Poster 20: Establishing a platform for investigating poly (adenosine diphosphate-ribose) polymerase (PARP) inhibitor sensitivity in PARP1 knockout cells

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Topic: Ovarian

Objectives

Poly (ADP-ribose) polymerase (PARP) inhibitors have a unique mechanism of action based upon synthetic lethality, theoretically leading to minimal side effects. However, significant adverse effects are seen with PARP inhibitor (PARPi) use. The FDA has approved three PARP inhibitors for ovarian cancer therapy (olaparib, niraparib, rucaparib). More recently, a next-generation highly selective PARP1 inhibitor (AZD5305) was shown to present anti-proliferative effects in BRCA-mutant xenograft models. Here we generate a genetically-defined platform to determine the effect of PARP1 knockout (KO) to PARP inhibitors.

Methods

CRISPR/Cas9-engineered PARP1 KO high-grade serous ovarian cancer cells (HEYA8) and human haploid chronic myelogenous leukemia (HAP1) cells were utilized. Isolated clones were confirmed by immunoblotting and DNA sequencing. For the viability assays, cells were subjected to short-term (4 days) and long-term (7 days) treatment with PARPis (olaparib, talazoparib, AZD5305) and viability was measured by MTT and Crystal Violet, respectively.

Results

Two independent HEYA8 PARP1 KO clones and a single HAP1 PARP1 KO clone were obtained. All clones harbor unique frameshift mutations at exon 3 of PARP1 and were confirmed to be negative for PARP1 protein expression. Overall, PARP1-deficient cells were more resistant to PARPi treatment when compared to their parental counterparts. HAP1 PARP1 KO cells presented less than 19% and 60% viability at high concentrations (10 μM) of talazoparib and AZD5305, respectively. Similar results were observed for HEYA8 cells with viability lower than 30% and 73% in PARP1-deficient clones, versus 7% and 42% viability in parental cells treated with 10 μM of talazoparib and AZD5305.

Conclusions

Our genetically-defined PARP1 KO platform shows that PARP1 KO cells exhibit resistance to overall PARP1 inhibition. However, reduced viability is observed in higher concentrations, suggesting a polypharmacology effect that could explain the adverse side effects of PARP inhibitors observed clinically.