The PARP-1-DDX21-ribosome biogenesis axis is an alternative pathway targeted by PARP inhibitors in ovarian cancer

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Topic: Ovarian

Objectives
Clinical trials have shown ovarian cancers respond to niraparib regardless of homologous recombination status suggesting a mechanism beyond DNA repair-based synthetic lethality. Prior work shows that the snoRNA-PARP-1-DDX21-ribosome biogenesis axis provides an alternative cellular pathway that can be targeted by PARPi for therapeutic benefits, irrespective of BRCA1/2 status in breast cancer cells. The objective of this study was to determine the effect niraparib has on the PARP-1-DDX21-ribosome biogenesis axis in homologous recombination proficient ovarian cancer cell lines.

Methods
All experiments were carried out using three HRp ovarian cancer cell lines: OVCAR 3, OVCAR 4, and HCC 5012. Western blotting was used to confirm the presence of DDX21 in ovarian cancer cells. DDX21 staining before and after treatment with niraparib was identified using immunofluorescent antibodies against DDX21 visualized on a confocal microscope. Level of transcription before and after treatment with niraparib was evaluated using qPCR analysis of the ribosomal subunits 18S rRNA, 28S rRNA, and 45S pre-rRNA. Translational effect before and after treatment with niraparib was evaluated using puromycin incorporation assay. Cell viability assays performed using multiple niraparib concentrations with a crystal violet staining assay after incubation for 48 hours. Statistics performed using GraphPad Prism version 9.0.

Results
DDX21 is expressed in ovarian cancer cells and did not show decreased expression after treatment with niraparib. Treatment of OVCAR 3, OVCAR 4, and HCC 5012 cells with niraparib caused decreased nucleolar translocation of DDX21 without disrupting the nucleolar membrane (Figure 1). Niraparib treatment caused a decrease in transcription of the ribosomal subunits 18S rRNA, 28S rRNA, and 45S pre-rRNA in all cell lines. Diminished transcription of ribosomal subunits lead to decreased protein translation in all cell lines. Increasing concentrations of niraparib were found to cause increasing levels of cell death in all ovarian cancer cell lines.

Conclusions
Our data provides a mechanism by which niraparib causes increased cell death in HRp ovarian cancer cells. We have shown that niraparib treatment causes delocalization of the RNA helicase DDX21 to the nucleolus causing impaired transcription of ribosomal subunits leading to diminished protein translation and, ultimately, cell death.

Abstract Table or Graph
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