

S supplementary Figure 1

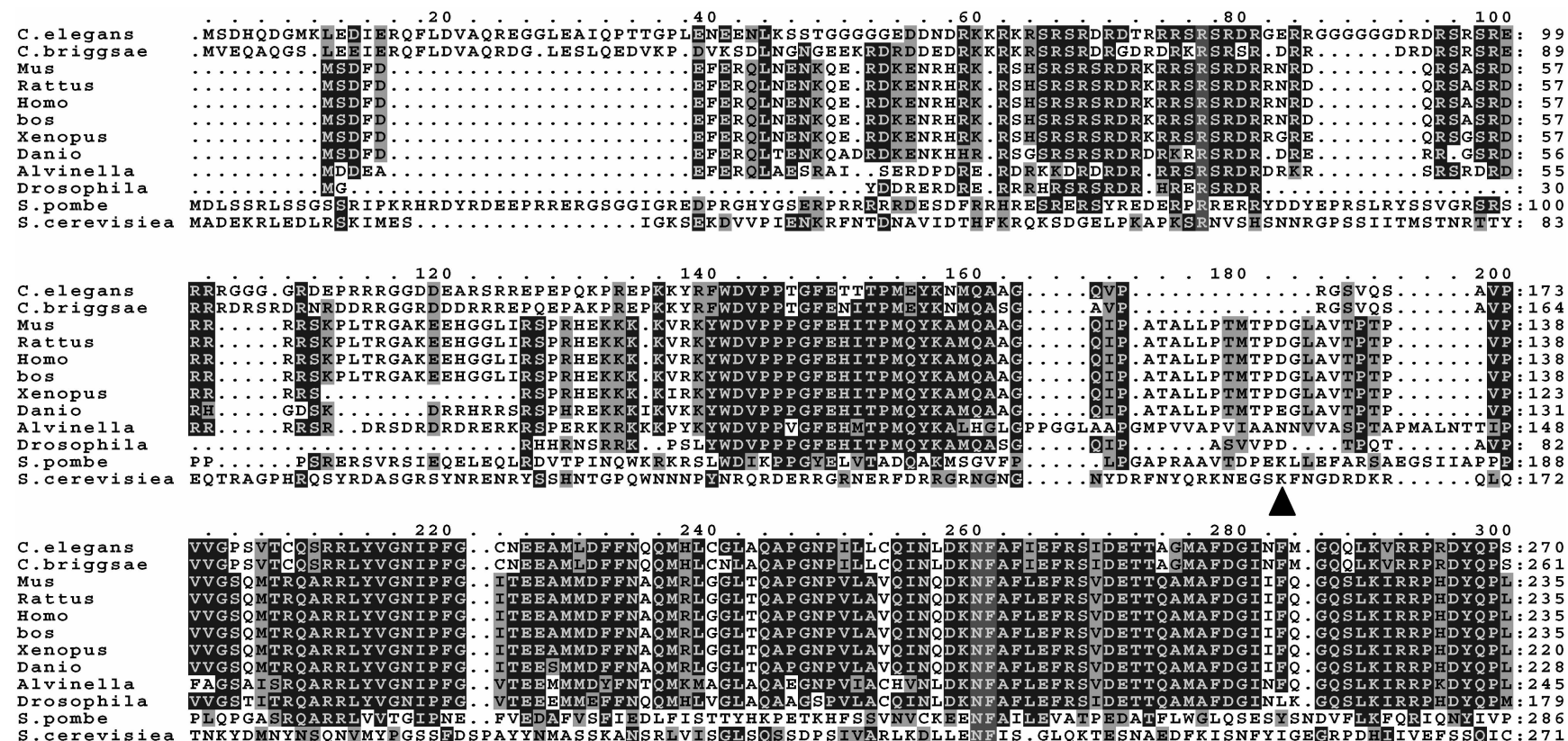


Figure 1. U2AF65 is cleaved by caspases at the aspartate 128. Evolutionary conservation of caspase cleavage site in U2AF65 homologues. The amino acid sequences of U2AF65 from several species were aligned using the Clustal program. The position of aspartate 128 is indicated by a black triangle. The NCBI protein accession numbers for these proteins are as follows: *C. elegans*: NP_497326; *C. briggsae*: U79145; *Mus*: CAA45874; *Rattus*: XP_218195; *Homo*: CAA45409; *Bos*: AAI14161; *Xenopus*: AAH67966; *Danio*: NP_991252; *Alvinella*: AY679693; *Drosophila*: NP_476891; *S. pombe*: L22577; *S. cerevisiae*: NP_012849.

S supplementary Figure 2

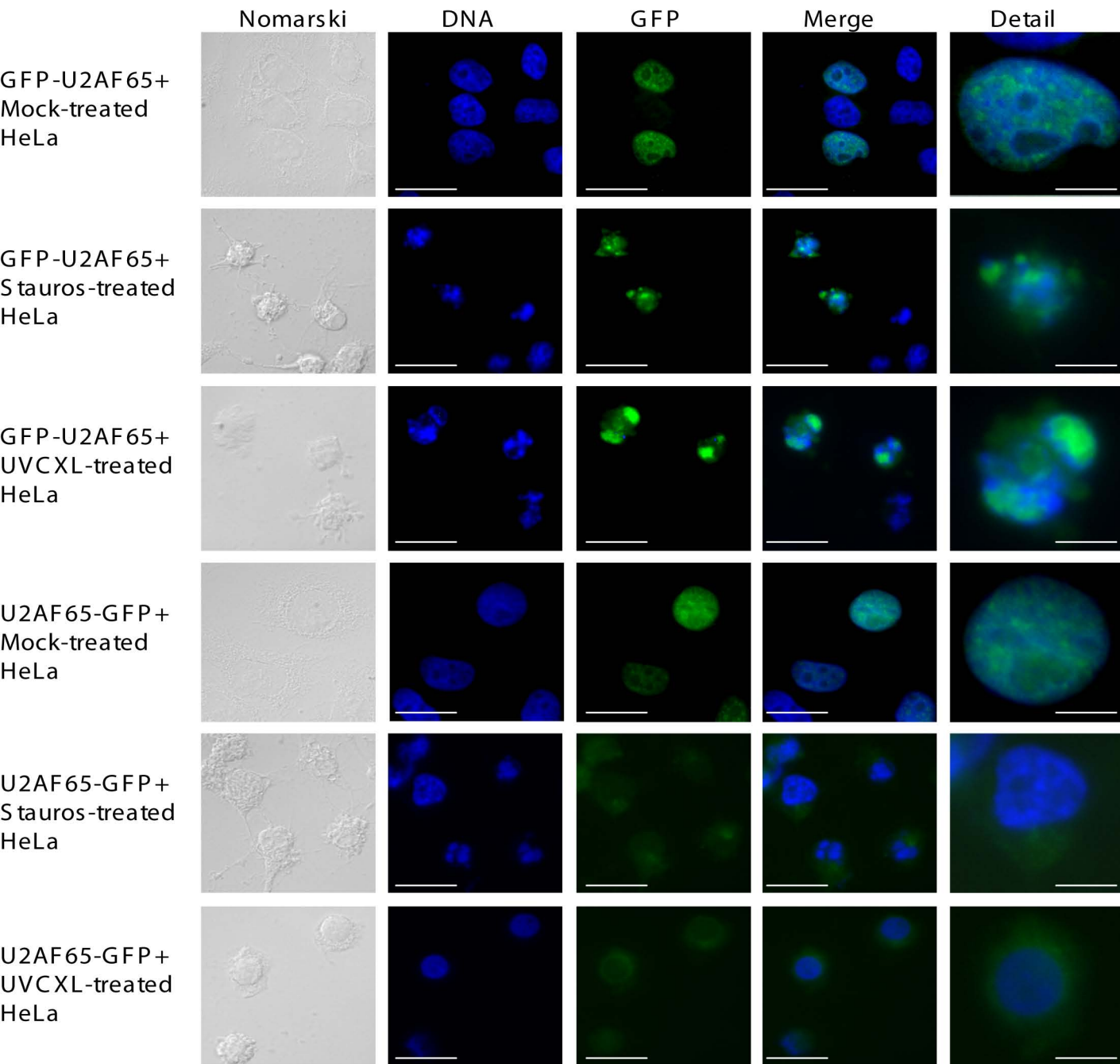


Figure 2. Subcellular localization of GFP-tagged U2AF65 fragments upon apoptosis induction. HeLa cells were treated with plasmids expressing GFP-fused with full-length U2AF65 at the C-terminal (GFP-U2AF65) or at the N-terminal (U2AF65-GFP) of GFP. Post-transfected HeLa cells were mock-treated or treated with staurosporine (2 μ M) or irradiated with ultraviolet light (100 mJ/cm²), cultured for 6 h and then stained with Hoechst for 15 min. From left to right, images show cells examined using Nomarski method (Nomarski), or stained with Hoechst through a DAPI filter (DNA), or fluorescence from GFP examined with a TRITC filter (GFP). The two right-hand images are the sum of DAPI and GFP images (Merge) as well as detailed images (Detail). In DNA, GFP, and Merge images, scale bars represent 30 μ m. In detailed images, scale bars represent 5 μ m.

S Supplementary Figure 3

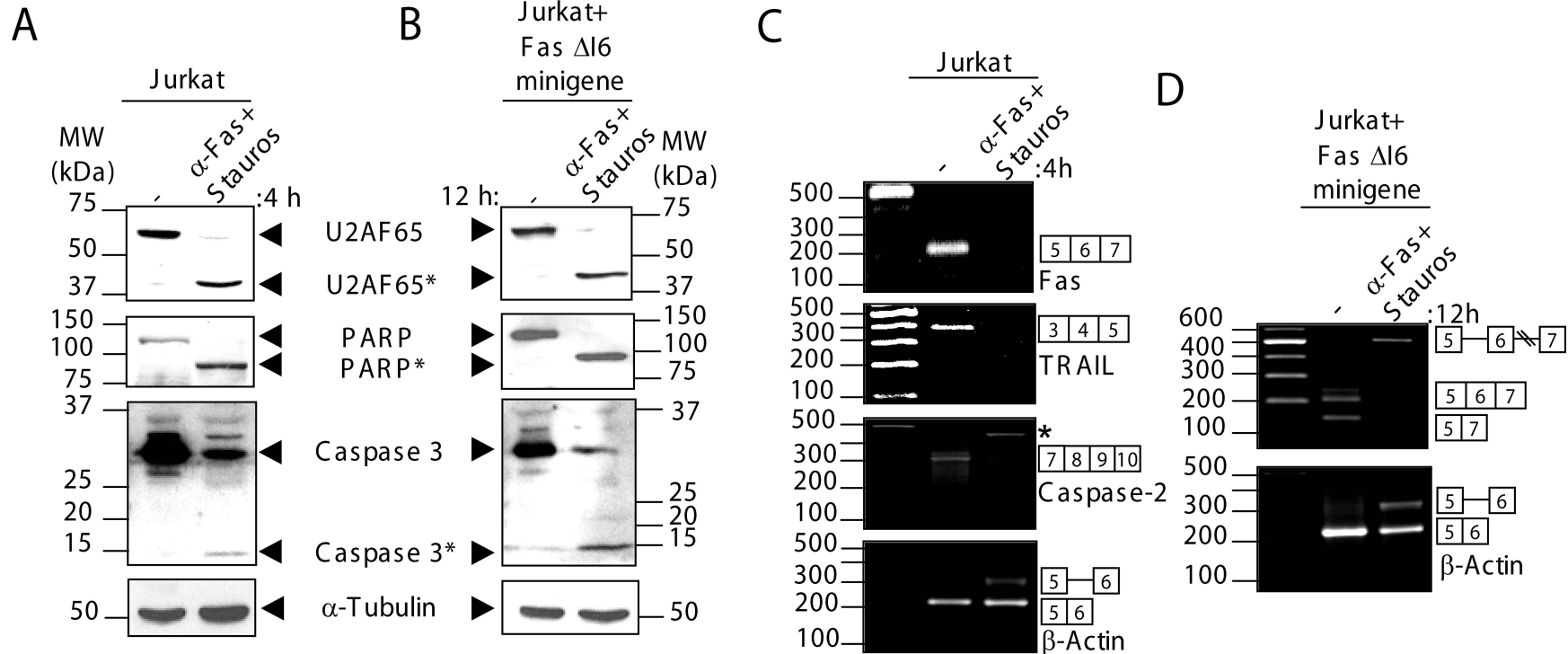


Figure 3. Complete U2AF65 cleavage compromises alternative splicing. (A and C) Protein extracts and total RNAs from mock-treated or treated Jurkat cells with the apoptosis-inducers indicated for 4 h were analyzed by Western blot (A) using antibodies referred and by RT-PCR (C) using specific primers to the genes tested. The identity of the products was confirmed by sequencing. The band indicated by an asterisk is a PCR artifact. (B and D) Jurkat cells transfected with Fas Δ 16 minigene reporter, were mock-treated or treated as before for 12 h. Protein extracts and total RNAs were analyzed by Western blot (B) and by RT-PCR (D) as described above. In all panels, the positions of molecular mass markers, protein bands, and predicted spliced products are schematically indicated.