Original Article

Microarray screening for key genes and prognosis factors in interferon regulatory factor 1-silenced ovarian cancer SKOV-3 cells

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Background: Ovarian cancer is a gynecological neoplastic disease with high mortality rate. Its early detection is difficult because of the lack of specific clinical symptoms. This study aimed to identify key genes and prognosis factors associated with ovarian cancer to provide new information and thus better understanding of ovarian cancer.

Methods: Microarray data from the Gene Expression Omnibus (GEO) database (accession number GSE38551) were used for analysis. Differentially expressed genes (DEGs) were screened, and functional enrichment and protein-protein interaction (PPI) network analyses for DEGs were performed. A subnetwork was constructed to gain further information regarding DEGs scored in the PPI network. Finally, we performed survival analysis.

Results: In total, 427 DEGs were obtained in interferon regulatory factor 1 (*IRF-1*)-silenced ovarian cancer SKOV-3 cell line samples compared to SKOV-3 samples without *IRF-1* silencing. DEGs were mainly enriched in metabolic pathways and systemic lupus erythematosus. Tumor necrosis factor (*TNF*) and cadherin 1 (*CDH1*; type 1, E-cadherin) were present in had higher degrees than others in both the PPI network and the subnetwork. The subnetwork results presented that *CDH1* was enriched in the epithelium morphogenesis and cancer pathways, and *TNF* was enriched in response to lipids. The Mir-30 family served as a tumor suppressor in ovarian cancer. Survival analysis revealed that *CDH1* was associated with ovarian cancer prognosis.

Conclusions: *TNF* and *CDH1* play important roles in ovarian cancer: *CDH1* is an important prognosis factor for ovarian cancer and may be involved mainly via epithelial morphogenesis and cancer pathways. *TNF* may be involved via response to lipids.

Keywords: Ovarian cancer; interferon regulatory factor 1 (*IRF-1*); differentially expressed genes (DEGs); pathways; protein-protein interaction (PPI) network

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Introduction

Ovarian cancer is a gynecological neoplastic disease and the fifth most common cause of cancer mortality in women (1). Survival of patients with ovarian cancer is reported to be highly related to the stage of cancer: 5-year survival rate for patients with early-stage cancer is 80–90%, whereas that for patients with advanced-stage disease is merely 25% (2).

Epithelial ovarian cancer, described as a "silent killer," is the most common type of ovarian cancer (3). Approximately 90% of ovarian cancers affect the single-cell epithelial layer of the ovarian surface (4). However, timely adoption of preventive measures for ovarian cancer is difficult because of the lack of obvious symptoms during the early stage and dearth of effective early-diagnostic tools.

In the past, several studies have used ultrasound (5) and cancer antigen 125 (CA 125) (6) as the primary test for ovarian cancer. The CA 125 assay was used as first-line screening because of its relatively noninvasive nature during blood sampling. Serum CA 125 levels increased in 23–50% of surgical stage I and 90% of stage II ovarian carcinomas (7). However, rather than a prognostic or diagnostic marker, CA 125 level is used only for following the response or progression of the disease (8).

Recently, large-scale gene expression analysis has been used to screen differentially expressed genes (DEGs) in ovarian cancer (9), especially for identifying potential tumor markers of early-stage diagnosis and ensuring timely treatment (10). Transcription factors regulate the expression of tumor-associated genes (*TAGs*), which provides insights for research regarding the key genes in ovarian cancer (11). Interferon regulatory factor 1 (*IRF-1*), a member of the interferon regulatory transcription factor family, activates the transcription of interferons alpha and beta. It is also a tumor suppressor gene (*TSG*) that prevents oncogene-mediated malignant transformation (12). *IRF-1* expression in tumors is an independent predictor of favorable clinical outcomes for ovarian cancer (13), and it is likely that gene expression could differ with *IRF-1* silencing.

In this study, epithelial ovarian cancer SKOV-3 cells that were separately transfected with *IRF-1* short hairpin ribonucleic acid (shRNA) and scrambled shRNA were used to analyze DEGs with *IRF-1* silencing to understand the mechanism of ovarian cancer.

Methods

Microarray data

Microarray expression data was obtained from the platform data of GPL10558 (IlluminaHumanHT-12 V4.0 expression beadchip) from the Gene Expression Omnibus (GEO) database (accession number GSE38551; http://www.ncbi.nlm.nih.gov/geo/), which was deposited by Pavan *et al.* (12). The microarray included 12 samples [3 SKOV-3 samples transfected with scrambled shRNA, 3 with scrambled shRNA with cis-diamminedichloroplatinum (CDDP), 3 with IRF-1 shRNA, and 3 with IRF-1 shRNA with CDDP]. For analysis, we used 3 SKOV-3 samples transfected with IRF-1 shRNA and 3 with scrambled shRNA.

Data preprocessing and DEG analysis

Data preprocessing (background correction, quantile normalization, probe summarization) was performed using the robust multi-array average algorithm (14) in the Limma software; the *t*-test (15) was used to identify significantly expressed DEGs in SKOV-3 samples transfected with IRF-1 shRNA and those transfected with scrambled shRNA. A false discovery rate (FDR) <0.05 and an absolute value of $\log_2$FC (fold change) >1 were used as thresholds.

Gene ontology (GO) and pathway enrichment analysis for DEGs

GO analysis, including biological process (BP), molecular function (MF), and cellular component, is used for the unification of biology (16). The Kyoto Encyclopedia of Genes and Genomes (KEGG) is a database used to classify relevant gene sets into their respective pathways (17). In this study, we used the Database for Annotation, Visualization, and Integrated Discovery (DAVID) to identify significant GO categories in BPs and significant pathways with P<0.05.

Functional annotation for DEGs

Using the transcription factor data, we screened and annotated DEGs to determine whether they could regulate transcription. TSG (18) and TAG databases (19) were used for screening TSGs and oncogenes.

Construction of a PPI network

The Search Tool for the Retrieval of Interacting Genes (STRING) database can provide both experimental and predicted interaction information of proteins (20). In this study, STRING was used for protein-protein interaction (PPI) network analysis and confidence value (combined score) >0.4 was regarded as the threshold. Cytoscape was used to construct the PPI network, and highly connected nodes (hubs) (21) were obtained.

Subnetwork construction and enrichment analyses

To obtain further information regarding DEGs scored in the PPI network, a subnetwork was constructed using the BioNet software (22) in R with FDR =0.0001. GO and KEGG enrichment analyses were performed for DEG-encoded proteins in the subnetwork.

MiRNA-target regulating analysis

We performed microRNA (miRNA) prediction using WebGestalt GAST (23) (http://www.webgestalt.org/option.php), and conducted miRNA-target enrichment prediction for DEGs in the PPI network by overrepresentation enrichment analysis (ORA). The species was Hsapiens, the minimum number of enriched DEGs was 2, and results with P<0.05 were obtained.

Survival analysis

DEGs related to survival and prognoses were searched in The Cancer Genome Atlas (TCGA) database. The DEGs were then grouped by the median into high- and low-expression genes. Age, gender, and cancer stage were adjusted using the Cox model; P<0.05 was considered to be significant. Hazard ratios (HRs) of these DEGs were predicted for survival. High-expression DEGs with HR >1 and low-expression DEGs with HR <1 were screened, and Kaplan–Meier survival curves were drawn.

Results

DEG selection

Results showed that 442 transcriptional factors were observed: 250 upregulated and 192 downregulated factors. In these 427 DEGs were obtained: 242 upregulated DEGs and 185 downregulated DEGs (*Figure 1*).

GO and KEGG enrichment analysis for DEGs

We performed GO and KEGG enrichment analyses using P<0.05 for the functional analysis of DEGs. Several GO categories were enriched among these DEGs, and *Table 1* lists the top ten categories for up- and downregulated DEGs. Three categories of BPs enriched most DEGs with a count >100. In upregulated DEGs, the BPs were single-organism process, single-organism cellular process, and response to stimulus, and in downregulated DEGs, the BPs were cellular process, single-organism process, and single-organism cellular process. *Table 1* also shows the pathways that were obtained by KEGG enrichment. In upregulated DEGs, a total of ten pathways with a count >2 were obtained, mainly metabolic pathways, systemic lupus erythematosus, and tight junctions. In downregulated genes, five pathways were obtained with a small count.

Functional annotation for DEGs

Results showed that eight transcriptional factors were upregulated and 11 downregulated (*Table 2*). In the upregulated factors, 26 genes were detected (2 oncogenes, 20 TSGs, and 4 genes with unknown functions), and in the downregulated factors, 19 genes were detected (3 oncogenes, 15 tumor genes, and 1 gene with whose unknown functions).

PPI network construction

We finally obtained 173 interaction pairs. In the PPI network (*Figure 2*), degrees of 14 proteins were >5: tumor necrosis factor (TNF), degree =28; CDH1, degree =20; matrix metallopeptidase 2 (MMP2), degree =13; collagen type I alpha 1 chain (COL1A1), degree =12; serpin family E-member 1 (SERPINE1), degree =12; MMP1, degree =9; gap junction protein alpha 1 (GJA1), degree =8; fibrillin 1 (FBN1), degree =8; Snail family transcriptional repressor 2 (SNAI2), degree =7; thrombospondin 1 (THBS1), degree =7; forkhead box O1 (FOXO1), degree =7; CCAAT/enhancer binding protein delta, degree =7; claudin 3 (CLDN3), degree =7; Dickkopf Wnt signaling pathway inhibitor 1 (DKK1), degree =7; integrin subunit beta 4 (ITGB4), degree =6; keratin 14 (KRT14), degree =6; tight junction protein 3 (TJP3), degree =6; KIT ligand (KITLG), degree =6; and MMP7, degree =6. The degrees of TNF and CDH1 were the top 2 nodes compared to other proteins.

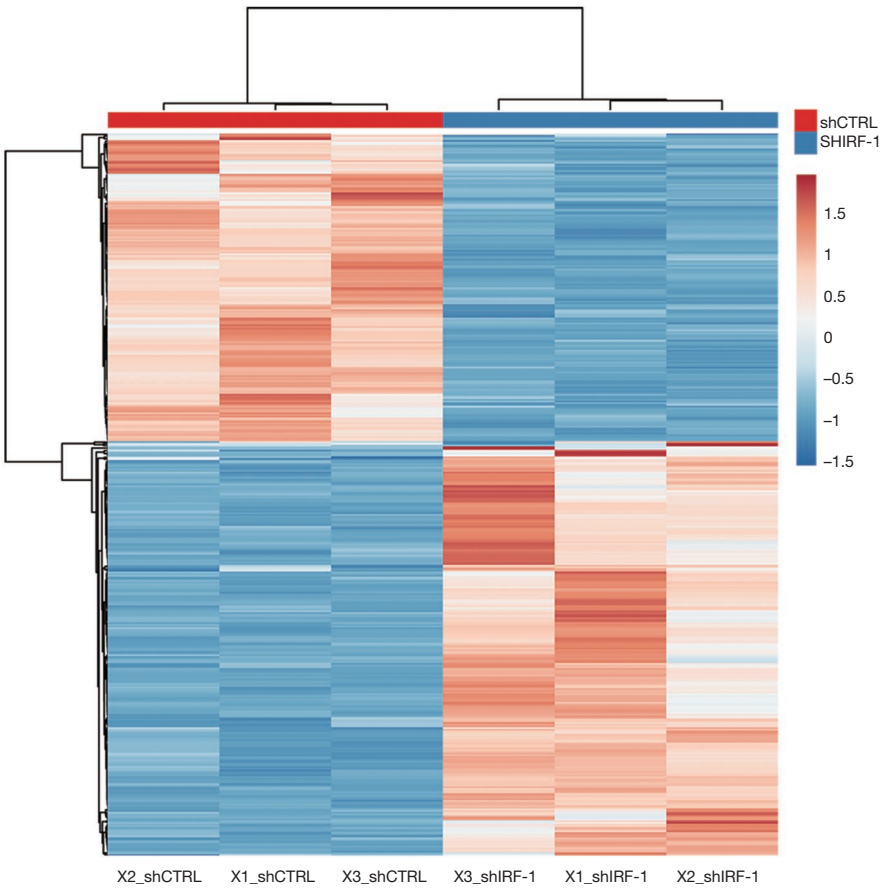

Figure 1 Heat map for DEGs identified in this study. DEGs, differentially expressed genes.

Subnetwork analyses

As shown in *Figure 3*, 48 nodes and 68 interaction pairs were included in the subnetwork. Among them, consistent with the results of the PPI network, the top 7 proteins with high degrees were TNF, CDH1, MMP2, GJA1, DKK1, THBS1, and SERPINE1. *Table 3* lists the GO terms and KEGG pathway enrichment for the subnetwork. For GO terms, DEGs were mainly enriched in the BP of epithelium morphogenesis (e.g., *CDH1*) and response to lipids (e.g., *TNF*). KEGG analysis revealed that DEGs were mainly enriched in cancer pathways (e.g., *CDH1* enriched), tight junctions, Wnt signaling pathway (e.g., *DKK1* enriched), bladder cancer (e.g., *CDH1*, *THBS1* enriched), and p53 signaling pathway (e.g., *THBS1*, *SERPINE1* enriched).

MiRNA-target-regulating analysis

Table 4 present results for miRNA-target-regulating analysis. In this study, we identified 15 miRNAs targeting DEGs. Among these miRNAs, miR-30A-5p, miR-30C, miR-30D, miR-30B, and miR-30E-5p targeted the TGTTTAC motif contained in 25 DEGs, including *FRMD6* , *FAM43A*, *EDNRA*, *EPHB2*, and *KHNYN*. MiR-498 targeted the GCTTGAA motif in nine DEGs, including *COL1A1*, *DUSP4*, *FLRT2*, *HBP1*, and *GJA1*. MiR-492 was predicted to target five DEGs: *TSKU*, *C17orf58*, *STC1*, *ZFP36*, and *SH3PXD2A*. MiR-489 targeted six DEGs: *ADAMTS5*, *PRSS23*, *FBN1*, *NRG1*, *SGK1*, and *WNT5A*. Both miR-27A and miR-27B were predicted to target 17 DEGs, including *CDH11*, *E2F7*, *DCP2*, *EDNRA*, *EPHB2*, and *FLRT2*.

Survival analysis

We isolated ovarian cancer prognosis-associated data from TCGA database and identified survival-correlated DEGs

Table 1 Results for gene ontology (GO) terms and Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway enrichment analysis

Category	Term	Description	Counts	P value
Up-regulated				
GO-BP	0044699	Single-organism process	150	4.02^{-4}
GO-BP	0044763	Single-organism cellular process	138	7.51^{-4}
GO-BP	0050896	Response to stimulus	103	6.03^{-4}
GO-BP	0044707	Single-multicellular organism process	79	4.38^{-2}
GO-BP	0007154	Cell communication	78	2.08^{-3}
GO-BP	0023052	Signaling	77	1.66^{-3}
GO-BP	0044700	Single organism signaling	77	1.66^{-3}
GO-BP	0051716	Cellular response to stimulus	77	1.76^{-2}
GO-BP	0032502	Developmental process	69	1.85^{-2}
GO-BP	0007165	Signal transduction	66	1.23^{-2}
KEGG	1100	Metabolic pathways	23	3.32^{-2}
KEGG	5322	Systemic lupus erythematosus	10	1.65^{-5}
KEGG	4530	Tight junction	5	3.67^{-2}
KEGG	4612	Antigen processing and presentation	4	2.12^{-2}
KEGG	5323	Rheumatoid arthritis	4	3.78^{-2}
KEGG	260	Glycine, serine and threonine metabolism	3	9.73^{-3}
KEGG	5143	African trypanosomiasis	3	1.24^{-2}
KEGG	4960	Aldosterone-regulated sodium reabsorption	3	2.03^{-2}
KEGG	330	Arginine and proline metabolism	3	3.91^{-2}
KEGG	920	Sulfur metabolism	2	1.36^{-2}
Down-regulated				
GO-BP	0009987	Cellular process	143	2.32^{-2}
GO-BP	0044699	Single-organism process	130	4.25^{-4}
GO-BP	0044763	Single-organism cellular process	120	6.77^{-4}
GO-BP	0032501	Multicellular organismal process	91	2.87^{-7}
GO-BP	0044707	Single-multicellular organism process	88	5.44^{-7}
GO-BP	0032502	Developmental process	76	1.18^{-6}
GO-BP	0048856	Anatomical structure development	74	2.43^{-8}
GO-BP	0007275	Multicellular organismal development	74	2.68^{-8}
GO-BP	0016043	Cellular component organization	69	3.86^{-5}
GO-BP	0071840	Cellular component organization or biogenesis	69	9.21^{-5}
KEGG	4360	Axon guidance	5	8.56^{-3}
KEGG	4974	Protein digestion and absorption	4	8.12^{-3}
KEGG	5219	Bladder cancer	3	7.99^{-3}
KEGG	4115	p53 signaling pathway	3	2.91^{-2}
KEGG	601	Glycosphingolipid biosynthesis-lacto and neolacto series	2	2.68^{-2}

Term, the identification number of GO term or KEGG; description represents the name of GO term or KEGG; counts, the number of genes enriched in GO term or KEGG. BP, biological process; GO, gene ontology; KEGG, Kyoto Encyclopedia of Genes and Genomes.

Table 2 Results of functional annotation for DEGs

Category	TF		TAG	
	Counts	Genes	Counts	Genes
Up	8	ARNT, CEBPD, EHF, LMO3, NR3C2, ONECUT2, SREBF1, VDR	26	TERC, WISP2, AMH, BEX2, BIK, CABLES1, CDH1, CDKN1C, CEBPD, EHF, FOXO1, HBP1, MAL, MTUS1, MUC1, NAPEPLD, PTPRD, SEMA3B, UNC5A, VWA5A, WNT5A, ZFP36, GRB7, ITGB4, TNF, ZFP36L2
Down	11	E2F7, FOXF2, HEY2, LHX1, MSC, SMAD5, SNAPC3, SOX7, TEAD4, TP63, TWIST2	19	CTTN, KIT, MLLT11, ABLIM3, CDH11, DKK1, EPHB2, GJA1, MT1G, SERPINB5, SFRP1, SOX7, SRPX, THBD, THBS1, TP63, TWIST2, UHRF2

DEGs, differentially expressed genes; TF, transcription factor; TAG, tumor-associated gene.

according to the selection criteria. As a result, 38 prognosis-associated genes (e.g., *CDH1*) were obtained. Specifically, *CDH1*, which was upregulated in IRF-1-silenced SKOV3 cells, was predicted to be negatively correlated with survival in patients with ovarian cancer (*Figure 4*).

Discussion

In this study, we found 427 DEGs (242 upregulated and 185 downregulated) and their function categories that altered in IRF-1-silenced SKOV-3 cell samples. Pathway enrichment analyses for all DEGs showed that genes were mainly enriched in metabolic pathways and systemic lupus erythematosus. TNF and CDH1 had a higher degree than others in both the PPI network and the subnetwork. Furthermore, GO and KEGG enrichment analyses for the subnetwork showed that *CDH1* was enriched in the BP of epithelium morphogenesis and cancer pathways, and *TNF* was enriched in response to lipids. Besides, survival analysis showed that *CDH1* was associated with ovarian cancer prognosis.

A previous study reported the significant role of cancer cell metabolism pathways in colorectal carcinomas (24), which suggested that cancer cells share common enzyme or transporter activities, suggestive of anaerobic metabolism with high ability for lactate extrusion and glucose absorption. Otherwise, it is difficult for the tumor to survive and grow. Systemic lupus erythematosus is a prototypical autoimmune disease characterized by the production of immunoglobulin G (IgG) autoantibodies that are specific for self-antigens. Bernatsky *et al.* (25) proved the association between systemic lupus erythematosus and cancer and indicated that certain cancers occur more frequently in patients with systemic lupus erythematosus than in those without. Our results of pathway enrichment analyses of all DEGs showed that the genes were mainly enriched in metabolic pathways and systemic lupus erythematosus. Thus, metabolic pathways and systemic lupus erythematosus may be involved in ovarian cancer.

TNF encodes a multifunctional pro-inflammatory cytokine belonging to the TNF superfamily from the CDH superfamily, and its high expression in ovarian cancer cells indicates its importance (26). Son *et al.* suggested that targeting pro-inflammatory chemokines induced by epidermal growth factor (EGF) or TNF, including CCL20, CXCL1-3, and CXCL8, may be a potential treatment for ovarian cancer with many epidermal growth factor receptor and *TNF* activation patterns (27).Tania *et al.* reported that lipid metabolism is related to ovarian cancer risk, and targeting enzymes of lysophosphatidic acid metabolism might be useful for further cancer therapy (28). In our study, *TNF* was significantly enriched in the BP of response to lipids and might play a significant role in ovarian cancer via this response.

CDH1, a classic cadherin, is expressed predominantly on epithelial cell surface and plays a key role in the maintenance and establishment of normal tissue architecture (29). It encodes a calcium-dependent cell-cell adhesion glycoprotein formed by five extracellular cadherin repeats. Studies have reported that transfection of human cancer cell lines with E-cadherin complementary deoxyribonucleic acid (cDNA) can decrease their invasiveness (30). This may explain why *CDH1* expression in epithelial ovarian cancer cells is upregulated with IRF-1 silencing. Combined detection of serum human epididymis protein 4 (HE4) and *CDH1* gene methylation levels could help differentiate ovarian endometriosis cysts from ovarian cancer during diagnosis (31). A previous meta-analysis indicated that *CDH1* promoter methylation could be a

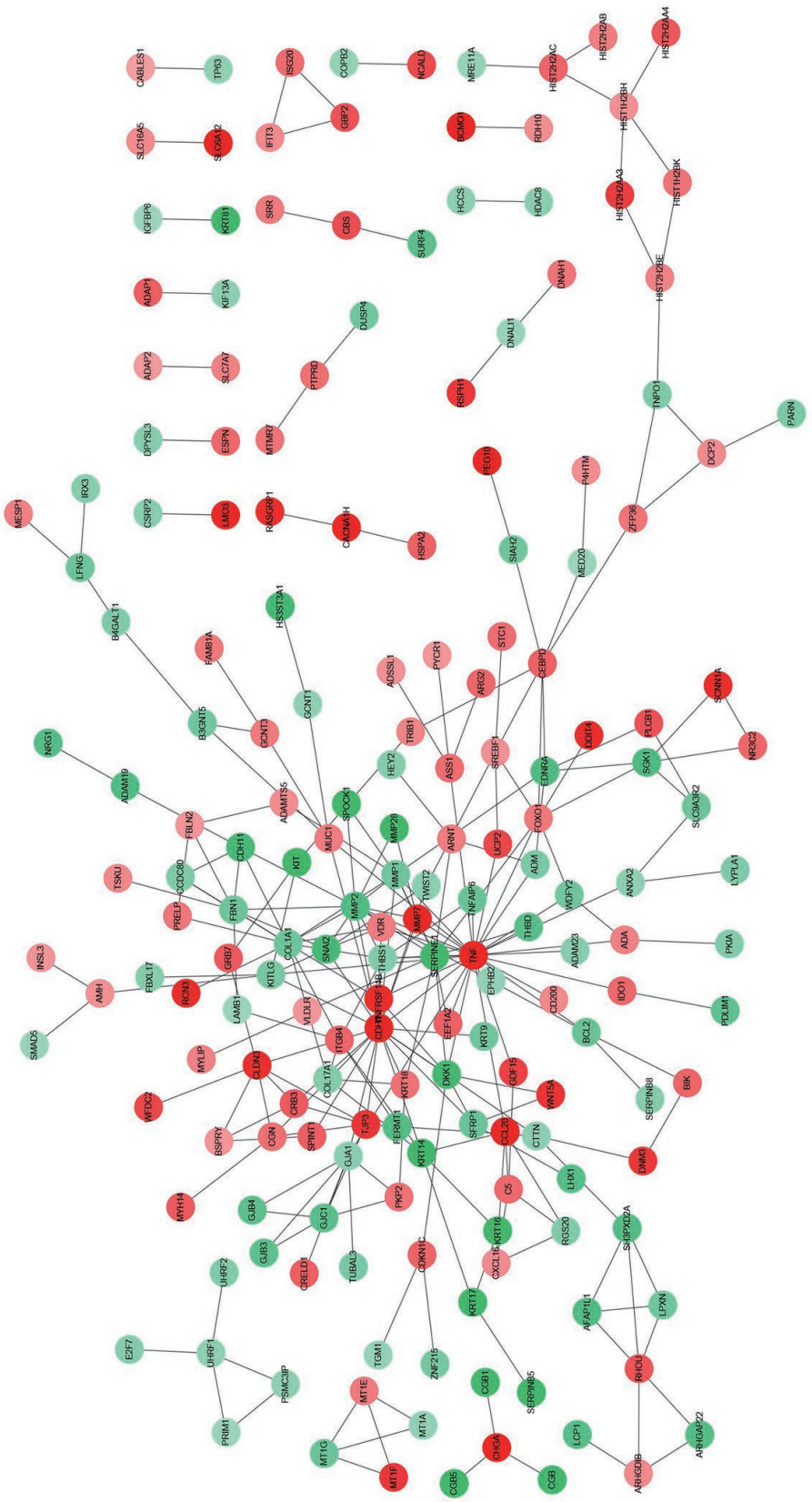

Figure 2 PPI network of DEGs. Red nodes, upregulated genes; green nodes, downregulated genes; a darker color means more significant DEGs. PPI, protein-protein interaction; DEGs, differentially expressed genes.

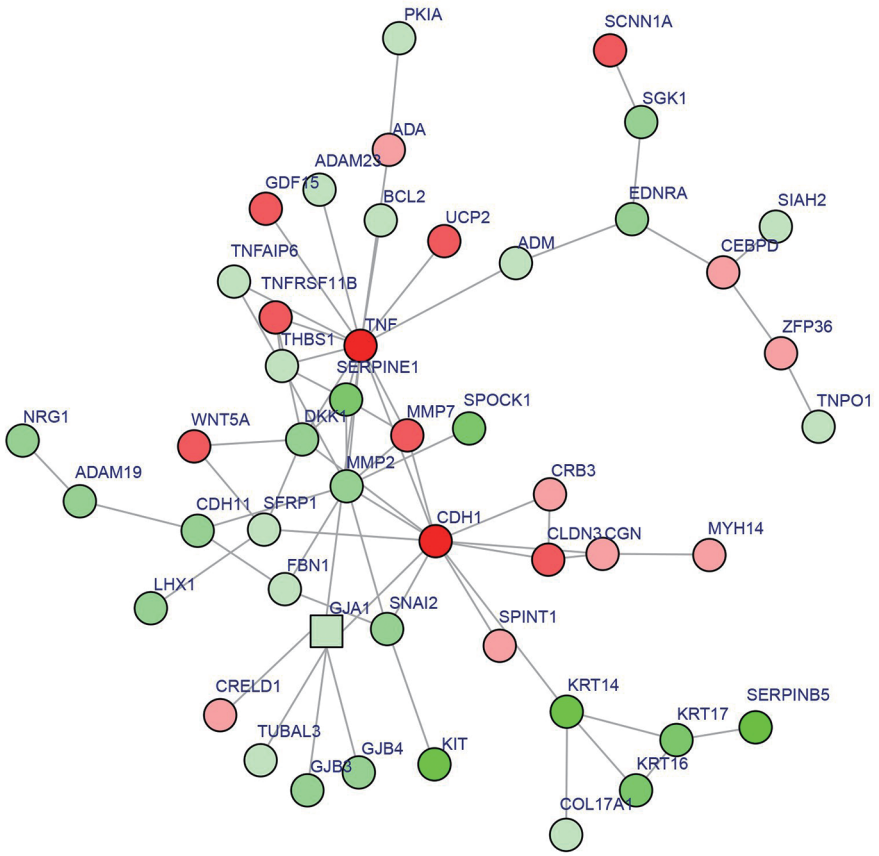

Figure 3 Results of subnetwork analysis. Red nodes, upregulated genes; green nodes, downregulated genes; a darker color means more significant DEGs; a square node means low importance in the subnetwork. DEGs, differentially expressed genes.

potential biomarker in ovarian cancer risk prediction (32). In our study, *CDH1* was enriched in the BP of epithelial morphogenesis and cancer pathways. In addition, survival analysis showed that *CDH1* is associated ovarian cancer prognosis. Therefore, our study further confirmed that *CDH1* is an important prognosis factor for ovarian cancer, using bioinformatics analysis. *CDH1* may be involved in this cancer mainly via the BP of epithelial morphogenesis and cancer pathways.

Previous studies have demonstrated that the miR-30 family plays a critical role in cancer pathogenesis. Ouzounova *et al.* documented that the miR-30 family regulates nonattachment growth of breast cancer cells (33). Another study reported that miR-30-5p acts as a tumor suppressor to regulate multiple myeloma pathogenesis by targeting the Wnt/β-Catenin/BCL-9 pathway (34). In addition, miR-30 also acts as a tumor suppressor to inhibit epithelial-mesenchymal transition (EMT) in prostate cancer via the EGF/Src tyrosine kinase pathway (35). In ovarian cancer, miR-30a overexpression could highly reduce the expression of proliferating cell nuclear antigen (PCNA), a common marker for proliferation, in human ovarian granulosa cells (36). Ye *et al.* reported that miR-30D suppresses transforming growth factor β1 (TGF-β1)-induced EMT by targeting Snail (a major determinant of ovarian cancer invasiveness at the transcription level) in ovarian cancer (37). Taken together, the miR-30 family may act as a tumor suppressor in ovarian cancer. In our study, the miR-30 family, including miR-30A, miR-30B, miR-30C, miR-30D, and miR-30E, was predicted to target several DEGs identified, such as ADAM19, FRMD6, and STC1. Therefore, it is important to further reveal the regulatory mechanism of the miR-30 family in ovarian cancer.

In conclusion, *TNF*, *CDH1*, and the miR-30 family may play significant roles in ovarian cancer. *CDH1* is a

Table 3 Gene ontology (GO) terms and Kyoto Encyclopedia of Genes and Genomes (KEGG) pathways analyses of sub-network

Category	Term	Description	Counts	P value
GO-BP	0002009	Morphogenesis of an epithelium	12	2.71^{-9}
GO-BP	0033993	Response to lipid	12	1.30^{-7}
GO-BP	0007507	Heart development	11	4.96^{-8}
GO-BP	0034329	Cell junction assembly	10	2.92^{-10}
GO-BP	0030198	Extracellular matrix organization	10	5.14^{-8}
GO-BP	0001666	Response to hypoxia	9	3.61^{-8}
GO-BP	0043588	Skin development	9	5.67^{-7}
GO-BP	0042493	Response to drug	9	1.34^{-6}
GO-BP	0016337	Cell-cell adhesion	9	6.22^{-6}
GO-BP	0010035	Response to inorganic substance	8	8.15^{-6}
KEGG	5200	Pathways in cancer	5	1.28^{-2}
KEGG	4530	Tight junction	4	2.48^{-3}
KEGG	4310	Wnt signaling pathway	4	3.95^{-3}
KEGG	5219	Bladder cancer	3	7.90^{-4}
KEGG	4115	p53 signaling pathway	3	3.19^{-3}
KEGG	4960	Aldosterone-regulated sodium reabsorption	2	1.45^{-2}
KEGG	5144	Malaria	2	2.10^{-2}
KEGG	5014	Amyotrophic lateral sclerosis (ALS)	2	2.26^{-2}
KEGG	5130	Pathogenic Escherichia coli infection	2	2.50^{-2}
KEGG	4520	Adherens junction	2	4.09^{-2}

Term, the identification number of GO term or KEGG; description represents the name of GO term or KEGG; counts, the number of genes enriched in GO term or KEGG. BP, biological process; GO, gene ontology; KEGG, Kyoto Encyclopedia of Genes and Genomes.

Table 4 The results for miRNA-target regulating analysis

miRNA	Count	P value	Targets
TGTTTAC, MIR-30A-5P, MIR-30C, MIR-30D, MIR-30B, MIR-30E-5P	25	1.60^{-3}	*FRMD6*; *FAM43A*; *EDNRA*; *EPHB2*; *KHNYN*; *GJA1*; *PCDH17*; *GLDC*; *YPEL2*; *SERPINE1*; *DDIT4*; *SEMA6A*; *FBXL17*; *RAPH1*; *STAC*; *STC1*; *KLF9*; *ESPN*; *B3GNT5*; *ARID5B*; *MEX3B*; *AFAP1L2*; *ADAM19*; *SH3PXD2A*; *RALGPS1*
GCTTGAA, MIR-498	8	5.50^{-3}	*COL1A1*; *DUSP4*; *FLRT2*; *HBP1*; *GJA1*; *YPEL2*; *EPB41L4B*; *C5*
CAGGTCC, MIR-492	5	1.50^{-2}	*TSKU*; *C17orf58*; *STC1*; *ZFP36*; *SH3PXD2A*
ATGTCAC, MIR-489	6	1.54^{-2}	*ADAMTS5*; *PRSS23*; *FBN1*; *NRG1*; *SGK1*; *WNT5A*
TACAATC, MIR-508	5	1.60^{-2}	*ABLIM3*; *HBP1*; *TNPO1*; *LMO3*; *RAPH1*
ACAACTT, MIR-382	5	2.31^{-2}	*HSPA2*; *HSPA14*; *MTUS1*; *SGK1*; *STC1*
TTGCACT, MIR-130A, MIR-301, MIR-130B	16	2.33^{-2}	*VPS37A*; *E2F7*; *FOXF2*; *PLCB1*; *TRIM2*; *NPNT*; *HBP1*; *GJA1*; *KIT*; *SMAD5*; *NR3C2*; *PSD*; *PMEPA1*; *STC1*; *MPPED2*; *TP63*
ACTGTGA, MIR-27A, MIR-27B	17	4.39^{-2}	*CDH11*; *E2F7*; *DCP2*; *EDNRA*; *EPHB2*; *FOXO1*; *FLRT2*; *PCDH17*; *MKNK2*; *MATN3*; *DNAJC27*; *PKIA*; *SEMA6A*; *GPAM*; *SFRP1*; *CA12*; *ADAM19*

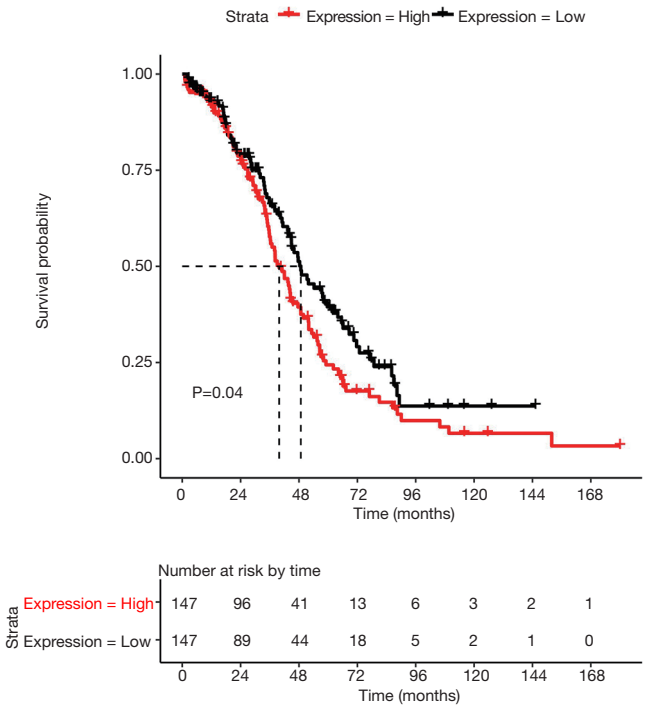

Figure 4 Survival curve for CDH1.

vital prognosis factor for ovarian cancer and might be involved via the BP of epithelium morphogenesis and cancer pathways. *TNF* plays a vital role via the BP of response to lipids, and the MiR-30 family may serve as a tumor suppressor in ovarian cancer pathogenesis. One of the limitations of this study is the lack of verification; thus, further verification experiments are required.

Acknowledgments

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Footnote

Conflicts of Interest: All authors have completed the ICMJE uniform disclosure form (available at http://dx.doi.org/10.21037/tcr.2018.03.10). 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). The institutional ethical approval and informed consent were waived.

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