Original Article

Identification of genes predicting chemoresistance and short survival in ovarian cancer

Cong Wang[1#], Cuilan Chen[1#], Xiaoying Chen[1], Jie Luo[1], Yuting Su[1], Xia Liu[2], Fuqiang Yin[1,3]

[1]Life Sciences Institute, Guangxi Medical University, Nanning, China; [2]Key Laboratory of Longevity and Ageing-Related Disease of Chinese Ministry of Education, Centre for Translational Medicine and School of Preclinical Medicine, Guangxi Medical University, Nanning, China; [3]Key Laboratory of High-Incidence-Tumor Prevention and Treatment (Guangxi Medical University), Ministry of Education, Nanning, China

Contributions: (I) Conception and design: X Liu, F Yin; (II) Administrative support: F Yin; (III) Provision of study materials or patients: F Yin; (IV) Collection and assembly of data: X Liu, C Wang, C Chen; (V) Data analysis and interpretation: X Liu, C Wang, C Chen, Y Su; (VI) Manuscript writing: All authors; (VII) Final approval of manuscript: All authors.

[#]These authors contributed equally to this work.

Correspondence to: Fuqiang Yin, PhD. Professor, Life Sciences Institute, Guangxi Medical University, 22 Shuangyong Rd., Nanning 530021, China; Key Laboratory of High-Incidence-Tumor Prevention and Treatment (Guangxi Medical University), Ministry of Education, Nanning 530021, China. Email: yinfq@mail2.sysu.edu.cn; Xia Liu, PhD. Professor, Key Laboratory of Longevity and Ageing-Related Disease of Chinese Ministry of Education, Centre for Translational Medicine and School of Preclinical Medicine, Guangxi Medical University, 22 Shuangyong Rd., Nanning 530021, China. Email: realliuxia@sina.com.

Background: Ovarian cancer (OC) is a kind of lethiferous cancer in gynecology, and the development of chemoresistance is the brief reason for treatment failure. The genes which contribute to chemoresistance are often leading to short survival. Thus, this study aims to identify predictive markers for chemoresistance and survival from chemoresistant-related genes.

Methods: Coremine was used to retrieve of genes linked to OC chemoresistance. The relationship of genes with patient survival was analyzed in 489 OC patients of The Cancer Genome Atlas (TCGA) cohort, which the subgroup of 90 resistant and 197 sensitive samples was used to determine gene expression. Kaplan-Meier (KM) plotter of 1,816 OC patients with survival data was retrieved for survival analysis. Survival analysis was carried out by the R survival package in R (version 3.3.1). KM and receiver operating characteristic (ROC) curve were respectively used to access the ability of a gene to predict survival and chemoresistance.

Results: In this study, a group of genes potentially linked to OC chemoresistance was identified, which dysregulated in 90 chemoresistant tissues compared with 197 sensitive tissues. Of them, thirteen genes could predict chemoresistance in 1,347 patients, especially *SOS1*, *MSH6*, *STAT5A* were excellent for predicting chemoresistance to any drugs, platin and taxane, *CASP2* and *PARD6B* for any drugs and platin, and *HSP90AA1* and *HSP90B1* for taxane. Meanwhile, 44 genes linked to OC chemoresistance could predict short overall survival (OS) and/or disease-free survival (DFS) in 489 OC patients, and 10 of them could predict short OS in large cohort of up to 1,657 patients. Finally, it is noteworthy that *CASP2* was down-regulated in 90 chemoresistant samples, and low expression of the gene predicted chemoresistance in 1,347 patients, short OS and DFS in 489 patients, and short OS and progression-free survival (PFS) in 1,657 patients.

Conclusions: The identified genes specifically the *CASP2* might be potentially used as predictive marker, prognostic marker and therapeutic target in management of OC.

Keywords: Ovarian cancer (OC); chemoresistance; predictive markers

Submitted Nov 22, 2023. Accepted for publication Jun 21, 2024. Published online Aug 06, 2024.
doi: 10.21037/tcr-23-2157
View this article at: https://dx.doi.org/10.21037/tcr-23-2157

Introduction

Ovarian cancer (OC) is a lethal gynecological malignancy, with about 19,710 new cases and 13,270 deaths every year in United States (1). The standard first-line treatment of OC is surgery followed with platinum and taxane centered chemotherapy (2). Although these therapies can achieve complete remission at the initial stage, most of patients are likely to suffer tumor recurrence predominantly owning to the emergence of chemoresistance, which finally leads to poor prognosis (3). Thus, the factors contribute to the chemoresistance are often the reason for short survival of the OC patients.

Lots of factors are participated in the modulation of chemoresistance and thus many kinds of molecules can be the potential biomarkers for chemoresistance and survival, such as microRNAs, cell cycle and mitosis-molecules, cancer stem cell related molecules, the immune response related molecules and other cancer-associated molecules (4). However, poor sensitivity and lack of specificity are the limitation for majority of biomarkers that have been studied (2). Therefore, there is an ongoing need to identify factors that affect chemoresistance and survival in OC.

Open data and bioinformatics can boost advancements in basic science (5), and reuse of open data is very powerful (6). For example, based on 2,579 tumors from The Cancer Genome Atlas (TCGA) of four gynecological types plus breast, five molecular subtypes have been developed to assess the survival status of patients (7). On the basis of big data of TCGA and Gene Expression Omnibus (GEO), we previously discovered a group of genes relevant to chemoresistance and outcome in OC (8), and developed the multi-gene prognostic signatures in liver cancer (9).

In the present study, based on big data mining and large sample analysis, we identified hundreds of chemoresistant-related genes in OC. The role of those genes in prediction of chemoresistance and short survival was evaluated. We present this article in accordance with the REMARK reporting checklist (available at https://tcr.amegroups.com/article/view/10.21037/tcr-23-2157/rc).

Highlight box

Key findings

- Our study found the *CASP2* might be potentially used as predictive marker, prognostic marker and therapeutic target in management of ovarian cancer (OC).

What is known and what is new?

- The development of chemoresistance is the main reason for the treatment failure and low survival rate of advanced OC patients.
- The study employed large sample analyses to identify genes predicting chemoresistance and short survival from chemoresistant-related genes in OC. And we found *CASP2* might be used as biomarker and therapeutic target in OC management.

What is the implication, and what should change now?

- The *CASP2* might be used as biomarker and therapeutic target in OC management. Further research is required to elucidate its functions in OC.

Methods

Text mining

Coremine (http://www.coremine.com/medical/) was used for retrieval of genes potentially linked to chemoresistance in OC, by combination of "ovarian cancer" ['Ovarian Neoplasms (alias Ovarian Cancer)' (mesh); 'Ovarian Neoplasms (alias Ovarian Cancer)' (mesh); 'Malignant neoplasm of ovary (alias Ovarian Cancer)' (disease)] and "drug resistance" ['drug resistance' (mesh); 'Drug Resistance, Neoplasm' (mesh)] ($P<0.05$).

Data acquisition and large samples

TCGA ovarian cohort (10) of 489 OC patients with clinical data and gene expression was retrieved from cBioPortal (http://www.cbioportal.org/) (11,12), including "staging", "grading", "overall survival" and "primary treatment outcome", in which a subgroup of 90 platinum resistant samples and 197 sensitive samples was included. Kaplan-Meier (KM) plotter (13) of 1,816 OC patients with survival data was retrieved for survival analysis, containing a subgroup of 1,656 patients of which the overall survival (OS) data were accessible, and 1,435 patients of whom the progression-free survival (PFS) data were available. The survival data of 1,816 OC patients with mRNA expressions were integrated from GEO (https://www.ncbi.nlm.nih.gov/geoprofiles/) (14,15) (GSE51373, GSE9891, GSE63885, GSE15622, GSE30161, GSE14764, GSE65986, GSE18520, GSE27651, GSE26712, GSE19829, GSE26193, GSE23554 and GSE3149) and the ovarian cohort of TCGA (10). The study was conducted in accordance with the Declaration of Helsinki (as revised in 2013).

Statistical analysis

Statistical analysis was performed with SPSS (v 20.0).

Student's *t*-test was used to determine the significant variance between the two sets of data. Survival analyses were carried out by the R survival package in R (version 3.3.1). The survival curves were evaluated by the KM method, which gene expression was divided into low and high expression according to the median value, in accordance with a previous study (16). The univariate survival analysis was applied to calculate the hazard ratio (HR) and 95% confidence intervals. The biological processes annotation and pathway enrichment were subsequently performed. Gene expression in prediction of chemoresistance was analyzed by receiver operating characteristic (ROC) plotter (http://www.rocplot.org/) (17), in which the transcriptomic data of 1,816 OC patients were included. The relapse status at six months was used as a cutoff for determination of patient's response to therapy. And patients relapsed within six months were considered as non-responders to drugs.

Results

Retrieval of genes that potentially affect OC chemoresistance

A total of 1,319 genes which potentially affect OC chemoresistance were retrieved from Coremine database by use of keywords "ovarian cancer" and "drug resistance" (P<0.05). A total of 1,298 genes potentially linked to chemoresistance in OC were obtained from a previous study (18). The above two groups of genes were combined and a total of 2,431 genes were obtained after deletion of duplicates. The transcript expression data of the 2,431 genes with clinical factors in 489 OC samples (with a subgroup of 90 chemoresistant samples and 197 sensitive samples) were retrieved to analyze the relationships of these genes with chemoresistance and prognosis, and 2,218 genes of which the data were available were used for subsequent analyses.

Identification of chemoresistant-related genes that predict chemoresistance

There were 301 genes differentially expressed in chemoresistant samples interacting with each other

Function annotation and enrichment were performed. There were 353 biological processes significantly annotated from the 301 genes and proteins [false discovery rate (FDR) <0.01]. And among the top 17 processes (FDR <1.0e–06), at least 185 genes (61.5%) were responded to stimulus, 166 (55.2%) were involved in regulation of biological processes, 118 (39.2%) were development related, and 115 (38.2%) were responded to chemical processes (*Figure 1A*). These results at least partially provide the links between those biological processes with chemoresistance in OC, specifically for the genes in response to stimulus.

Pathway-based identification of 26 novel and key genes which contributed to chemoresistance

Fifty-three pathways were significantly enriched from the 301 genes (FDR <0.01), and among the top 16 pathways (FDR <0.001), 10 of them were typical pathways playing critical roles in OC resistance, which including PI3K-Akt signaling, apoptosis, platinum chemoresistance signaling, Ras signaling, mammalian target of rapamycin (mTOR) signaling, ErbB signaling, etc. (*Figure 1B*).

Gene distribution on the above 10 pathways related to chemoresistance in OC was comprehensively analyzed. Combination of all the genes in the 10 pathways indicated that 51 of the 301 genes were distributed in the 10 pathways. Further analysis based on previous studies was performed to reveal the associations of the 51 genes with cancer development, and the results indicated that the roles of 26 genes in OC chemoresistance were less reported. The transcriptional levels of the 26 genes in 90 chemoresistant OC samples and 197 sensitive samples are shown in *Figure 2*. Among these, 14 genes including *MSH6*, *CASP2*, *EIF2AK3*, *SOS1*, *EIF2AK2*, *FLT1*, *FZD5*, *HSP90AA1*, *HSP90B1*, *KDR*, *PAX6*, *PCK1*, *SDC1* and *WNT7A* were significantly down-regulated in 90 resistant samples, and 12 genes including *PARD6B*, *AKT1S1*, *CALML3*, *CSF3*, *GNG7*, *IHH*, *NRG1*, *PIK3CD*, *RIN1*, *RPS6KA1*, *STAT5A* and *TBP* were significantly up-regulated.

Large sample-based identification of 13 genes that predicted chemoresistance

The roles of the above 26 genes in prediction of chemoresistance in OC were evaluated in a large sample of 1,347 OC patients. As shown in *Figure 3*, 13 of the 26 genes were identified to be potential predictive biomarkers of chemoresistance in OC. Consistent with their expressions in chemoresistant samples (*Figure 2*), low expression of *SOS1*, *CASP2*, *MSH6*, *HSP90AA1*, *HSP90B1* and *FLT1*, and high expression of *PARD6B*, *STAT5A*, *RPS6KA1*, *RIN1*, *PIK3CD*, *CALML3* and *NRG1*, could predict the emergence of chemoresistance (*Figure 3A*). In particular, on one hand, low expression of *SOS1*, *CASP2* and *MSH6*, and high expression of *PARD6B* and *STAT5A* could be more excellent for predicting drug resistance to any drugs

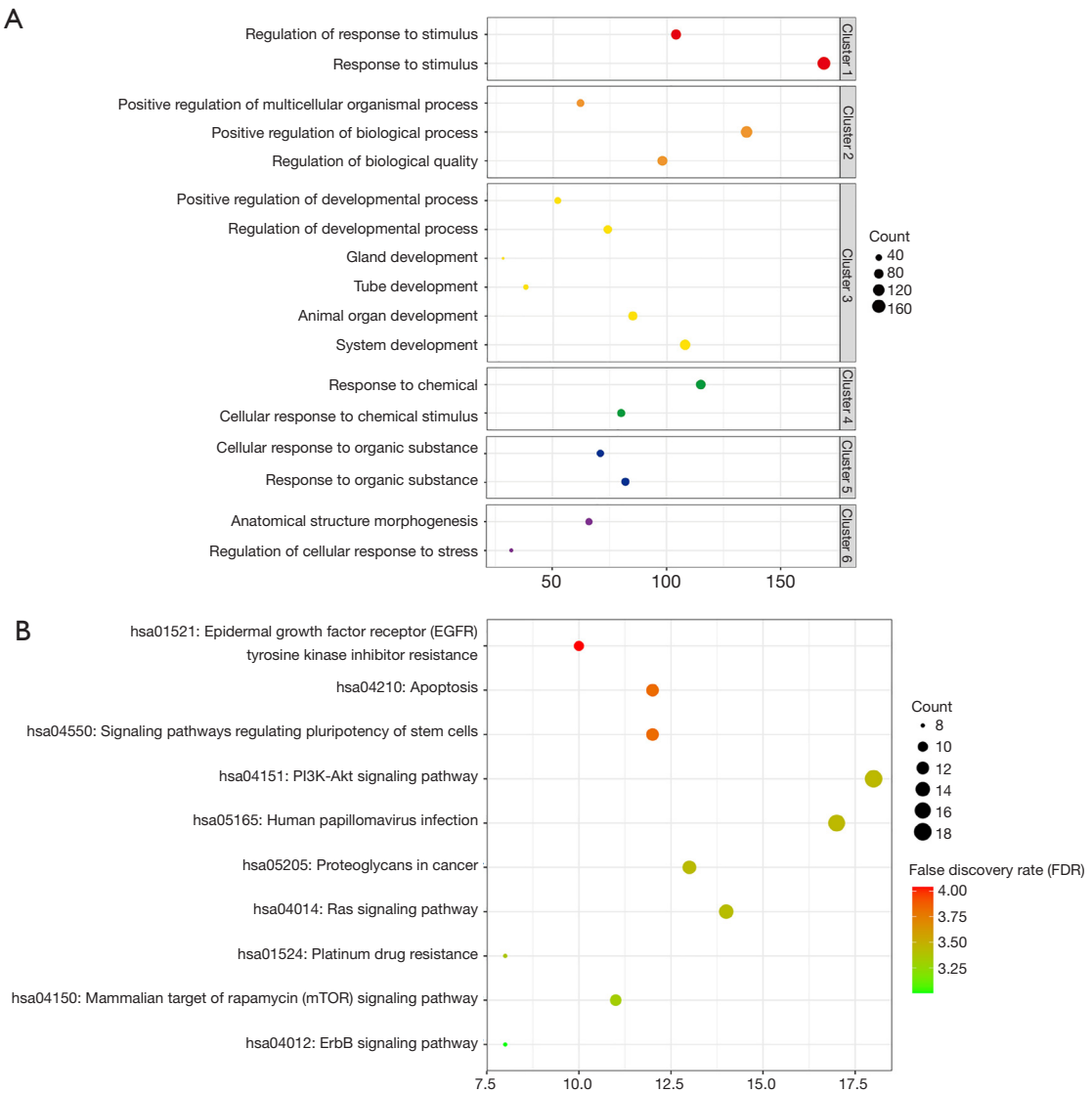

Figure 1 The 301 genes differentially expressed in 90 chemoresistant samples in contrast to 197 sensitive samples according to TCGA ovarian cancer cohort. (A) Function annotation and enrichment analyses revealed the top 17 biological processes (FDR <1×10^{-6}) that were differentially expressed, bubbles in the same cluster are represented by the same color; (B) enriched top 10 typical pathways that correlated with ovarian cancer chemoresistance. The X-axis represents the number of genes. TCGA, The Cancer Genome Atlas; FDR, false discovery rate.

and platin [P<0.01, area under the curve (AUC) >0.6] (*Figure 3B*); while low expression of *SOS1*, *MSH6*, *HSP90AA1* and *HSP90B1*, and high expression of *STAT5A* could be more excellent for prediction of taxane resistance (P<0.01, AUC >0.6) (*Figure 3C*). On the other hand, three genes including *SOS1*, *MSH6* and *STAT5A* were excellent for predicting chemoresistance to any drugs, platin and taxane (P<0.01, AUC >0.6).

Identification of chemoresistant-related genes for predicting short survival

Forty-four genes which dysregulated in chemoresistant samples could predict short OS and disease-free survival (DFS)

The roles of the 2,218 genes in prediction of DFS and OS were determined in 489 OC samples of TCGA cohort.

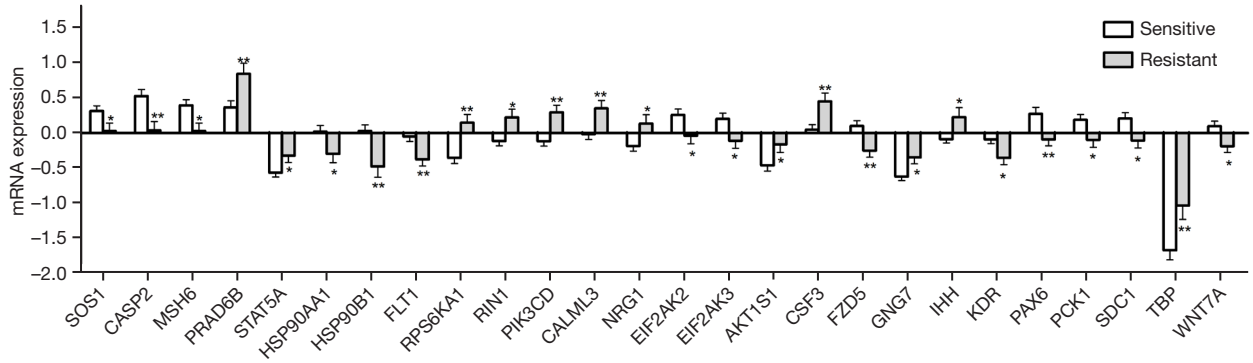

Figure 2 Twenty-six genes significantly and differentially expressed in chemoresistant samples in contrast to the sensitive samples, in accordance with TCGA ovarian cancer cohort. Sensitive: 197 platinum sensitive ovarian cancer samples; resistant: 90 resistant samples. *, P<0.05; **, P<0.01. TCGA, The Cancer Genome Atlas.

Of which, 207 genes were related to DFS (P<0.05), 249 genes were related to OS (P<0.05), and collectively a total of 380 genes were related to DFS and/or OS. Then, an intersection of these 380 genes with the 301 genes dysregulated in 90 chemoresistant samples were performed, and total of 84 common genes were identified.

Further analyses on these 84 genes were performed. On one hand, the expression of the gene in chemoresistant samples should match the poor status of prognosis. For example, a gene would be retained if it was highly expressed in 90 chemoresistant samples in contrast to 197 sensitive samples, and the high expression predicted a short survival. On the other hand, a gene would be retained if its association with prognosis in OC was poorly studied. After the selection, total of 44 genes which significantly dysregulated in chemoresistant samples and predicted short survival were identified (*Table 1*).

Among the 44 genes, 17 of them were prominently associated with both OS and DFS in 489 OC patients. Among them, low expression of 12 genes (*CASP2*, *CHIT1*, *STAT1*, *MSH6*, *AADAC*, *CAPN13*, *GCH1*, *OR6F1*, *PHGDH*, *RNF148*, *SLAMF7*, and *WDR45B*) which were down-regulated in resistant samples were correlated with poor prognosis, and high expression of five genes (*LAYN*, *PARD6B*, *GDF6*, *LIPC* and *TENM3*) which were up-regulated in resistant samples predicted poor prognosis (*Tables 1*,*2*, *Figure 4*). Ten genes were only significantly associated with OS, among those, low expression of six genes (*ALDH5A1*, *TREML2*, *MRS2*, *TRIM27*, *CXCR4* and *KCNE3*) which were down-regulated in resistant samples predicted short OS, and high expression of four genes (*SPOCK2*, *RPL23*, *TCF15* and *ACSS3*) which were up-regulated in resistant samples associated with short OS (*Tables 1*,*3*). Seventeen genes were only significantly relevant to DFS, among those, low expression of 10 genes (*NCOA1*, *KCNJ16*, *C16ORF89*, *HIPK1*, *LARP4*, *LGR5*, *PSMD1*, *VTCN1*, *EIF2AK3* and *SOS1*) which were down-regulated in resistant samples predicted short DFS and high expression of seven genes (*GRYAB*, *GAP43*, *ICAM5*, *CATSPERD*, *PSG1*, *RSL24D1* and *SNHG29*) which were up-regulated in chemoresistant samples predicted short DFS (*Tables 1*,*4*).

Large sample-based identification of 10 genes which significantly predicted short OS

The roles of the above 44 genes in prediction of prognosis were further verified in large samples of 1,816 OC patients, which included a subgroup of 1,656 specimens with OS data, and 1,435 specimens with PFS data. Firstly, among the 17 genes which were relevant to OS in TCGA cohort of 489 patients (*Figure 4*, *Table 3*), 10 of them (*CASP2*, *CH1T1*, *SPOCK2*, *TREML2*, *RPL23*, *TCF15*, *ALDH5A1*, *MRS2*, *TRIM27* and *STAT1*) were consistently related to OS in 1,656 OC samples (*Figure 5A*), of which, four genes including *CASP2*, *CHIT1*, *SPOCK2* and *TREML2* were also significantly associated with PFS (*Figure 5A*). The relationships of the 17 genes associated with DFS in the 489 patients (*Table 4*) were also submitted to the analysis, and five of them including *CRYAB*, *GAP43*, *NCOA1*, *ICAM5* and *KCNJ16* were consistently associated with prognosis in the 1,816 patients, and the former three were associated with both OS and PFS (*Figure 5B*).

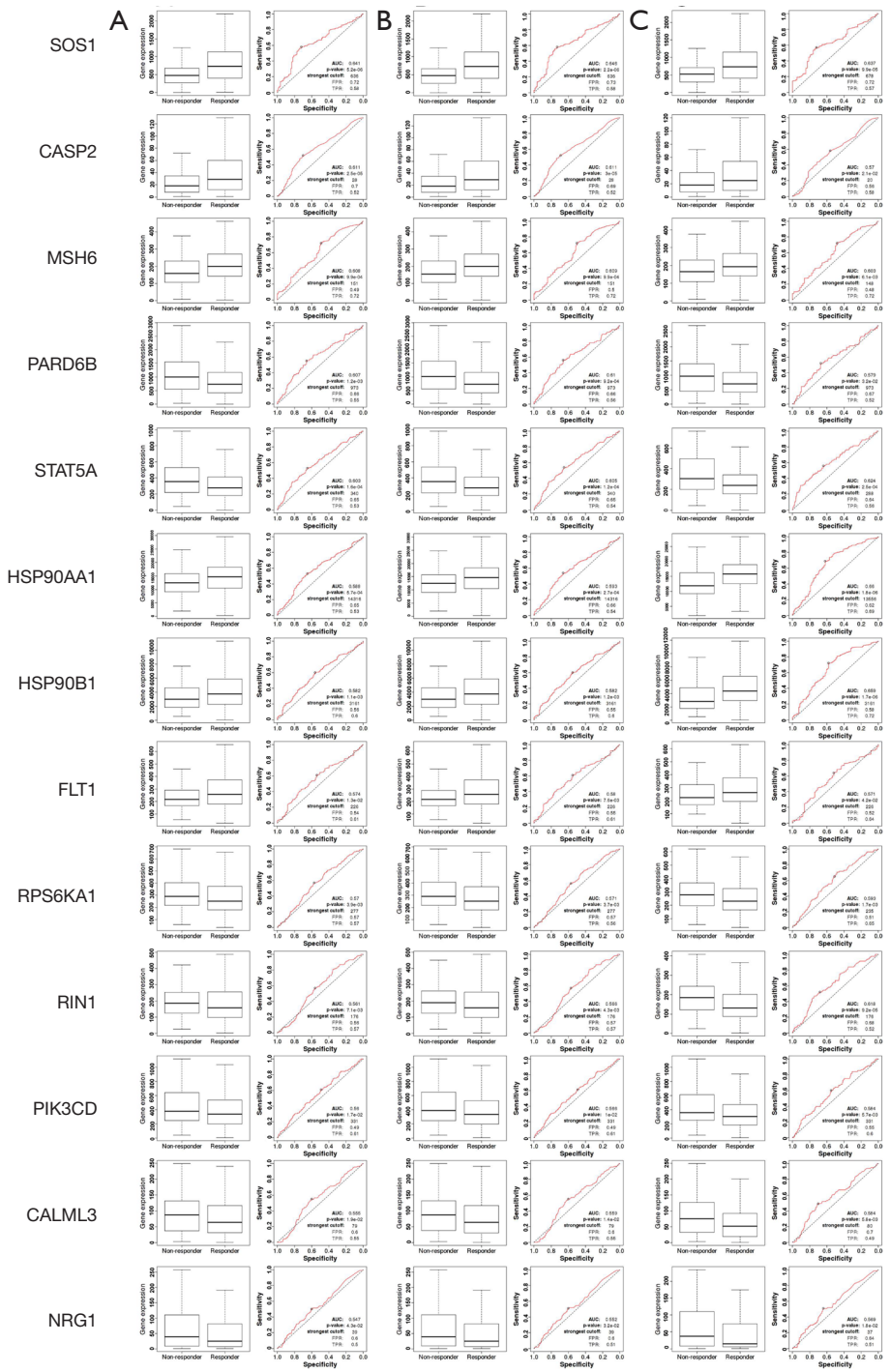

Figure 3 Role of genes in prediction of chemoresistance in ovarian cancer. Transcriptome-level data of 1,347 ovarian cancer patients were included according to ROC plotter. The relapse status at 6 months was used as a cut off for definition of patient's response to therapy, and those relapsed within the 6 months were considered as non-responders. (A) Abnormal expression of genes predicts chemoresistance to any drugs (includes platin, taxane, docetaxel, paclitaxel, gemcitabine, topotecan and avastin), in which 130 non-responders and 1,217 responders were included. (B) Abnormal expression of genes predicts chemoresistance to platin, in which 114 non-responders and 1,095 responders were included. (C) Abnormal expression of genes predicts chemoresistance to taxane, in which 81 non-responders and 807 responders were included. AUC, area under the curve; FPR, false positive rate; TPR, true positive rate; ROC, receiver operating characteristic.

Table 1 Forty-four genes which dysregulated in chemoresistant samples were relevant to DFS and OS in ovarian cancer

Gene	Up/down-regulated in drug resistant samples* (P<0.05)		High/low expression predicts poor survival# (P<0.05)			
	Up	Down	High	Low	DFS	OS
CASP2		√		√	√	√
CHIT1		√		√	√	√
STAT1		√		√	√	√
MSH6		√		√	√	√
AADAC		√		√	√	√
CAPN13		√		√	√	√
GCH1		√		√	√	√
OR6F1		√		√	√	√
PHGDH		√		√	√	√
RNF148		√		√	√	√
SLAMF7		√		√	√	√
WDR45B		√		√	√	√
LAYN	√		√		√	√
PARD6B	√		√		√	√
GDF6	√		√		√	√
LIPC	√		√		√	√
TENM3	√		√		√	√
ALDH5A1		√		√		√
TREML2		√		√		√
MRS2		√		√		√
TRIM27		√		√		√
CXCR4		√		√		√
KCNE3		√		√		√
SPOCK2	√		√			√
RPL23	√		√			√
TCF15	√		√			√
ACSS3	√		√			√
NCOA1		√		√	√	
KCNJ16		√		√	√	
C16ORF89		√		√	√	
HIPK1		√		√	√	
LARP4		√		√	√	
LGR5		√		√	√	

Table 1 (*continued*)

Table 1 (*continued*)

Gene	Up/down-regulated in drug resistant samples* (P<0.05)		High/low expression predicts poor survival# (P<0.05)			
	Up	Down	High	Low	DFS	OS
PSMD1		√		√	√	
VTCN1		√		√	√	
EIF2AK3		√		√	√	
SOS1		√		√	√	
CRYAB	√		√		√	
GAP43	√		√		√	
ICAM5	√		√		√	
CATSPERD	√		√		√	
PSG1	√		√		√	
RSL24D1	√		√		√	
SNHG29	√		√		√	

The expression of the gene in chemoresistant samples and their relationships with survival in ovarian cancer was determined based on the TCGA ovarian cohort: (*) 90 chemoresistant samples and 197 sensitive samples were used for determine gene expression, and (#) 489 samples were used for prognosis analysis. Kaplan-Meier method was used for survival analysis; gene expression was divided into low and high by the median value. DFS, disease-free survival; OS, overall survival; TCGA, The Cancer Genome Atlas.

Table 2 Kaplan-Meier analyses revealed that seventeen genes were relevant to overall survival and disease-free survival in ovarian cancer, based on TCGA cohort of 489 patients

mRNA expression	Expression level	Disease-free survival					Overall survival				
		Estimate	Standard error	95% confidence interval		P	Estimate	Standard error	95% confidence interval		P
				Lower	Upper				Lower	Upper	
CASP2	High	18.960	1.127	16.751	21.169	0.002	48.720	2.501	43.817	53.623	<0.001
	Low	14.460	1.080	12.342	16.578		36.890	2.217	32.545	41.235	
	All	16.850	0.743	15.394	18.306		43.790	2.117	39.640	47.940	
CHIT1	High	18.040	0.864	16.347	19.733	0.02	47.570	2.223	43.212	51.928	0.006
	Low	15.280	1.036	13.250	17.310		40.380	2.656	35.174	45.586	
	All	16.850	0.743	15.394	18.306		43.790	2.117	39.640	47.940	
STAT1	High	17.970	0.869	16.267	19.673	0.03	47.370	2.423	42.620	52.120	0.03
	Low	14.720	1.396	11.984	17.456		41.000	2.997	35.126	46.874	
	All	16.850	0.743	15.394	18.306		43.790	2.117	39.640	47.940	
MSH6	High	19.120	0.993	17.173	21.067	0.01	45.300	3.434	38.569	52.031	0.04
	Low	15.150	0.931	13.326	16.974		40.970	2.933	35.220	46.720	
	All	16.850	0.743	15.394	18.306		43.790	2.117	39.640	47.940	
AADAC	High	17.840	1.158	15.571	20.109	0.02	48.290	3.223	41.973	54.607	0.001
	Low	16.000	1.021	13.999	18.001		39.560	2.730	34.210	44.910	
	All	16.850	0.743	15.394	18.306		43.790	2.117	39.640	47.940	

Table 2 (*continued*)

Table 2 (*continued*)

mRNA expression	Expression level	Disease-free survival					Overall survival				
		Estimate	Standard error	95% confidence interval		P	Estimate	Standard error	95% confidence interval		P
				Lower	Upper				Lower	Upper	
CAPN13	High	17.970	0.852	16.299	19.641	0.045	47.670	2.468	42.833	52.507	0.041
	Low	15.380	1.044	13.333	17.427		37.910	2.935	32.157	43.663	
	All	16.850	0.743	15.394	18.306		43.790	2.117	39.640	47.940	
GCH1	High	18.960	1.264	16.483	21.437	0.01	47.670	2.331	43.101	52.239	0.049
	Low	15.080	1.063	12.996	17.164		40.970	2.778	35.525	46.415	
	All	16.850	0.743	15.394	18.306		43.790	2.117	39.640	47.940	
OR6F1	High	17.970	1.274	15.474	20.466	0.007	45.110	3.046	39.139	51.081	0.03
	Low	15.640	1.018	13.644	17.636		41.360	2.862	35.750	46.970	
	All	16.850	0.743	15.394	18.306		43.790	2.117	39.640	47.940	
PHGDH	High	19.120	1.009	17.143	21.097	0.003	44.880	1.928	41.100	48.660	0.02
	Low	15.110	0.705	13.728	16.492		39.360	2.760	33.951	44.769	
	All	16.850	0.743	15.394	18.306		43.790	2.117	39.640	47.940	
RNF148	High	19.150	1.271	16.658	21.642	0.02	48.290	2.795	42.812	53.768	<0.001
	Low	15.110	1.128	12.900	17.320		36.240	2.347	31.639	40.841	
	All	16.850	0.743	15.394	18.306		43.790	2.117	39.640	47.940	
SLAMF7	High	18.960	1.439	16.140	21.780	<0.001	47.370	2.753	41.974	52.766	0.007
	Low	14.780	0.927	12.963	16.597		41.530	2.247	37.127	45.933	
	All	16.850	0.743	15.394	18.306		43.790	2.117	39.640	47.940	
WDR45B	High	18.170	1.669	14.899	21.441	0.02	48.750	3.009	42.852	54.648	0.005
	Low	16.130	0.788	14.586	17.674		39.360	3.042	33.398	45.322	
	All	16.850	0.743	15.394	18.306		43.790	2.117	39.640	47.940	
LAYN	High	16.690	1.245	14.250	19.130	0.01	40.970	2.386	36.293	45.647	0.044
	Low	17.510	1.042	15.468	19.552		47.370	2.732	42.016	52.724	
	All	16.850	0.743	15.394	18.306		43.790	2.117	39.640	47.940	
PARD6B	High	14.690	0.788	13.146	16.234	0.02	38.410	2.599	33.317	43.503	0.044
	Low	18.660	0.853	16.987	20.333		47.670	2.345	43.075	52.265	
	All	16.850	0.743	15.394	18.306		43.790	2.117	39.640	47.940	
GDF6	High	15.440	1.405	12.685	18.195	0.050	39.850	3.080	33.813	45.887	0.02
	Low	17.640	0.968	15.743	19.537		47.510	2.155	43.286	51.734	
	All	16.850	0.743	15.394	18.306		43.790	2.117	39.640	47.940	
LIPC	High	15.280	0.630	14.044	16.516	0.005	39.360	2.581	34.301	44.419	0.004
	Low	19.150	1.016	17.158	21.142		49.020	3.563	42.037	56.003	
	All	16.850	0.743	15.394	18.306		43.790	2.117	39.640	47.940	
TENM3	High	15.440	1.146	13.194	17.686	0.03	39.360	3.224	33.041	45.679	0.040
	Low	17.970	1.543	14.945	20.995		44.880	2.200	40.568	49.192	
	All	16.850	0.743	15.394	18.306		43.790	2.117	39.640	47.940	

Gene expression was divided into low (L) and high (H) by the median value. TCGA, The Cancer Genome Atlas.

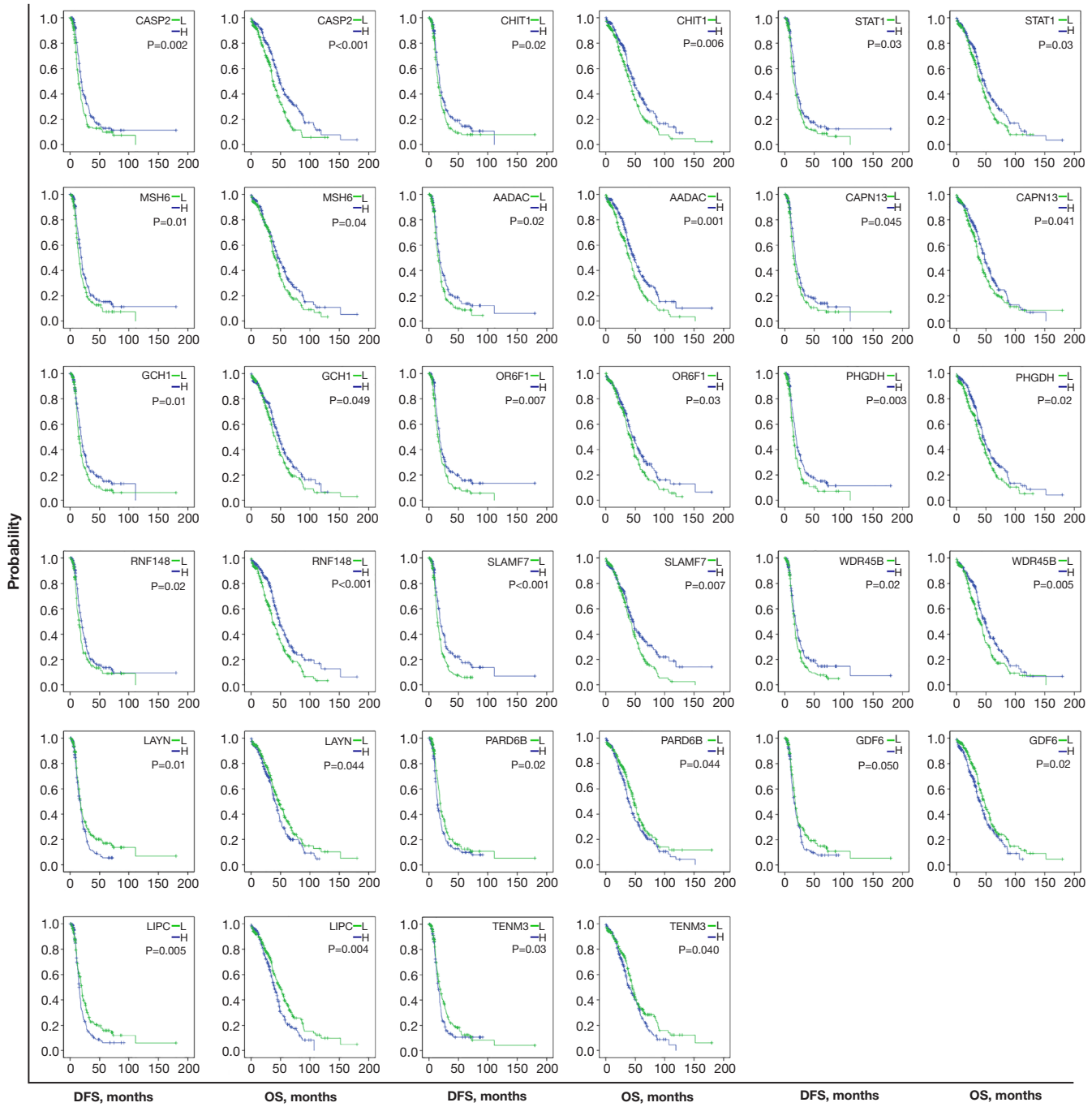

Figure 4 Kaplan-Meier analyses determined that seventeen genes were relevant to overall survival and disease-free survival in ovarian cancer, based on TCGA cohort of 489 patients. Gene expression was divided into low (L) and high (H) by the median value. DFS, disease-specific survival; OS, overall survival; TCGA, The Cancer Genome Atlas.

Table 3 Kaplan-Meier analyses revealed that ten genes were correlated with overall survival in ovarian cancer, based on TCGA cohort of 489 patients

mRNA expression	Expression level	Overall survival				
		Estimate	Standard error	95% confidence interval		P
				Lower	Upper	
ALDH5A1	High	48.290	4.063	40.326	56.254	0.002
	Low	39.560	2.748	34.173	44.947	
	All	43.790	2.117	39.640	47.940	
TREML2	High	49.580	2.877	43.942	55.218	<0.001
	Low	36.340	2.287	31.857	40.823	
	All	43.790	2.117	39.640	47.940	
MRS2	High	44.980	2.983	39.133	50.827	0.045
	Low	41.000	2.290	36.513	45.487	
	All	43.790	2.117	39.640	47.940	
TRIM27	High	49.810	4.298	41.387	58.233	0.004
	Low	39.360	2.686	34.096	44.624	
	All	43.790	2.117	39.640	47.940	
KCNE3	High	47.510	2.085	43.423	51.597	0.04
	Low	39.000	2.227	34.635	43.365	
	All	43.790	2.117	39.640	47.940	
CXCR4	High	48.030	3.115	41.925	54.135	0.04
	Low	39.850	2.723	34.512	45.188	
	All	43.790	2.117	39.640	47.940	
SPOCK2	High	38.410	2.886	32.754	44.066	0.01
	Low	48.720	3.001	42.838	54.602	
	All	43.790	2.117	39.640	47.940	
RPL23	High	41.000	2.268	36.556	45.444	0.02
	Low	47.670	2.346	43.072	52.268	
	All	43.790	2.117	39.640	47.940	
TCF15	High	39.850	2.037	35.858	43.842	0.02
	Low	48.030	3.258	41.645	54.415	
	All	43.790	2.117	39.640	47.940	
ACSS3	High	40.970	2.799	35.483	46.457	0.044
	Low	45.300	2.743	39.923	50.677	
	All	43.790	2.117	39.640	47.940	

Gene expression was divided into low (L) and high (H) by the median value. TCGA, The Cancer Genome Atlas.

Table 4 Kaplan-Meier analyses showed that seventeen genes were correlated to disease-free survival in ovarian cancer, based on TCGA cohort of 489 patients

mRNA expression	Expression level	Disease-free survival				
		Estimate	Standard error	95% confidence interval		
				Lower	Upper	P
NCOA1	High	18.660	1.214	16.280	21.040	0.003
	Low	15.280	1.180	12.967	17.593	
	All	16.850	0.743	15.394	18.306	
KCNJ16	High	17.640	1.391	14.914	20.366	0.02
	Low	16.300	1.371	13.613	18.987	
	All	16.850	0.743	15.394	18.306	
C16ORF89	High	18.140	1.413	15.370	20.910	0.02
	Low	15.410	0.746	13.948	16.872	
	All	16.850	0.743	15.394	18.306	
HIPK1	High	18.100	1.126	15.894	20.306	<0.001
	Low	14.720	0.968	12.823	16.617	
	All	16.850	0.743	15.394	18.306	
LARP4	High	18.170	1.312	15.598	20.742	0.03
	Low	15.380	1.238	12.953	17.807	
	All	16.850	0.743	15.394	18.306	
LGR5	High	17.970	1.308	15.406	20.534	0.02
	Low	15.540	1.245	13.099	17.981	
	All	16.850	0.743	15.394	18.306	
PSMD1	High	18.860	0.871	17.154	20.566	0.003
	Low	14.780	0.921	12.975	16.585	
	All	16.850	0.743	15.394	18.306	
VTCN1	High	17.840	1.151	15.584	20.096	0.03
	Low	15.640	1.432	12.833	18.447	
	All	16.850	0.743	15.394	18.306	
EIF2AK3	High	18.170	1.296	15.630	20.710	0.047
	Low	15.540	1.008	13.564	17.516	
	All	16.850	0.743	15.394	18.306	
SOS1	High	18.170	1.000	16.209	20.131	0.001
	Low	14.720	0.933	12.892	16.548	
	All	16.850	0.743	15.394	18.306	
CRYAB	High	15.640	0.953	13.773	17.507	0.046
	Low	18.040	1.284	15.524	20.556	
	All	16.850	0.743	15.394	18.306	

Table 4 (*continued*)

Table 4 (*continued*)

mRNA expression	Expression level	Disease-free survival				
		Estimate	Standard error	95% confidence interval		
				Lower	Upper	P
GAP43	High	14.460	1.248	12.013	16.907	0.02
	Low	18.170	1.092	16.030	20.310	
	All	16.850	0.743	15.394	18.306	
ICAM5	High	16.130	1.111	13.953	18.307	0.03
	Low	17.840	0.986	15.908	19.772	
	All	16.850	0.743	15.394	18.306	
CATSPERD	High	15.150	1.177	12.842	17.458	0.01
	Low	18.140	1.110	15.964	20.316	
	All	16.850	0.743	15.394	18.306	
PSG1	High	14.880	1.100	12.724	17.036	0.02
	Low	18.860	1.051	16.799	20.921	
	All	16.850	0.743	15.394	18.306	
RSL24D1	High	15.380	1.284	12.864	17.896	0.04
	Low	18.040	1.178	15.732	20.348	
	All	16.850	0.743	15.394	18.306	
SNHG29	High	16.300	1.192	13.963	18.637	0.008
	Low	18.040	1.153	15.779	20.301	
	All	16.850	0.743	15.394	18.306	

Gene expression was divided into low (L) and high (H) by the median value. TCGA, The Cancer Genome Atlas.

Discussion

In this study, a total of 2,218 genes which potentially affected chemoresistance in OC were identified based on text mining, and 301 of them were significantly dysregulated in 90 chemoresistant OC samples in contrast to 197 sensitive samples. Analysis by biological processes annotation suggested those genes could potentially respond to stimulus (*Figure 1A*). It has been proved that response to stimulus is implicated in regulation of chemoresistance in cancers (19,20). Furthermore, pathway enrichment of the 301 genes was performed and 10 pathways such as PI3K-Akt signaling, apoptosis, and platinum chemoresistance signaling were significantly enriched (*Figure 1B*), which are typical pathways involved in modulation of chemoresistance in OC. The results strongly supported the relevance of those genes with chemoresistance. Intersection of all genes of the 10 pathways with the 301 genes identified 51 genes, which included some typical chemoresistant related genes in OC, such as *AKT1*, *PIK3CA* and *MAPK1* (21). Among the 51 genes, the associations of 26 genes (*Figure 2*) with chemoresistance in OC have been rarely reported. The results above suggested that, probably via interactions with the typical 10 drug-resistant pathways in OC, the 26 genes might be new targets for management of OC, particularly in chemoresistant patients.

Because of the heterogeneity of OC, identifying predictive biomarkers are important for the selection of suitable treatments to improve patient survival (22). The use of gene expression signatures of key pathways that contribute to chemoresistance as predictive biomarkers is a reasonable approach (22). Several genes were previously identified to predict chemoresistance. For example, a low-RAS signature was shown to associate with sensitivity to

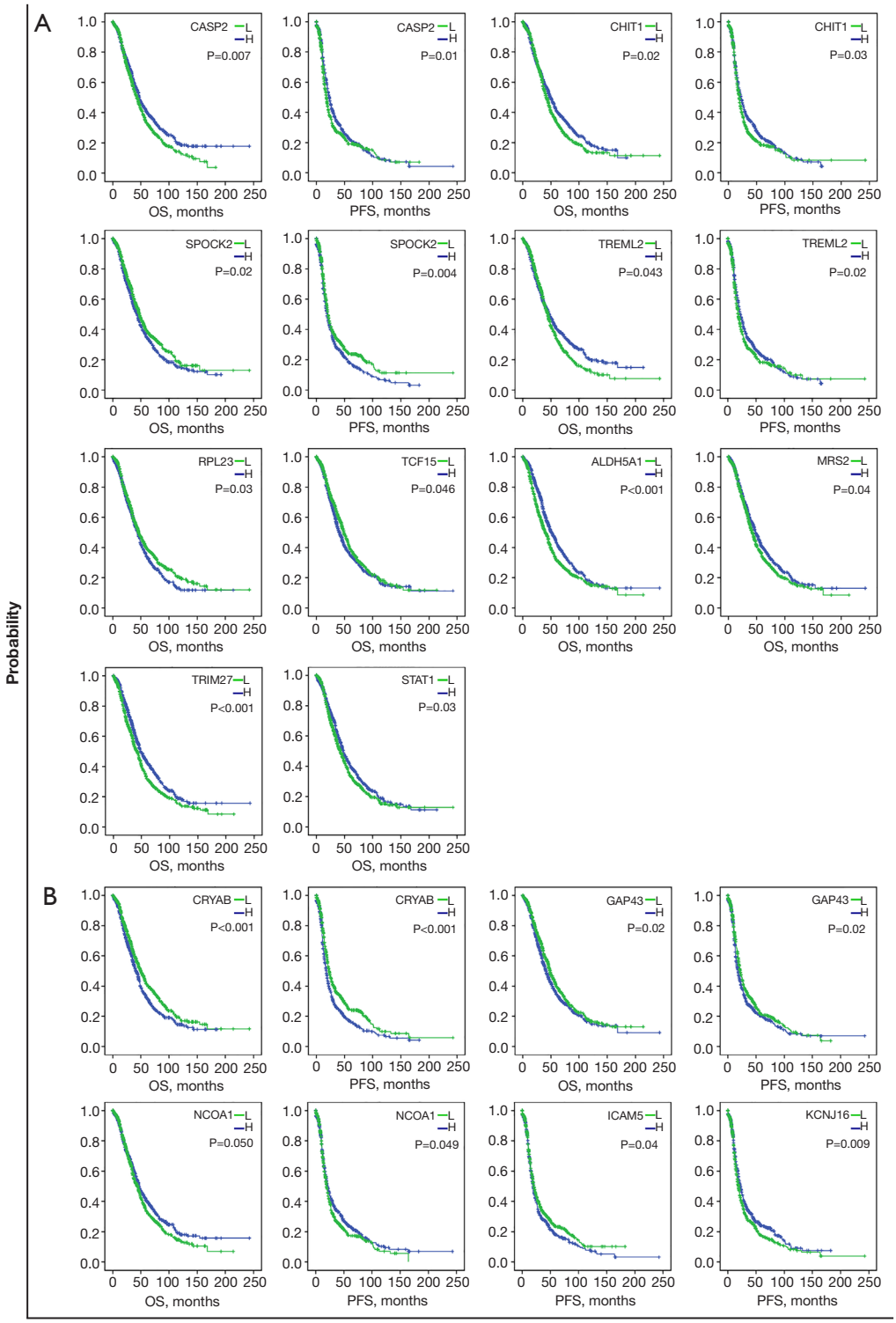

Figure 5 Genes correlated with OS and PFS in 1,816 ovarian cancer patients based on the KM plotters collection. Gene expression was divided into low (L) and high (H) by the median value. (A) Genes related to OS in 489 patients of TCGA cohort consistently predicted OS and/or PFS in 1,816 patients of KM plotter collection. (B) Genes related to DFS in 489 patients was also relevant to OS and/or PFS in 1,816 patients. OS, overall survival; PFS, progression-free survival; KM, Kaplan-Meier; TCGA, The Cancer Genome Atlas.

the AKT inhibitor MK2206 (23), and BRCAness gene-expression in OC was associated with responsiveness to platinum-based chemotherapy (24,25). Despite of those findings, the biomarkers for prediction of chemoresistance are still less understood. In this study, the above identified 26 genes were distributed in 10 chemoresistant-related pathways, and based on the large sample analysis, 13 of them were potentially the predictive biomarkers for chemoresistance (*Figure 3*). Especially the low expression of *SOS1*, *CASP2* and *MSH6*, and high expression of *PARD6B* and *STAT5A* were excellent for predicting chemoresistance to any drugs and platin; low expression of SOS1, MSH6, HSP90AA1 and HSP90B1, and high expression of STAT5A were excellent for predicting taxane resistance. The associations of *SOS1*, *CASP2*, *PARD6B*, *STAT5A*, *HSP90AA1* and *HSP90B1* with chemoresistance in OC were rarely known, although a research suggested that the silence of *MSH6* in *Saccharomyces cerevisiae* could increase the strain resistance to cisplatin, doxorubicin, and carboplatin (26). This is consistent with our findings. However, the roles of those genes in prediction of chemoresistance in OC have not been reported so far.

Chemoresistance is a main factor that contributes to short survival in OC. Thus, further analyses of 2,218 genes in 489 OC patients were performed to identify the candidates that affected both chemoresistance and survival, and 44 genes were identified to be relevant to OS and/or DFS (*Table 1*), and their relationships with chemoresistance and survival were limited. The correlation of the above 44 genes with survival of OC patients was further verified in large cohort of up to 1,816 specimens, and 10 genes including *CASP2*, *CH1T1*, *SPOCK2*, *TREML2*, *RPL23*, *TCF15*, *ALDH5A1*, *MRS2*, *TRIM27* and *STAT1* were consistently related to OS (*Figure 5*). The results are consistent with the findings in previous studies. For example, low expression of *CASP2* associated with poor OS in gastric carcinoma (27), and high expression of *SPOCK2* predicted poor prognosis in OC (28).

Among all the genes identified in this study, *CASP2* was the only one that the results were positive in all of the analyses. In the TCGA cohort of 489 OC patients, *CASP2* was significantly down-regulated in 90 chemoresistant samples in contrast to 197 sensitive samples, and its low expression predicted short DFS and OS. In large cohort up to thousands of OC patients, *CASP2* expression was consistently lower in chemoresistant samples and its low expression predicted chemoresistance to any drugs and platin, and short OS and PFS as well. *CASP2* is one of *CASPS*, which often acts as intrinsic initiators of apoptosis (29,30). *CASP2* plays important roles in apoptotic as well as nonapoptotic processes including apoptosis, cell cycle, autophagy, DNA repair, regulation of oxidant levels and lipid biosynthesis (31). The roles of *CASP2* in cancer remain a matter of controversy, because the gene normally produces two mRNA splice variants *CASP2L* and *CASP2S*. *CASP2L* normally promotes apoptosis, while *CASP2S* normally inhibits apoptosis (32). In OC, *CASP2* is the target of miR-383, which is overexpressed in OC cells and samples. High level of miR-383 and low expression of the *CASP2* improve cell invasion, cell cycle progression and cell proliferation (33). These results are basically consistent with the findings in this study.

Taken together, 13 genes that affected chemoresistance and predicted chemoresistance in OC were identified, especially the six excellent candidate genes *SOS1*, *CASP2*, *PARD6B*, *STAT5A*, *HSP90AA1* and *HSP90B1*. A total of 44 genes which potentially contributed to chemoresistance and related to prognosis were identified, especially 10 genes including *CASP2*, *CH1T1*, *SPOCK2*, *TREML2*, *RPL23*, *TCF15*, *ALDH5A1*, *MRS2*, *TRIM27* and *STAT1* were consistently related to overall survival in a group of 1,656 patients. Finally, it is noteworthy that *CASP2* was the only gene that the results were positively and consistently in all analyses. The genes discovered in this study might be developed to be predictive markers, prognostic markers and therapeutic targets in the clinical management of OC. Studies have shown that, the caspase-2 regulatory mechanism can induce OC cell death (34), Han *et al.* found that casp-2S affects cellular apoptosis through its interaction with membrane-associated cytoskeletal Fodrin protein (35). Future work will investigate the function of *CASP2*.

OC cells interact with their surrounding microenvironment through a complex communication mechanism, impacting the tumor's response to drugs. OC cells can change the composition of their surrounding microenvironment (immune cells, stromal cells and vascular endothelial cells, etc.) through various means, such as releasing cytokines, VEGF and other angiogenic factors (36), secreting exosomes (37), etc., thus forming a favorable environment that promotes tumor growth and invasion. The poorly metabolized tumor microenvironment affects clinical prognosis by forming a barrier to tumor-infiltrating immune cells (38).

The genes related to chemotherapy resistance in OC show some correlation with tumor immune infiltration, tumor mutation burden, and microsatellite instability. Some studies suggest that chemo-resistant cancer cells

often have higher levels of tumor immune infiltration and tumor mutation burden (39-41). Additionally, microsatellite instability is also associated with chemotherapy resistance. An increase in mutation burden and microsatellite instability could potentially impact tumor growth, replication, and treatment response (42). Future work will explore the association between *CASP2* and tumor immune invasion, tumor mutation burden, and microsatellite instability to provide reference for OC treatment strategies.

Conclusions

The identified genes specifically the *CASP2* might be potentially used as predictive markers, prognostic markers and therapeutic targets in management of OC.

Acknowledgments

Funding: This work was supported by the National Natural Science Foundation of China (Nos. 81860458, 81903644, and 82260721), Guangxi Natural Science Foundation (Nos. 2024GXNSFDA010045, 2021GXNSFAA075002, and 2018GXNSFAA281227), Program Foundation of Key Laboratory of High-Incidence Tumor Prevention and Treatment, Ministry of Education (Nos. GKE-ZZ202148, and GKE-ZZ202017) and Advanced Innovation Teams and Xinghu Scholars Program of Guangxi Medical University, Innovation Project of Guangxi Graduate Education (No. YCSW2023237).

Footnote

Reporting Checklist: The authors have completed the REMARK reporting checklist. Available at https://tcr.amegroups.com/article/view/10.21037/tcr-23-2157/rc

Peer Review File: Available at https://tcr.amegroups.com/article/view/10.21037/tcr-23-2157/prf

Conflicts of Interest: All authors have completed the ICMJE uniform disclosure form (available at https://tcr.amegroups.com/article/view/10.21037/tcr-23-2157/coif). The authors have no conflicts of interest to declare.

Ethical Statement: The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. The study was conducted in accordance with the Declaration of Helsinki (as revised in 2013).

References

1. Siegel RL, Miller KD, Wagle NS, et al. Cancer statistics, 2023. CA Cancer J Clin 2023;73:17-48.
2. Chandra A, Pius C, Nabeel M, et al. Ovarian cancer: Current status and strategies for improving therapeutic outcomes. Cancer Med 2019;8:7018-31.
3. Xiang Y, Chen YJ, Yan YB, et al. MiR-186 bidirectionally regulates cisplatin sensitivity of ovarian cancer cells via suppressing targets PIK3R3 and PTEN and upregulating APAF1 expression. J Cancer 2020;11:3446-53.
4. Davidson B. Biomarkers of drug resistance in ovarian cancer - an update. Expert Rev Mol Diagn 2019;19:469-76.
5. Mangul S, Martin LS, Langmead B, et al. How bioinformatics and open data can boost basic science in countries and universities with limited resources. Nat Biotechnol 2019;37:324-6.
6. Rung J, Brazma A. Reuse of public genome-wide gene expression data. Nat Rev Genet 2013;14:89-99.
7. Berger AC, Korkut A, Kanchi RS, et al. A Comprehensive Pan-Cancer Molecular Study of Gynecologic and Breast Cancers. Cancer Cell 2018;33:690-705.e9.
8. Yin F, Yi S, Wei L, et al. Microarray-based identification of genes associated with prognosis and drug resistance in ovarian cancer. J Cell Biochem 2019;120:6057-70.
9. Liu M, Liu X, Liu S, et al. Big Data-Based Identification of Multi-Gene Prognostic Signatures in Liver Cancer. Front Oncol 2020;10:847.
10. Integrated genomic analyses of ovarian carcinoma. Nature 2011;474:609-15.
11. Cerami E, Gao J, Dogrusoz U, et al. The cBio cancer genomics portal: an open platform for exploring multidimensional cancer genomics data. Cancer Discov 2012;2:401-4.
12. Gao J, Aksoy BA, Dogrusoz U, et al. Integrative analysis

of complex cancer genomics and clinical profiles using the cBioPortal. Sci Signal 2013;6:pl1.
13. Gyorffy B, Lánczky A, Szállási Z. Implementing an online tool for genome-wide validation of survival-associated biomarkers in ovarian-cancer using microarray data from 1287 patients. Endocr Relat Cancer 2012;19:197-208.
14. Barrett T, Wilhite SE, Ledoux P, et al. NCBI GEO: archive for functional genomics data sets--update. Nucleic Acids Res 2013;41:D991-5.
15. Barrett T, Suzek TO, Troup DB, et al. NCBI GEO: mining millions of expression profiles--database and tools. Nucleic Acids Res 2005;33:D562-6.
16. Henderson MJ, Haber M, Porro A, et al. ABCC multidrug transporters in childhood neuroblastoma: clinical and biological effects independent of cytotoxic drug efflux. J Natl Cancer Inst 2011;103:1236-51.
17. Fekete JT, Ősz Á, Pete I, et al. Predictive biomarkers of platinum and taxane resistance using the transcriptomic data of 1816 ovarian cancer patients. Gynecol Oncol 2020;156:654-61.
18. Lloyd KL, Cree IA, Savage RS. Prediction of resistance to chemotherapy in ovarian cancer: a systematic review. BMC Cancer 2015;15:117.
19. Liu D, Wang L, Zhong R, et al. Parallel microfluidic networks for studying cellular response to chemical modulation. J Biotechnol 2007;131:286-92.
20. Lahiani MH, Eassa S, Parnell C, et al. Carbon nanotubes as carriers of Panax ginseng metabolites and enhancers of ginsenosides Rb1 and Rg1 anti-cancer activity. Nanotechnology 2017;28:015101.
21. Liu X, Gao Y, Lu Y, et al. Oncogenes associated with drug resistance in ovarian cancer. J Cancer Res Clin Oncol 2015;141:381-95.
22. Cheaib B, Auguste A, Leary A. The PI3K/Akt/mTOR pathway in ovarian cancer: therapeutic opportunities and challenges. Chin J Cancer 2015;34:4-16.
23. Loboda A, Nebozhyn M, Klinghoffer R, et al. A gene expression signature of RAS pathway dependence predicts response to PI3K and RAS pathway inhibitors and expands the population of RAS pathway activated tumors. BMC Med Genomics 2010;3:26.
24. Konstantinopoulos PA, Spentzos D, Karlan BY, et al. Gene expression profile of BRCAness that correlates with responsiveness to chemotherapy and with outcome in patients with epithelial ovarian cancer. J Clin Oncol 2010;28:3555-61.
25. Weberpals JI, Clark-Knowles KV, Vanderhyden BC. Sporadic epithelial ovarian cancer: clinical relevance of BRCA1 inhibition in the DNA damage and repair pathway. J Clin Oncol 2008;26:3259-67.
26. Durant ST, Morris MM, Illand M, et al. Dependence on RAD52 and RAD1 for anticancer drug resistance mediated by inactivation of mismatch repair genes. Curr Biol 1999;9:51-4.
27. Wang Z, Ni F, Yu F, et al. Prognostic significance of mRNA expression of CASPs in gastric cancer. Oncol Lett 2019;18:4535-54.
28. Lou W, Ding B, Zhong G, et al. Dysregulation of pseudogene/lncRNA-hsa-miR-363-3p-SPOCK2 pathway fuels stage progression of ovarian cancer. Aging (Albany NY) 2019;11:11416-39.
29. Kim YR, Kim KM, Yoo NJ, et al. Mutational analysis of CASP1, 2, 3, 4, 5, 6, 7, 8, 9, 10, and 14 genes in gastrointestinal stromal tumors. Hum Pathol 2009;40:868-71.
30. Kumar S. Mechanisms mediating caspase activation in cell death. Cell Death Differ 1999;6:1060-6.
31. Tiwari M, Sharma LK, Vanegas D, et al. A nonapoptotic role for CASP2/caspase 2: modulation of autophagy. Autophagy 2014;10:1054-70.
32. Solier S, Logette E, Desoche L, et al. Nonsense-mediated mRNA decay among human caspases: the caspase-2S putative protein is encoded by an extremely short-lived mRNA. Cell Death Differ 2005;12:687-9.
33. Liu J, Dou Y, Sheng M. Inhibition of microRNA-383 has tumor suppressive effect in human epithelial ovarian cancer through the action on caspase-2 gene. Biomed Pharmacother 2016;83:1286-94.
34. Yang CS, Matsuura K, Huang NJ, et al. Fatty acid synthase inhibition engages a novel caspase-2 regulatory mechanism to induce ovarian cancer cell death. Oncogene 2015;34:3264-72.
35. Han C, Zhao R, Kroger J, et al. Caspase-2 short isoform interacts with membrane-associated cytoskeleton proteins to inhibit apoptosis. PLoS One 2013;8:e67033.
36. Nunes SC. Tumor Microenvironment - Selective Pressures Boosting Cancer Progression. Adv Exp Med Biol 2020;1219:35-49.
37. Nakamura K, Sawada K, Kinose Y, et al. Exosomes Promote Ovarian Cancer Cell Invasion through Transfer of CD44 to Peritoneal Mesothelial Cells. Mol Cancer Res 2017;15:78-92.
38. Kao KC, Vilbois S, Tsai CH, et al. Metabolic communication in the tumour-immune microenvironment. Nat Cell Biol 2022;24:1574-83.
39. Fan S, Gao X, Qin Q, et al. Association between tumor

mutation burden and immune infiltration in ovarian cancer. Int Immunopharmacol 2020;89:107126.
40. Gao C, Li H, Liu C, et al. Tumor Mutation Burden and Immune Invasion Characteristics in Triple Negative Breast Cancer: Genome High-Throughput Data Analysis. Front Immunol 2021;12:650491.
41. Wang ZM, Xu QR, Kaul D, et al. Significance of tumor mutation burden and immune infiltration in thymic epithelial tumors. Thorac Cancer 2021;12:1995-2006.
42. Chen J, Apizi A, Wang L, et al. TCGA database analysis of the tumor mutation burden and its clinical significance in colon cancer. J Gastrointest Oncol 2021;12:2244-59.

Cite this article as: Wang C, Chen C, Chen X, Luo J, Su Y, Liu X, Yin F. Identification of genes predicting chemoresistance and short survival in ovarian cancer. Transl Cancer Res 2024;13(8):4354-4371. doi: 10.21037/tcr-23-2157

