

## INDIANA UNIVERSITY

SCHOOL OF MEDICINE

# Epigenome and Genome Alterations in Platinum Resistant Ovarian Cancer

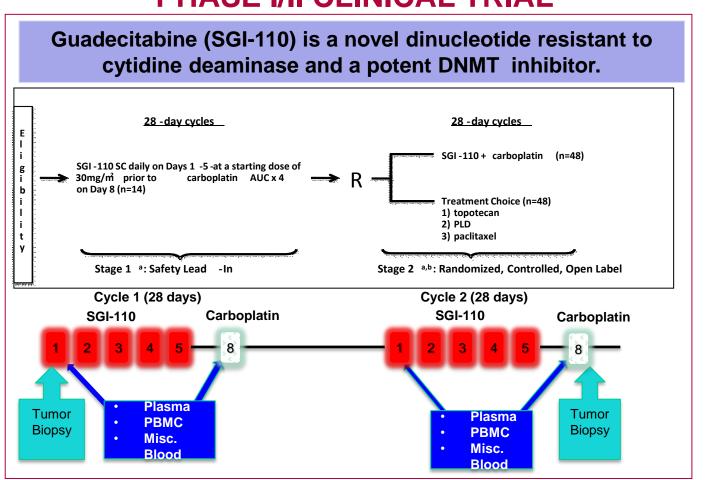
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## **ABSTRACT**

**Background**: Epigenetic changes, particularly in DNA methylation, have been implicated in acquired resistance to platinum in ovarian cancer (OC). **Methods**: An ongoing phase I/II multi-institutional clinical trial uses the novel DNA methyltransferase (DNMT) inhibitor guadecitabine (SGI-110) to re-sensitize recurrent platinum resistant OC to carboplatin. Patients enrolled in this trial had recurrent platinum resistant OC and multiple lines of prior therapy. Tumor biopsies were collected at baseline and after two cycles of guadecitabine administered daily for 5 days in low dose (30mg/m<sup>2</sup>). The goal of the current study was to analyze and integrate global RNA expression and DNA methylation profiles of platinum resistant tumors and to measure genomic and epigenomic changes induced by guadecitabine in tumors. RNA and DNA were extracted from 48 and 57 baseline tumors and analyzed using next generation sequencing (RNAseq) and Infinium Human Methylation450 (HM450) arrays, respectively. Differential gene expression and DNA methylation profiles were generated and used for Ingenuity Pathway Analysis (IPA) to identify the top altered pathways in response to guadecitabine. Results: Analysis of a limited number of paired samples before and after treatment (n=8) revealed significant changes in global gene expression profiles induced by SGI-110, with 960 altered genes representing immunopathway enrichment including: cytokine production in macrophages and T helper cells by IL-17A and IL-17F, granulocyte /agranulocyte adhesion and inflammation, IL-8 signaling, p38 MAPK signaling, cAMP-mediated signaling, and innate immunity. HM450 analysis showed a greater number of hypermethylated genes in baseline tumors compared to primary OC samples in The Cancer Genome Atlas (TCGA) and demethylation (decreased β-values relative to baseline) of a large number of loci (381 gene promoters) after guadecitabine treatment. IPA analysis of baseline tumor transcriptome and methylome demonstrated significant enrichment in a wide range of pathways associated with cancer, stem cells, inflammation and the immune system. **Conclusions:** These data suggest that treatment with a DNMT inhibitor induces a reactivation of immune responses in human OC. Correlations between methylation changes and expression profiles are being explored.

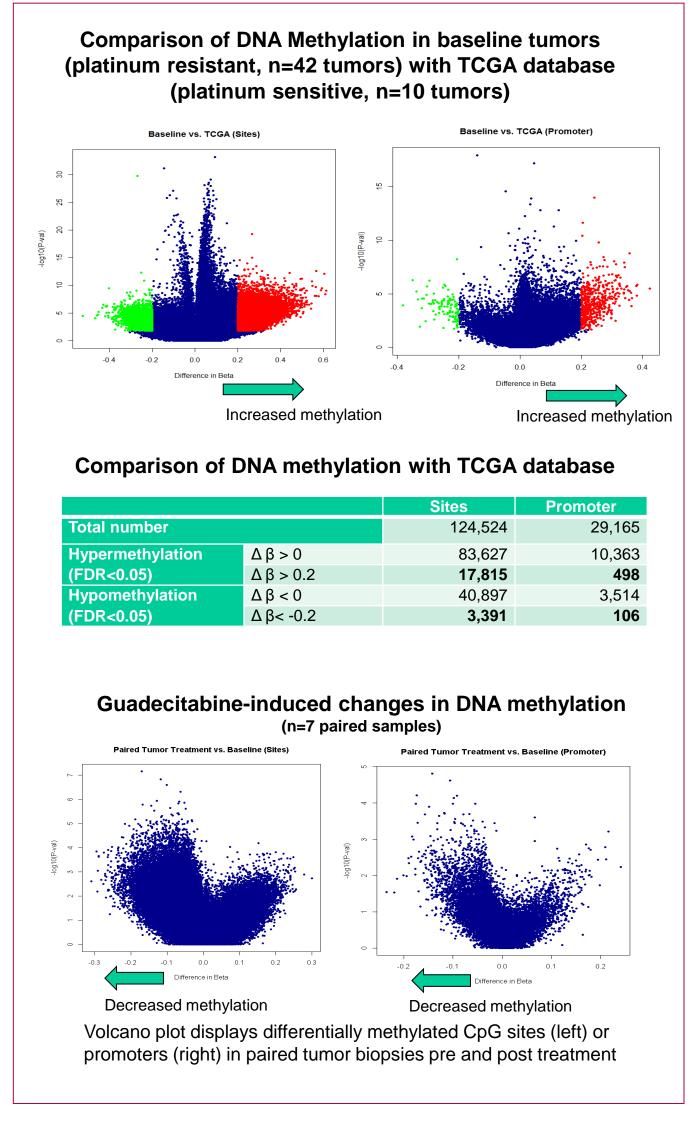
## PHASE I/II CLINICAL TRIAL

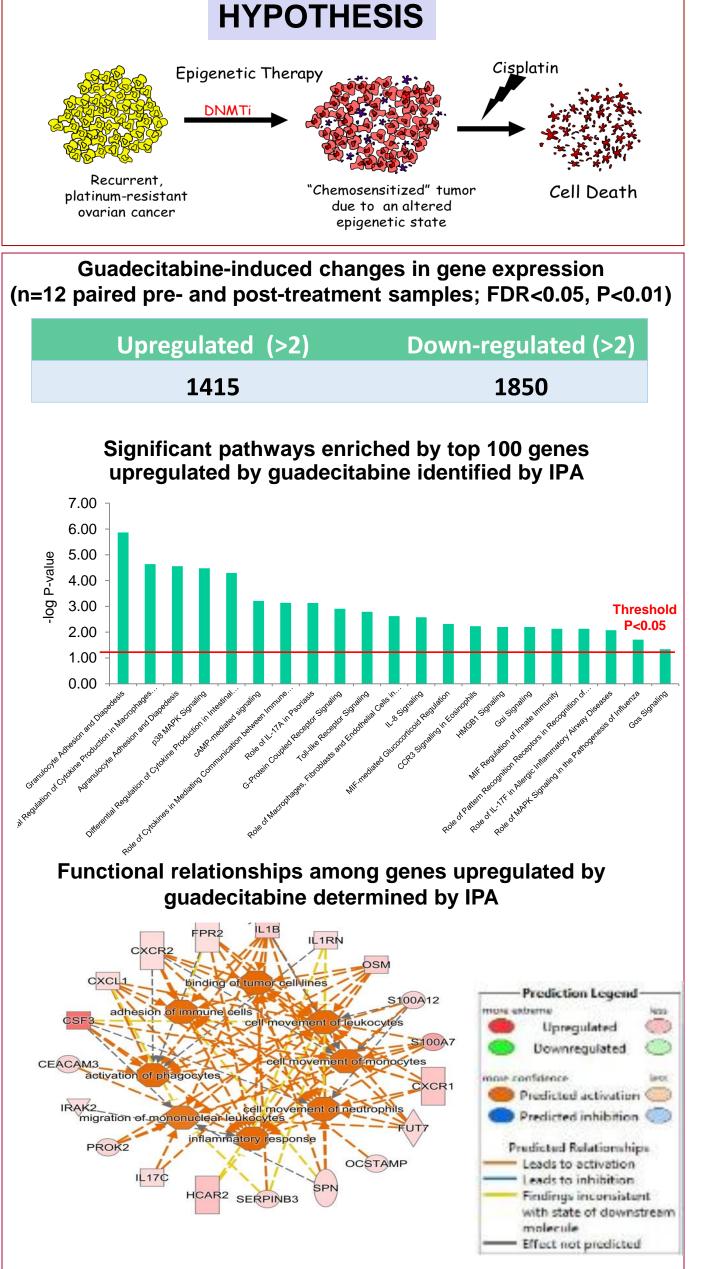


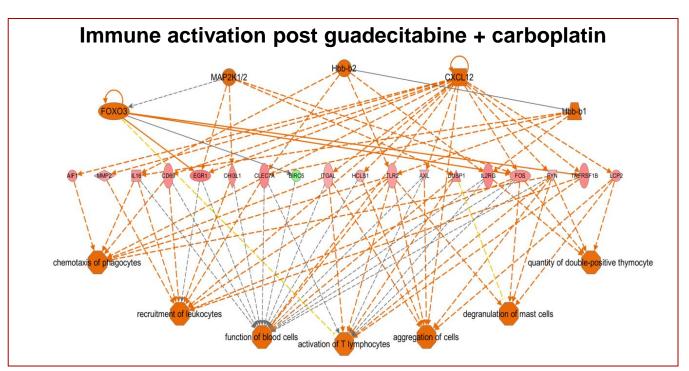
#### MATERIALS AND METHODS

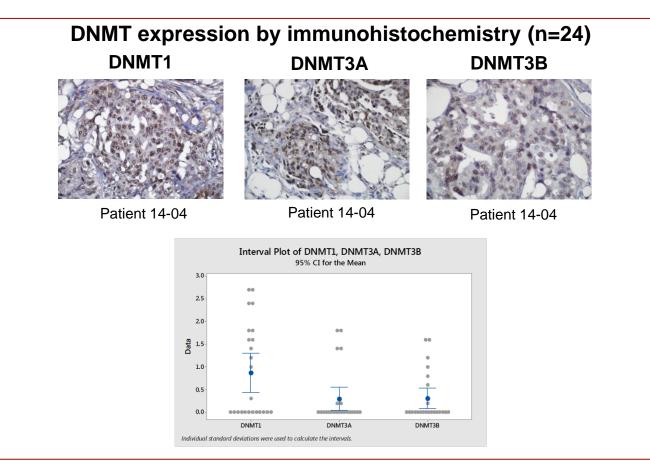
- DNA Methylation measured by Infinium HumanMethylation450 array (485,577 methylation sites) and by pyrosequencing.
- mRNA expression levels measured by RNAseq and real-time RT-PCR.
- DNMT protein expression measured by immunohistochemistry.

## **RESULTS**









## **CONCLUSIONS**

- 1. Increased DNA methylation is observed in platinum resistant tumors collected in this trial compared to platinum naïve tumors analyzed by the TCGA project.
- 2. Guadecitabine induced significant hypomethylation and alterations in gene expression in ovarian tumors.
- 3. Treatment with guadecitabine induced a reactivation of immune pathways in ovarian tumors.
- 4. Baseline DNMT expression levels are detectable, but highly variable among sampled tumors.

#### **ACKNOWLEDGMENTS**

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