

Supplementary Materials for
The Kinesin Protein Kif7 Is a Critical Regulator of Gli Transcription Factors in Mammalian Hedgehog Signaling

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Fig. S2. Gene targeting of *Kif7*.

Fig. S3. Kif7 inhibits SmoM2-induced Gli transcriptional activity in ovo.

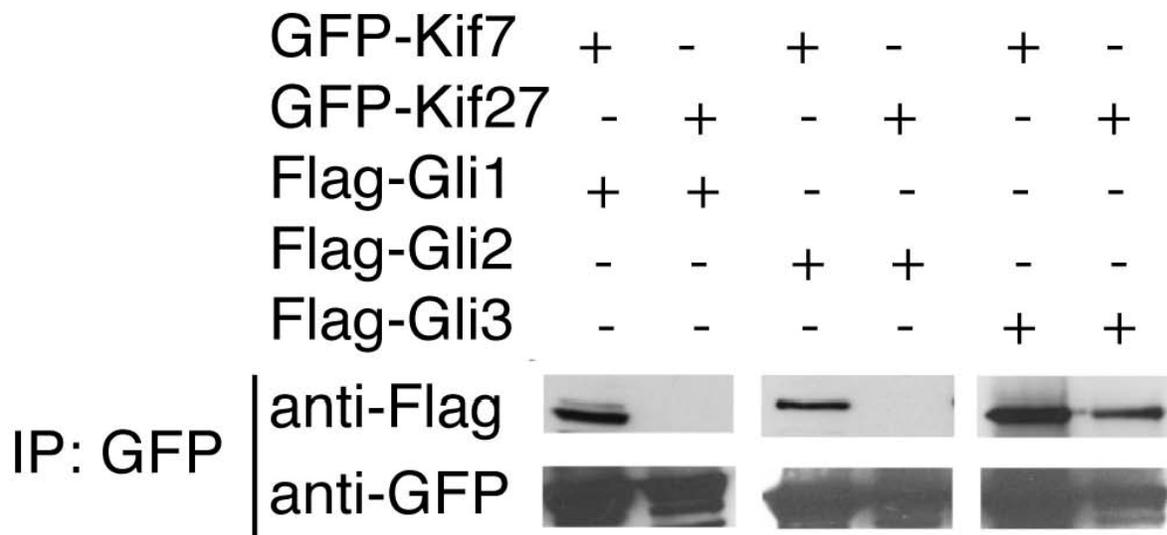


Fig. S1. Interaction of Kif7 and Kif27 with Gli transcription factors.

Coimmunoprecipitation experiments with lysates from transfected COS-7 cells revealed that Kif7 interacts with Gli1, Gli2, and Gli3, whereas Kif27 only interacts with Gli3.

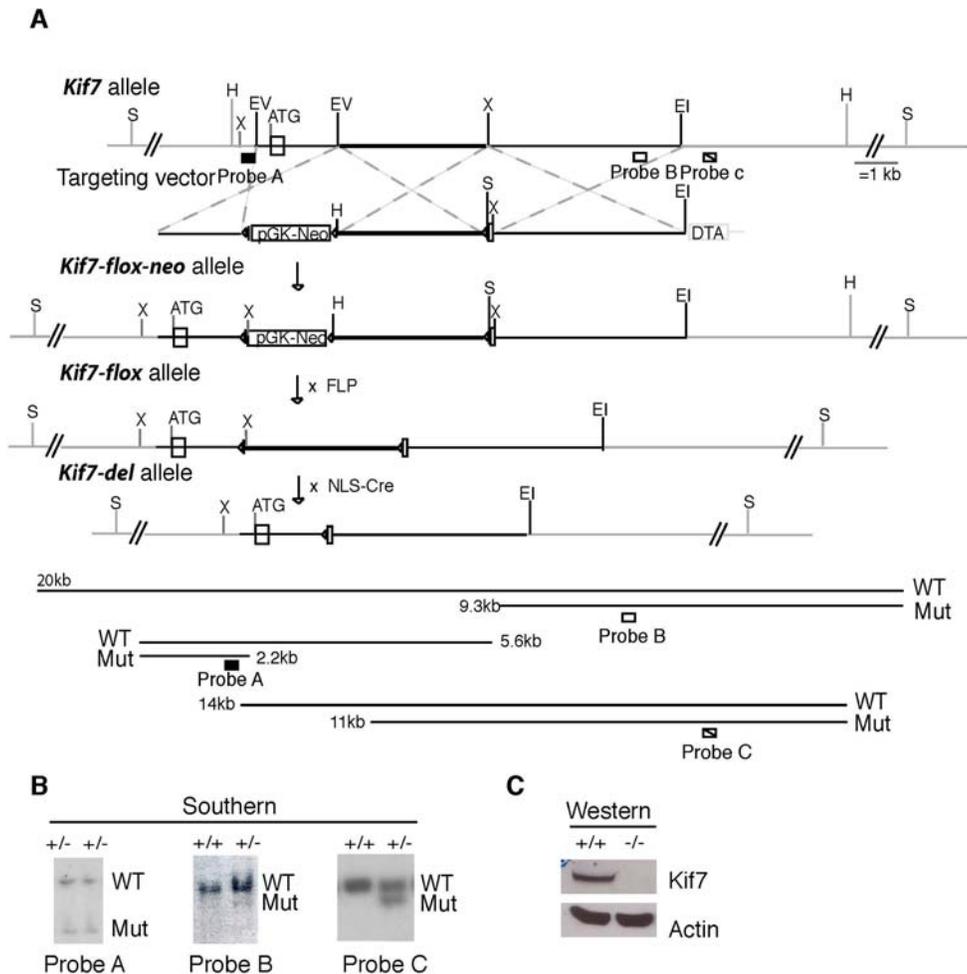


Fig. S2. Gene targeting of *Kif7*. **(A)** A conditional mutant allele of *Kif7* was generated by gene targeting in embryonic stem cells. The targeting vector contains a pair of *loxP* sites flanking exons 2-4 as well as a *frt*-flanked neomycin selection cassette (PGK-neo) inserted 5' to exon 2. Three probes used for screening are indicated and the corresponding predicted size of wild-type and mutant bands are shown at the bottom. EI: *EcoRI*, EV: *EcoRV*, H: *HindIII*, S: *SpeI*, X: *XbaI*. **(B)** Southern blotting results using probes A, B, or C all indicate correct recombination of the targeting constructs. *Kif7^{flox/flox}* mice do not exhibit any phenotype. By crossing *Kif7^{flox/flox}* mice with *NLS-Cre* mice, *Kif7^{del/+}* mice carrying a deletion of exons 2 to 4 of *Kif7* were generated. *Kif7^{del/+}* mice are viable, fertile, and have no discernible phenotype. *Kif7^{del/+}* is denoted as *Kif7^{+/-}*. **(C)** Western blot analysis of E10.5 total embryo lysate revealed no detectable Kif7 protein in *Kif7^{-/-}* mice. Actin was used as a loading control.

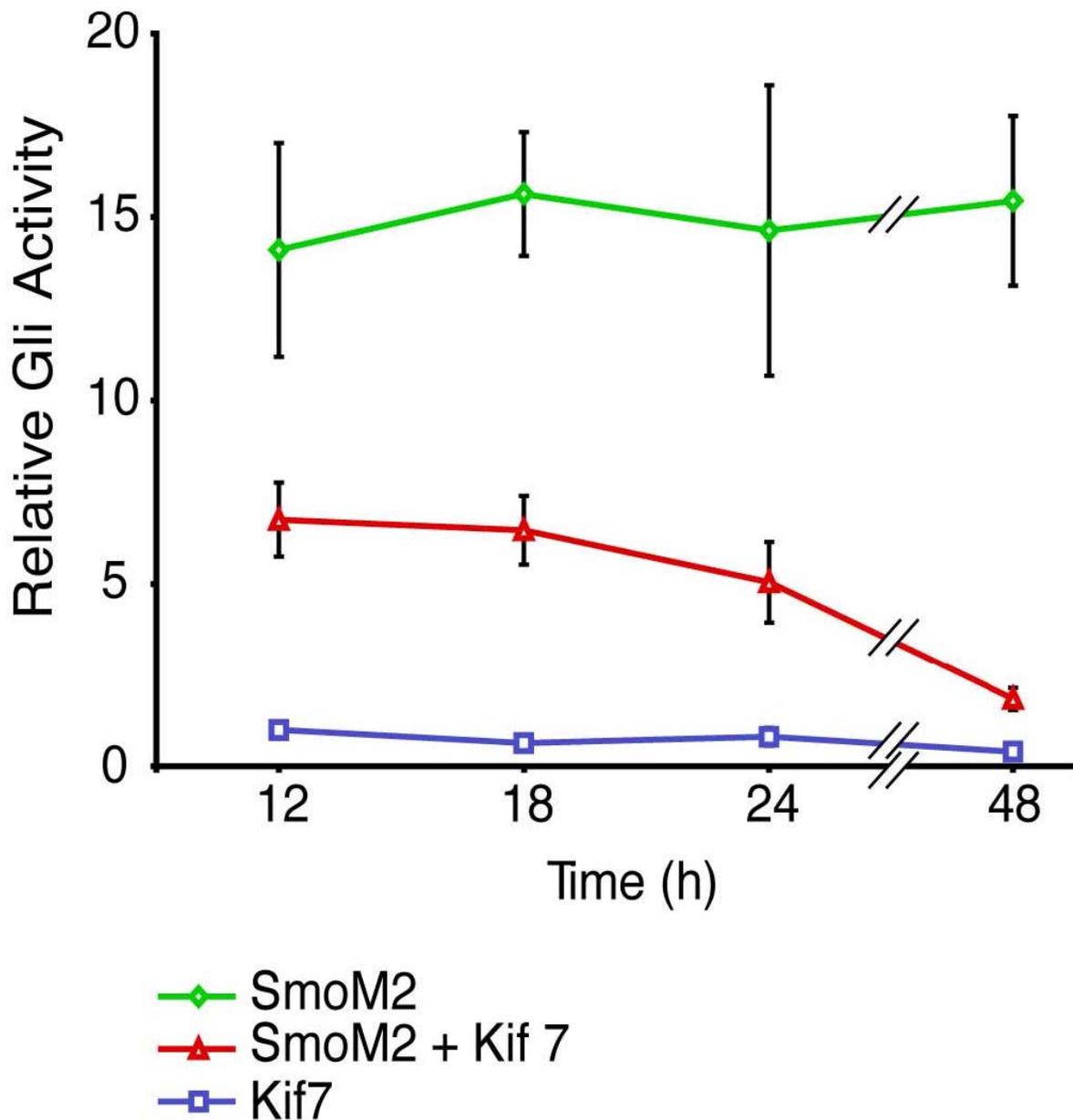


Fig. S3. Kif7 inhibits SmoM2-induced Gli transcriptional activity in ovo. A Gli-luciferase reporter gene was electroporated in ovo with Kif7 (blue), SmoM2 (green), or both (red), and luciferase activity was measured at different times post-electroporation as indicated.