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Chapter

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 ± 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 $\geq 1.5$ or $\leq -1.5$, 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 control ovaries. 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   | Aminosyrl-tRNA biosynthesis                  | RT | 24    | 1.0  | 4.30E−04  | 6.60E−03  |
## Mitochondria and Brain Disorders

| Category | Term | RT | Count | %  | P-value | Benjamini |
|----------|------|----|-------|----|---------|-----------|
| KEGG_PATHWAY | Biosynthesis of amino acids | RT | 41 | 1.0 | 1.1E−03 | 1.50E−02 |
| KEGG_PATHWAY | Terpenoid backbone biosynthesis | RT | 10 | 0.4 | 7.80E−03 | 1.60E−02 |
| KEGG_PATHWAY | Proteasome | RT | 20 | 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 | 66 | 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 | Staphylococcus aureus 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 |
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       |
| Q96H1S1          | 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        | 1               | 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.
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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

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

| 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 |
| A0A024RSZ9    | 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 (OGDHHL) | 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 |

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

Modified from Li et al. [14], with permission from Elsevier Inc., 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

| Acronym | Definition |
|---------|------------|
| 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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