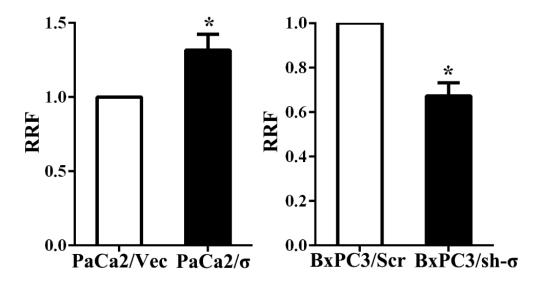
## **Supplemental Figures**



**Figure S1. 14-3-3\sigma contributes to IR resistance.** Relative resistance factor (RRF) was derived using IC<sub>50</sub> from colony formation assay survival curves shown in Figure 1. REF=IC<sub>50</sub>(MiaPaCa-2/ $\sigma$ )/IC<sub>50</sub>(MiaPaCa-2/Vec) or IC<sub>50</sub>(BxPC-3/Sh- $\sigma$ )/IC<sub>50</sub>(BxPC-3/Scr). (\*p<0.05; n=3).

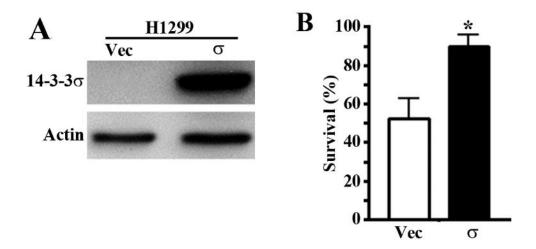


Figure S2. 14-3-3 $\sigma$  contributes to IR resistance in H1299 cells. (A) Western blot analysis of 14-3-3 $\sigma$  expression in stable H1299/ $\sigma$  cells with 14-3-3 $\sigma$  over-expression and vector-transfected control H1299/Vec cells. Actin was used as a loading control. (B) Colony formation assay was performed to determine the survival of H1299/ $\sigma$  and H1299/Vec cells following treatments with 5 Gy IR (\*p< 0.05; n=3).

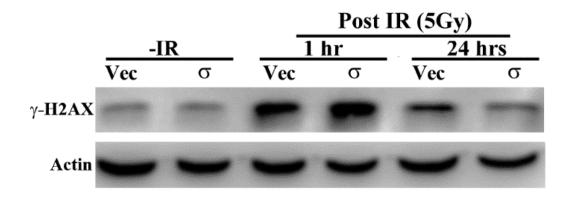


Figure S3. 14-3-3σ modulates IR-induced  $\gamma$ -H2AX in H1299 cells.  $\gamma$ -H2AX accumulation in H1299/σ and H1299/Vec cells following IR treatment was determined using Western blot analysis. Actin was used as a loading control.

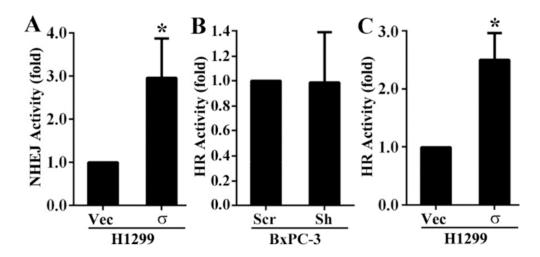
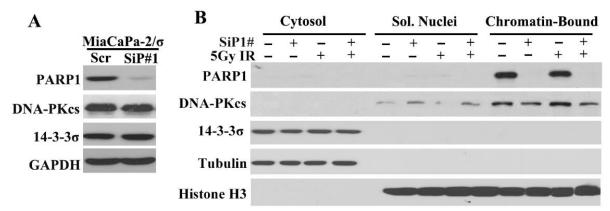


Figure S4. 14-3-3 $\sigma$  promotes NHEJ but not HR repair of DSBs. Host cell reactivation assays of NHEJ (A) and HR (B, C) activity were performed using reporter plasmids in H1299/ $\sigma$  and the control H1299/Vec cells (A, C) and BxPC-3/Sh- $\sigma$  and BxPC-3/Scr cells (B) as described in Materials and Methods (\*p< 0.05; n=3).



**Figure S5. Role of PARP1 in DNA-PKcs recruitment.** (A) Western blot analysis of PARP1 in MiaPaCa-2/σ cells after transient PARP1 knockdown. (B) Western blot analysis of subcellular localization of DNA-PKcs in MiaPaCa-2 cells following transient PARP1 knockdown and IR treatments.

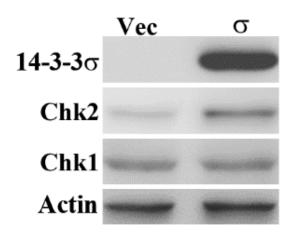
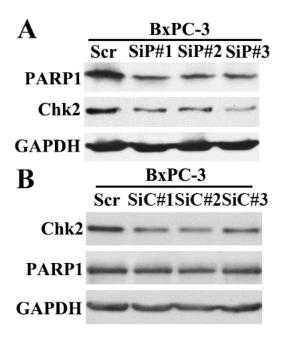


Figure S6. 14-3-3 $\sigma$  regulation of Chk2 expression in H1299 cells. Western blot analysis was performed to determine the expression of Chk2 and Chk1 in H1299/ $\sigma$  and H1299/Vec cells.



**Figure S7. PARP1 regulates Chk2 expression but not vice versa.** Western blot analyses were performed to determine expressions of PARP1, Chk2, and GAPDH loading control following knockdown of PARP1 (A) or Chk2 (B) using different siRNAs for PARP1 (SiP) or Chk2 (SiC).