

Predictive Analysis and Significance of Potential Drug Resistance Genes of Cisplatin in Ovarian Cancer

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Abstract

Objective: To use bioinformatics methods to screen the differentially expressed genes of cisplatin resistance in ovarian cancer, and to conduct relevant biological analysis of the differentially expressed genes to obtain molecular markers related to cisplatin resistance in ovarian cancer. **Methods:** The GSE45553, GSE33482 and GSE739353 ovarian cancer cisplatin resistance-related microarray data sets were found from the GEO database, and the GEO2R online analysis tool was used to draw a volcano map and screen the genes with poor expression up-regulated. Obtain from the DAVID database, perform GO function enrichment analysis on significantly up-regulated differential genes to understand their molecular functions, participating biological processes and pathways, and cellular components and locations. At the same time, perform KEGG signaling pathway enrichment analysis to predict their possible The mechanism of action, and a Venn diagram to obtain the intersection; construct a protein-protein interaction (PPI) network diagram for genes with significant differential expression up-regulation, and obtain the hub gene (Hub gene) after analysis; use Kaplan-MeierPlotter to analyze the Hub gene in the ovary online Survival curve in cancer. **Results:** Three data sets were screened out and 77 up-regulated differential genes were analyzed and enriched. Through analysis, we obtained 5 Hub genes: NCOR2, PIK3R1, VCL, NCOA3, ZNF217. Survival curve analysis showed that the increased expression of the three genes NCOR2, NCOA3, and ZNF217 significantly reduced the overall survival rate of patients with ovarian cancer ($P < 0.05$). **Conclusion:** The three genes NCOR2, NCOA3, and ZNF217 have increased expression of cisplatin resistance in ovarian cancer, which may be molecular markers of cisplatin resistance in ovarian cancer, and they can be used for the diagnosis and treatment of cisplatin resistance in ovarian cancer. The prognosis judgment provides a reference.

Keywords

Ovarian cancer; Cisplatin resistance.

1. Introduction

According to estimates by the World Health Organization, by 2020, the global incidence of ovarian cancer will be 308,069, and the total mortality rate of ovarian cancer will be 193,811 [1]. Ovarian cancer is one of the major gynecological diseases with high mortality worldwide. As one of the most common gynecological cancers, they have the highest mortality rate, ranking third, second only to cervical cancer and uterine cancer [2]. At present, it has made extensive progress in the field of chemotherapy and surgery, but the anatomical location of the ovary is special, and the onset of ovarian cancer is hidden. However, due to lack of early disease or non-specific symptoms, there is a lack of early clinical screening methods, although it is necessary to develop effective treatment methods. Various efforts have been made, but more than 75% of ovarian cancer cases are not detected until the advanced stage, the 5-year survival rate is low [3], and its overall survival rate (OS) is still less than 50%. The main first-line drugs for conventional chemotherapy are Paclitaxel and platinum drugs. Multidrug resistance (MDR)

means that while cells are resistant to a drug, they are also resistant to other anti-tumor drugs with similar or different structures and targets [4-6]. At the same time, the stubborn drug resistance of ovarian cancer makes traditional treatments very limited, and the molecular mechanism of tumor progression is still unclear. Therefore, more basic research on ovarian cancer and ovarian cancer drug resistance is needed.

Cisplatin, a common chemotherapeutic drug, is a drug often used to treat metastatic BC that exerts its effects by inducing the formation of interstrand crosslinks between DNA chains[7, 8]. Cisplatin causes DNA intrastrand and interstrand cross-links (ICLs) by forming cisplatin-DNA adducts. Intrastrand cross-links are removed by the nucleotide excision repair (NER) mechanism [9]. Resistance to cisplatin can be attributed to three molecular mechanisms, including altered accumulation of drug in cells, cytosolic inactivation of drug and altered DNA damage response with enhanced DNA repair process. Among these mechanisms, enhanced DNA repair is the most important cause of cisplatin resistance [10].

Bioinformatics is based on computer simulation methods to compare and screen gene chip data for gene mining and function prediction [11]. Gene chip (gene chip) is also called biochip. The basic principle of gene chip detection is hybridization, that is, the principle of base complementary pairing. A matches T and C matches G. If the sequences of two nucleic acid strands can be matched, they form base complementary pairings. In molecular biology, it is called hybridization. In recent years, due to the rapid development of computer science, biology, etc., various databases have emerged, and bioinformatics analysis has also developed rapidly. Gene Expressed Omnibus (GEO), as one of the largest collections of tumor genome data, not only includes a large amount of clinical data, but also contains a large amount of related genetic information. In this article, we use bioinformatics methods to identify genes related to cisplatin resistance in ovarian cancer chemotherapy and predict their target genes to analyze potential targets for cisplatin resistance in ovarian cancer.

2. Materials and methods

2.1. Data Set Acquisition

Search the GEO database (<https://www.ncbi.nlm.nih.gov/geo/>) with the keywords "ovarian cancer" and "drug resistance" and screen cell lines with cisplatin resistance and sensitivity as controls for ovarian cancer and The species "human" is the research object. After screening, we obtained the following three databases: GSE45553, GSE33482 and GSE73935. DEGs were obtained based on the GEO database. GSE45553 selected two cisplatin-resistant groups and one cisplatin-sensitive group, and GSE33482 selected three cisplatin-resistant groups and three in the cisplatin-sensitive group, GSE73935 selects six cisplatin-resistant groups and three cisplatin-sensitive groups. The screening criteria for significant DEGs in each data set were set as: ① $P < 0.05$; ② $\text{Log}_2|\text{FC}|(\text{foldchange}) > 1$ (that is, the multiple of two-fold difference). Use (<http://bioinformatics.psb.ugent.be/webtools/Venn/>) online software to analyze the significant DEGs of the above three data sets to make the intersection of the Venn diagram.

2.2. Enrichment Analysis of Biological Process and Pathway

Use DAVID (<https://david.ncifcrf.gov/>) online analysis tool. Perform GO functional enrichment analysis on significant DEGs to understand their molecular functions, biological processes and pathways involved, and cellular components and location. At the same time, perform KEGG signaling pathway enrichment analysis to predict their possible mechanism of action, and $P < 0.05$ As the inclusion criteria.

2.3. Construct PPI Network Diagram and Screen Hub Gene

The significant DEGs obtained were imported into the Sting database (<https://cn.string-db.org/>), the screening criteria for the existence of high correlation between proteins were set

as: $P < 0.05$ and Cytoscape 3.7.2 was used to construct potential targets and disease targets Point of protein-protein interaction (PPI) network.

2.4. Survival Analysis of Hub Gene

Kaplan-Meier Plotter (<http://kmplot.com/analysis/>) was used to analyze the relationship between Hub gene and ovarian cancer survival online. The survival analysis results were based on $P < 0.05$ as the inclusion criteria.

3. Results and Discussion

3.1. Identification and Functional Identification of up-regulated DEGs in Cisplatin-resistant Ovarian Cancer Cells

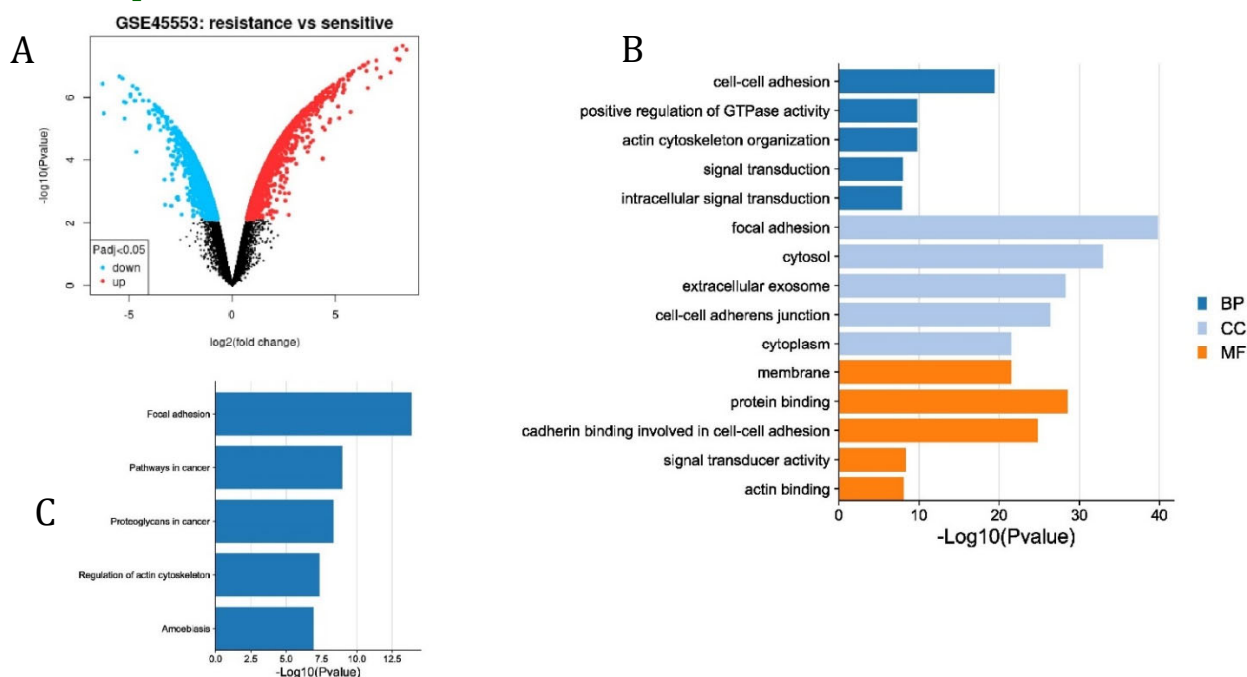


Figure 1. Identification and characterization of DEGs from the GSE45553 dataset. (A) The DEGs volcano graph between the cisplatin resistant group and the sensitive group in ovarian cancer. Red dots are significantly up-regulated DEGs; blue dots are significantly down-regulated DEGs; black dots are genes with no significant difference ($P < 0.05$ and $|\log_2FC| > 2$ are used as thresholds). (B) Enrichment analysis of cisplatin-resistant up-regulated gene Go in ovarian cancer. (C) And KEGG signaling pathway enrichment.

In order to determine the potential genes that cause cisplatin resistance in ovarian cancer, we use ① $P < 0.05$; ② $\log_2|FC|$ (foldchange) > 1 (double difference multiple) as the standard setting for screening differential genes, and perform GSE45553, GSE33482 and GSE73935 Research and draw a volcano map of gene expression in the dataset, with red for up-regulated genes and blue for down-regulated genes. By processing the GSE45553 data set, we screened 7553 DEGs, of which 3571 were up-regulated and 3982 were down-regulated (figure 1A). In order to explore the functions of these significantly up-regulated DEGs, we performed GO function enrichment analysis and KEGG signal pathway analysis. GO functional enrichment analysis results show that significant DEGs are mainly involved in: cell-cell adhesion, positive regulation of GTPase activity, actin cytoskeleton organization, signal transduction, intracellular signal transduction; focal adhesion, cytosol, extracellular exosome, cell-cell adherens junction, cytoplasm, Membrane, protein binding, cadherin binding involved in cell-cell adhesion, signal

transducer activity, actin binding (figure 1B), KEGG signaling pathway analysis showed that these significantly up-regulated DEGs were mainly found in Focal adhesion, Pathways in cancer, Proteoglycans in cancer, Regulation of actin cytoskeleton, Amoebiasis (figure 1C).

Next we conducted research on GSE33482. A total of 10777 DEGs were screened, of which 6563 were up-regulated and 4214 were down-regulated (figure 2A). We performed GO function enrichment analysis and KEGG signal pathway analysis on the up-regulated DEGs. GO functional enrichment analysis showed that significant DEGs are mainly involved in transcription, DNA-templated, regulation of transcription, DNA-templated, negative regulation of transcription from RNA polymerase II, promoter, nucleus, MHC class I protein complex, intracellular, protein binding, DNA Binding, metal ion binding (figure 2B), and KEGG signaling pathway analysis showed that these significantly up-regulated DEGs were mainly found in Pathways in cancer and Herpes simplex infection (figure 2C).

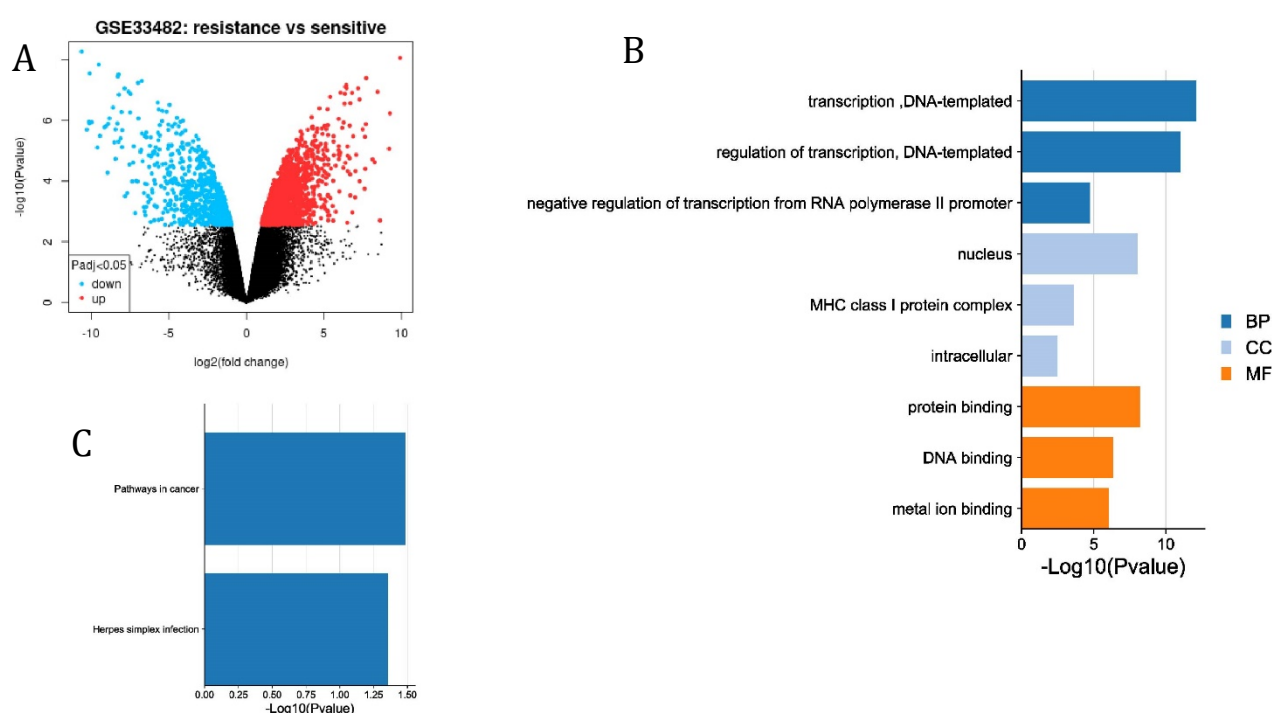


Figure 2. Identification and characterization of DEGs from the GSE33482 dataset. (A) The DEGs volcano graph between the cisplatin resistant group and the sensitive group in ovarian cancer. Red dots are significantly up-regulated DEGs; blue dots are significantly down-regulated DEGs; black dots are genes with no significant difference ($P < 0.05$ and $|\log_2FC| > 2$ are used as thresholds). (B) Enrichment analysis of cisplatin-resistant up-regulated gene Go in ovarian cancer. (C) And KEGG signaling pathway enrichment.

Finally, we conducted research on GSE73935. A total of 9906 DEGs were screened, of which 5205 DEGs were found to be significantly up-regulated and 4701 DEGs were down-regulated (figure 3A). We found that GO function enrichment analysis results of up-regulated DEGs showed significant DEGs were mainly involved in cell-cell adhesion, positive regulation of I-kappaB kinase/NF-kappaB signaling, negative regulation of transcription from RNA polymerase II promoter, xenobiotic glucuronidation, cell migration, Cytosol, membrane, extracellular exosome, cell-cell adherens junction, cytoplasm, protein binding, cadherin binding involved in cell-cell adhesion, ubiquitin protein ligase binding, signal transducer activity, actin binding (figure 3B). KEGG signaling pathway analysis showed that these significantly up-regulated DEGs were mainly found in Pathways in cancer, Regulation of actin cytoskeleton, p53 signaling pathway, B cell receptor signaling pathway, Acute myeloid leukemia. Through the

above analysis, the three data sets were intersected with significantly up-regulated differential genes to make a Venn diagram, and finally 77 co-expressed significantly differential genes were obtained (figure 3C).

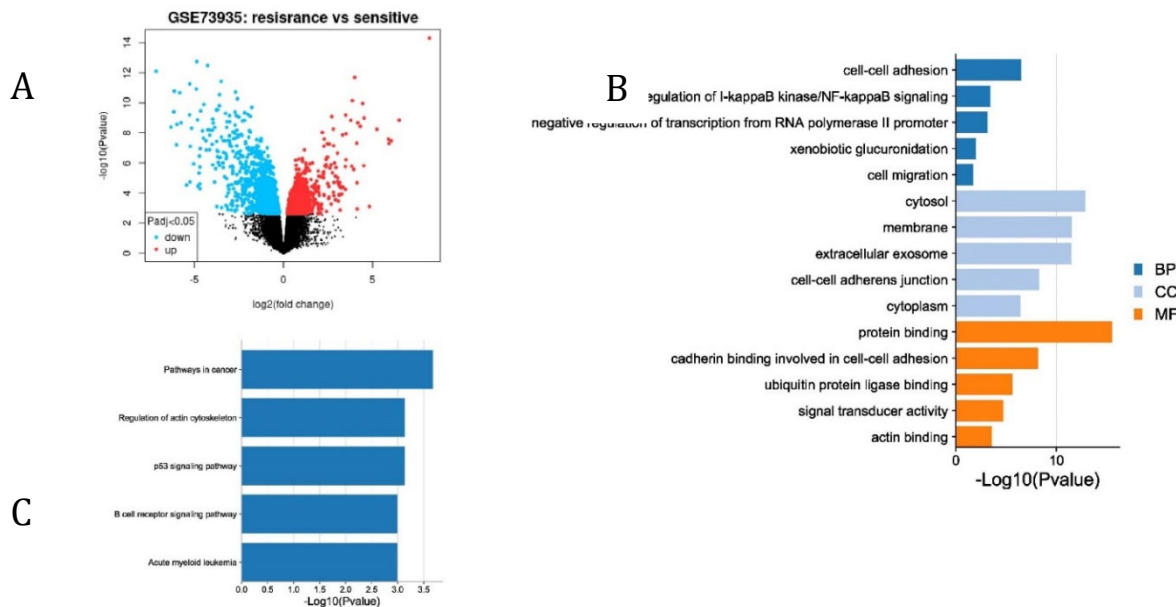


Figure 3. Identification and characterization of DEGs from the GSE73925 dataset. (A) The DEGs volcano graph between the cisplatin resistant group and the sensitive group in ovarian cancer. Red dots are significantly up-regulated DEGs; blue dots are significantly down-regulated DEGs; black dots are genes with no significant difference ($P < 0.05$ and $|\log_2FC| > 2$ are used as thresholds). (B) Enrichment analysis of cisplatin-resistant up-regulated gene Go in ovarian cancer. (C) And KEGG signaling pathway enrichment.

3.2. Significantly up-regulate DEGs to Construct A Protein Interaction Network and Analyze the 5 Hub Genes Obtained

Using the String database to analyze and screen the 77 significantly up-regulated DEGs, obtain the PPI network diagram with the interaction relationship, and analyze the gene modules with the highest degree of tightness (figure 4A). Finally, the Cytoscape software was used to process the PPI network to obtain 5 Hub genes: NCOR2, PIK3R1, VCL, NCOA3, ZNF217. These genes may be potential targets for cisplatin resistance in ovarian cancer and are related to the poor prognosis of patients (figure 4B).

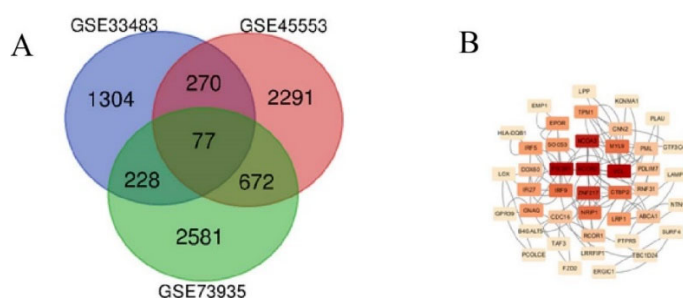


Figure 4. (A) Venn diagrams of significantly up-regulated DEGs from the GSE45553, GSE33482, and GSE73925 data sets. (B) The protein interaction network diagram and the gene module with the highest degree of tight connection. The darker the color, the higher the degree of connection.

3.3. Correlation between Hub gene and Overall Survival Rate

Using the Kaplan-Meier Plotter online website to analyze the correlation between the Hub gene and the survival time of ovarian cancer patients, it was found that NCOR2, NCOA3, and ZNF217 were significantly related to the survival time of ovarian cancer ($P < 0.05$) (figure 5A-E). It indicates that these genes may be potential targets for cisplatin resistance in ovarian cancer and are related to the poor prognosis of patients.

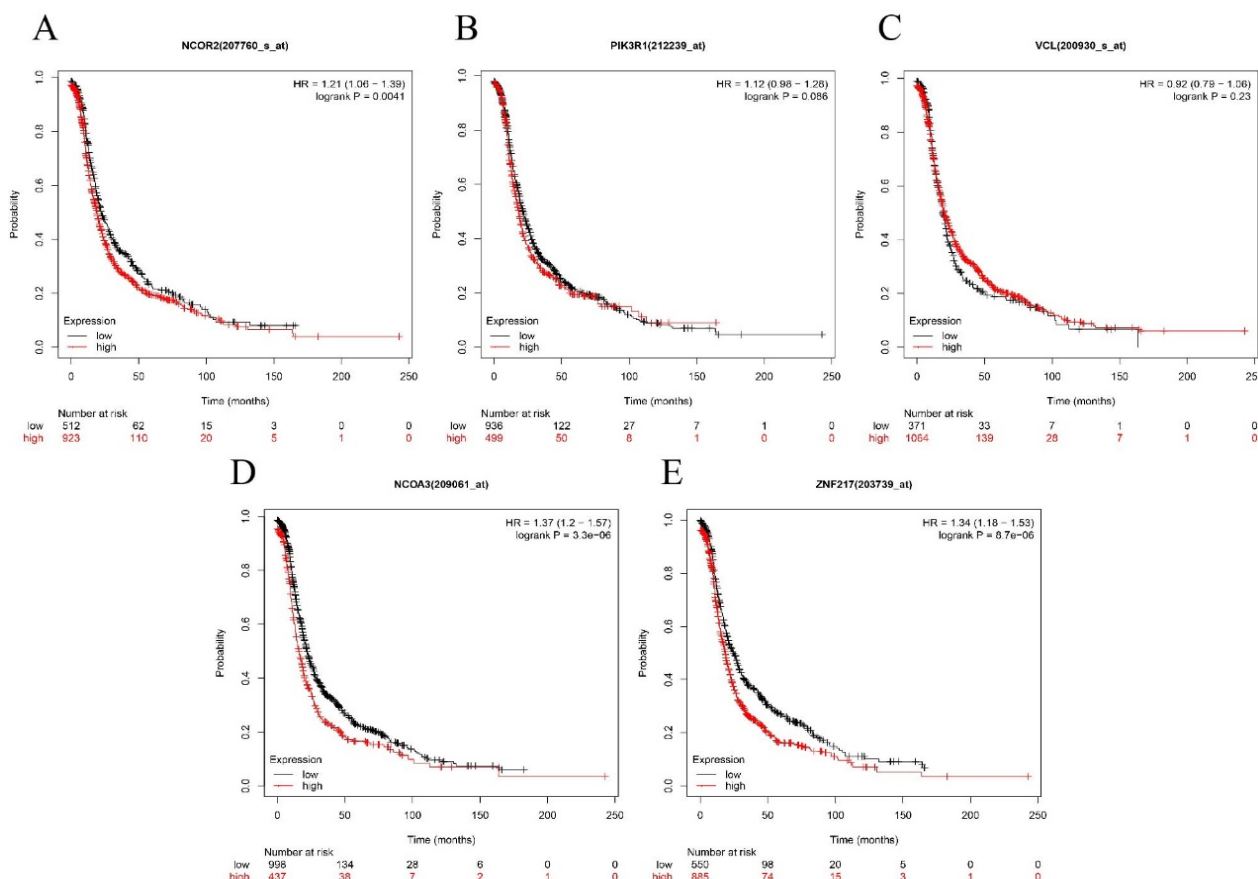


Figure 5. Kaplan-Meier Plotter analyzes the overall survival rate of patients. A to E are the genes NCOR2, PIK3R1, VCL, NCOA3, and ZNF217. Survival curves in ovarian cancer.

4. Conclusions

To sum up, this article analyzes the cisplatin resistance gene chip in ovarian cancer through GEO2R of GEO database, combined with DAVID online analysis tool, SRING database, Cytoscape software, Kaplan-Meier Plotter online website and other analysis methods, and finally obtained NCOR2, NCOA3, ZNF217. Genes are significantly related to the survival risk of ovarian cancer, and may be involved in cisplatin resistance. NCOR2, NCOA3, and ZNF217 genes may serve as potential targets for cisplatin resistance in ovarian cancer and are related to the poor prognosis of patients. Down-regulating the expression of these genes or finding effective inhibitors may improve the resistance of patients and increase patients' resistance to cisplatin. The sensitivity of platinum chemotherapy gives patients greater hope of survival.

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