Comprehensive assessment of gene mutations revealed overlapping responses for PARPi and chemotherapy in ovarian cancer cells

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**Background:** PARP inhibitors (PARPi) have revolutionized the therapeutic landscape of epithelial ovarian cancer (EOC), showing outstanding benefits in regard to progression-free survival, especially in patients carrying BRCA1/2 mutations or harboring defects in homologous recombination (HR) repair. However, it remains uncertain which PARPi to choose and how to select responders by using clinical and molecular characteristics, especially in frontline therapy when platinum sensitivity is unknown.

**Materials and Methods:** Through a systematic literature review and the exploration of publicly available CRISPR-Cas9 library screens and Genomics of Drug Sensitivity in Cancer data, we identified potential genes linked with PARPi response. Using a CRISPR-Cas9 mutagenesis assay, we functionally tested 33 genes for PARPi and carboplatin response in six EOC cell lines.

**Results:** ATM was the only tested gene that induced olaparib sensitivity in a cell line-independent manner. Acquired olaparib sensitivity was also observed upon Cas9-mediated loss of MUS81, NBN, RAD51B/C, RNASEH2A, PALB2, XRCC1, and XRCC3 in at least 3 out of 6 cell lines. As the major survival benefit of PARPi treatment was reported in chemo-sensitive tumors, we next assessed the effect of top candidate genes on olaparib, niraparib, talazoparib, and carboplatin response. Interestingly, we observed identical effects in a gene- and drug compound-independent manner on acquired drug sensitivity, supporting the strong correlation of cancer cell response to PARPi and chemotherapy. In contrast, we identified CDK12 as an essential gene for cell proliferation/survival in ovarian cancer cells, independent of PARPi.

**Conclusions:**
- General mechanism of response to PARPi and chemotherapy as demonstrated by various overlapping gene dependencies
- The genetic screen of the genes identified as correlated with PARPi sensitivity may allow better stratification of patients with increased benefit to this treatment

**Figure 1:** Depiction of the CRISPR-Cas9 cell competition assay. Including assessment and quantification of alterations induced and PARPi response confirmed by the gene editing. Dropout values represent the fold-change of EGFP+ cells for up to six continuous cell culture passages evaluated by flow cytometry every 4 days relative to the 60G% percentage at passage 0. Percentage was measured 3 days after transient transfection with gRNAs. Cutoff WTs triggering the human genome wide barley DKH5 (non-essential) and RNAs for KI/KO potential genes were used as negative and positive controls throughout all experiments performed, respectively.