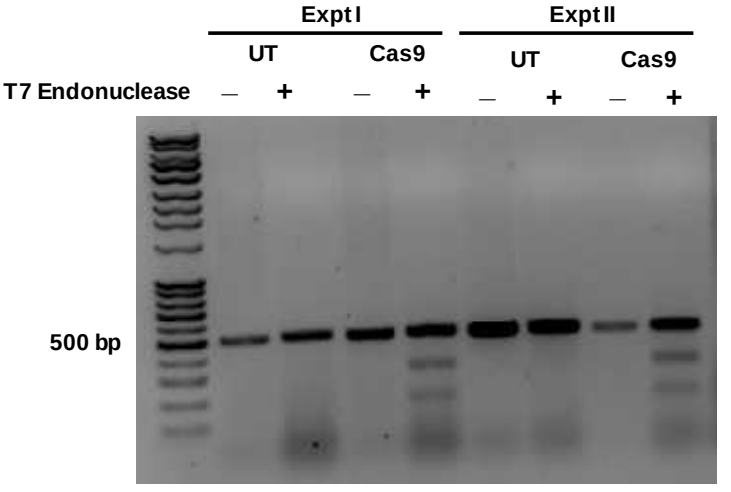
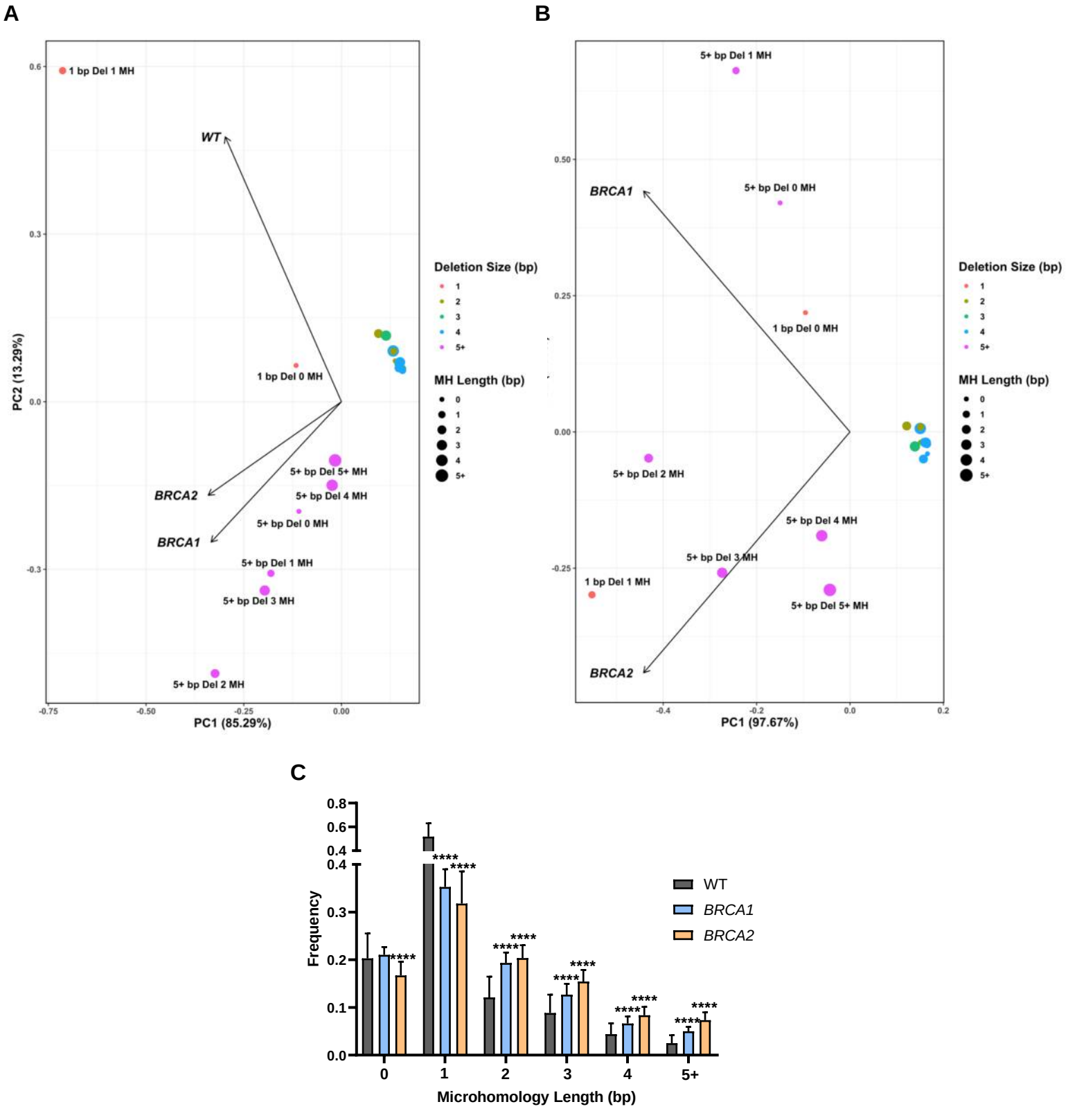
Sup. Figure 1



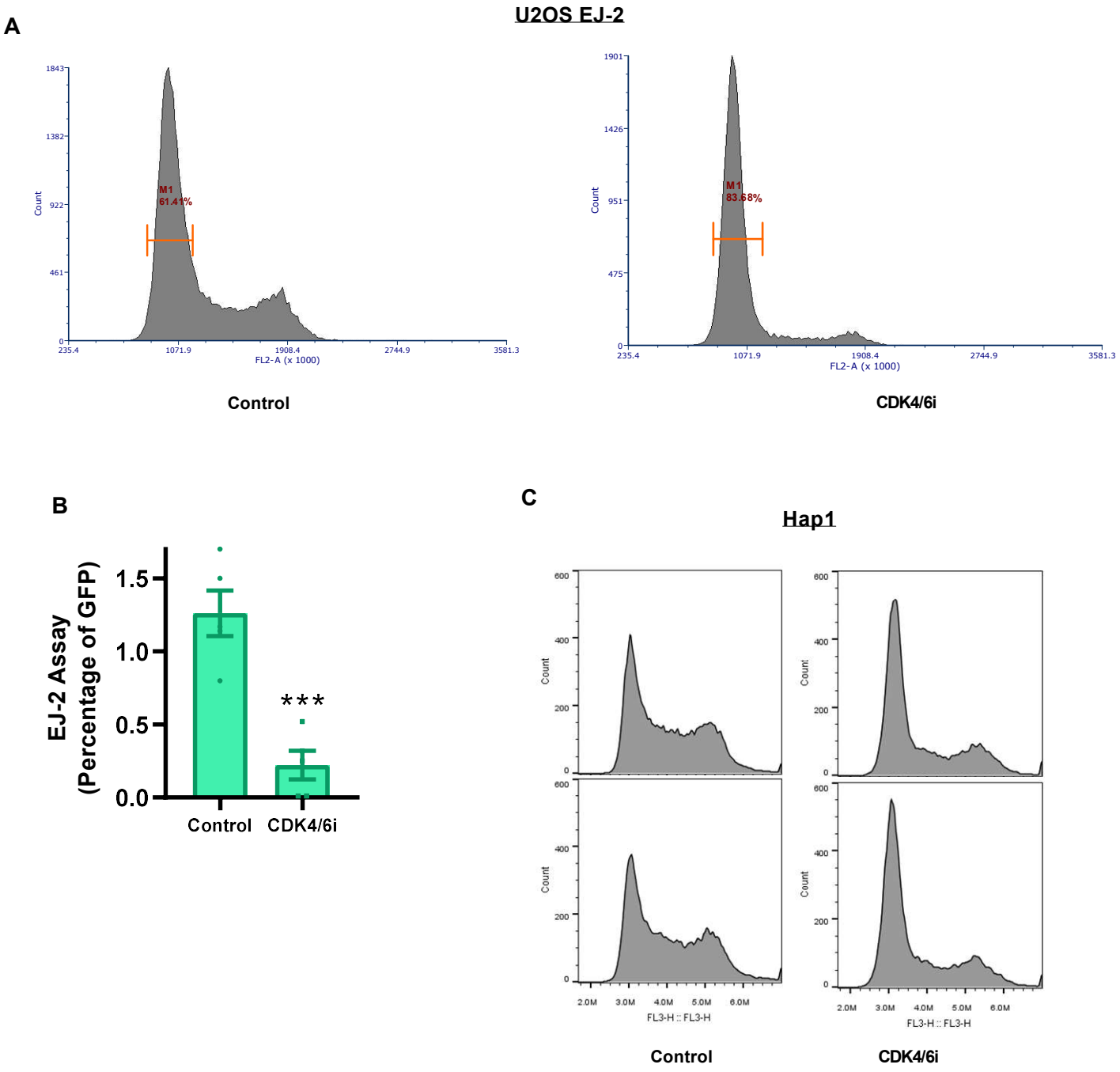
Supplmentary Figure 1. Confirmation of Cas9 cleavage at AAVS1 locus using the T7 endonuclease assay. T7 assay performed in untransfected and AAVS1-Cas9 plasmid treated HEK293T cells confirms cleavage only in the Cas9 treated sample.

Sup. Figure 2



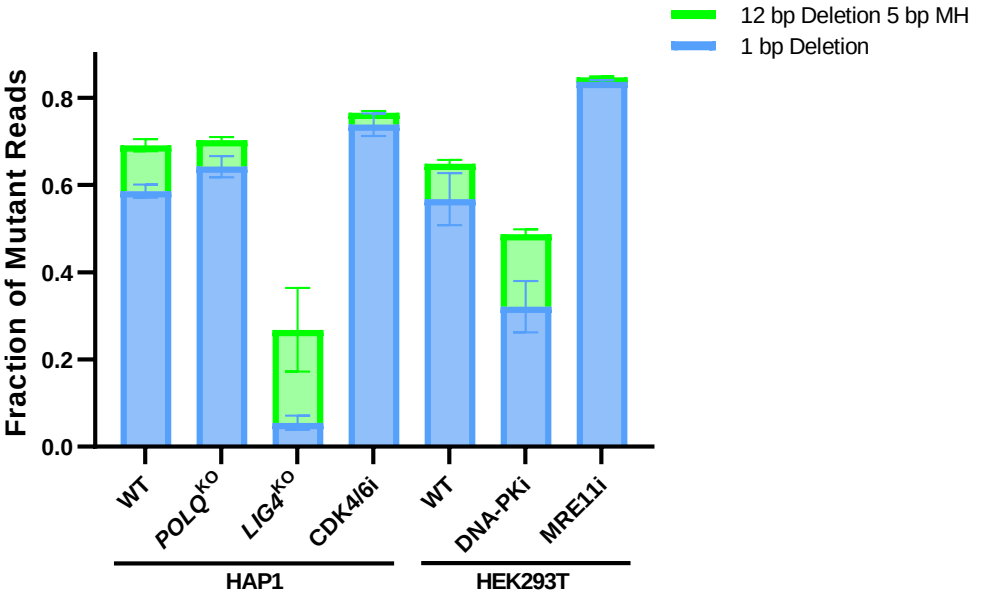
Supplementary Figure 2. Deletions size and microhomology length analysis in *BRCA1* and *BRCA2* biallelic mutated cancer patients. A) PCA of deletion size and microhomology length identified by PCAWG in *BRCA1* and *BRCA2* biallelic mutated prostate, pancreatic, breast, and ovarian cancer patients. **B**) Frequency of deletions with varying microhomology lengths identified by PCAWG in WT, *BRCA1*, and *BRCA2* biallelic mutated prostate, pancreatic, breast, and ovarian cancer patients. Asterisks signify t-tests as follows: * P≤0.05, ** P≤0.01, *** P≤0.001, **** P≤0.0001. **C**) Principal component analysis (PCA) of deletion size and microhomology length identified by the Pan-Cancer Analysis of Whole-Genomes (PCAWG) consortium in WT, *BRCA1*, and *BRCA2* biallelic mutated prostate, pancreatic, breast, and ovarian cancer patients.

Sup. Figure 3



Supplmentary Figure 3. CDK4/6i arrests cells in G1 and causes an Alt-EJ defect in EJ-2 assay. A) Cell cycle analyis performed in U20S EJ-2 controls cells and after treatment with 1 μM CDK4/6i for 48 hours. **B)** EJ-2 assay perfomed in U2OS EJ-2 cells in control cells and after treatment with 1 μm CDK4/6i for 48 hours after transfection with the I-Sce-I plasmid. **C)** Cell cycle analysis performed in Hap1 WT control cells and after treatment with 1 μM CDK4/6i for 48 hours.

Sup. Figure 4



Supplmentary Figure 4. Fractions of 1 bp deletion and 12 bp deletions obtained from Next Generation Sequencing used to calculate effect of the various genetic and pharmacoogical controls used in Figure 3E and 3F.