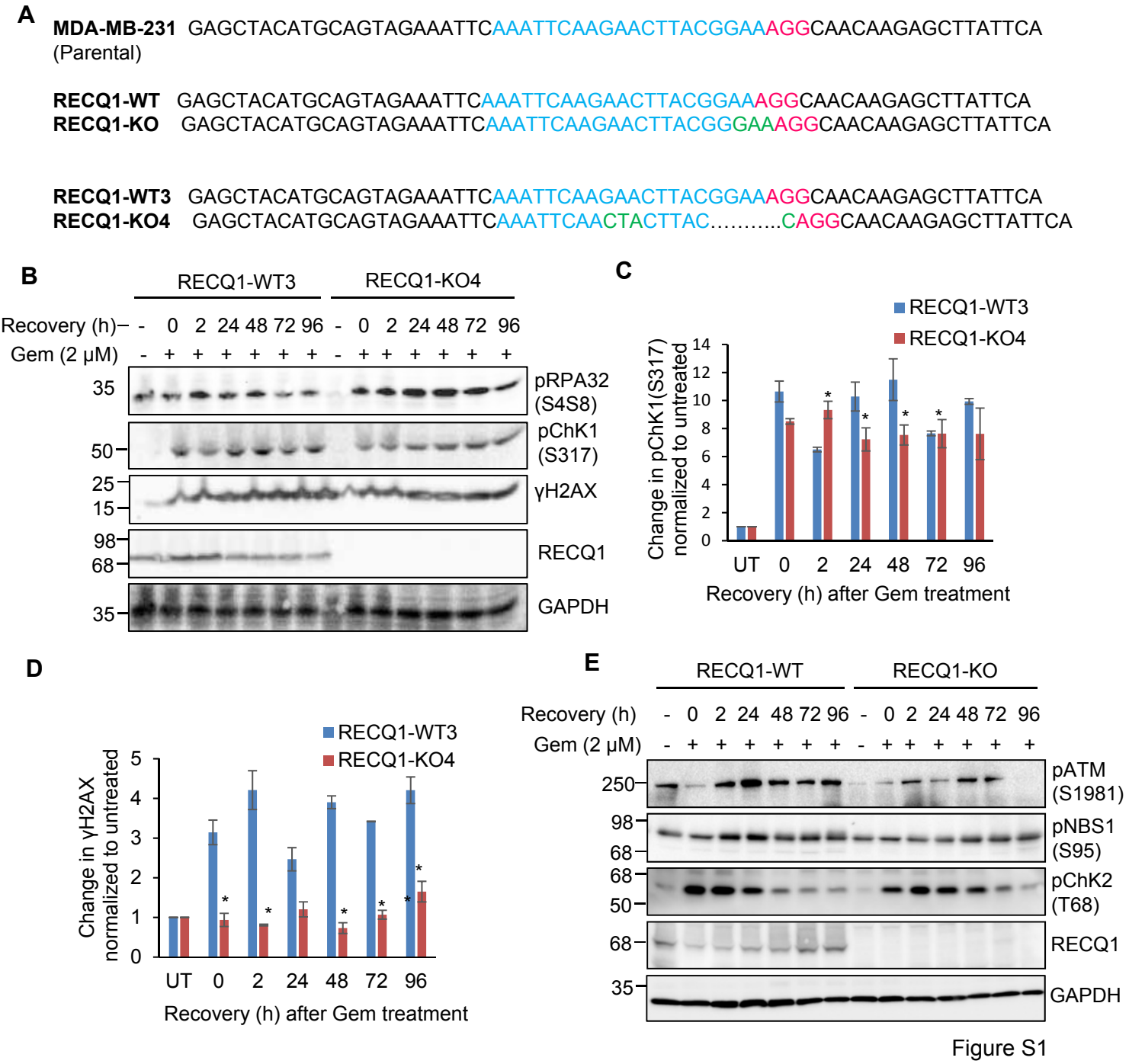

Figure S1

Figure S1: (A) Sequence information from individual clones at CRISPR-Cas9 target loci obtained by Sanger sequencing. The PAM sequence is shown in pink, target site is shown in blue and the change in sequences at target site in RECQ1-KO clones is indicated in green. (B) Western blot analysis of pRPA32(S4/S8), pChK1(S317), γH2AX and RECQ1 in RECQ1-WT3/RECQ1-KO4 cells treated with gemcitabine (2 μM for 2 h) followed by recovery in drug-free medium for the indicated time points. GAPDH is used as a loading control. (C) and (D) Quantitative representation of western blot signal intensities for pChK1(S317) and γH2AX proteins (in panel B) using Image J. (E) Western blot analysis of pATM(S1981), pNBS1(S95) and pChK2(T68) in RECQ1-WT/RECQ1-KO cells treated with gemcitabine (2 μM for 2 h) followed by recovery in drug-free medium at the indicated time points. Same cell lysates were used in Figure 3A. RECQ1 panel from Figure 3A is shown here to indicate RECQ1 protein status in RECQ1-WT and RECQ1-KO cell lysates. GAPDH is used as a loading control. Molecular mass (in kDa) is shown to the left of the Western blots.

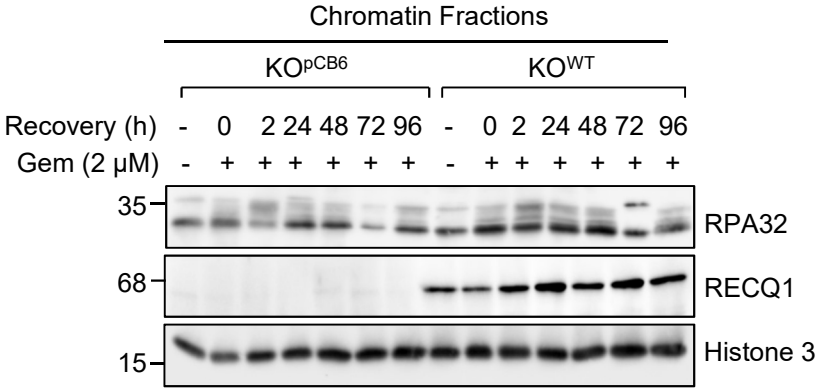

Figure S2: Decreased RPA recruitment on chromatin in RECQ1-KO cells is rescued by complementation of wildtype RECQ1 (KO^{WT}) upon gemcitabine treatment. Western blot analysis of RPA32 and RECQ1 in chromatin enriched fractions of KO^{pCB6} and KO^{WT} cells treated with gemcitabine (2 µM for 2 h) followed by recovery in drug-free medium as indicated. Histone 3 is used as loading control. Molecular mass (in kDa) is shown to the left of the western blots.

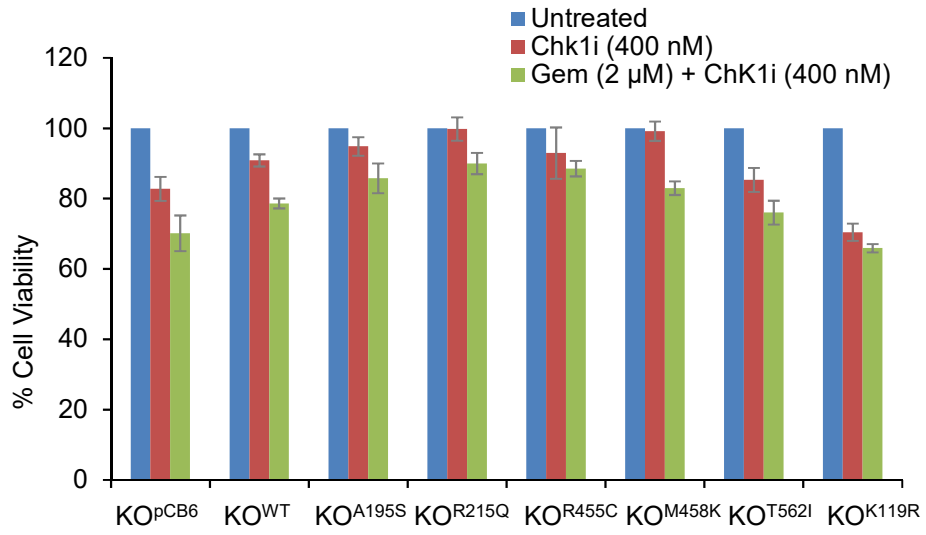

Figure S3: Sensitivity to ChK1i alone or a combination of Gemcitabine ± ChK1i in RECQ1-KO cells complemented with vectors encoding RECQ1 wildtype or missense mutants. For ChK1i treatment the cells were treated with ChK1i (400 nM for 24 h), for combination treatments, the cells were treated with gemcitabine (2 μM for 2 h) followed by subsequent treatment with ChK1i (400 nM for 24 h). Cell viability was measured by CCK-8 reagent and percent cell viability was calculated by normalizing to untreated.