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Mitochondrial Proteomic and Molecular Network Alterations in Human Ovarian Cancers

Xianquan Zhan and Na Li

Abstract

Mitochondrion is a multi-functional organelle, which plays important role in human ovarian cancers. Mitochondrial quantitative proteomics was used to detect, identify, and quantify proteins from mitochondrial samples prepared from ovarian cancer and normal control ovary tissues. A total of 5115 mitochondrial proteins and 1198 mitochondrial differentially expressed proteins (mtDEPs) were identified in human ovarian cancer compared to control tissues. Pathway network analysis revealed multiple pathway network changes to involve those mitochondrial proteins and mtDEPs. These findings provide the scientific data about the role of mitochondria plays in ovarian cancer, and offer the source for discovery of mitochondrial biomarker for ovarian cancers.

Keywords: mitochondrial proteome, proteomics, molecular networks, biomarker, ovarian cancer

1. Introduction

Mitochondrion is a multi-functional organelle, which is the center of cell energy metabolism, cell signaling, and oxidative stress [1, 2]. Mitochondrial dysfunction is a hallmark in human ovarian cancers, and plays important roles in ovarian carcinogenesis, which has been looked as the cause, biomarker, and therapeutic target for ovarian cancers [3–5]. First, a study finds mitochondrial morphology is significantly changed in ovarian cancers compared to controls. Electron microscopy morphology study shows that mitochondria are abundant and large volume in ovarian cancer cells and tissues [6, 7]. Second, mitochondrial ribosomal protein-encoding genes might be the anti-oncogenes to serve as new biomarkers and therapeutic targets. For example, bcl-2-interacting mitochondrial ribosomal protein L41 (MRPL41) is differentially expressed in carcinomas to associate with various epigenetic states [8]. Mitochondrial ribosomal protein S23 (MRPS23) is involved in cancer cell proliferation, which might serve as the therapeutic target [9]. MRPS15 is significantly upregulated in epithelial breast cells and tissues [10]. Mitochondrial COX1 is expressed abnormally in multiple cancers [11–13]. Many cancer-relevant communication signaling pathways are linked to mitochondrial proteins. Third, mitochondria are the center of oxidative stress, which might be the ‘fuel’ center for a cancer metabolism [10]. The abnormal energy metabolism, namely the Warburg and reverse-Warburg effects, is the important characteristics in cancers [14].

Therefore, mitochondria play important roles in tumorigenesis, proliferation, angiogenesis, invasiveness, and metastasis of cancer cells [14, 15]. Proteins are the important performer in maintaining mitochondrial morphology and functions. It emphasizes the important scientific merits of mitochondrial proteomics in ovarian cancer research and clinical practice [16–22]. Mitochondrial proteins function in mutually interacted molecular pathway network system, which fits the real situation of ovarian cancer that is a multi-cause, multi-process, and multi-result disease [23–25]. It is very difficult to use single-parameter biomarker to predict, diagnose, and prognostic assess ovarian cancer, thus multi-parameter biomarkers or molecule pattern biomarker is necessary for ovarian cancer prediction, prevention, and treatment [26, 27]. Mitochondrial proteomics is an effective approach to systematically investigate the role of mitochondria in ovarian cancer for discovery of reliable mitochondrial protein biomarkers to insight into the molecular mechanism and determination of therapeutic target to mitochondria for ovarian cancers. Quantitative proteomic methods commonly include two-dimensional gel electrophoresis (2DGE) [28, 29] or two-dimensional difference in-gel electrophoresis (2D DIGE) [30] comparative proteomics, and gel-free-based quantitative proteomics [14, 15], for example, isobaric tags for relative and absolute quantification (iTRAQ) [31, 32], tandem mass tag (TMT) [33], or label-free-based quantitative proteomics [34, 35], with different advantages and disadvantages, respectively. Those quantitative proteomic methods can achieve a high-throughput and high-sensitive identification of mitochondrial proteins and post-translational modifications. Currently, stable isotopic labeled large-scale 2DGE coupled with high-sensitivity liquid chromatography-tandem mass spectrometry (LC-MS/MS) is able to detect, identify, and quantify up to least 500,000 protein proteoforms in human tissue proteoforms [36, 37]. iTRAQ, TMT, or label-free is commonly coupled with two-dimensional LC-MS/MS (2DLC-MS/MS), which enables detect, identify, and quantify up to several thousands of proteins and PTMs, even though these gel-free methods are unable to discriminate proteoforms and homolog proteins [38].

Ovarian cancer is a malignant cancer with high morbidity and mortality [39, 40] and without clear molecular mechanisms and effectively reliable biomarkers for its early-stage diagnosis to improve its prognosis. This book chapter used iTRAQ-labeled strong cation exchange chromatography (SCX)-LC-MS/MS method to detect, identify, and quantify mitochondrial proteins and mitochondrial differentially expressed proteins (mtDEPs) between human ovarian cancer and control ovary tissues. The identified mitochondrial proteins and differentially expressed proteins were subject to gene ontology (GO) and Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway network analysis for revealing pathway network alteration in ovarian cancers compared to controls. Those findings provide the scientific data to establish mitochondrial proteomic reference map of ovarian cancer, mtDEP profile and the corresponding pathway network alterations to link with ovarian cancer pathogenesis, which is the resource for discovery of potential biomarkers and mitochondria-targeting drug targets for ovarian cancers.

2. Methods

2.1 Ovarian cancer tissues and preparation of mitochondria protein samples

Seven ovarian cancer tissues and eleven control ovaries with benign gynecologic disease were used in this study. Mitochondria were isolated and purified from ovarian cancer and control tissues with differential-speed centrifugation and

Nycodenz density gradient centrifugation. The purified mitochondria were verified with electron microscopy, and Western blot with different antibodies specific to different subcellular organelles, including COX4I1 (mitochondrion), flotillin-1 (cytomembrane), GM130 (Golgi apparatus), catalase (peroxisomes), cathepsin B (lysosome), and lamin B (cell nucleus). The proteins were extracted from purified mitochondrial samples for iTRAQ-labeled quantitative proteomic analysis. The detailed procedure was described in our previous publications [14, 15].

2.2 iTRAQ-based quantitative proteomics analysis

The prepared mitochondrial proteins (200 µg/each sample) were treated with *N*-hydroxysuccinimide (SDT), followed by reduction, alkylation, digestion with trypsin, and desalination. The tryptic peptide (100 µg/each sample) was labeled with iTRAQ reagent, and each sample was labeled three times. The six labeled tryptic peptide samples were mixed, followed by peptide fractionation with strong cation exchange (SCX) chromatography. Each SCX-fractionated sample was subject to LC-MS/MS analysis on a Q Exactive mass spectrometer (Thermo Scientific) within a 60-min LC separation gradient to obtain MS/MS data. The MS/MS data were used for identity of proteins with MASCOT search engine. The iTRAQ reporter-ion intensities were used to quantify each protein and determine each mtDEPs. The detailed procedure was described in our previous publications [14, 15].

2.3 Bioinformatics and pathway network analysis

The identified proteins and DEPs in mitochondrial samples were subject to GO and KEGG pathway enrichment analysis with Cytoscape, and DAVID online software (<https://david.ncifcrf.gov/home.jsp>). Multiple Experiment Viewer (<https://sourceforge.net/projects/mev-tm4/files/mev-tm4/>) was used to make heat map. GO analysis included cellular component (CC), molecular function (MF), and biological process (BP). PANTHER (<http://www.pantherdb.org/>) was used to further enrich GO CC.

2.4 Validation of mtDEPs and molecular networks in cell models and mitochondrial tissues

Ovarian cancer cells TOV-21G and control cells IOSE80 were used to extract RNAs and proteins. Quantitative real-time PCR (qRT-PCR) was used to measure the mRNA expression levels of GLDC, PCK2, IDH2, CPT2 and HMGCS2 in TOV-21G cells compared to IOSE80 cells. Western blot was used to measure the protein expression levels of GLDC, PCK2, IDH2, CPT2 and HMGCS2 in TOV-21G cells compared to IOSE80 cells, and in ovarian cancer mitochondrial samples compared to control mitochondrial samples; and β -actin was used as internal standard for Western blot analysis.

2.5 Statistical analysis

For GO and KEGG enrichment analyses, *p* values were corrected with Benjamini-Hochberg (FDR) for multiple testing. For qRT-PCR and Western blot, the student's *t*-test was used to measure between-group difference with SPSS software 13.0, and data was presented as the mean $\pm$ SD with *p* < 0.05. Each experiment was repeated at least three times.

3. Results and discussion

3.1 Mitochondrial proteomic profile in human ovarian cancer tissue

iTRAQ-labeling coupled with SCX-LC-MS/MS identified 5115 proteins in mitochondrial samples prepared from human ovarian cancer and control ovary tissues, with at least one peptide sequence matches (PSMs). All of identified proteins was collected in the supplemental Table 1 in our previous publication [15]. Those 5115 proteins mainly distributed within pI 3.81–12.25 and molecular weight (MW) 2.6–1158.2 kDa, and in multiple cell components including cell junction (0.8%), cell part (42.7%), extracellular matrix (0.6%), macromolecular complex (17.8%), organelle (28.2%), and synapse (0.3%) (**Figure 1**). Of them, 2565 (50.14%) were increased, and 2550 (49.86%) were decreased in the abundance in ovarian cancers compared to control ovaries. Furthermore, statistical significance analysis revealed 1198 mtDEPs in human ovarian cancers compared to control ovaries, including 523 (43.66%) upregulated proteins and 675 (56.34%) downregulated proteins, with fold-change ≥ 1.5 or ≤ -1.5 , and $p < 0.05$. Those 1198 mtDEPs were collected in the supplemental Table 1 in our previous publication [14]. Those mtDEPs might be directly linked to ovarian cancer pathogenesis, and the potential resource for biomarkers. From a systemic molecular network angle, one must realize that those non-significant difference proteins might be also important in ovarian cancer pathogenesis because they might be the hub-molecule in a network, because some studies have found that some hub-molecules changed smaller than those boundary molecules in a molecular network in a given condition.

3.2 Pathway networks involved in mitochondrial proteins in ovarian cancer

KEGG pathway network analysis revealed 52 statistically significant pathways to involve mitochondrial proteins including mtDEPs in ovarian cancers compared to

Figure 1. Subcellular location of 5115 proteins analyzed with PANTHER. Modified from Li et al. [15], with permission from Bioscientifica Ltd., copyright 2018.

Category	Term	RT	Count	%	P-value	Benjamini
KEGG_PATHWAY	Lysosome	RT	52	1.3	3.70E-02	2.00E-01
KEGG_PATHWAY	Peroxisome	RT	53	1.5	8.00E-08	4.60E-06
KEGG_PATHWAY	Valine, leucine and isoleucine degradation	RT	41	1.0	1.10E-07	5.50E-06
KEGG_PATHWAY	Phagosome	RT	77	2.1	1.20E-05	2.90E-04
KEGG_PATHWAY	Citrate cycle (TCA cycle)	RT	19	0.8	1.80E-07	7.50E-06
KEGG_PATHWAY	Oxidative phosphorylation	RT	94	2.0	3.40E-07	1.10E-05
KEGG_PATHWAY	Glycolysis/ Gluconeogenesis	RT	33	0.8	1.60E-02	1.20E-01
KEGG_PATHWAY	Fatty acid metabolism	RT	29	0.8	1.90E-03	2.20E-02
KEGG_PATHWAY	Prion diseases	RT	14	0.6	2.20E-03	2.40E-02
KEGG_PATHWAY	Propanoate metabolism	RT	13	0.5	1.40E-03	1.60E-02
KEGG_PATHWAY	Sulfur metabolism	RT	7	0.3	2.90E-03	3.10E-02
KEGG_PATHWAY	Pyruvate metabolism	RT	15	0.6	5.30E-03	4.90E-02
KEGG_PATHWAY	beta-Alanine metabolism	RT	11	0.5	3.10E-02	2.00E-01
KEGG_PATHWAY	Butanoate metabolism	RT	10	0.4	3.30E-02	2.00E-01
KEGG_PATHWAY	Tryptophan metabolism	RT	13	0.5	3.30E-02	2.00E-01
KEGG_PATHWAY	Arginine and proline metabolism	RT	15	0.6	4.00E-02	2.10E-01
KEGG_PATHWAY	Metabolic pathways	RT	524	12.6	1.30E-12	1.80E-10
KEGG_PATHWAY	Carbon metabolism	RT	75	2.2	3.80E-12	3.70E-10
KEGG_PATHWAY	2-Oxocarboxylic acid metabolism	RT	9	0.4	4.30E-03	4.30E-02
KEGG_PATHWAY	Glutathione metabolism	RT	33	0.9	4.90E-05	1.00E-03
KEGG_PATHWAY	Glyoxylate and dicarboxylate metabolism	RT	15	0.6	4.20E-05	9.40E-04
KEGG_PATHWAY	Porphyrin and chlorophyll metabolism	RT	14	0.6	2.10E-02	1.50E-01
KEGG_PATHWAY	Ribosome	RT	110	3.0	3.00E-20	8.80E-18
KEGG_PATHWAY	Biosynthesis of antibiotics	RT	124	3.2	3.50E-11	2.60E-09
KEGG_PATHWAY	Aminoacyl-tRNA biosynthesis	RT	24	1.0	4.30E-04	6.60E-03

Category	Term	RT	Count	%	P-value	Benjamini
KEGG_PATHWAY	Biosynthesis of amino acids	RT	41	1.0	1.10E-03	1.50E-02
KEGG_PATHWAY	Terpenoid backbone biosynthesis	RT	10	0.4	7.80E-03	6.60E-02
KEGG_PATHWAY	Proteasome	RT	30	0.6	3.10E-02	2.00E-01
KEGG_PATHWAY	Protein digestion and absorption	RT	24	1.0	2.30E-02	1.60E-01
KEGG_PATHWAY	Fatty acid degradation	RT	27	0.7	5.30E-03	5.00E-02
KEGG_PATHWAY	Protein processing in endoplasmic reticulum	RT	86	2.4	3.20E-07	1.10E-05
KEGG_PATHWAY	PPAR signaling pathway	RT	20	0.8	1.60E-02	1.20E-01
KEGG_PATHWAY	ECM-receptor interaction	RT	46	1.3	2.00E-04	3.20E-03
KEGG_PATHWAY	Pentose phosphate pathway	RT	11	0.5	1.90E-02	1.40E-01
KEGG_PATHWAY	Focal adhesion	RT	88	2.3	1.30E-03	1.70E-02
KEGG_PATHWAY	Protein export	RT	19	0.5	3.00E-03	3.10E-02
KEGG_PATHWAY	Parkinson's disease	RT	97	2.1	1.20E-06	3.30E-05
KEGG_PATHWAY	Alzheimer's disease	RT	99	2.3	3.60E-06	9.40E-05
KEGG_PATHWAY	Huntington's disease	RT	101	2.3	5.30E-05	1.00E-03
KEGG_PATHWAY	Amoebiasis	RT	36	1.5	5.60E-05	1.00E-03
KEGG_PATHWAY	Complement and coagulation cascades	RT	26	1.1	1.20E-04	2.10E-03
KEGG_PATHWAY	Viral myocarditis	RT	21	0.9	9.10E-04	1.30E-02
KEGG_PATHWAY	Cardiac muscle contraction	RT	25	1.0	1.30E-03	1.60E-02
KEGG_PATHWAY	<i>Staphylococcus aureus</i> infection	RT	18	0.8	7.60E-03	6.70E-02
KEGG_PATHWAY	Bacterial invasion of epithelial cells	RT	38	1.0	1.10E-02	9.00E-02
KEGG_PATHWAY	Vasopressin-regulated water reabsorption	RT	15	0.6	1.30E-02	1.10E-01
KEGG_PATHWAY	Arrhythmogenic right ventricular cardiomyopathy (ARVC)	RT	21	0.9	1.50E-02	1.10E-01
KEGG_PATHWAY	Platelet activation	RT	58	1.3	3.40E-02	2.00E-01

Category	Term	RT	Count	%	P-value	Benjamini
KEGG_PATHWAY	Regulation of actin cytoskeleton	RT	87	2.0	3.50E-02	2.00E-01
KEGG_PATHWAY	Legionellosis	RT	16	0.7	3.70E-02	2.10E-01
KEGG_PATHWAY	Toxoplasmosis	RT	29	1.2	4.50E-02	2.30E-01
KEGG_PATHWAY	Systemic lupus erythematosus	RT	32	1.3	4.90E-02	2.50E-01

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Table 1.
52 statistically significant KEGG pathways enriched from 5115 proteins in ovarian cancers.

Figure 2.
52 statistically significant KEGG pathways enriched from 5115 proteins in ovarian cancers. Modified from Li et al. [15], with permission from Bioscientifica Ltd., copyright 2018.

control ovaries (**Table 1** and **Figure 2**), including phagosome, peroxisome, valine, leucine and isoleucine degradation, lysosome, fatty acid metabolism, citrate cycle (TCA cycle), oxidative phosphorylation, glycolysis/gluconeogenesis, metabolic

Accession number	Protein name	Gene name	Coverage (%)	Unique peptides	PSMs	Ratio (T/N)	t-test p-value
Q8IVP5	FUN14 domain-containing protein 1	FUNDC1	10.97	1	1	1.16	4.82E-2
B4E164	cDNA FLJ56613, highly similar to Serine/threonine-protein kinase TBK1 (EC 2.7.11.1)	TBK1	2.42	1	1	1.25	1.12E-2
O60313	Dynamin-like 120 kDa protein, mitochondrial	OPA1	51.15	44	130	1.19	3.72E-4
Q99623	Prohibitin-2	PHB2	81.61	24	220	1.26	4.44E-4
B4E3V2	cDNA FLJ52854, highly similar to Sequestosome-1	p62	10.47	1	1	1.10	1.96E-1
H0YBC7	BCL2/adenovirus E1B 19 kDa protein-interacting protein 3-like (Fragment)	BNIP3L (NIX)	9.19	1	2	0.77	2.31E-3
A0A0S2Z5I6	Optineurin isoform 3	OPTN	7.94	2	2	0.62	1.01E-2
E7EU96	Casein kinase II subunit alpha	CSNK2A1 (CK)	25.45	6	8	0.84	1.20E-2
Q96HS1	Serine/threonine-protein phosphatase PGAM5, mitochondrial	PGAM5	32.53	10	37	1.49	3.32E-3
B7Z737	cDNA FLJ52784, highly similar to Bcl-2-like 13 protein	Bcl2-L13	13.17	2	2	0.81	3.99E-2

PSMs = peptide sequence matches; MW = molecular weight; Ratio (T/N) = ratio of tumors to normal controls. Reproduced from Li et al. [15], with permission from Bioscientifica Ltd., copyright 2018.

Table 2.
Mitophagy adaptors and regulatory molecules involved the identified proteins in ovarian cancer biological system.

pathways, carbon metabolism, glyoxylate and dicarboxylate metabolism, glutathione metabolism, propanoate metabolism, sulfur metabolism, 2-oxocarboxylic acid metabolism, pyruvate metabolism, porphyrin and chlorophyll metabolism, beta-alanine metabolism, butanoate metabolism, tryptophan metabolism, arginine and proline metabolism, ribosome, protein processing in endoplasmic reticulum, biosynthesis of amino acids, aminoacyl-tRNA biosynthesis, proteasome, protein

Figure 3.
Energy metabolism pathway changed in ovarian cancer. Reproduced from Li et al. [14], with permission from Elsevier Inc., copyright 2018.

digestion and absorption, ECM-receptor interaction, focal adhesion, protein export, signaling pathway, complement and coagulation cascades, platelet activation, PPAR pentose phosphate pathway, fatty acid degradation, vasopressin-regulated water reabsorption, and regulation of actin cytoskeleton. Those pathway systems provided an overall molecular network changes in ovarian cancers, which might be important in ovarian cancer pathogenesis.

Among those altered pathway systems, especially interested is that mitophagy pathway and energy metabolism pathway were significantly changed in ovarian cancers compared to controls. The changed mitophagy pathway in ovarian cancer included phagosome, peroxisome, valine, leucine and isoleucine degradation, lysosome, and fatty acid metabolism pathways [15]. Mitophagy is to engulf any

Accession no.	Protein	Unique peptide	Coverage (%)	PSMs	Ratio (T/N)	p value (t test)
Q01813	Phosphofructokinase, platelet (PFKP)				1.90	2.28E-02
P11177	Pyruvate dehydrogenase E1 component subunit beta (PDHB)	14	52.92	79	1.51	3.25E-03
A0A024R5Z9	Pyruvate kinase (PKM)				2.38	1.50E-04
O43837	Isocitrate dehydrogenase [NAD] subunit beta (IDH3B)	13	41.56	43	1.75	8.69E-03
B4DJV2	Citrate synthase (CS)	13	26.93	73	1.59	4.65E-03
P50213	Isocitrate dehydrogenase [NAD] subunit alpha (IDH3A)	18	47.81	53	1.60	2.27E-02
P48735	Isocitrate dehydrogenase [NADP] (IDH2)	27	56.64	355	2.02	2.07E-03
A0A0A0QN99	Cytochrome b reductase 1 (CYB)	14	4.21	4	1.71	7.60E-03
Q9ULD0	2-oxoglutarate dehydrogenase-like (OGDHL)	13	26.83	58	1.55	1.25E-03
A0A096WB60	NADH-ubiquinone oxidoreductase chain 5 (MT-ND5)	1	5.14	6	0.38	3.34E-04
P07919	Cytochrome b-c1 complex subunit 6 (QCR 6)	5	51.65	18	1.59	1.63E-02
A0A059T3A1	NADH-ubiquinone oxidoreductase chain 2 (MT-ND2)	1	4.61	2	0.38	6.03E-04
P38919	Eukaryotic initiation factor 4A-III (EIF4AIII)	4	11.92	9	0.71	1.48E-02

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Table 3.
Differentially expressed glycolysis/Kreb’s cycle/mitochondrial respiratory chain/RNA binding proteins in EOC.

material in autophagosome, and subsequently fuses with lysosomes to release high-energy substance such as fatty acid and amino acid. Autophagosome also commonly contains mitochondria, proteins, or peroxisome. Mitophagy processes are involved in autophagy machinery, mitophagy adaptors, and regulatory molecules such as Bcl2-L12, p62, OPTN, prohibitin 2, OPA1, CK, PGAM5, BNIP3L(NIX), and FUNDC1 (**Table 2**). These findings were consistent with previous studies. The changed energy metabolism pathway in ovarian cancers included citrate cycle (TCA cycle), oxidative phosphorylation, and glycolysis (**Figure 3**) [14], and the important molecules were significantly changed in three energy metabolism pathways, including PFKM, PKM, PDHB, CS, and IDH2 (**Table 3**). It clearly demonstrated the Warburg and reverse-Warburg effects coexisted in ovarian cancers.

Figure 4.
Validation of potential biomarkers (GLDC, PCK2, IDH2, CPT2 and HMGCS2) in ovarian cancer cell model with qRT-PCR (A) and Western blot (B), and in human mitochondrial samples with Western blot (C). β -actin was used as internal standard. Reproduced from Li et al. [15], with permission from Bioscientifica Ltd., copyright 2018.

3.3 Potential biomarkers for ovarian cancers

Those 5115 mitochondrial proteins including 1198 mtDEPs were the resource of potential biomarkers for ovarian cancers. For example, mtDEPs in mitophagy pathway and energy metabolism pathway might be effective biomarkers and therapeutic targets for ovarian cancer. Five mtDEPs, including GLDC, PCK2, and IDH2 in peroxisome pathway, CPT2 in fatty acid degradation pathway, and HMGCS2 in the valine, leucine and isoleucine degradation pathway were further validated by qRT-PCR and Western blot in ovarian cancer cells compared to normal control cells (Figure 4A and B), and by Western blot in the ovarian cancer tissue mitochondrial samples (Figure 4C). These results also confirmed the results of iTRAQ quantitative proteomics.

4. Conclusions

iTRAQ-labeled SCX-LC-MS/MS quantitative proteomics was an effective method to detect, identify, and quantify mitochondrial proteins and mtDEPs in mitochondrial samples prepared from human ovarian cancer and control ovary tissues. Totally 5115 mitochondrial proteins including 1198 mtDEPs were identified in ovarian

cancers, and 52 statistically significant pathways were identified to involve those mtDEPs. More interested is that this study found mitophagy pathway (phagosome, peroxisome, valine, leucine and isoleucine degradation, lysosome, and fatty acid metabolism), and energy metabolism pathways (citrate cycle, oxidative phosphorylation, and glycolysis) were significantly changed in ovarian cancers. The important molecules Bcl2-L12, p62, OPTN, prohibitin 2, OPA1, CK, PGAM5, BNIP3L(NIX), and FUNDC1 in mitophagy pathway, and PFKM, PKM, PDHB, CS, and IDH2 in energy metabolism pathways were significantly changed. It clearly demonstrated the changed mitophagy and energy metabolism pathways played important roles in ovarian cancers. These findings provide the large-scale proteomic variation profiles and molecular network alterations for ovarian cancer, which are the important scientific data to insight into the roles of mitochondrial dysfunction in ovarian cancer.

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Conflict of interest

We declare that we have no financial and personal relationships with other people or organizations.

Author’s contributions

X.Z. conceived the concept, designed the manuscript, wrote and critically revised the manuscript, coordinated and was responsible for the correspondence work and financial support. N.L. participated in the literature analysis, data analysis, and prepared figures.

Acronyms and abbreviations

GO	gene ontology
iTRAQ	isobaric tags for relative and absolute quantification
KEGG	kyoto encyclopedia of genes and genomes
LC	liquid chromatography
MRPL41	Bcl-2-interacting mitochondrial ribosomal protein L41
MRPS23	mitochondrial ribosomal protein S23
MS/MS	tandem mass spectrometry
PSMs	peptide sequence matches
SCX	strong cation exchange
TMT	tandem mass tag
2DGE	two-dimensional gel electrophoresis
2D DIGE	two-dimensional difference in-gel electrophoresis
2DLC	two-dimensional liquid chromatography

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References

- [1] Deng P, Haynes CM. Mitochondrial dysfunction in cancer: Potential roles of ATF5 and the mitochondrial UPR. *Seminars in Cancer Biology*. 2017;**47**:43-49
- [2] Georgieva E, Ivanova D, Zhelev Z, Bakalova R, Gulubova M, Aoki I. Mitochondrial dysfunction and redox imbalance as a diagnostic marker of "free radical diseases". *Anticancer Research*. 2017;**37**(10):5373-5381
- [3] Kingnate C, Charoenkwan K, Kumfu S, Chattipakorn N, Chattipakorn SC. Possible roles of mitochondrial dynamics and the effects of pharmacological interventions in chemoresistant ovarian cancer. *eBioMedicine*. 2018;**34**:256-266
- [4] Lim W, Ryu S, Bazer FW, Kim SM, Song G. Chrysin attenuates progression of ovarian cancer cells by regulating signaling cascades and mitochondrial dysfunction. *Journal of Cellular Physiology*. 2018;**233**(4):3129-3140
- [5] Chowdhury SR, Ray U, Chatterjee BP, Roy SS. Targeted apoptosis in ovarian cancer cells through mitochondrial dysfunction in response to *Sambucus nigra* agglutinin. *Cell Death & Disease*. 2017;**8**(5):e2762
- [6] Salazar H, Merkow LP, Walter WS, Pardo M. Human ovarian neoplasms: Light and electron microscopic correlations. II. The clear cell tumor. *Obstetrics and Gynecology*. 1974;**44**(4):551-563
- [7] Saitou M, Isonishi S, Hamada T, Kiyokawa T, Tachibana T, Ishikawa H, et al. Mitochondrial ultrastructure-associated chemotherapy response in ovarian cancer. *Oncology Reports*. 2009;**21**(1):199-204
- [8] Kim TW, Kim B, Kim JH, Kang S, Park SB, Jeong G, et al. Nuclear-encoded mitochondrial MTO1 and MRPL41 are regulated in an opposite epigenetic mode based on estrogen receptor status in breast cancer. *BMC Cancer*. 2013;**13**:502
- [9] Pu M, Wang J, Huang Q, Zhao G, Xia C, Shang R, et al. High MRPS23 expression contributes to hepatocellular carcinoma proliferation and indicates poor survival outcomes. *Tumor Biology*. 2017;**39**:1393380537
- [10] Sotgia F, Whitaker-Menezes D, Martinez-Outschoorn UE, Salem AF, Tsirigos A, Lamb R, et al. Mitochondria "fuel" breast cancer metabolism: Fifteen markers of mitochondrial biogenesis label epithelial cancer cells, but are excluded from adjacent stromal cells. *Cell Cycle*. 2012;**11**:4390-4401
- [11] Ksiezakowska-Lakoma K, Kulczycka-Wojdala D, Kulig A, Baum M, Wilczynski JR. The presence of A5935G, G5949A, G6081A, G6267A, T9540C mutations in MT-CO1 and MT-CO3 genes and other variants of MT-CO1 and MT-CO3 gene fragments in the study population diagnosed with endometrial cancer. *Ginekologia Polska*. 2017;**88**:343-348
- [12] Huhta H, Helminen O, Palomaki S, Kauppila JH, Saarnio J, Lehenkari PP, et al. Intratumoral lactate metabolism in Barrett's esophagus and adenocarcinoma. *Oncotarget*. 2017;**8**:22894-22902
- [13] Michalak S, Rybacka-Mossakowska J, Gazdulska J, Golda-Gocka I, Ramlau R. The effect on cognition of mitochondrial respiratory system proteins in peripheral blood mononuclear cells in the course of lung cancer. *Advances in Experimental Medicine and Biology*. 2016;**911**:45-52
- [14] Li N, Zhan X, Zhan X. The lncRNA SNHG3 regulates energy metabolism of ovarian cancer by an analysis of

mitochondrial proteomes. *Gynecologic Oncology*. 2018;**150**:343-354

[15] Li N, Li H, Cao L, Zhan X. Quantitative analysis of the mitochondrial proteome in human ovarian carcinomas. *Endocrine-Related Cancer*. 2018;**25**:909-931

[16] Zhan X, Zhou T, Li N, Li H. The differentially mitochondrial proteomic dataset in human ovarian cancer relative to control tissues. *Data in Brief*. 2018;**20**:459-462

[17] Chen M, Huang H, He H, Ying W, Liu X, Dai Z, et al. Quantitative proteomic analysis of mitochondria from human ovarian cancer cells and their paclitaxel-resistant sublines. *Cancer Science*. 2015;**106**(8):1075-1083

[18] Chen X, Wei S, Ma Y, Lu J, Niu G, Xue Y, et al. Quantitative proteomics analysis identifies mitochondria as therapeutic targets of multidrug-resistance in ovarian cancer. *Theranostics*. 2014;**4**(12):1164-1175

[19] Chappell NP, Teng PN, Hood BL, Wang G, Darcy KM, Hamilton CA, et al. Mitochondrial proteomic analysis of cisplatin resistance in ovarian cancer. *Journal of Proteome Research*. 2012;**11**(9):4605-4614

[20] Dai Z, Yin J, He H, Li W, Hou C, Qian X, et al. Mitochondrial comparative proteomics of human ovarian cancer cells and their platinum-resistant sublines. *Proteomics*. 2010;**10**(21):3789-3799

[21] Tian Y, Tan AC, Sun X, Olson MT, Xie Z, Jinawath N, et al. Quantitative proteomic analysis of ovarian cancer cells identified mitochondrial proteins associated with paclitaxel resistance. *Proteomics. Clinical Applications*. 2009;**3**(11):1288-1295

[22] Young TW, Mei FC, Yang G, Thompson-Lanza JA, Liu J, Cheng X. Activation of antioxidant pathways in

ras-mediated oncogenic transformation of human surface ovarian epithelial cells revealed by functional proteomics and mass spectrometry. *Cancer Research*. 2004;**64**(13):4577-4584

[23] Cheng T, Zhan X. Pattern recognition for predictive, preventive, and personalized medicine in cancer. *The EPMA Journal*. 2017;**8**:51-60

[24] Zhan X, Long Y, Zhan X, Mu Y. Consideration of statistical vs. biological significances for omics data-based pathway network analysis. *Med One*. 2017;**2**:e170002

[25] Hu R, Wang X, Zhan X. Multi-parameter systematic strategies for predictive, preventive and personalised medicine in cancer. *The EPMA Journal*. 2013;**4**:2

[26] Gonzalez-Angulo AM, Iwamoto T, Liu S, Chen H, Do KA, Hortobagyi GN, et al. Gene expression, molecular class changes, and pathway analysis after neoadjuvant systemic therapy for breast cancer. *Clinical Cancer Research*. 2012;**18**:1109-1119

[27] Lu M, Zhan X. The crucial role of multiomic approach in cancer research and clinically relevant outcomes. *The EPMA Journal*. 2018;**9**(1):77-102

[28] Wang X, Guo T, Peng F, Long Y, Mu Y, Yang H, et al. Proteomic and functional profiles of a follicle-stimulating hormone positive human nonfunctional pituitary adenoma. *Electrophoresis*. 2015;**36**:1289-1304

[29] Zhan X, Wang X, Long Y, Desiderio DM. Heterogeneity analysis of the proteomes in clinically nonfunctional pituitary adenomas. *BMC Medical Genomics*. 2014;**7**:69

[30] Liu J, Zhan X, Li M, Li G, Zhang P, Xiao Z, et al. Mitochondrial proteomics of nasopharyngeal carcinoma metastasis. *BMC Medical Genomics*. 2012;**5**:62

- [31] Karabudak AA, Hafner J, Shetty V, Chen S, Secord AA, Morse MA, et al. Autoantibody biomarkers identified by proteomics methods distinguish ovarian cancer from non-ovarian cancer with various CA-125 levels. *Journal of Cancer Research and Clinical Oncology*. 2013;**139**:1757-1770
- [32] Nie S, Lo A, Zhu J, Wu J, Ruffin MT, Lubman DM. Isobaric protein-level labeling strategy for serum glycoprotein quantification analysis by liquid chromatography-tandem mass spectrometry. *Analytical Chemistry*. 2013;**85**:5353-5357
- [33] Wang Z, Liu F, Ye S, Jiang P, Yu X, Xu J, et al. Plasma proteome profiling of high-altitude polycythemia using TMT-based quantitative proteomics approach. *Journal of Proteomics*. 2019;**194**:60-69
- [34] Russell JD, Scalf M, Book AJ, Ladrer DT, Vierstra RD, Smith LM, et al. Characterization and quantification of intact 26S proteasome proteins by real-time measurement of intrinsic fluorescence prior to top-down mass spectrometry. *PLoS ONE*. 2013;**8**:e58157
- [35] Merl J, Deeg CA, Swadzba ME, Ueffing M, Hauck SM. Identification of autoantigens in body fluids by combining pull-downs and organic precipitations of intact immune complexes with quantitative label-free mass spectrometry. *Journal of Proteome Research*. 2013;**12**:5656-5665
- [36] Zhan X, Yang H, Peng F, Li J, Mu Y, Long Y, et al. How many proteins can be identified in a 2DE gel spot within an analysis of a complex human cancer tissue proteome? *Electrophoresis*. 2018;**39**(7):965-980
- [37] Zhan X, Li N, Zhan X, Qian S. Revival of 2DE-LC/MS in proteomics and its potential for large-scale study of human proteoforms. *Med One*. 2018;**3**(5):e180008
- [38] Zhan X, Long Y, Lu M. Exploration of variations in proteome and metabolome for predictive diagnostics and personalized treatment algorithms: Innovative approach and examples for potential clinical application. *Journal of Proteomics*. 2018;**188**:30-40
- [39] Torre LA, Trabert B, DeSantis CE, Miller KD, Samimi G, Runowicz CD, et al. Ovarian cancer statistics 2018. *CA: A Cancer Journal for Clinicians*. 2018;**68**(4):284-296
- [40] Pinsky PF, Miller EA, Zhu CS, Prorok PC. Overall mortality in men and women in the randomized prostate, lung, colorectal, and ovarian cancer screening trial. *Journal of Medical Screening*. 2019;**2019**:969141319839097