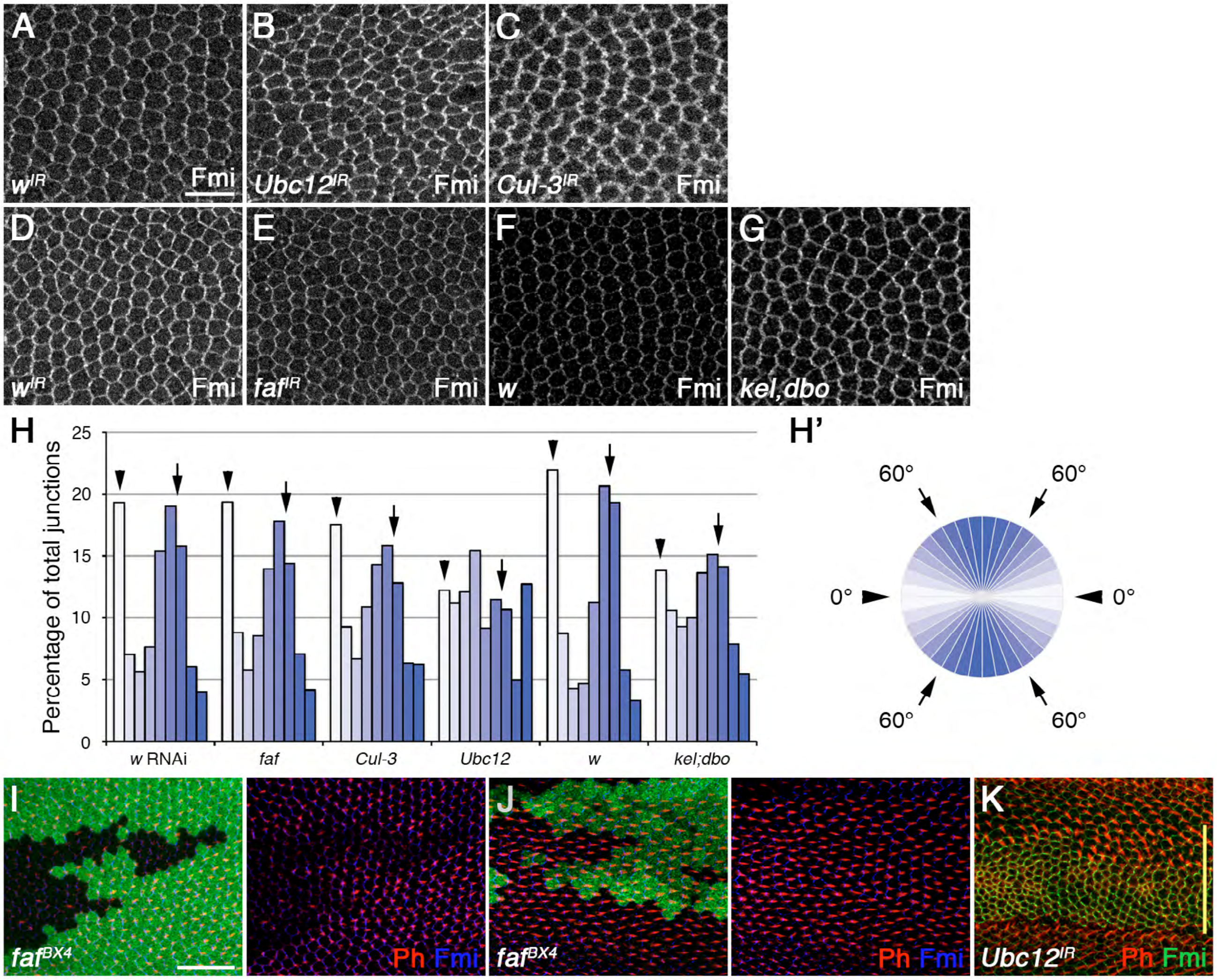

Fig. S3. Loss of asymmetry and trichome phenotypes of *Ubc12, Cul-3* and *faf.* (**A-E**) Male wings expressing $w^{IR\text{-}30033}$ (A,D), $Ubc12^{IR\text{-}7375R\text{-}3}$ (B), $Cul\text{-}3^{IR\text{-}109415}$ (C) and $faf^{IR\text{-}2956}$ (E) using *MS1096-GAL4*, stained with Fmi. (**F**,**G**) Pupal wings from w^{1118} (F) or kel^{DE1}; $dbo^{\Delta 25.1}$ double mutants (G), stained for Fmi. Sets of wings (A-C, D,E and F,G) were dissected, stained and imaged in parallel using the same confocal settings. All images are below vein 4. (**H**,**H′**) The percentage of junctions oriented at different angles, where 0° is horizontal and 90° is vertical, in the above genotypes. Angles are binned into 10° categories, where 0-10° is white and 80-90° is dark blue. The colour scheme of the differently oriented junctions is shown in H′. For a hexagonal array of cells, most junctions would be expected to be oriented at around 0° (arrowheads) or 60° (arrows). Cell packing is very irregular in wings expressing *Ubc12* RNAi, which appears to have pleiotropic effects. By contrast, cell packing is only mildly affected in *Cul-3*, *dbo; kel* and *faf* wings, in line with the known role of core proteins in regulating junctional rearrangement. (**I**,**J**) faf^{BX4} pupal wing clones stained for Fmi (blue) and with phalloidin (red); clones are marked by loss of β-gal (green), just after trichomes emerge in wild-type tissue (I) or when trichomes are slightly older (J). (**K**) *ptc-GAL4/+; Ubc12*$^{IR\text{-}7375R\text{-}3}$/+ pupal wings stained for Fmi (green) and with phalloidin (red). Note trichomes are very delayed where high levels of *Ubc12* RNAi are expressed, and trichomes swirl towards the compartment boundary. Yellow bars mark the *ptc-GAL4* domain. Scale bars: 10 µm in A-G; 20 µm in I-K.

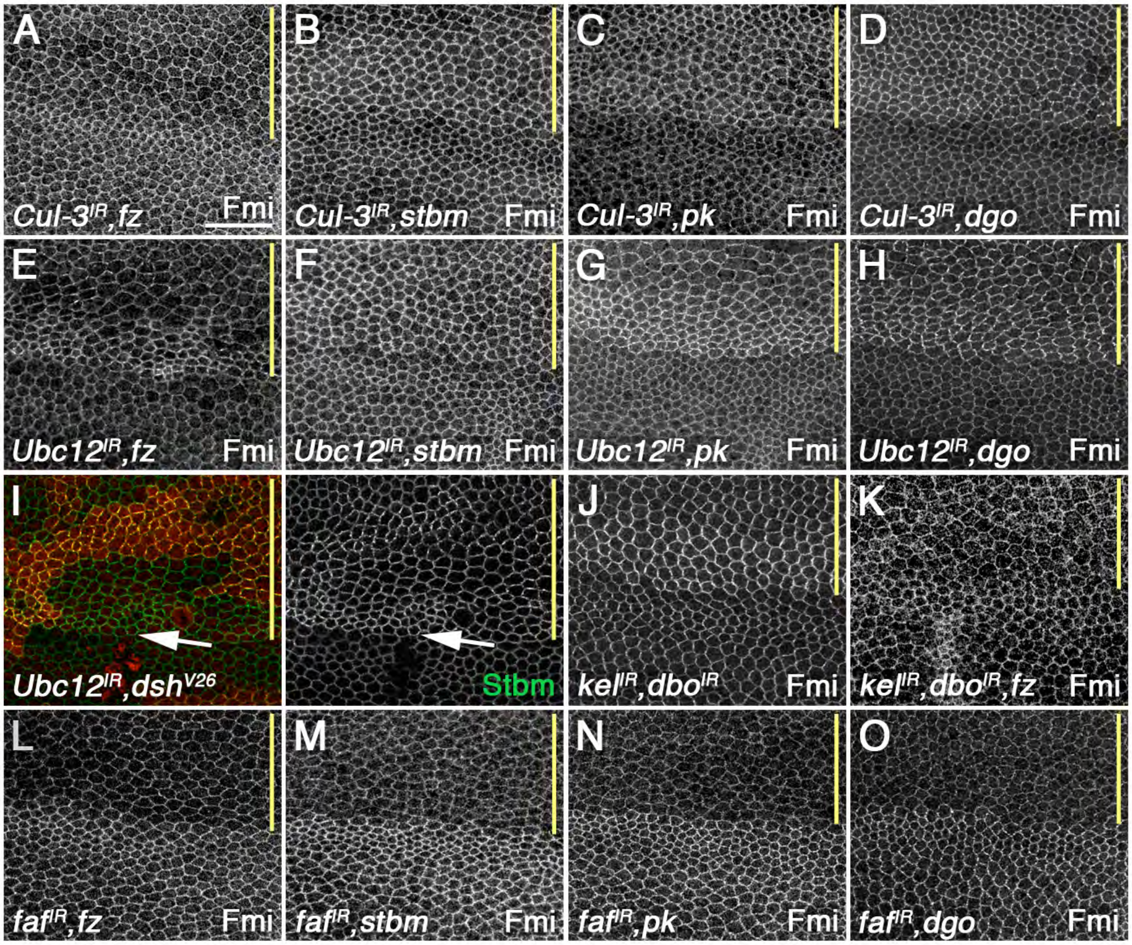

Fig. S4. Identification of the targets of Dbo/Kel and Faf. (**A-O**) *ptc-GAL4/Cul-3*$^{IR-109415}$*; fz*^{P21}*/fz*^{P21} (A), *ptc-GAL4, stbm*^{6}*/Cul-3*$^{IR-109415}$*, stbm*^{6} (B), *ptc-GAL4, pk*$^{pk-sple13}$*/Cul-3*$^{IR-109415}$*, pk*$^{pk-sple13}$ (C), *ptc-GAL4, dgo*^{380}*/Cul-3*$^{IR-109415}$*, dgo*^{380} (D), *ptc-GAL4/+; Ubc12*$^{IR-7375R-3}$*, fz*^{P21}*/fz*^{P21} (E), *ptc-GAL4, stbm*^{6}*/stbm*^{6}*; Ubc12*$^{IR-7375R-3}$*/+* (F), *ptc-GAL4, pk*$^{pk-sple13}$*/pk*$^{pk-sple13}$*; Ubc12*$^{IR-7375R-3}$*/+* (G), *ptc-GAL4, dgo*^{380}*/dgo*^{380}*; Ubc12*$^{IR-7375R-3}$*/+* (H), *dsh*^{V26} *FRT19A/FRT19A; ptc-GAL4/+; Ubc12*$^{IR-7375R-3}$*, Ubx-FLP/+* (I), *ptc-GAL4/dbo*$^{IR-105407}$*; kel*$^{IR-JF01768}$*/+* (J), *ptc-GAL4/dbo*$^{IR-105407}$*; kel*$^{IR-JF01768}$*, fz*^{P21}*/fz*^{P21} (K), *ptc-GAL4/+; faf*$^{IR-2956}$*, fz*^{P21}*/fz*^{P21} (L), *ptc-GAL4, stbm*^{6}*/stbm*^{6}*; faf*$^{IR-2956}$*/+* (M), *ptc-GAL4, pk*$^{pk-sple13}$*/pk*$^{pk-sple13}$*; faf*$^{IR-2956}$*/+* (N) and *ptc-GAL4, dgo*^{380}*/dgo*^{380}*; faf*$^{IR-2956}$*/+* (O). Pupal wings are stained for Fmi, except for I which is stained for Stbm (green) and Dsh (red). Yellow bars mark the *ptc-GAL4* domain. Note that the *dsh* clone in I is very proximal in the wing and overlaps the AP compartment boundary, where *Ubc12* knockdown is strongest. *dsh* clones in more distal wing regions (where *Ubc12* knockdown is weaker) show complete suppression of the increase in core protein levels. In this proximal clone, *Ubc12* knockdown still induces a small increase in Stbm levels at junctions compared with wild-type tissue (below the arrow). Scale bar: 20 µm.

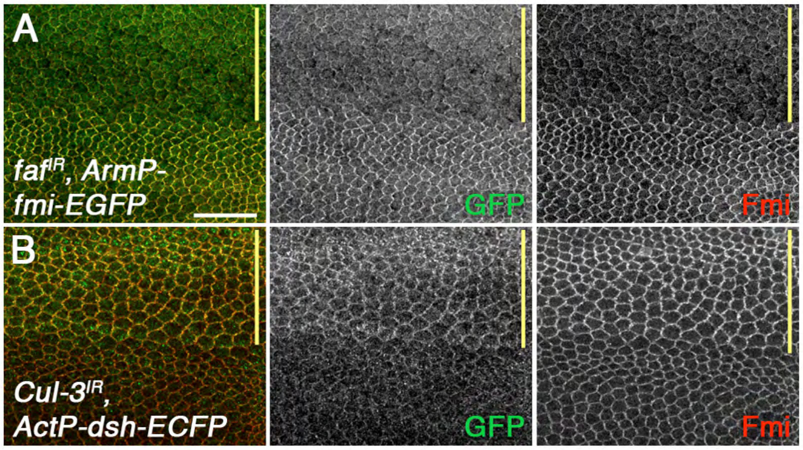

Fig. S5. Alteration of core protein levels is post-transcriptional. *ptc-GAL4/ArmP-Fmi-EGFP; faf*$^{IR-2956}$*/+* (**A**) and *Ubx-FLP; ptc-GAL4/Cul3*$^{IR-109415}$*; ActP-FRT-polyA-FRT-Dsh-ECFP/+* (**B**) pupal wings, stained for GFP (green) and Fmi (red). Yellow bars mark the *ptc-GAL4* domain. Scale bar: 20 µm.

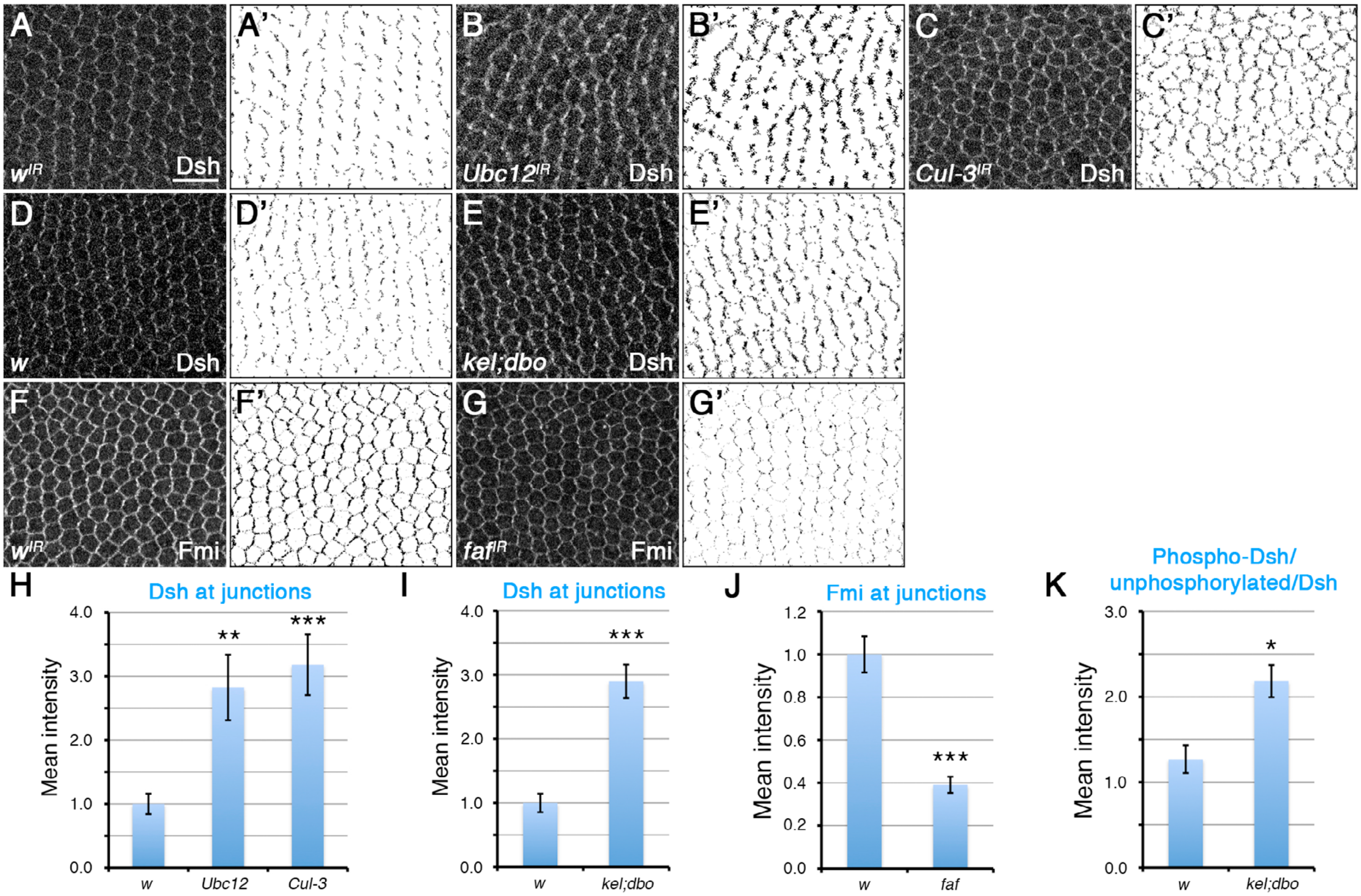

Fig. S6. Analysis of junctional levels of Dsh and Fmi. (**A-C**) Male pupal wings expressing *w*$^{IR-30033}$ (A), *Ubc12*$^{IR-7375R-3}$ (B) and *Cul-3*$^{IR-109415}$ (C) using *MS1096-GAL4*, stained for Dsh. (**D**,**E**) Pupal wings from *w*^{1118} and *kel*^{DE1}*; dbo*$^{\Delta 25.1}$ double mutants, stained for Dsh. (**F**,**G**) Male pupal wings expressing *w*$^{IR-30033}$ (F) and *faf*$^{IR-2956}$ (G) using *MS1096-GAL4*, stained for Fmi. Sets of wings (A-C, D,E and F,G) were dissected, stained and imaged in parallel using the same confocal settings. All images are below vein 4. Scale bar 10 µm. (**A′-G′**) Masks showing the area measured for intensity after thresholding the image (see Materials and methods). (**H-J**) Quantitation of junctional Dsh staining (H,I) or junctional Fmi staining (J) for the above genotypes. Error bars are s.e.m.; ****P*<0.001, ***P*<0.01. (**K**) Quantitation of phosphorylated Dsh relative to unphosphorylated Dsh from western blots of *w*^{1118} and *kel*^{DE1}*; dbo*$^{\Delta 25.1}$ double mutants (see Fig. 5C). Quantitation is from western blots of three biological replicates. Error bars are s.e.m.; **P*=0.02.

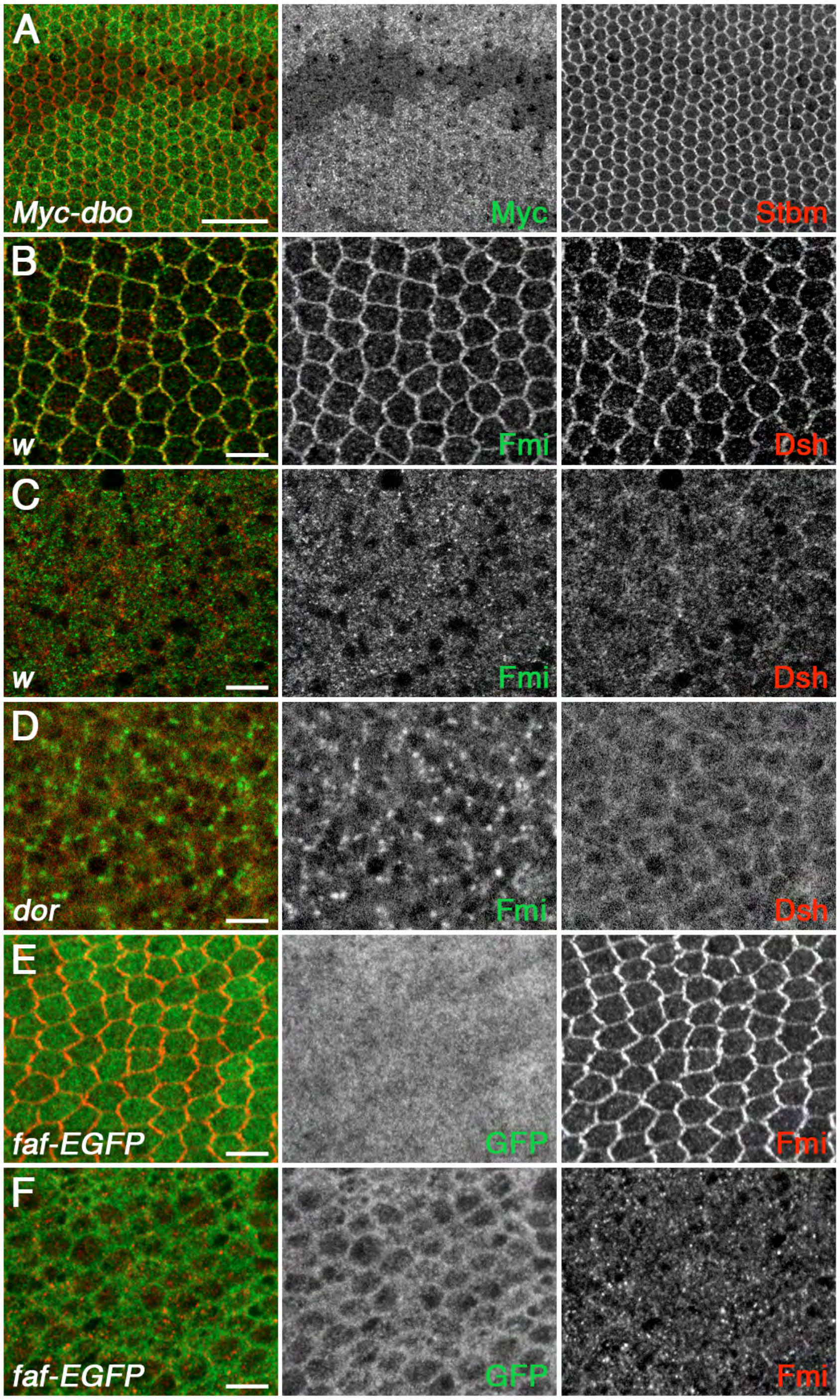

Fig. S7. Localisation of Dbo and Faf in pupal wings. (**A**) *Ubx-FLP; ActP-FRT-polyA-FRT-Myc-dbo/*Δ*2-3* pupal wings stained for Myc (green) and Stbm (red). (**B**,**C**) *w*^{1118} pupal wings stained for Fmi (green) and Dsh (red), with apical (B) and subapical (C) sections. (**D**) *dor*$^{IR\text{-}33733}$*/w; ptc-GAL4/+* pupal wings, stained for Fmi (green) and Dsh (red); subapical section (see Fig. 5L for junctional staining). Image is within the *ptc-GAL4* domain. (**E**,**F**) *Ubx-FLP; ActP-FRT-polyA-FRT-faf-EGFP/+* pupal wings stained for GFP (green) and Fmi (red), with apical (E) and subapical (F) staining. The *faf-EGFP* transgene rescues the *faf* mutant phenotype (not shown). Scale bars: 20 µm in A; 5 µm in B-F.

Table S1. Adult and pupal wing screen of genes encoding predicted E2 ubiquitin-conjugating enzymes

CG number	Gene symbol	RNAi line	***MS1096-GAL4*** **adult wing**	***ptc-GAL4*** **pupal wing**
CG10254	CG10254	GD 15992	wild type	wild type
		NIG 10254R-1	wild type	wild type
		NIG 10254R-2	wild type	wild type
		KK 108657	wild type	-
CG10536	*crbx*	GD 16078	wild type	wild type
		KK 101755	wild type	-
CG10640	*Uev1A*	GD 30890	variable hair orientation[#]	cell packing defect*
		GD 32267	variable hair orientation[#]	cell packing defect*
CG10682	*vih*	GD 27306	wild type	wild type
		KK 107720	vein defect	-
CG10862	CG10862	GD 31372	wild type	wild type
		GD 31373	variable hair orientation	wild type
		KK 101113	no polarity phenotype[‡]	wild type with narrow intervein
CG12799	*Ubc84D*	GD 20260	variable hair orientation	wild type
		NIG 12799R-2	variable hair orientation	-
		KK 106363	vein defect	-
CG14739	CG14739	GD 18358	wild type	wild type
		NIG 14739R-1	wild type	-
		NIG 14739R-2	wild type	-
		KK 105594	wild type	-
CG15437	CG15437	GD 11090	wild type	wild type
CG16894	CG16894	GD 10067	wild type	wild type
		GD 108983	wild type	-
CG17030	CG17030	GD 32827	variable hair orientation	wild type
		GD 32828	variable hair orientation	-
		NIG 17030R-1	variable hair orientation	-
		NIG 17031R-2	wild type	-
		KK 108804	wild type	-
CG18319	*ben*	GD 9413	wings disrupted	cell packing defect*
CG2013	CG2013	GD 23229	lethal	large, irregular cells
		GD 23230	lethal	large, irregular cells
		GD 46927	lethal	large, irregular cells
CG2257	CG2257	GD 33509	wings disrupted	cell packing defect*
		GD 33510	wings disrupted	cell packing defect*
CG2574	CG2574	GD 40173	wild type	wild type
		NIG 2574R-2	wild type	-
		NIG 2574R-3	wild type	-
		KK 105725	wild type	-
CG2924	CG2924	NIG 2924R-3	wild type	-
		KK 104482	wild type	-
CG3018	*lwr*	GD 33684	lethal	cell packing defect*
		GD 33685	lethal	cell packing defect*

CG number	**Gene symbol**	**RNAi line**	***MS1096-GAL4*** **adult wing**	***ptc-GAL4*** **pupal wing**
CG3473	CG3473	GD 26201	wild type	wild type
		NIG 3473R-2	wild type	-
		NIG 3473R-7	wild type	-
		KK 104207	no polarity phenotype[‡]	lethal
CG40045	CG40045	KK 109167	vein defect	-
CG4443	*crl*	GD 34109	variable hair orientation	wild type
		GD 34111	variable hair orientation	wild type
		NIG 4443R-1	variable hair orientation	-
		NIG 4443R-2	variable hair orientation	-
		KK 104440	wild type	-
CG4502	CG4502	GD 34855	wings disrupted	cell packing defect*
		GD 34858	wild type	wild type
CG5440	CG5440	GD 49029	wings disrupted	cell packing defect*
		GD 49030	-	wild type
		NIG 5440R-3	variable hair orientation	-
CG5788	CG5788	GD 27515	wings disrupted	cell packing defect*
		GD 48145	vein defect	cell packing defect*
		GD 48146	wings disrupted	cell packing defect*
CG5823	CG5823	GD 8301	wild type	wild type
		GD 33260	wild type	wild type
		NIG 5823R-1	wild type	-
		NIG 5823R-3	wild type	-
		KK 108292	wild type	-
CG6303	*Bruce*	NIG 6303R-1	wild type	-
		NIG 6303R-2	wild type	-
		KK 107620	ectopic bristles	-
CG6720	*UbcD2*	GD 31158	vein defect, possible swirls below vein 4	wild type
CG7220	CG7220	GD 34198	wild type	wild type
		GD 34199	wild type	wild type
		KK 104478	wild type	-
CG7375	*Ubc12*	GD 35219	wings disrupted	accumulation of Fmi
		GD 35220	wings disrupted	accumulation of Fmi
		NIG 7375R-2	wild type	wild type
		NIG 7375R-3	wings disrupted	accumulation of Fmi
		KK 100761	wings disrupted	accumulation of Fmi, cell morphology abnormal
CG7425	*eff*	GD 26011	late larval lethal	lethal
		NIG 7425R-1	wings disrupted	wild type
		NIG 7425R-2	wings disrupted	wild type
CG7656	CG7656	GD 26880	wild type	wild type
		GD 26881	variable hair orientation	cell packing defect*
		NIG 7656R-2	wild type	-
CG8188	CG8188	NIG 8188R-1	wild type	-
		NIG 8188R-2	wild type	-
		KK 103362	wings curly	wild type
CG8284	CG8284	GD 35872	wild type	wild type
		GD 35873	wild type	wild type
		KK 106600	ectopic bristles	-

CG number	**Gene symbol**	**RNAi line**	***MS1096-GAL4* adult wing**	***ptc-GAL4* pupal wing**
CG9602	CG9602	GD 29498	wild type	wild type
		GD 29499	vein defect	wild type
		NIG 9602R-2	wild type	-
		NIG 9602R-3	wild type	-

GD, GD collection of RNAi lines from VDRC.
KK, KK collection of RNAi lines from VDRC.
NIG, RNAi lines from NIG-FLY.
Wings disrupted: hair polarity could not be assessed as wings could not be mounted flat.
Cell packing defect: cells not organised in the normal hexagonal array.
Irregular cells: cell size and packing disturbed.
#Using ptc-GAL4.
‡Male wings not mountable, hinge defect in female wings.
*Fmi asymmetric in hexagonal cells.

Table S2. Adult and pupal wing screen of genes encoding neddylation pathway components

CG number	**Gene symbol**	**RNAi line**	***MS1096-GAL4*** **adult wing**	***ptc-GAL4*** **pupal wing**
CG10679	*Nedd8*	GD 28444	lethal	lethal
		GD 28445	lethal	lethal
CG13343	*Uba3*	GD 17137	wild type	wild type
		GD 17139	wild type	accumulation of Fmi
		NIG 13343R-4	wings disrupted	accumulation of Fmi
CG7828	*APP-BP1*	GD 7728	wild type	-
		NIG 7828R-2	wings slightly curly	-
		NIG 7828R-3	wild type	-
CG16982	*Roc1a*‡	GD 32399	lethal	accumulation of Fmi
CG16988	*Roc1b*‡	GD 32797	lethal	Cell packing defect, Fmi levels increase slightly?

GD, GD collection of RNAi lines from VDRC.
NIG, RNAi lines from NIG-FLY.
Wings disrupted: hair polarity could not be assessed as wings could not be mounted flat.
Cell packing defect: cells not organised in the normal hexagonal array.
‡Roc proteins are not specific to the neddylation pathway, as they are also subunits of Cullin E3 ligase complexes.

Table S3. Adult and pupal wing screen of genes encoding Cullins

CG number	**Gene symbol**	**RNAi line**	***MS1096-GAL4*** **adult wing**	***ptc-GAL4*** **pupal wing**
CG1877	*lin19/Cul-1*	GD 33406	lethal	wild type
		GD 33407	wild type	embryonic lethal 29°C
		GD 42445	lethal	pupal lethal 18°C
CG1512	*Cul-2*	GD 19297	wings disrupted	cell packing defect*
		GD 19298	wings disrupted	wild type
CG42616	*Cul-3*	GD 16331	wings disrupted	accumulation of Fmi
		NIG 11861R-1	wings disrupted	accumulation of Fmi
		NIG 11861R-2	wings disrupted	accumulation of Fmi
		KK 109415	wings disrupted	accumulation of Fmi
CG8711	*Cul-4*	NIG 8711R-1	wings disrupted	wild type
CG1401	*Cul-5*	GD 52175	wild type	wild type
		GD 52176	wild type	wid type
		KK 108817	lethal	wild type

GD, GD collection of RNAi lines from VDRC.
KK, KK collection of RNAi lines from VDRC.
NIG, RNAi lines from NIG-FLY.
Wings disrupted: hair polarity could not be assessed as wings could not be mounted flat.
Cell packing defect: cells not organised in the normal hexagonal array.
*Fmi asymmetric in hexagonal cells.

Table S4. Adult and pupal wing screen of genes encoding BTB domain proteins

CG number	**Gene symbol**	**RNAi line**	***MS1096-GAL4*** **adult wing**	***ptc-GAL4*** **pupal wing**
CG10465	CG10465	KK 107131	variable hair orientation	wild type
CG10752	CG10752	GD 30007	wild type	-
		NIG 10752R-1	wild type	-
		NIG 10752R-3	wild type	-
		KK 106721	wings disrupted	wild type
CG10801	CG10801	GD 45748	wild type	-
		GD 45749	wild type	-
		KK 100397	wings slightly disrupted, possible swirl	wild type
CG11275	CG11275	NIG 11275R-1	wild type	-
		NIG 11275R-2	wild type	-
CG11491	*br*	GD 13705	lethal	disrupted
		GD 38526	wings disrupted	wild type
		NIG 11514R-1	wild type	-
CG11714	CG11714	GD 31116	wild type	-
		GD 31117	wild type	-
		GD 50558	wild type	-
		GD 50559	wild type	-
CG12052	*lola*	GD 12573	wild type	-
		GD 20634	wild type	wild type
		GD 21057	wild type	-
		GD 25333	wings disrupted	wild type
		GD 41415	lethal	large cells
CG12236	CG12236	NIG 12236R-1	disrupted	large cells
CG12423	*klhl10*	GD 35721	wild type	-
		GD 35722	wild type	-
		KK 109083	wings disrupted	wild type
CG12537	*rdx*	GD 28798	wings disrupted	wild type
		GD 28800	wings disrupted	wild type
		GD 45759	wild type	-
		GD 45761	wings disrupted	wild type
CG12692	CG12692	GD 17292	wild type	-
		KK 109683	wild type	-
CG12857	CG12857	GD 45029	wild type	-
		GD 45030	wild type	-
		NIG 12857R-2	wild type	-
		NIG 12857R-3	wild type	-
		KK 101639	wild type	-
CG13917	CG13917	GD 32082	wings disrupted	cell packing defect
CG14260	CG14260	GD 50132	wild type	-
		NIG 14260R-4	wild type	-
CG14262	CG14262	GD 24855	wild type	-
		GD 24856	wild type	-
CG14307	*fru*	KK 105005	wings slightly disrupted, possible swirl, multiple wing hairs	wild type
CG14785	CG14785	GD 43594	wings disrupted	wild type

CG number	**Gene symbol**	**RNAi line**	***MS1096-GAL4* adult wing**	***ptc-GAL4* pupal wing**
CG15097	CG15097	KK 109428	wings slightly disrupted, possible swirl	slightly large cells
CG15725	CG15725	GD 38868	wild type	-
		KK 103687	wild type	-
		KK 106953	wild type	-
CG15812	*pfk*	GD 13361	wings disrupted	wild type
		GD 14020	wings disrupted	wild type
CG16778	CG16778	GD 10739	wild type	-
CG16952	CG16952	GD 10149	wild type	-
		GD 19166	wild type	-
CG17068	CG17068	GD 32849	wild type	-
		GD 32850	wild type	-
		NIG 17068R-1	wild type	-
		NIG 17068R-4	wild type	-
CG17754	CG17754	GD 47274	weak multiple wing hairs	wild type
		GD 47275	wild type	-
		GD 47276	wild type	-
		GD 47277	wild type	-
		KK 104337	wings disrupted	wild type
CG1812	CG1812	GD 15491	wild type	-
CG1826	CG1826	GD 33049	wild type	-
CG18471	*gprs*	GD 41673	wild type	-
CG1856	*ttk*	KK 10855	wild type	-
CG2368	*psq*	GD 30591	wild type	-
		KK 106404	wings disrupted	lethal
CG30357	CG30357	GD 21144	wild type	-
		KK 108708	wild type	-
CG31666	*chinmo*	NIG 17156R-1	vein defects	wild type
		NIG 17156R-2	variable hair orientation	-
CG32121	CG32121	GD 34404	wild type	-
		GD 34405	wings disrupted	cell packing defect
CG32491	*mod (mdg4)*	GD 52268	lethal	cell packing defect
		NIG 15802R-1	lethal	cell packing defect
		NIG 15802R-3	wild type	-
		NIG 32491R-1	wings disrupted	wild type
CG33261	*Trl*	GD 41095	wings disrupted	cell packing defect*
		NIG 9343R-1	hair morphology defect	wild type
		NIG 9343R-2	wings curly	slightly large cells
		KK 106433	wings disrupted	cell packing defect*
CG33291	CG33291	GD 35501	wild type	-
		GD 35503	wild type	-
		KK 107170	ectopic bristles	-

CG number	**Gene symbol**	**RNAi line**	***MS1096-GAL4* adult wing**	***ptc-GAL4* pupal wing**
CG34346	*mamo*	GD 29874	wild type	wild type
		GD 29875	wild type	wild type
		GD 38223	wings disrupted	wild type
		GD 45349	wings disrupted	cell packing defect
		GD 48679	wild type	-
		GD 48680	wild type	-
		NIG 11082R-2	wild type	-
		NIG 11082R-3	wild type	-
CG34376	CG34376	GD 24804	wild type	-
		GD 24805	wings disrupted	wild type
		GD 25038	wild type	-
		NIG 31160R-1	wild type	-
		NIG 31160R-3	wings disrupted	wild type
		KK 102795	wings disrupted	wild type
CG3571	*KLHL18*	GD 12052	wild type	-
		GD 43777	wild type	-
		GD 43778	wild type	-
CG3711	CG3711	GD 11164	wild type	-
		GD 11166	wild type	-
		NIG 3711R-2	wild type	-
		NIG 3711R-3	wild type	-
CG3726	CG3726	GD 41090	wild type	-
		GD 41091	wild type	-
CG3962	*Keap1*	KK 107052	wings curly	wild type
CG4069	CG4069	GD 21783	wings disrupted	wild type
CG41099	CG41099	GD 32927	wild type	-
		GD 32928	wild type	-
		GD 40076	wild type	-
CG43226	*lute*	GD 34986	wild type	-
		GD 34987	wild type	-
		NIG 5319R-3	wild type	wild type
		NIG 5319R-4	wild type	-
		KK 108082	disrupted	slightly large cells
CG43365	*BtbVII*	KK 106063	wings disrupted	cell packing defect
CG4807	*ab*	GD 41005	lethal	cell packing defect
CG5186	*slim*	GD 15185	wild type	-
		NIG 5186R-1	wild type	-
		NIG 5186R-2	wings disrupted	lethal
CG5575	*ken*	GD 48596	lethal	disrupted
CG5701	*RhoBTB*	KK 100815	wings disrupted	wild type
CG5738	*lolal*	GD 9571	lethal	lethal
		GD 9572	lethal	cell packing defect
		GD 9573	wings disrupted	cell packing defect
CG6118	CG6118	GD 38061	lethal	wild type
		GD 38062	lethal	wild type

CG number	**Gene symbol**	**RNAi line**	***MS1096-GAL4*** **adult wing**	***ptc-GAL4*** **pupal wing**
CG6224	*dbo*	GD 22476	wild type	wild type[‡]
		NIG 6224R-1	variable hair orientation	wild type[‡]
		NIG 6224R-3	variable hair orientation	wild type[‡]
		KK 105407	wings disrupted	slight accumulation of Fmi?[‡]
G6384	*Cp190*	GD 35077	wild type	-
		GD 35078	wings disrupted	wild type
		NIG 6384R-1	wild type	-
CG6758	CG6758	GD 43606	wild type	-
CG6765	CG6765	GD 22424	wild type	-
		NIG 6765R-1	wild type	-
		NIG 6765R-2	wild type	-
		KK 100851	wings slightly disrupted	wild type
CG6792	CG6792	GD 35118	vein defects	wild type
		GD 35119	wild type	wild type
		NIG 6792R-2	wild type	-
		NIG 6792R-3	wild type	-
CG7058	CG7058	GD 38230	wild type	-
		GD 38231	wild type	-
		NIG 7058R-1	wild type	-
		NIG 7058R-3	weak multiple wing hairs	-
CG7102	CG7102	GD 20571	wild type	-
		KK 107887	wings disrupted	wild type
CG7210	*kel*	KK 105397	wings disrupted	slight accumulation of Fmi?
CG7230	*rib*	GD 15900	wild type	-
		NIG 7230R-2	wild type	-
		NIG 7230R-4	wild type	wild type
		KK 103977	wild type	-
CG7837	CG7837	GD 22573	wild type	-
		GD 23099	wild type	-
		KK 109784	ectopic bristles	-
CG8060	CG8060	GD 22684	wild type	-
		GD 46990	wild type	-
		GD 46991	wild type	-
		NIG 8060R-1	wings disrupted	wild type
		NIG 8069R-2	wild type	-
CG8260	CG8260	GD 45257	wings disrupted	wild type
CG8411	*gcl*	GD 28896	wild type	-
		GD 28897	wild type	-
		NIG 8411R-2	wild type	-
		NIG 8411R-3	wild type	-
CG8811	*muskelin*	GD 29774	wild type	-
		KK 107043	wings disrupted	lethal
CG8924	CG8924	GD 48041	wild type	-
		GD 48042	wild type[#]	wild type
		NIG 8924R-1	wild type	-
		NIG 8924R-2	wild type	-

CG number	**Gene symbol**	**RNAi line**	***MS1096-GAL4*** **adult wing**	***ptc-GAL4*** **pupal wing**
CG9097	*bab1*	GD 6960	lethal	cell packing defect
		GD 6961	lethal	cell packing defect
		GD 24496	wild type	-
		GD 50285	wild type	-
		GD 50286	wild type	-
CG9102	*bab2*	GD 8943	wild type	-
		GD 49042	wild type	wild type
		GD 49043	wild type	-
CG9426	CG9426	GD 10843	wild type	-
		NIG 9426R-2	wild type	-
CG9467	CG9467	GD 45806	ectopic bristles	wild type
		GD 45807	wild type	-
CG9970	CG9970	GD 45817	wild type	-

GD, GD collection of RNAi lines from VDRC.
KK, KK collection of RNAi lines from VDRC.
NIG, RNAi lines from NIG-FLY.
Wings disrupted: hair polarity could not be assessed as wings could not be mounted flat.
Cell packing defect: cells not organised in the normal hexagonal array.
Irregular cells: cell size and packing disturbed.
[#]Using *ptc-GAL4*.
*Fmi asymmetric in hexagonal cells.
[‡]Strong accumulation of Fmi in combination with $kel^{IR\text{-}JF01768}$. Note that the hairpins in the existing RNAi lines targeting *dbo* all overlap.

Table S5. Summary of epistasis experiments

	Ubc12$^{IR-30033}$	*Cul-3*$^{IR-109415}$	*faf*$^{IR-2956}$
wt	Fmi Fz Stbm Pk Dsh strong increase	Fmi Fz Stbm Pk Dsh strong increase	Fmi Fz Stbm Pk Dsh strong decrease
fz^{P21}	Fmi Stbm Pk slight increase	Fmi Stbm Pk wild type levels	Fmi Stbm Pk strong decrease
stbm^{6}	Fmi Fz Dsh slight increase	Fmi Fz Dsh slight increase	Fmi Fz Dsh strong decrease
pk$^{pk-sple13}$	Fmi Fz Stbm Dsh strong increase	Fmi Fz Stbm Dsh strong increase	Fmi Fz Stbm Dsh strong decrease
dgo^{380}	Fmi Fz Stbm Dsh strong increase	Fmi Fz Stbm Dsh strong increase	Fmi Fz Stbm Dsh strong decrease
dsh^{V26}	Fmi (wt) Stbm (wt) wild type levels*	Fmi Stbm wild type levels	Fmi Stbm strong decrease

ptc-GAL4 was used to express RNAi against *Ubc12*, *Cul-3* or *faf* in the various mutant backgrounds, and the levels of junctional proteins were assessed relative to non-RNAi-expressing tissue within the same wing. The table shows all the core proteins that were tested for any particular genotype. If a protein is not listed, this indicates that its junctional levels were not analysed in this genotype.
*Distal *dsh* clones within the *Ubc12* RNAi-expressing domain completely suppress the accumulation of core proteins at junctions caused by the RNAi, as the RNAi expression is less well expressed distally. Proximal *dsh* clones (in regions where the *Ubc12* RNAi is strongly expressed) do not completely suppress the accumulation phenotype. This argues that *Ubc12* has a second target that affects core protein accumulation at junctions.