

Expanded View Figures

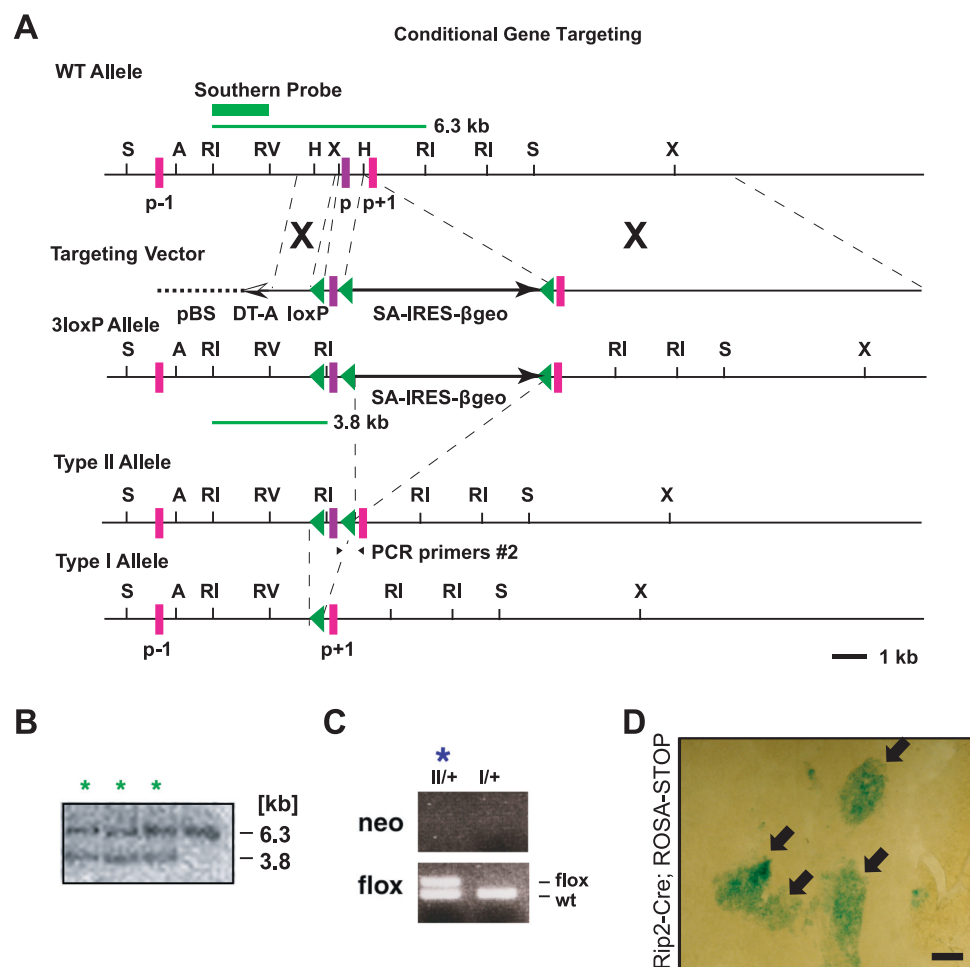


Figure EV1. Conditional knockout of mouse *Kif5b* gene.

(A–D) Establishment of beta-cell-specific *Kif5b* gene conditional knockout (cKO) mice, represented by a gene targeting strategy in mouse ES cells (A), Southern blotting screening for homologous recombinants (B; asterisks); genotyping PCR for the floxed allele (C: asterisk); and characterization of Rip2-Cre activity in a pancreas section detected by a LacZ reporter, ROSA-STOP mice (D). p, the 74 bp P-loop exon flanked by *loxP* sites (green triangles). S, *Sall*; A, *Apal*; RI, *EcoRI*; RV, *EcoRV*; H, *HindIII*; X, *XbaI*. Arrows in (D), specific Cre/*loxP* recombination sites in the pancreas of a *Rip2-Cre* ROSA-STOP double heterozygous mouse. Scale bar, 100 μ m. Corresponding to Fig. 1A–C.

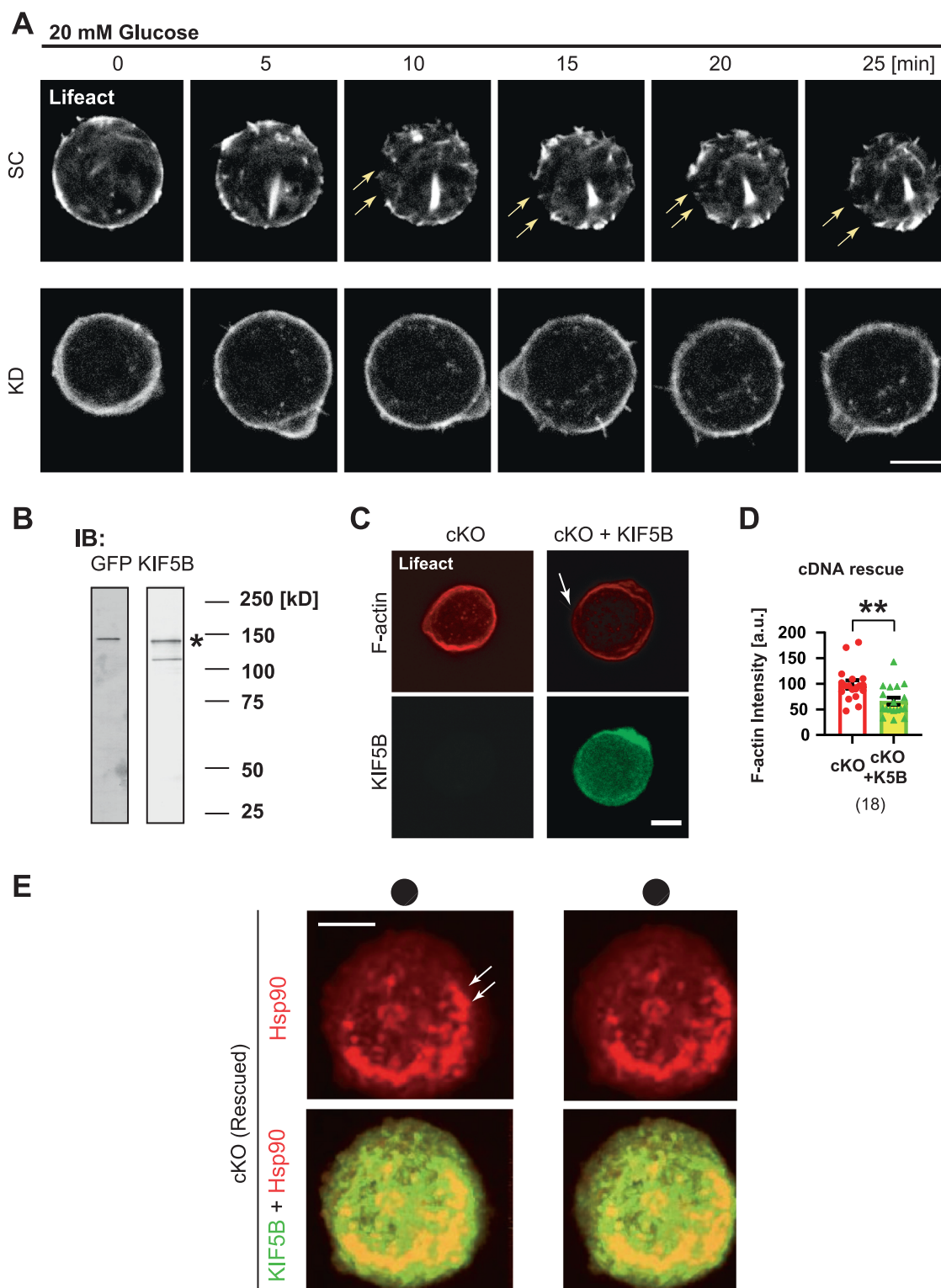


Figure EV2. KIF5B facilitates cortical actin remodeling.

(A) Time-lapse study of glucose-stimulated actin remodeling of primary beta cells from *Lifeact-mCherry* transgenic mouse pancreas, transduced with scrambled-control (SC) or KIF5B-knockdown (KD) miRNA expression vectors. Scale bar, 5 μ m. Arrows, actin remodeling. Corresponding to Fig. 4A and Movie EV3. (B–D) Rescue study of the glucose-stimulated actin remodeling in cKO primary beta cells by transducing KIF5B-EYFP, represented by immunoblotting of the expressed proteins in Ins1 cells using a mouse anti-GFP antibody and a rabbit anti-KIF5B antibody (B), *Lifeact-mCherry* transgene labeling (C), and F-actin quantification (D). Scale bar, 5 μ m. Asterisks in (B), bands for tagged KIF5B. Arrow in (C), actin remodeling. $^{**}P = 0.0042$, two-sided unpaired Welch's *t* test; *n* = 18. Corresponding to Fig. 4A. (E) Stereoscopic fluorescence microscopy of a cKO primary beta-cell expressing tagRFP-Hsp90 (red) and KIF5B-EYFP (green). Scale bar, 5 μ m. Data are represented by the mean $\pm$ SEM. Corresponding to Fig. 8A.