Figure S1. Characterization of GFP-hFbh1–expressing cell lines. (A) U2OS/GFP-hFbh1 WT cells were induced or not with DOX, cultured for up to 5 d, and counted to monitor cell proliferation. (B) U2OS/GFP-hFbh1 WT cells induced with DOX for 24 h were harvested at the indicated times after exposure to IR (6 Gy). The cells were fixed, and the GFP signal was visualized by confocal microscopy. Bar, 10 µm. (C) Parental U2OS cells or stable U2OS/NLS–GFP-hFbh1 cell lines were treated with DOX for 24 h. Cells were lysed and subjected to GFP immunoprecipitation followed by immunoblotting with GFP and Skp1 antibodies. WCE, whole cell extract; IP, immunoprecipitation. (D) U2OS/NLS–GFP-hFbh1 cell lines were left untreated or induced with DOX for 48 h. The cells were harvested, stained with propidium iodide, and processed for flow cytometric analysis to determine the DNA content.
Figure S2. **Characterization of hFbh1 mutants.** (A) U2OS/GFP-hFbh1 cells were induced with DOX for 24 h and irradiated with 100 J/m² UV-C through a polycarbonate filter with 5-µm pores. 30 min later, cells were fixed and immunostained with cyclin A antibody. (B) U2OS/GFP-hFbh1 cells were induced with DOX for 24 h followed by addition of nocodazole for 16 h. Mitotic cells were shaken off and plated on glass coverslips. After 4 h, cells were either left untreated or UV irradiated as in A. Cells were preextracted, fixed, and stained with DAPI and proliferating cell nuclear antigen (PCNA) antibody. (C) EMSA of WT or mutant GST-hFbh1 proteins, using 32P-labeled ssDNA as a probe, is shown. The experiment was performed as in Fig. 2 C. (D) U2OS cells were transfected with the indicated combinations of GFP- and Flag-tagged hFbh1 for 24 h. The cells were lysed and subjected to Flag immunoprecipitation (IP) followed by immunoblotting with Flag and GFP antibodies. Monoclonal anti-Flag antibody was obtained from Sigma-Aldrich. WCE, whole cell lysate. Bars, 10 µm.

Figure S3. **Efficiency of RNAi-mediated knockdown of hFbh1 in BJ cells.** BJ cells were transfected with control or hFbh1 siRNAs for 48 h and processed for immunoblotting with the indicated antibodies.