Supplementary Information, Figure S1. Interaction of Plk3 and CD95.

(A) HeLa cells were treated with CD95L and CHX for different time periods. Endogenous CD95 was immunoprecipitated from cell lysates with anti-CD95 mouse antibodies and subsequently immunoblotted for Plk3 and CD95. Normal mouse IgG was used as a control. (B) HeLa cells were transfected with CD95 siRNA (siCD95) or a control siRNA (siC) and then treated with CD95L and CHX for the indicated time periods. Endogenous Plk3 was immunoprecipitated from cell lysates with anti-Plk3 rabbit (Abcam) antibody. Normal rabbit IgG was used as a control. The precipitated proteins were immunoblotted for Plk3, CD95 and GAPDH. (C) Co-localization of Plk3 and CD95 with or without CD95L treatment in HeLa cells was monitored via immunofluorescence microscopy. Cells were fixed and stained for Plk3, CD95 and DNA. Representative image acquisition was performed using a laser-scanning confocal microscope (CLSM) and rendering of confocal z stacks was performed using the LAS AF software. Scale bar: 2 µm. (D) Bright-field images of Hela (upper panel) and Jurkat (lower panel) cells without (0 min) or with CD95L/CHX treatment (15 min). Scale bar: 50 µm (upper panel), 10 µm (lower panel).