Li et al., http://www.jcb.org/cgi/content/full/jcb.201203150/DC1

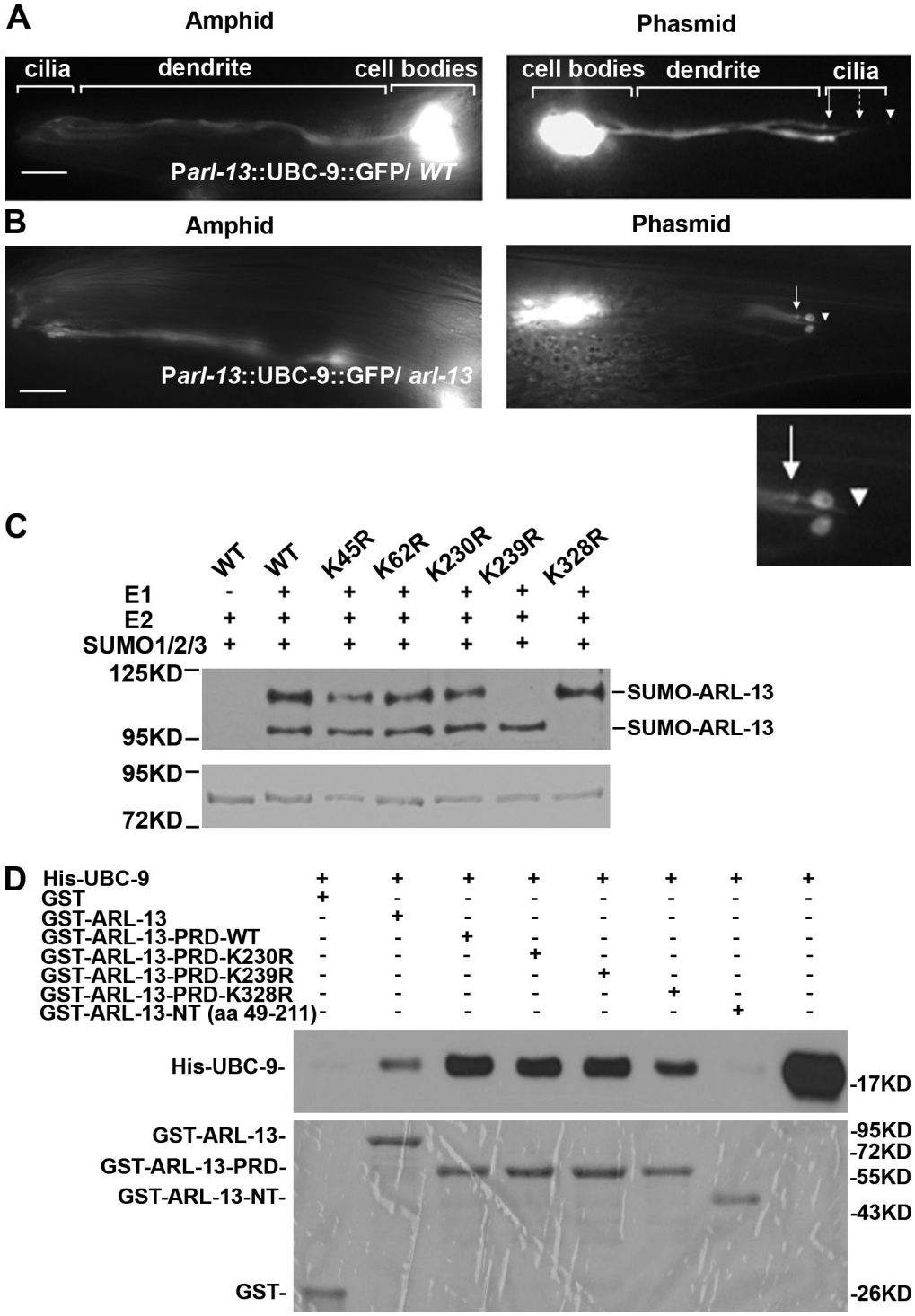

Figure S1. **The ciliary targeting of UBC-9 is independent of ARL-13.** (A and B) Localization of UBC-9::GFP in wild-type and *arl-13* mutants. Note that UBC-9 accumulates in the abnormal bulges in *arl-13* cilia. Arrows with solid lines, ciliary base; arrow with broken line, middle-distal junction; arrowheads, ciliary tip. Bar, 10 µm. (C) In vitro SUMOylation assay. Anti-SUMO antibody was used to detect SUMOylated ARL-13. (D) SUMOylation-deficient ARL-13 can still bind to UBC-9 in a GST pull-down assay.

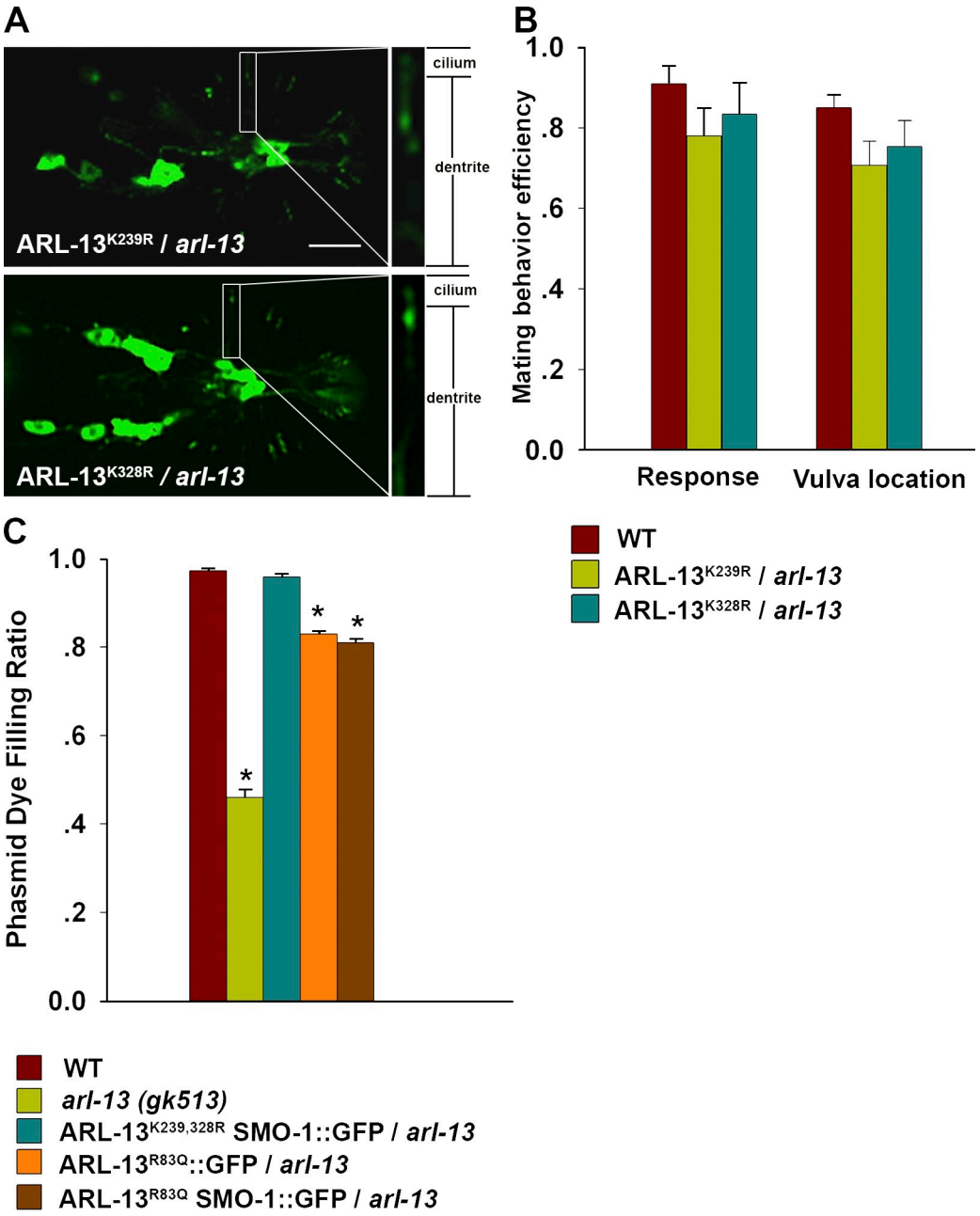

Figure S2. **The effects of SUMOylation on worm ARL-13.** (A) The ciliary localization of PKD-2::GFP is mostly rescued by reintroducing either ARL-13^{K239R} or ARL-13^{K328R} in *arl-13* mutant. Bar, 10 µm. (B) The mating defects are mostly rescued in ARL-13^{K239R}/*arl-13* and ARL-13^{K328R}/*arl-13* worms. (C) A dye-filling assay was used to examine the ciliogenesis. The data were analyzed using an unpaired Student's *t* test and are presented as mean ± SEM (error bars); *, $P < 0.01$.

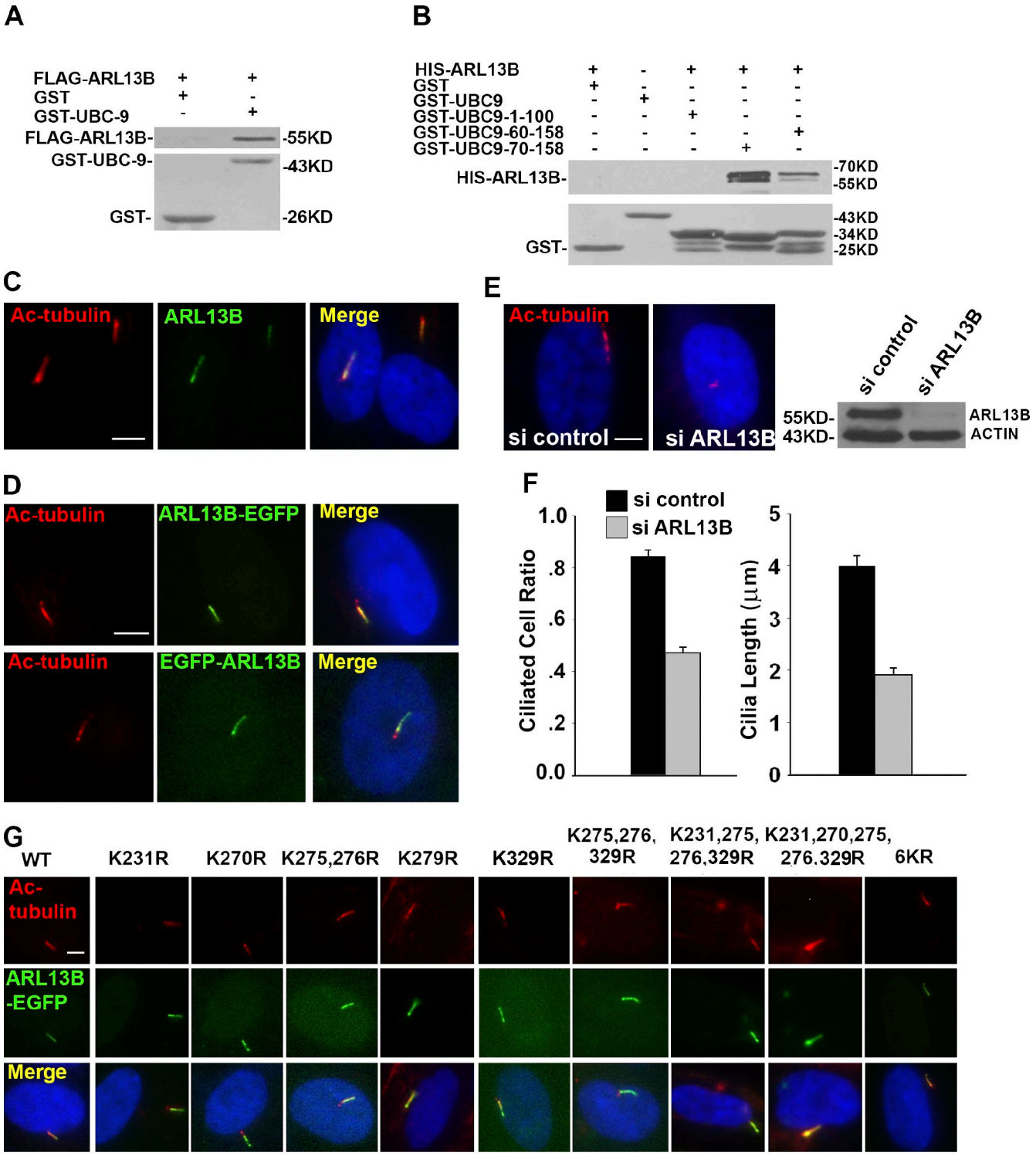

Figure S3. **The effect of SUMOylation on human ARL13B.** (A) GST-UBC-9 can pull down FLAG-tagged ARL13B overexpressed in HEK293 cells. (top) Western blot with anti-FLAG antibody. (bottom) Ponceau S staining. (B) Mapping down the ARL13B interaction region in UBC-9. (C) Endogenous ARL13B localizes specifically to cilia in hTERT-RPE1 cells. (D) EGFP-tagged ARL13B present on cilia in hTERT-RPE1 cells. (E) hTERT-RPE1 cells were treated with siRNAs, and down-regulation of ARL13B was detected by Western blotting. (F) Ciliated cell ratio and mean cilia length were measured. The data were analyzed using an unpaired Student's *t* test and presented as mean ± SEM (error bars). $P < 0.01$. (G) A set of EGFP-tagged ARL13B variants were transfected into hTERT-RPE1 cells. All ARL13B variants show normal ciliary localization. Bars: (C) 4 μm; (D, E, and G) 2 μm.

Table S1. **Strains used in this study**

Strain name	Genotype
ZP404	*pha-1(e2123); him-5(e1490); jhuEx [Parl-13::UBC-9::GFP; pBX]*
ZP444	*arl-13(gk513); pha-1(e2123); him-5(e1490); jhuEx [Parl-13::UBC-9::GFP; pBX]*
ZP449	*N2; jhuEx [Parl-13::UBC-9::GFP; pRF4]*
JH145	*arl-13(gk513)*
ZP406	*pha-1(e2123); him-5(e1490); jhuEx [Parl-13::ARL-13^{K239R}::GFP; pBX]]*
ZP352	*arl-13(gk513); pha-1(e2123); him-5(e1490); jhuEx [Parl-13::ARL-13::GFP; pBX]*
ZP800	*arl-13(gk513); pha-1(e2123); him-5(e1490); jhuEx [Parl-13::ARL-13^{K239R}::GFP; pBX]*
ZP801	*arl-13(gk513); pha-1(e2123); him-5(e1490); jhuEx [Parl-13::ARL-13^{K328R}::GFP; pBX]*
ZP798	*arl-13(gk513); pha-1(e2123); him-5(e1490); jhuEx [Parl-13::ARL-13^{K239,328R}::GFP; pBX]*
ZP799	*arl-13(gk513); pha-1(e2123); IsOsm6; jhuEx [Parl-13::ARL-13^{K239,328R}::SMO-1::GFP; pBX]*
ZP986	*arl-13(gk513); pha-1(e2123); Is [PKD-2::GFP; CC::GFP]*
ZP988	*arl-13(gk513); pha-1(e2123); Is [PKD-2::GFP; CC::GFP]; jhuEx[Parl-13::ARL-13::GFP; pBX]*
ZP853	*arl-13(gk513); pha-1(e2123); Is [PKD-2::GFP; CC::GFP]; jhuEx [Parl-13::ARL-13^{K239,328R}; pBX]*
ZP847	*arl-13(gk513); pha-1(e2123); jhuEx [Podr-10::ODR-10::GFP; pBX]*
ZP852	*arl-13(gk513); pha-1(e2123); jhuEx [Podr-10::ODR-10::GFP; Parl-13::ARL-13; pBX]*
ZP910	*arl-13(gk513); pha-1(e2123); jhuEx [Podr-10::ODR-10::GFP; Parl-13::ARL-13^{K239,328R}; pBX]*
ZP613	*N2; jhuEx [Parl-13::ARL-13^{K239R}::GFP; pRF4]*
ZP614	*N2; jhuEx [Parl-13::ARL-13^{K328R}::GFP; pRF4]*
ZP615	*N2; jhuEx [Parl-13::ARL-13^{K239,328R}::GFP; pRF4]*
ZP1130	*arl-13(gk513); pha-1(e2123); IsOsm6; jhuEx [Parl-13::ARL-13^{K239,328R}; pBX]*
ZP1131	*arl-13(gk513); pha-1(e2123); IsOsm6; jhuEx [Parl-13::ARL-13; pBX]*
ZP317	*arl-13(gk513); IsOSM-6*
ZP917	*arl-13(gk513); pha-1(e2123); Is [PKD-2::GFP; CC::GFP]; jhuEx [Parl-13:: ARL-13^{K239,328R}::SMO-1; pBX]*
ZP928	*arl-13(gk513);); pha-1(e2123); Is [PKD-2::GFP; CC::GFP]; jhuEx [Podr-10::ODR-10::GFP; Parl-13:: ARL-13^{K239,328R}::SMO-1; pBX]*
JH81	*pha-1(e2123); him-5(e1490;) myEx [Podr-10::ODR-10::GFP; pBX]*
ZP353	*arl-13(gk513); pha-1(e2123); him-5(e1490); jhuEx [Parl-13::ARL-13^{R83Q}::GFP; pBX]*
ZP1132	*arl-13(gk513); pha-1(e2123); him-5(e1490); jhuEx [Parl-13::ARL-13^{R83Q} SMO-1::GFP; pBX]*

Table S2. **Yeast two-hybrid candidates**

Cosmid name	Gene name	Hits
F29B9.6	*ubc-9*	12
ZK85.4	*mel-26*	5
C18A11.7	*dim-1*	2
F01G10.8	*daf-14*	3
C36B1.4	*pas-4*	3
Y43B11AR4	*rps-4*	1