Figure S1

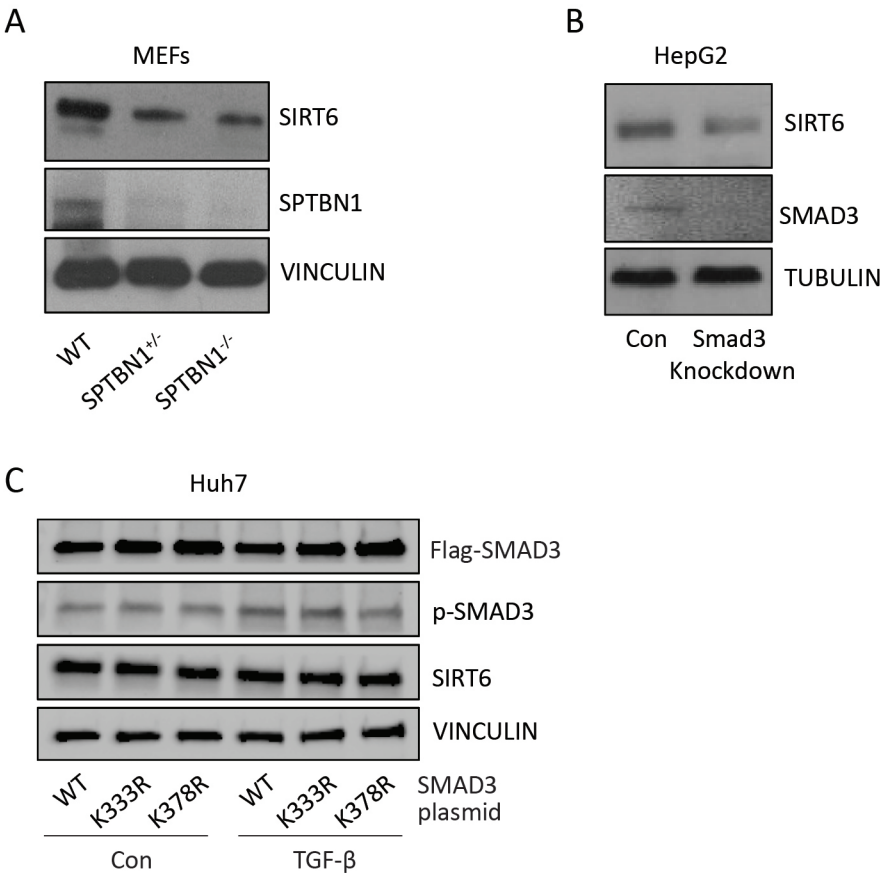

FIGURE S1. Deficiency of SPTBN1 or SMAD3 decreases the abundance of SIRT6. (A, B) SIRT6 was detected by Western blotting in MEFs and HepG2 stable cell lines with or without SMAD3 knockdown. VINCULIN or TUBULIN served as the loading control. (C) SIRT6 was detected by Western blotting in Huh7 cells expressing the indicated SMAD3 constructs. Cells were transfected with the indicated plasmids for 48 h, after 24 h starvation, then exposed to TGF-β (200 pM) for 24 h. Data shown are from 1 of 2 independent experiments.

Figure S2

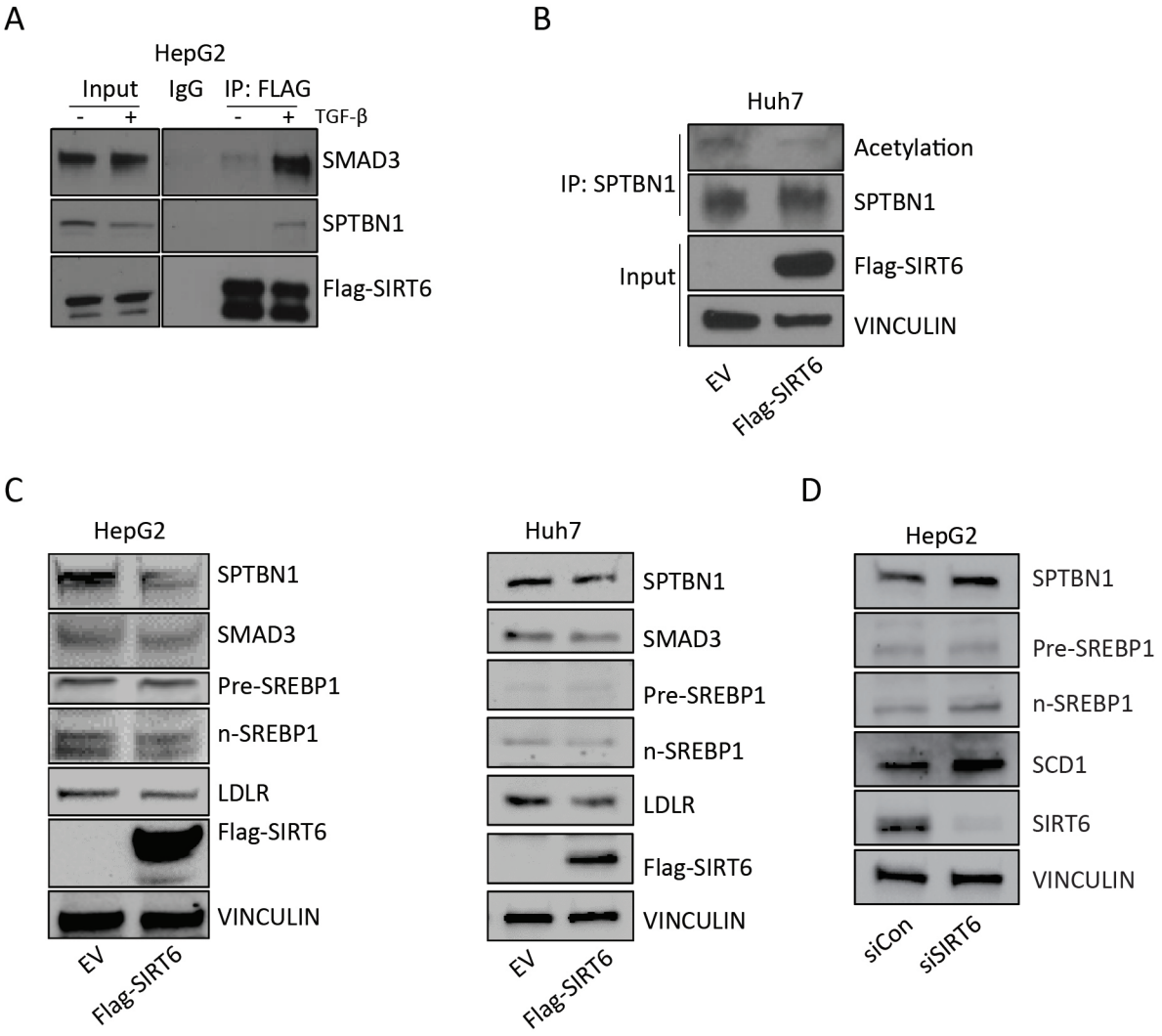

FIGURE S2. SIRT6 interacts with SMAD3 and SPTBN1 and regulates their abundance. (A) Co-immunoprecipitation of SMAD3, SPTBN1, and Flag-SIRT6 was performed in HepG2 cells with or without exposure to 200 pM TGF-β for 3 h, using an irrelevant IgG antibody or anti-FLAG antibody. IP, immunoprecipitation. (B) The effect of SIRT6 overexpression on SPTBN1 acetylation in Huh7 cells was determined by immunoprecipitating SPTBN1 and Western blotting for the presence of acetylation with SPTBN1. (C) The effect of SIRT6 overexpression on SPTBN1, SMAD3, Pre-SREBP1, n-SREBP1, and LDLR abundance in HepG2 cells and Huh7 cells was determined by Western blotting. VINCULIN served as the loading control. EV, empty vector. (D) The effect of SIRT6 knockdown on SPTBN1, Pre-SREBP1, n-SREBP1, and SCD1 abundance in HepG2 cells was determined by Western blotting. VINCULIN served as the loading control. siCon, control siRNA. All data shown are from 1 of 2 - 3 independent experiments.

Figure S3

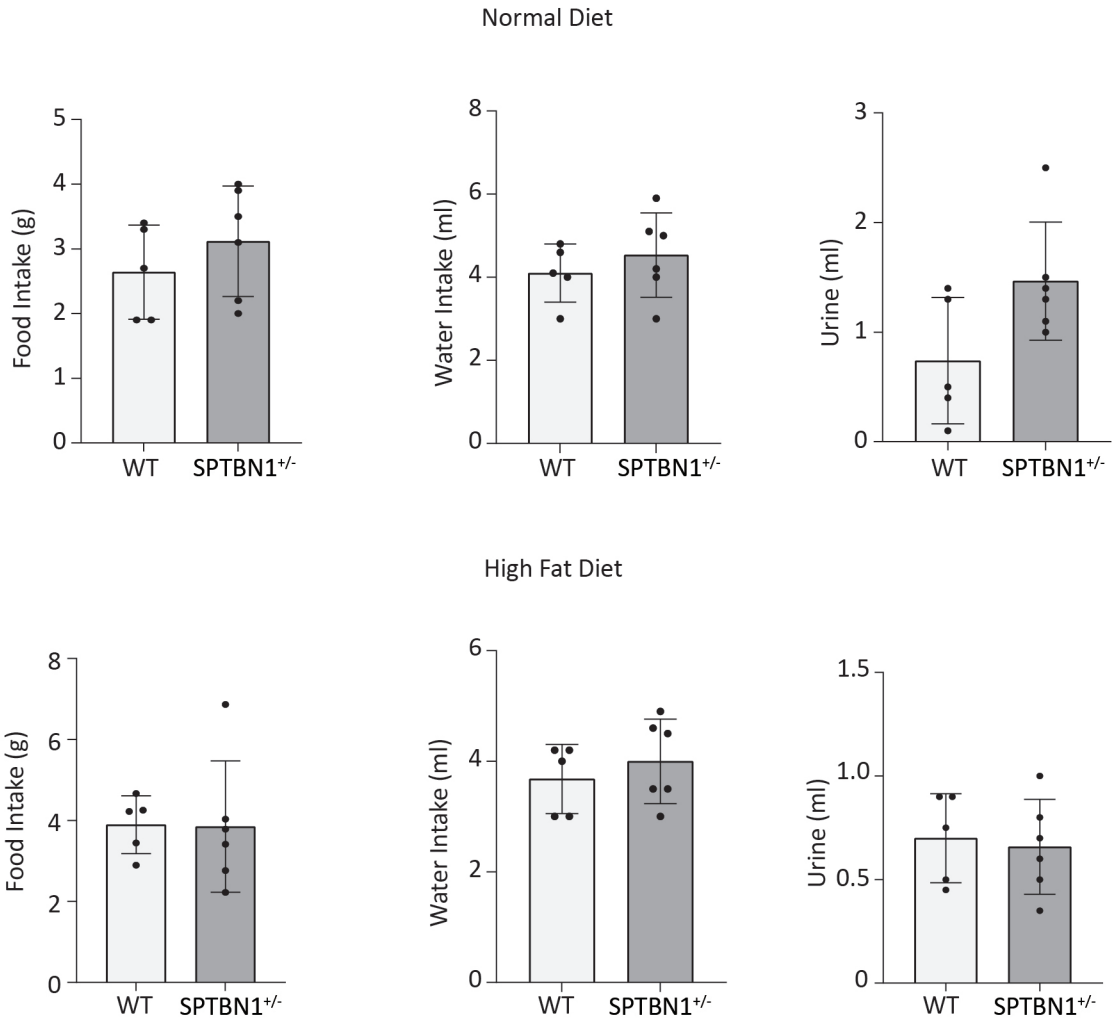

FIGURE S3. Effects of SPTBN1 heterozygosity on food intake, water intake, and urine output. WT or *SPTBN1*$^{+/-}$ mice (10 – 12 weeks old) were fed a normal chow diet or a high-fat diet for 12 – 16 weeks. Data are presented as mean ± SEM of 5 – 6 mice per group.