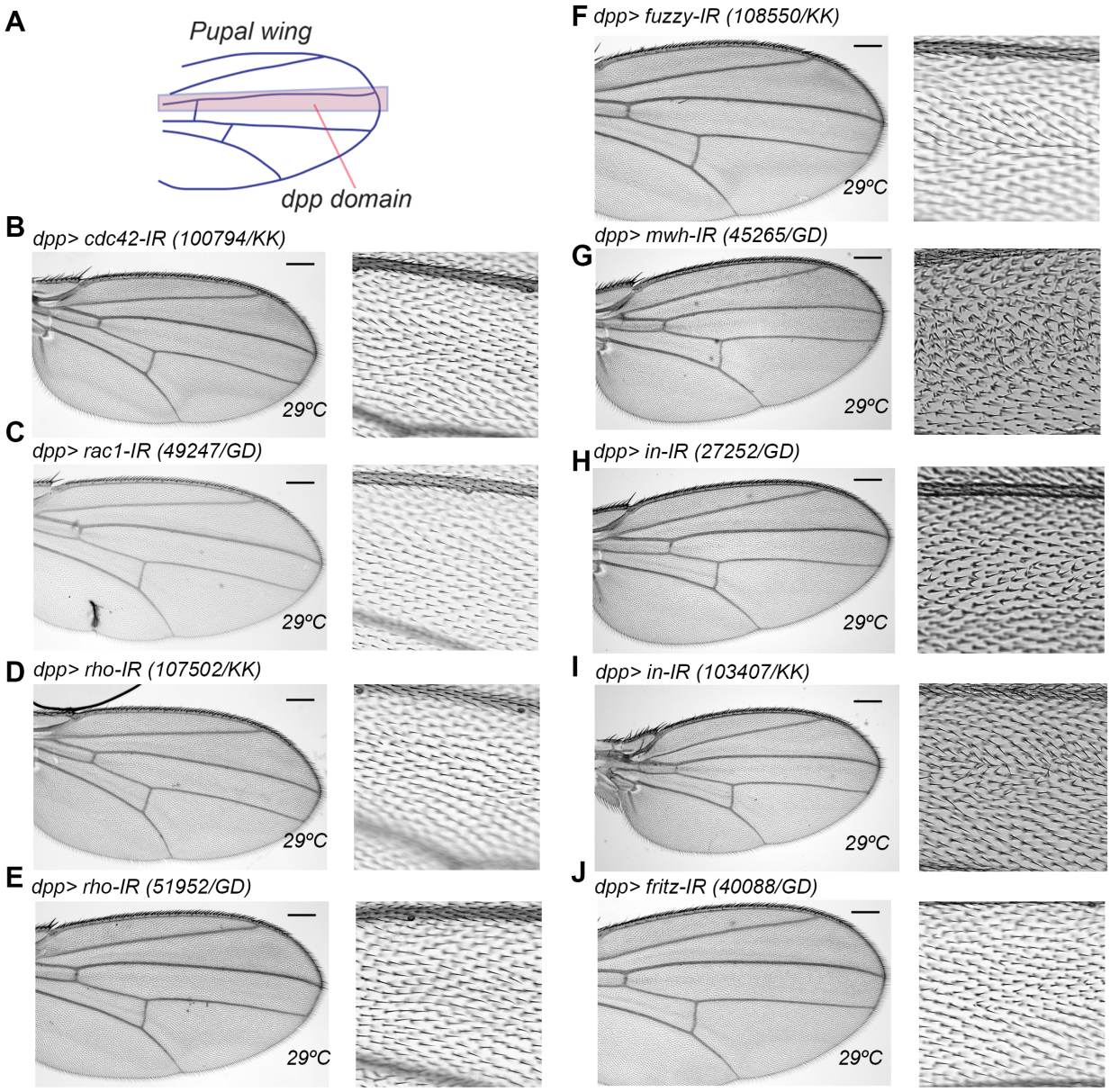

Figure S1. Adult wing phenotypes observed in different PPE RNAi lines.
(**A**) Illustration of a pupal wing highlighting the dpp (*decapentaplegic*) domain. (**B-F**) Adult wing aged 29°C using *dpp>gal4* for RNAi expression against actin regulators that do not generate robust multi hairs cell phenotypes under these conditions (except *mwh* RNAi lines panel **G**). Mhc phenotype caused by *Inturned* RNAi lines (**H,I**) was very mild and it was not used for further analyses. (**G**)The RNAi line 45265/GD against *mwh* showed mhc phenotypes up to the level of the RNAi lines use in Figure 1. Scale bars represent 250μm.

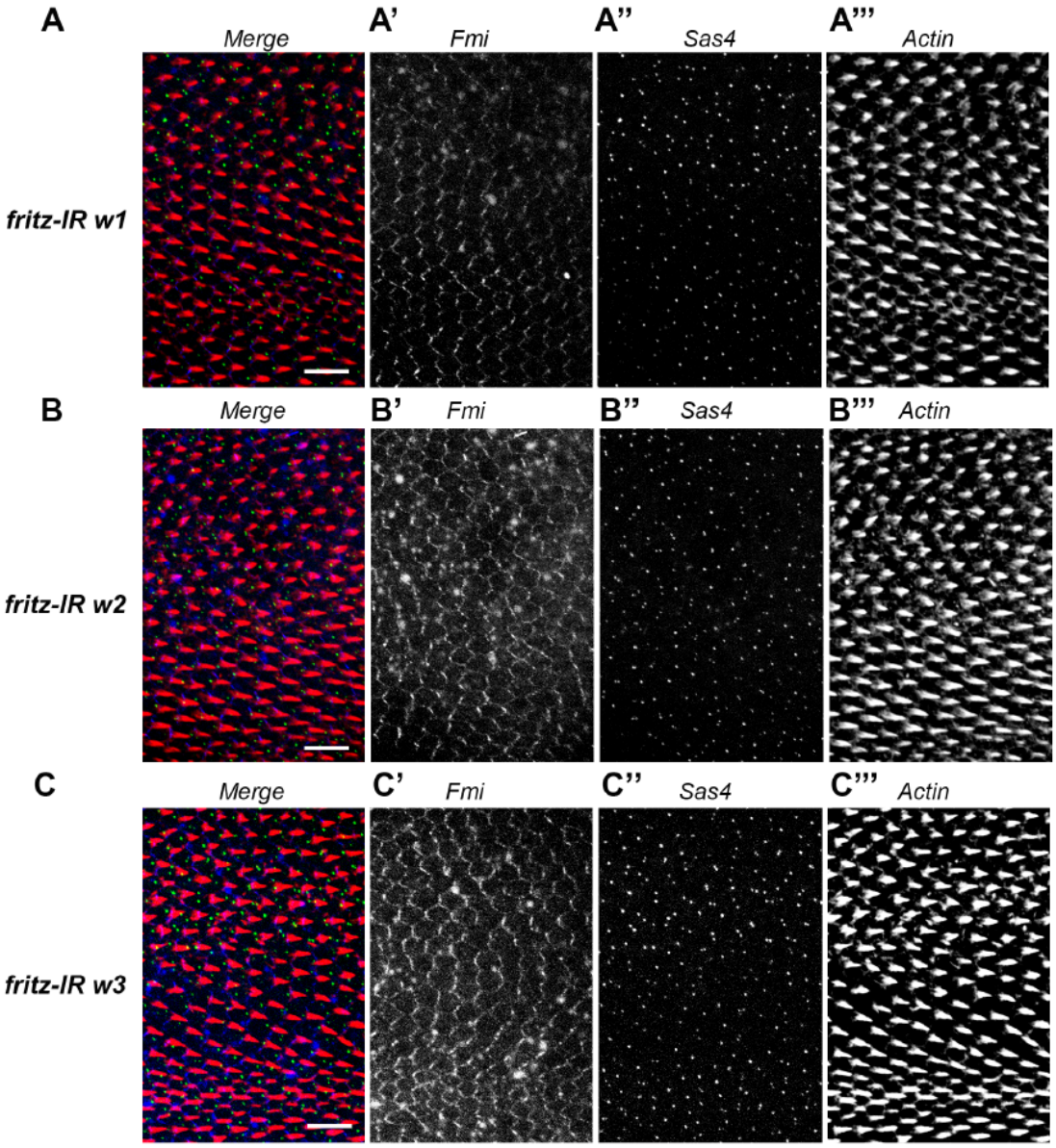

Figure S2. *frtz*I RNAi causes a delayed hair formation.
(**A-C**) Confocal images from three different pupal wings using *dpp* as a driver to express RNAi against *frtz* and *sas4* couple to GFP (green in **A, B, C** and monochrome in **A'', B''** and **C''**) and labeled with *Fmi* (blue in **A, B, C** and monochrome in **A', B'** and **C'**) and actin (*phalloidin*) (red in **A, B, C**, monochrome in **A''', B'''** and **C'''**). Scale bars represent 10μm.

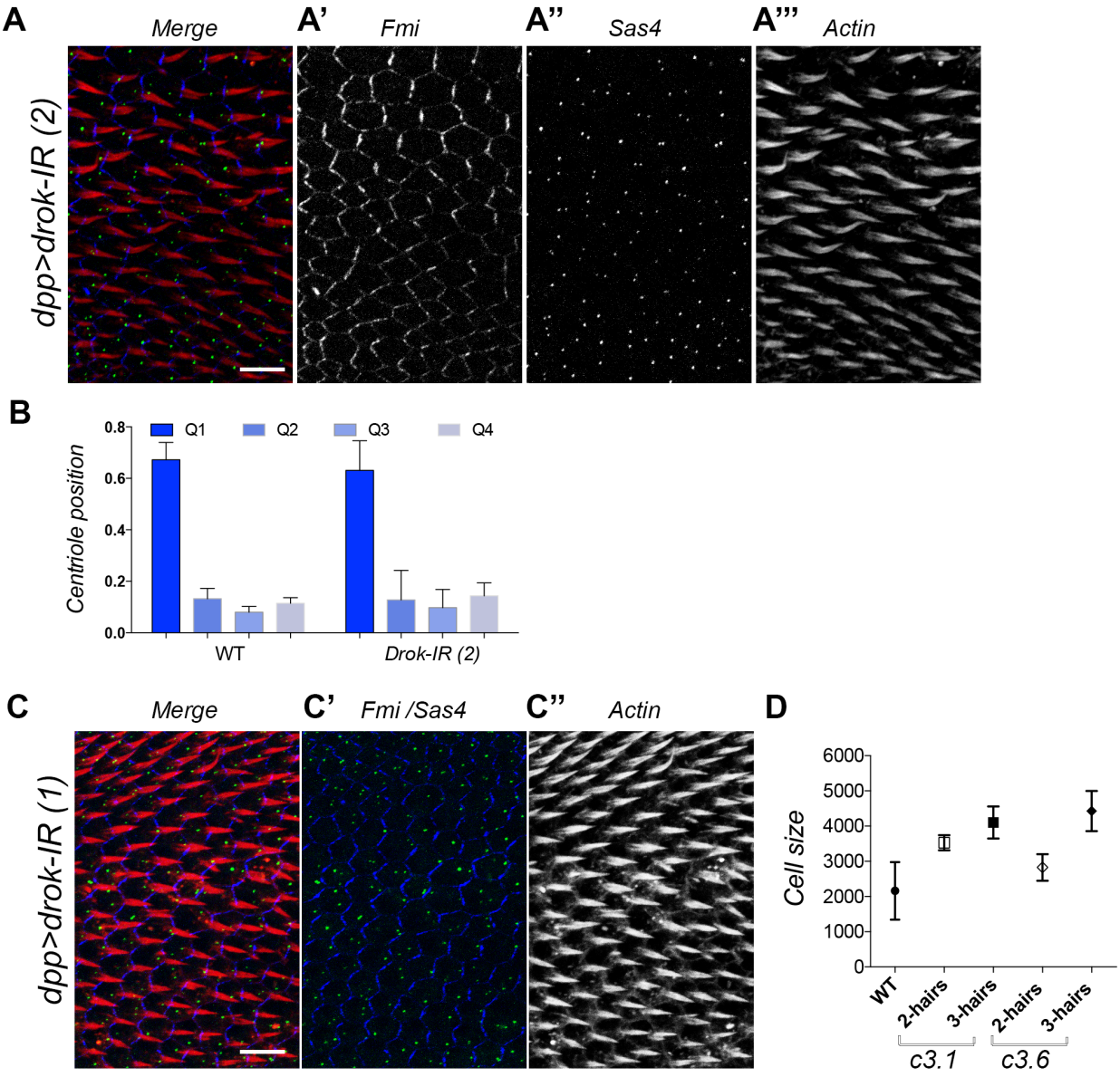

Figure S3. Mhc phenotypes and increased cell size observed in *Droki IR lines* does not affect centrioles polarization.

(**A**) Pupal wing showing mhc and using *dpp* as a driver to express *drok-IR (2)* (VDRC line 3793/GD) and expressing *sas4* coupled to GFP (green in **A** and monochrome in **A**'') and labeled with f*mi* (blue in **A** and monochrome in **A'**) and actin (*phalloidin*) (red in **A** and monochrome in **A'''**). (**B**) Centriole Q position analyses in *drok-IR (2)* (VDRC line 3793/GD). (**C**) Multiple hair cells generated by *drok-IR (1)* (VDRC line 104675/KK). *Sas4* couple to GFP (green in **e** and e'), labeled with *fmi* (blue in **C** and **C'**) and actin (*phalloidin*) (red in **C** and monochrome in **C''**). (**D**) Cell size quantification in *dpp>drok-IR* lines confirming an increased cell size in cells with two or three-hairs cells when compare to WT areas of the same wings. Scale bars represent 10μm.

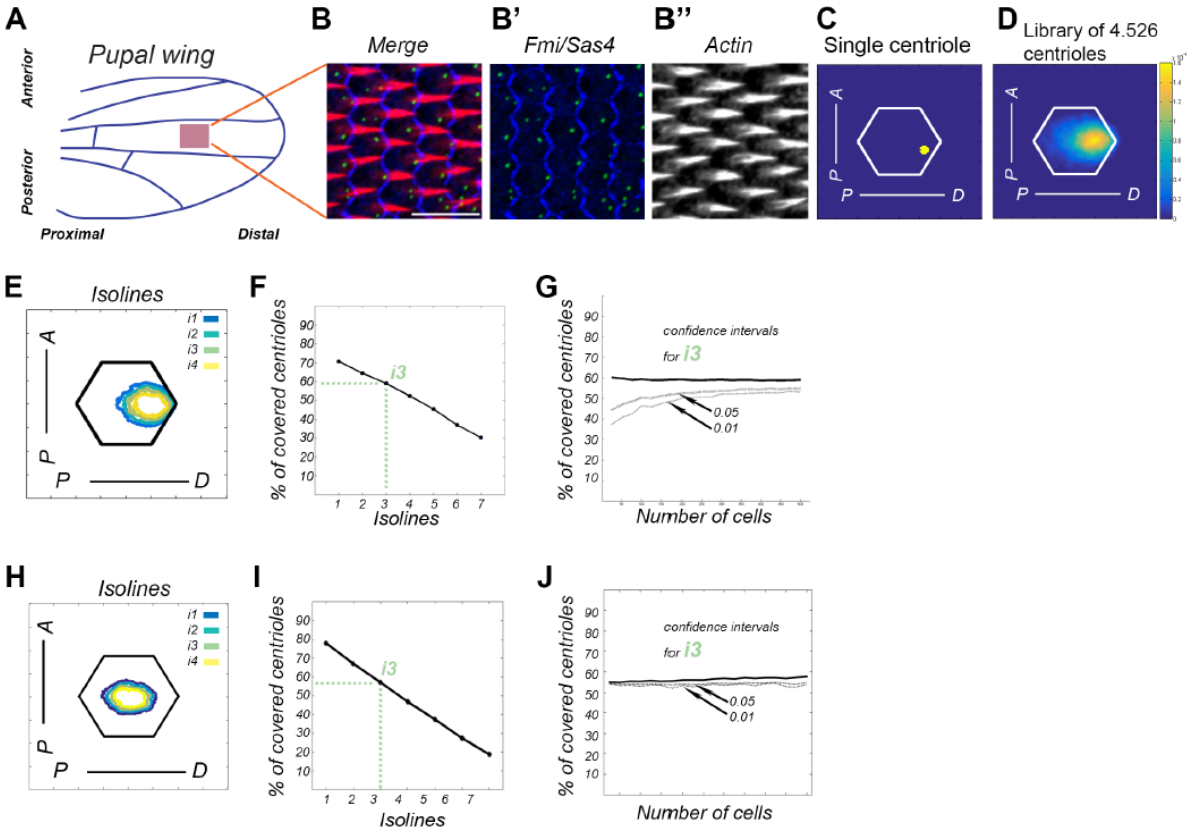

Figure S4. Representative polarized distribution of centrioles (RPCD) is a complementary imaging tool to measure and compare centrioles distribution in twol dimensions.

(**A**) Illustration of a pupal wing and the orientation used along this study. (**BI B''**) Pupal wing expressing Sas4 couple to green fluorescent protein (GFP) (in green) and labeled with Fmi in blue and actin (phalloidin) in red (**B**) and monochrome in **B''**. Scale bar represents 10μm. (**C**) Single centriole position in a cell. (**D**) Density map for centrioles localization generated out of the library with more than 4,000 centrioles in WT pupal wings aged for 28.5 hours at 29° C. (**E**) Representation of 4 isolines delimiting different areas of the centrioles library density map. (**F**) Percentage of centrioles covered (70% to 30% of centrioles) from the centrioles library for 7 isolines i1 to i7). (**G**) Confidence interval calculation for i3 at 0.05 or 0.01 threshold for a number of cells ranging from 25 to 500. (**H**) Representation of 4 isolines delimiting different areas of the centrioles library density map for Representative Nonn polarized distribution of centrioles (RNCD**)**. (**I**) Percentage of centrioles covered (80% to 20% of centrioles) from the centrioles library for 7 isolines i1 to i7). (**J**) Confidence interval calculation for i3 at 0.05 or 0.01 threshold for a number of cells ranging from 25 to 500.

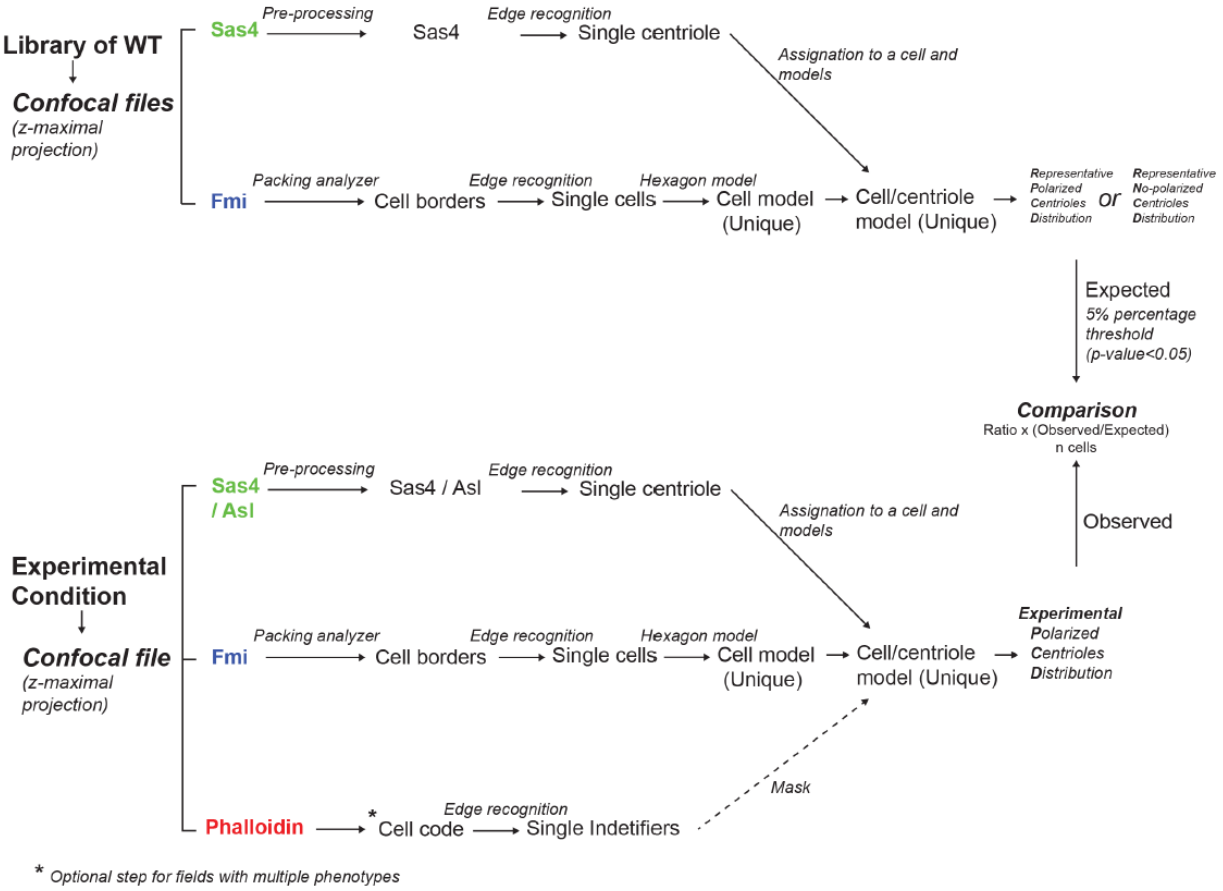

Figure S5. Diagram of the workflow followed to compare Representative Polarized Centrioles Distribution (RPCD) or Representative NonI polarized Centrioles Distribution (RNCD) with centriole distributions in experimental conditions in two-dimensions

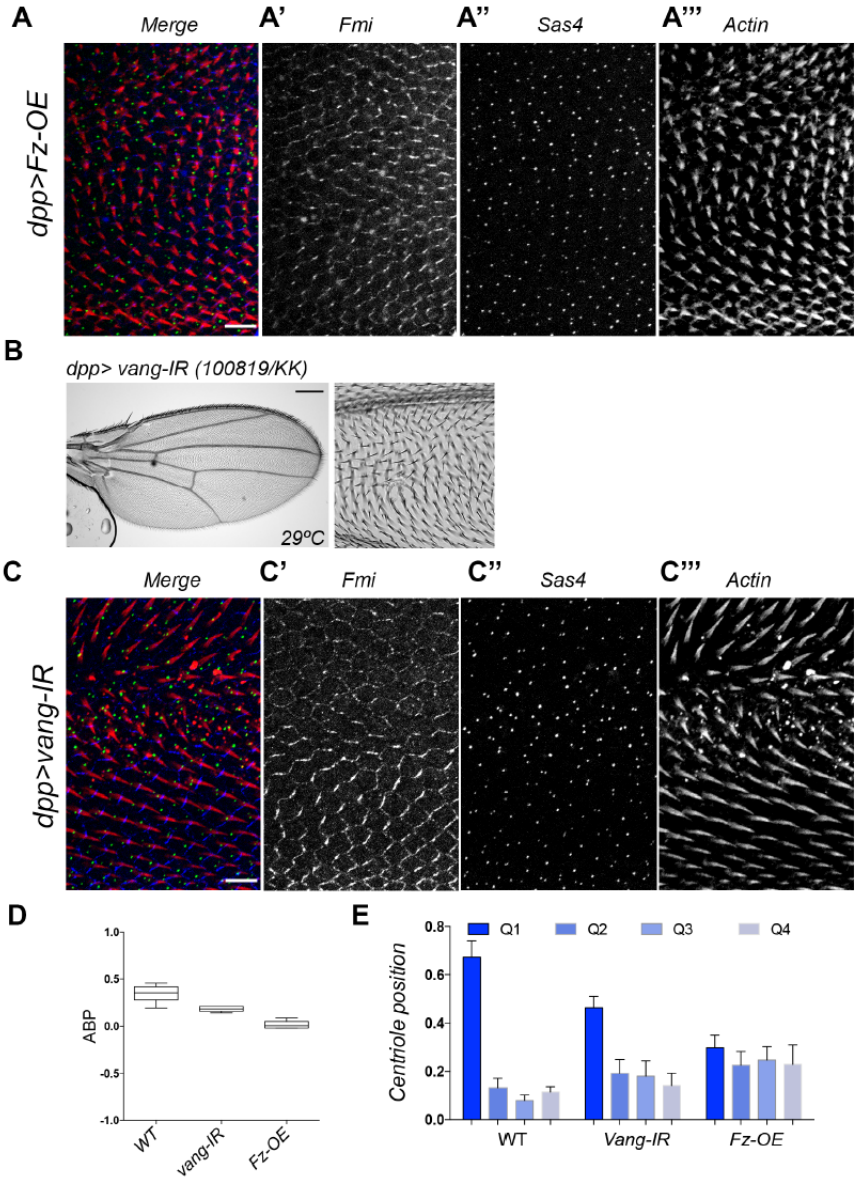

Figure S6. Centrioles polarization is impaired in *Fzi OE* and *vangi IR* conditions. (**A**) Confocal image showing a hair misn orientation phenotype in pupal wing using *dpp* as a driver to overexpress *frizzled* and expressing *sas4* coupled to GFP (green in a and monochrome in a'') and labeled with *Fmi* (blue in **A**, monochrome in **A'**) and actin (*phalloidin*) (red in **A** and monochrome in **A'''**). Scale bars represent 10μm. (**B**) Adult wing aged at 29°C using *dpp>gal4* and vang-IR to suppress v*ang* expression*. dpp>vang-IR* generates mis-orientation of wing hairs. Scale bars represent 250μm. (**C**) Hair orientation disruption is also present in *vang-iR* pupal wings. *Sas4* couple to GFP (green in **C** and monochrome in **C''**), labeled with *fmi* (blue in **C**, monochrome in **C'**) and actin (*phalloidin*) (red in **C** and monochrome in **C'''**). (**D-E**) Centriole polarity analyses using ABP and Q methods in *dpp>vang-IR* and Fz-OE wings. Scale bars represent 10μm. Statistical analyses among experimental groups: ANOVA (see values in Supplementary Table III).

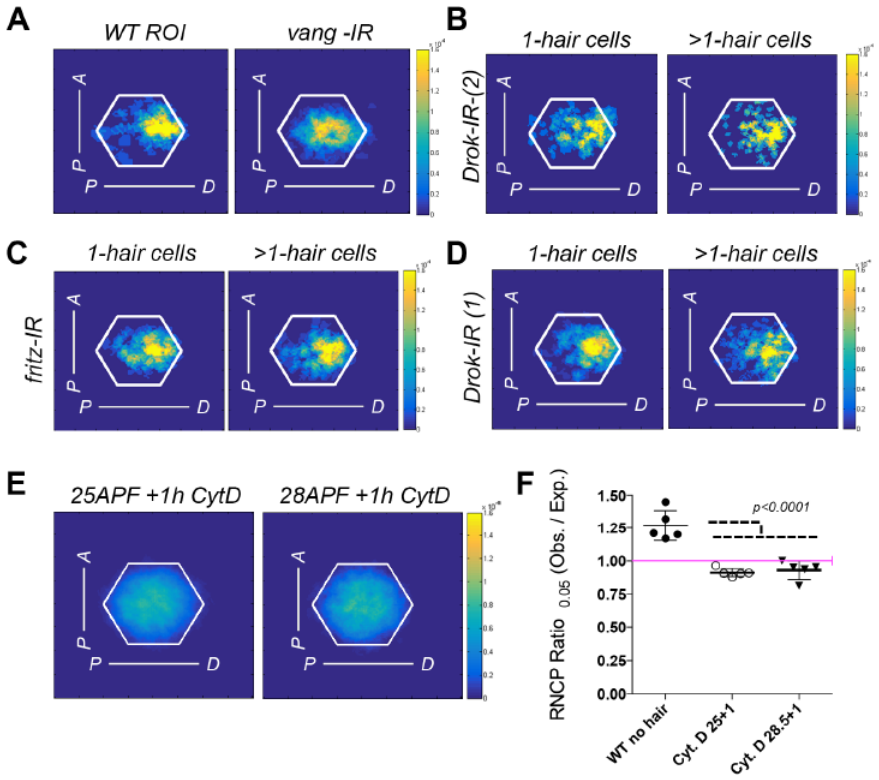

Figure S7. Density maps for centriole distribution analyses in different experimental conditions.

(**A-D**) Centriole density maps for paired WT and different PCP LOF conditions. (E) Centriole density maps for wings treated with Cytochalasin D *in vitro* for one an hour in wing aged for 25APF (before hair formation) or 28.5 APF (after hair formation).

Table S1. Statistical analysis of ABP and Q1 centrioles analyses using ANOVA followed by Bonferroni test for multiple comparisons.

	WT	***fritz-IR***	***mwh-IR***	***Drok (1)***
ABP				
Mean	0.36	0.25	0.35	0.26
Std	0.09	0.05	0.08	0.09
Q1				
Mean	0.67	0.46	0.68	0.59
Std	0.07	0.05	0.12	0.09
	WT	***fritz-IR***	***mwh-IR***	***Drok (1)***
Q1	***ANOVA***	***p=0.0002***		
Bonferroni's test				
WT		p<0.01	n.s.	n.s.
fritz-IR			p<0.001	n.s.
mwh-IR				n.s.
ABP	***ANOVA***	***p>0.05***		

Table S2. Statistical analysis of ABP and Q1 centrioles analyses using ANOVA follow by Bonferroni test for multiple comparisons.

	25 (WT)	***28.5 (WT)***	***25+10Mock***	***25+10 CytD***
ABP				
Mean	0.02	0.35	0.17	0.01
Std	0.02	0.09	0.05	0.08
Q1				
Mean	0.33	0.71	0.45	0.30
Std	0.02	0.06	0.06	0.08
	25 (WT)	***28.5 (WT)***	***25+10Mock***	***25+10 CytD***
Q1	***ANOVA***	***p<0.0001***		
Bonferroni's test				
25 (WT)		p<0.0001	p<0.01	n.s.
28.5 (WT)			p<0.0001	p<0.0001
25+10Mock				p<0.01
	25 (WT)	***28.5 (WT)***	***25+10Mock***	***25+10 CytD***
ABP	***ANOVA***	***p<0.0001***		
Bonferroni's test				
25 (WT)		p<0.0001	p<0.01	n.s.
28.5 (WT)			p<0.001	p<0.0001
25+10Mock				p<0.01

Table S3. Statistical analysis of ABP and Q1 centrioles analyses using ANOVA follow by Bonferroni test for multiple comparisons.

	WT	***vang-IR***	***Fz-OE***
ABP			
Mean	0.35	0.18	0.02
Std	0.09	0.03	0.04
Q1			
Mean	0.67	0.46	0.30
Std	0.07	0.05	0.05

	WT	***vang-IR***	***Fz-OE***
Q1	***ANOVA***	***p<0.0001***	
Bonferroni's test			
WT		p<0.0001	p<0.0001
vang-IR			p<0.0001

	WT	***vang-IR***	***Fz-OE***
ABP	***ANOVA***	***p<0.0001***	
Bonferroni's test			
WT		p<0.001	p<0.0001
vang-IR			p<0.001