

Supplemental material

Takezaki et al., <https://doi.org/10.1084/jem.20182351>

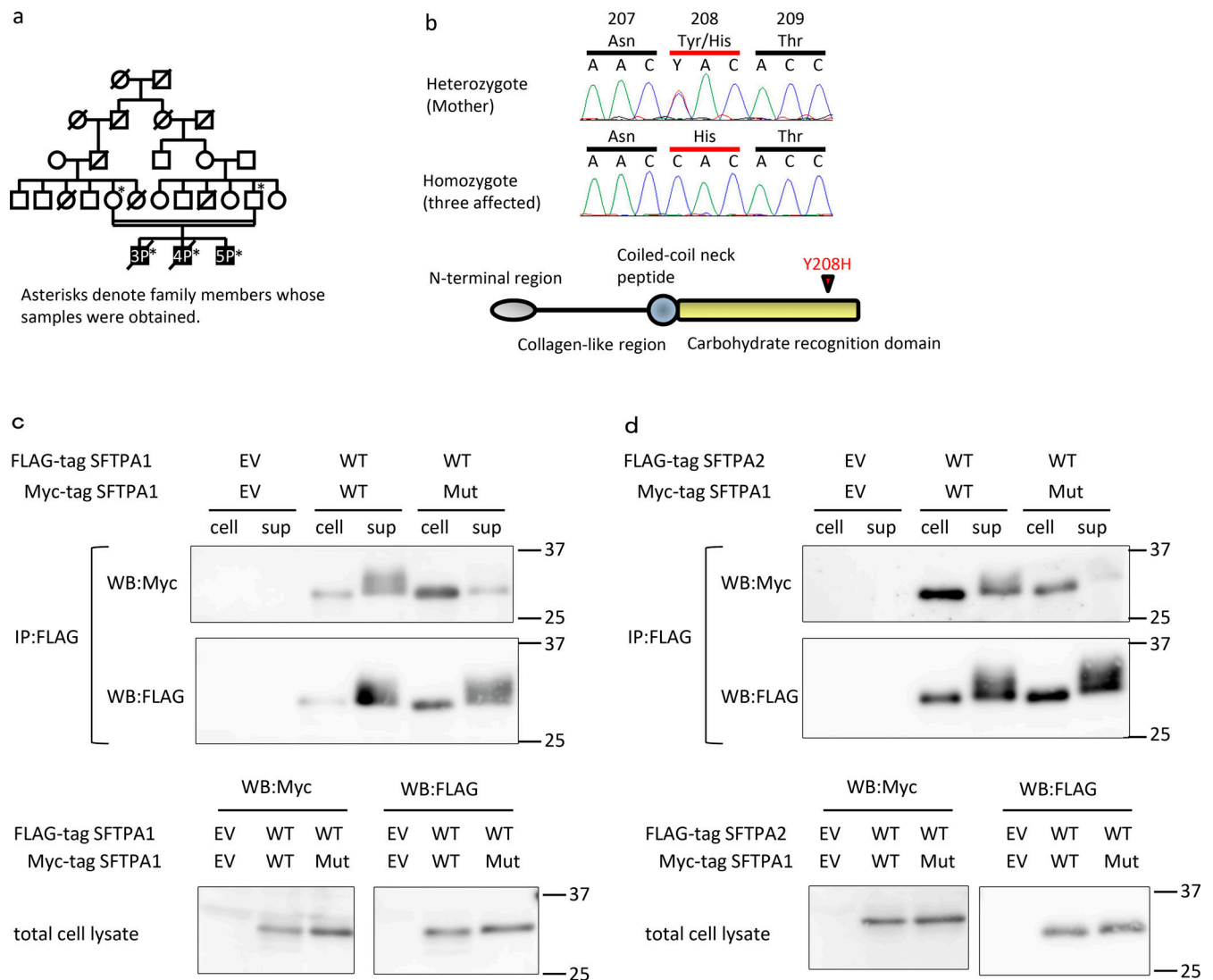


Figure S1. **Identification of an SFTP1 mutation in IPF patients and its effect on the secretion of SFTP1.** (a) Pedigree of a Japanese IPF family: double lines indicate consanguinity, filled black indicates IPF patients, and diagonal lines indicate deceased siblings. P, patient. (b) The heterozygous (parents) or homozygous (patients) missense mutation in *SFTP1* (Y208H) as shown by capillary sequencing. *SFTP1* protein domains: the red arrowhead indicates position of the missense mutation. (c and d) The Flag-tagged *SFTP1* and Myc-tagged *SFTP1* (c; WT or mutant [Mut]) or Flag-tagged *SFTP2* and Myc-tagged *SFTP1* (d; WT or Mut) cDNA was transfected into A549 cells. As a negative control, empty vector (EV) was transfected. 1 d after transfection, the cell pellets (cell) or supernatant (sup) were immunoprecipitated with anti-Flag antibody, and the immunoprecipitates were blotted with an anti-Flag or Myc antibody. As a control, total cell lysates were blotted with the anti-Flag or Myc antibody. WB, Western blot. Data shown are representative of five (c and d) independent experiments.

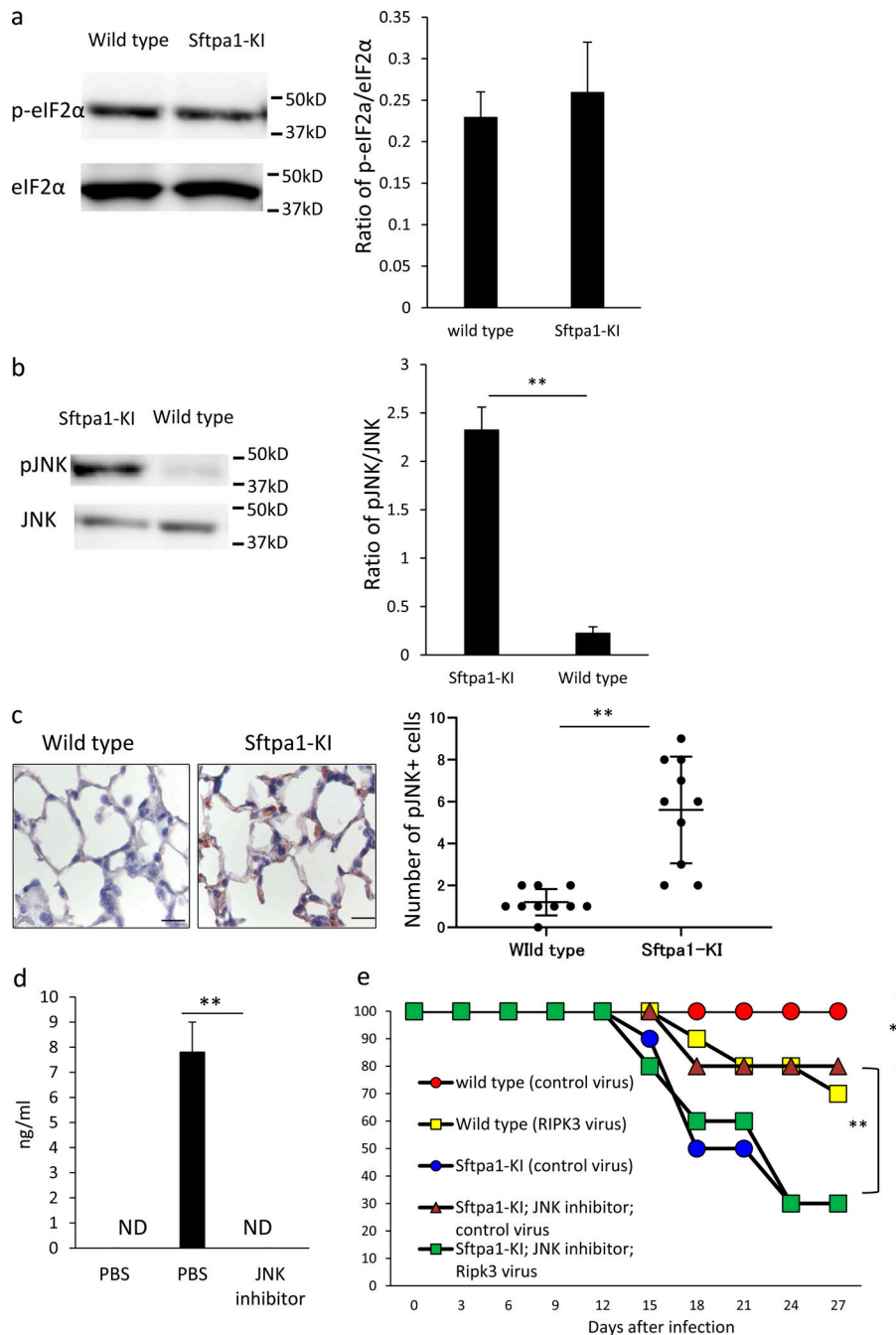


Figure S3. The inhibition of JNK blocks pulmonary fibrosis. (a and b) The expression of phosphorylated eIF2 α (a) or phosphorylated JNK (b) in WT or Sftpa1-KI mice at the age of 35 wk was evaluated by Western blotting. Data are means \pm SD. **, $P < 0.01$. (c) The expression of pJNK in WT or Sftpa1-KI mice at the age of 30 wk was evaluated by immunohistochemistry. The pJNK-positive cells (average of the number of pJNK-positive cells in 10 independent fields in right lung) was counted. Data are means \pm SD. **, $P < 0.01$. The scale bar indicates 20 μ m. (d) WT mice were injected with LPS (50 μ g), and serum TNF α was measured 3 h after LPS injection. The JNK inhibitor (30 mg/kg, s.c.) was administered 3 h before LPS injection. Data are means \pm SD. **, $P < 0.01$. The data are representative of two independent experiments. (e) This is an additional set of experiments for Fig. 3 b. Survival of 20-wk-old WT (red circles), WT infected with adenovirus carrying *Ripk3* (yellow squares), Sftpa1-KI mice infected with control virus (blue circles), Sftpa1-KI mice infected with control virus and treated with a JNK inhibitor (brown triangles), or Sftpa1-KI mice infected with *Ripk3*-adenovirus and treated with a JNK inhibitor (green squares; number of mice in each independent experiment is 10). *, $P < 0.05$; **, $P < 0.01$ by log rank test. Data shown are representative of five (a–d) independent experiments.