Discovering New Drug Targets for Ovarian Cancer Patients

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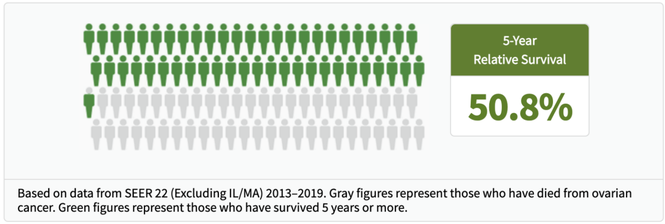
INTRODUCTION

- Ovarian cancer accounts for 2.5% of cancers in women
- It is the 11th most common cancer among women
- It is the 5th leading cause of cancer-related deaths in women. (1)



Based on data from SEER 22 (Excluding IL/MA) 2013–2019. Gray figures represent those who have died from ovarian cancer. Green figures represent those who have survived 5 years or more.

Due to drug resistance, majority of metastatic disease is incurable
Preclinical models have emerged as a useful resource to study cancer mechanisms and predict efficacy of anticancer drugs
Multicellular spheroids (MCS) serve as an alternative to patient-derived xenograft models for drug testing. (2)

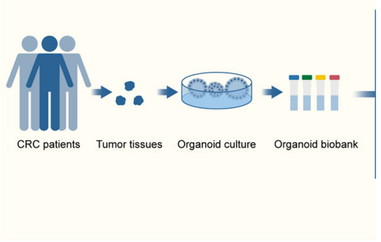


Figure 2: Flow diagram of collection of organoids. Adapted from Frontiers (3).

Through RNA-seq analysis of MCSs, a 2020 study found that there was significant up-regulation of genes related to cellular proliferation, epithelial-mesenchymal transition, and KRAS signaling pathways (4)
These four genes are reported to be most commonly mutated in epithelial ovarian cancer TP53, BRCA1/2, PIK3CA, and KRAS(5)

HYPOTHESIS + RESEARCH QUESTION

We predict that genes involved in Reactome Pathways of Disease, Gene Expression, and Signal Transduction would be the most dysregulated (pathways common to 4 genes in intro).
Are any of the top dysregulated genes not current drug targets for cancer treatment?

METHODS

THE DATA
Read counts of genes through RNA-sequencing at two time points (Day 0 and Day 6) for four different samples (A778, A820, A870, and A899).

Data Cleaning + Processing
1. 8 TSV files combined into 1 table
2. Count and meta data made for DESeq analysis
3. DESeq Analysis run with R
4. Genes ordered by most to least differentially expressed

Analysis + Visualizations
1. Plots for Exploration
   a. Volcano plots
   b. Mean-Difference Plots
   c. Heatmaps
2. Find Reactome Pathways (6) of most differentially expressed genes
3. Using Drug Gene Interaction Database (7), identify genes (with padj < 0.001) with no known drug interactions

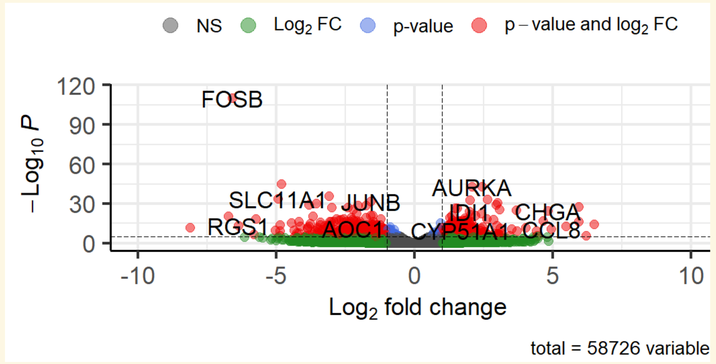


Figure 3: Volcano Plot showing the Distribution of Up and Down Regulated Genes in MCS of Ovarian Cancer Patients.
Red values show which genes had both a significant p-value and a significant log2FC. This volcano plot is labelled the genes that were most significantly differentially expressed. The total number of genes is also seen at the bottom, with 58, 726 genes examined.

RESULTS

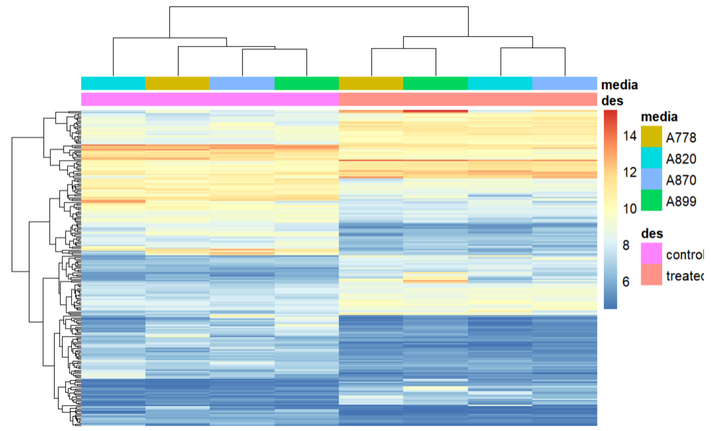


Figure 4: Heatmap Comparing Gene Expression Levels Between Samples
The figure above depicts the heatmap for the count matrix of the gene expression levels in multicellular spheroids of ovarian cancer patients. In this figure we can see that many genes had different expression levels between the control (the pink columns) and the treatment group (salmon columns). Especially visible in the upper half of the heatmap, we can see that the top section of genes were more highly expressed in the treated group, and the section slightly below that was less expressed.

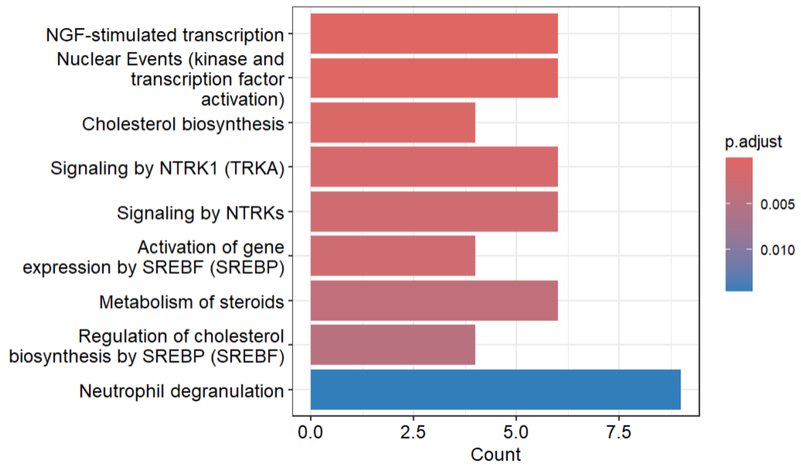


Figure 5: Bar Graph Representing Count of Genes in Each Reactome Pathway
The graph above shows the 9 subcategories of Reactome Pathways that the top 74 most differentially expressed genes take part in. These 9 subcategories fit into 3 main categories, Signal Transduction, Metabolism, and Immune System.

CONCLUSION

74 genes had an adjusted p value of <0.001 (Figure 3)
Using the ReactomePA R package, can see that the most dysregulated genes were those involved in Neutrophil Degranulation (Immune System pathway), NGF-stimulated transcription (Signal Transduction), and Metabolism of Steroids (Metabolism pathway) -> one of which matches out hypothesis (Figure 5).
According to the Drug Gene Interaction Database, 46 of the top 74 genes don't have any drug interactions.
Further research should be done to explore these genes as potential targets for ovarian cancer treatment

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