Expanded View Figures

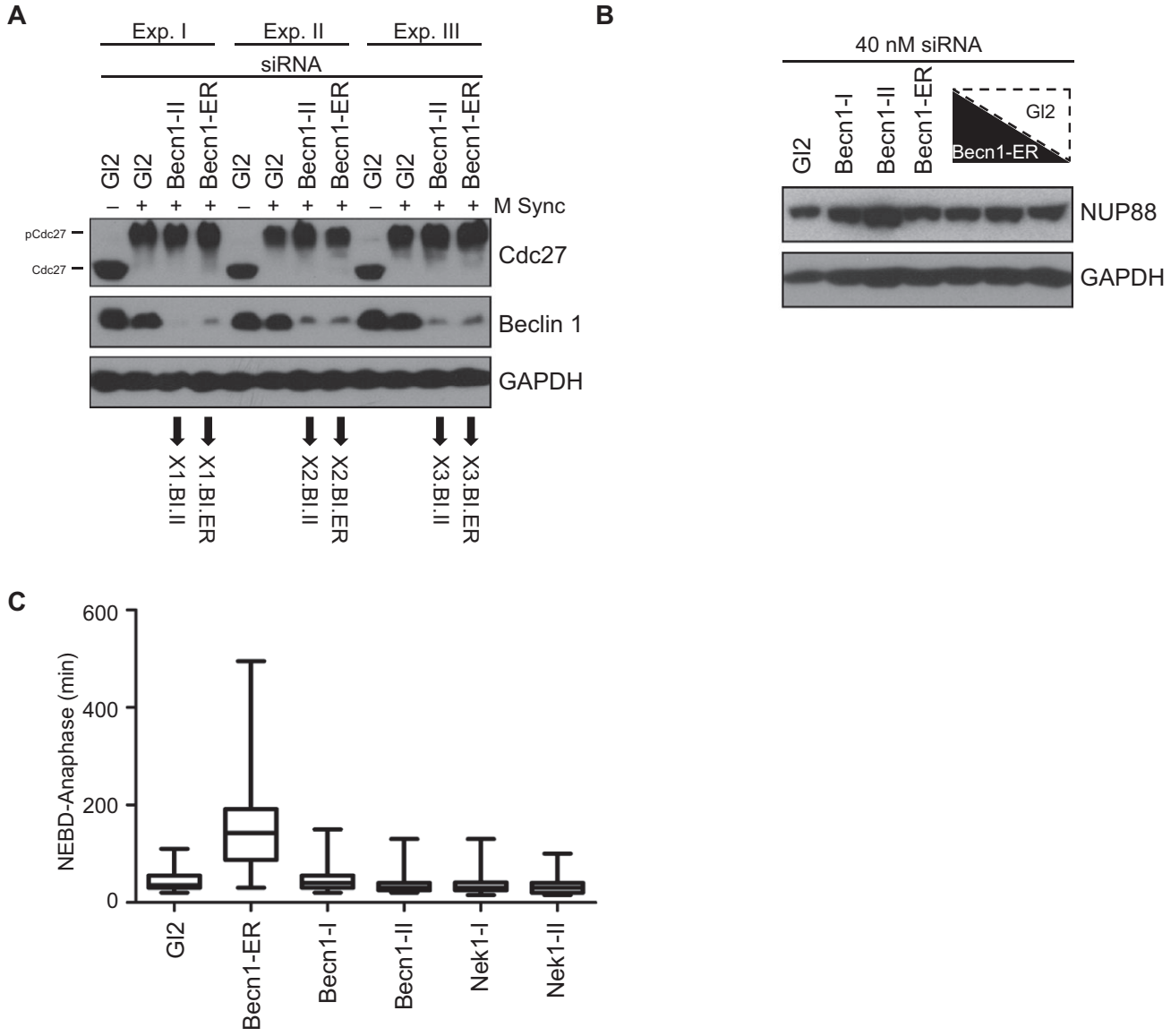

Figure EV1. Search for possible off-target effects of the Beclin 1-ER siRNA.
(A) HeLaS3 cells have been either left asynchronous (M Sync −) or synchronized in mitosis by a single thymidine arrest, released in the presence of nocodazole, followed by mitotic shake-off (M Sync +). SiRNA with the indicated sequences was performed for a total of 48 h. Part of the cells obtained in three independent experiments was subjected to protein extraction and immunoblot with the indicated antibodies, whereas the remaining cells were subjected to RNA extraction followed by hybridization on the gene chips indicated. The results of the transcriptome profiling are depicted in Fig 1H and Table EV1. (B) HeLaS3-H2B-GFP cells were subjected to siRNA transfection with a constant amount of RNA duplex composed by either the indicated sequence or a mixture of decreasing Becn1-ER (50, 25 and 12.5%) and increasing amount of Gl2 siRNA (50, 75 and 88.5%, from left to right). Please note that the lowest panel (GAPDH, representing a loading control) has been duplicated from Fig 1F (which has been generated using the same samples). The NUP88 immunoblot is shown in addition. (C) HeLaS3-H2B-GFP cells were subjected to time-lapse video microscopy following transfection with the indicated siRNAs for 48 h. Box (interquartile range) and whisker (min to max) plots showing the elapsed time (min) between NEBD and anaphase for 50 individual cells ($n = 1$).