Identification of endometriosis-associated genes and pathways based on bioinformatic analysis

Ting Wang, MD, Ruoan Jiang, MD, Yingsha Yao, MD, Linhua Qian, MD, Yu Zhao, MD, Xiufeng Huang, MD

Abstract
Endometriosis is associated with dysmenorrhea, chronic pelvic pain, and infertility. The specific mechanism of endometriosis remains unclear. The aim of this study was to apply a bioinformatics approach to reveal related pathways or genes involved in the development of endometriosis.

The gene expression profiles of GSE25628, GSE5108, and GSE7305 were downloaded from the gene expression omnibus (GEO) database. Differentially expressed gene (DEG) analysis was performed using GEO2R. The database for annotation, visualization, and integrated discovery (DAVID) was utilized to analyze the functional enrichment, gene ontology (GO) and kyoto encyclopedia of genes and genomes (KEGG) pathway of the differentially expressed genes. A protein-protein interaction (PPI) network was constructed and module analysis was performed using search tool for the retrieval of interacting genes and cytoscape.

A total of 119 common differentially expressed genes were extracted, consisting of 51 downregulated genes and 68 upregulated genes. The enriched functions and pathways of the DEGs and hub genes include DNA strand separation, cellular proliferation, degradation of the extracellular matrix, encoding of smooth muscle myosin as a major contractile protein, exiting the proliferative cycle and entering quiescence, growth regulation, and implication in a wide variety of biological processes.

A bioinformatics approach combined with cell experiments in this study revealed that identifying DEGs and hub genes leads to better understanding of the molecular mechanisms underlying the progression of endometriosis, and efficient biomarkers underlying this pathway need to be further investigated.

Abbreviations: DAVID = database for annotation, visualization, and integrated discovery, DEG = differentially expressed gene, GEO = gene expression omnibus, GO = gene ontology, KEGG = kyoto encyclopedia of genes and genomes, LAMA4 = laminin subunit alpha 4, MCODE = molecular complex detection, MYH11 = myosin heavy chain 11, PPI = protein-protein interaction, QSOX1 = quiescin sulfhydryl oxidase 1.

Keywords: bioinformatic analysis, endometriosis, genes, pathways

1. Introduction
Endometriosis is a debilitating disease with features of chronic inflammation and is defined as the presence of functional endometrial glands and stroma outside the uterine cavity, the most common locations for the ectopic endometrial implants being the ovaries, the fossa ovarica, the uterosacral ligaments, and the posterior cul-de-sac.[1] Endometriosis currently affects approximately 5.5 million reproductive-aged women in the United States.[2] Worldwide, it represents a significant cause of morbidity in approximately 10% to 15% of women in their reproductive years.[3] Endometriosis is one of the major causes of economic burden and compromised quality of life in a very large percentage of Asian women.[4] While it is perceived as a benign condition, recent research has shown that it may be a significant cause of infertility and metastatic cancer.[5]

Although the cause of endometriosis remains unclear, genetic,[6,7] hormonal, and immunological factors[8] as well as endometrial progenitor cells have been implicated in the development of lesions.[9] Endometriosis-associated genes and pathways still remain unclear. This present study aimed to identify critical genes and pathways contributing to endometriosis.

2. Materials and methods
2.1. Microarray data
The gene expression omnibus (GEO) database (http://www.ncbi.nlm.nih.gov/geo) is a public functional genomics data repository of high-throughput gene expression data, chips, and microarrays.[10] Three gene expression datasets [GSE25628 (Platforms:
Differentially expressed gene (DEG) analysis for the 3 datasets were carried out using GEO2R (http://www.ncbi.nlm.nih.gov/geo/geo2r/). GEO2R is an interactive web tool that allows users to compare 2 or more datasets in a GEO series in order to identify DEGs across experimental conditions. For the 3 gene expression datasets, an adjusted 
P
value of <.01 and a ≥ fourfold change were set as the cutoff to be considered a statistically significant differentially expressed gene. The common differentially expressed genes of the 3 datasets were selected using the Draw Venn Diagram web tool (http://bioinformatics.psb.ugent.be/webtools/Venn/).

2.3. Kyoto encyclopedia of genes and genomes and gene ontology enrichment analyses of differentially expressed genes

The database for annotation, visualization, and integrated discovery (DAVID; http://david.ncifcrf.gov) (version 6.8) is an online biological information database that integrates biological data and analysis tools, and provides a comprehensive set of functional annotation information of genes and proteins for users to extract biological information. KEGG is a database resource for understanding high-level functions and biological systems from large-scale molecular datasets generated by high-throughput experimental technologies. GO is a major bioinformatics tool to annotate genes and analyze the biological processes of these genes. In order to analyze the function of DEGs, biological analyzes were performed using the DAVID online database. 
P
value of <.0001 for GO functional enrichment analysis and 
P
value of <.05 for KEGG pathway enrichment analysis were considered statistically significant.

2.4. Protein-protein interaction network construction and module analysis

The PPI network was predicted using the search tool for the retrieval of interacting genes (STRING; http://string-db.org) (version 11.0) online database. Analyzing the functional interactions between proteins may provide insights into the mechanisms of generation or development of diseases. In the present study, a PPI network of DEGs was constructed using the search tool for the retrieval of interacting genes database, and an interaction with a combined score of >0.4 was considered statistically significant. Cytoscape (version 3.8.0) is an open-source bioinformatics software platform for visualizing molecular interaction networks. The molecular complex detection (MCODE) (version 1.6.1) plug-in of cytoscape is an application for clustering a given network based on topology to discover densely connected regions. The PPI networks were drawn using cytoscape and the most significant module in the PPI networks was identified using MCODE. The criteria for selection were as follows: MCODE score >5, degree of cut-off=2, node score cut-off=0.2, Max depth=100, and k-score=2. Subsequently, the KEGG and GO analyzes for genes in this module were performed using DAVID.

2.5. Hub gene selection and analysis

Genes with a connectivity degree ≥10 were selected as hub genes. Biological process analysis of hub genes was performed and visualized using the Biological Networks Gene Oncology tool (version 3.0.4) plugin of cytoscape.

The Institutional Review Board or Ethics Committee approval was not needed.

3. Results

3.1. Overview of the gene expression omnibus microarray data and identification of differentially expressed genes

For GSE25628, GSE5108, and GSE7305, an adjusted 
P
value of <.01 and a ≥ fourfold change were set as the cutoff to be considered a statistically significant differentially expressed gene. A total of 2514 elements in GSE25628, 2767 elements in GSE5108, and 2403 elements in GSE7305 were selected. Using Draw Venn Diagram, 2001, 2131, and 1678 unique elements were identified in GSE25628, GSE5108, and GSE7305, respectively. A total of 119 common DEGs were extracted from the 3 groups after comparing (Fig. 1), and consisted of 51 downregulated genes and 68 upregulated genes.

3.2. Gene ontology functional enrichment analysis

Three categories of GO functional annotation analysis were performed on these potential target genes mentioned above, including biological process, cellular component, and molecular function. As shown in Table 1, the GO analysis results for the

Figure 1. Venn diagram. Differentially expressed genes with an adjusted 
P
value of <.01 and a ≥ four-fold change were selected from among the gene expression profiling sets GSE25628, GSE5108, and GSE7305. The 3 datasets showed an overlap of 119 genes.
common DEGs indicated circulatory system development and regulation of the meiotic cell cycle in the biological process category; extracellular region, bicellular, and apical junction in the cellular component category; and protein binding, metallo-carboxypeptidase activity, and carboxypeptidase activity in the molecular function category.

3.3. Kyoto encyclopedia of genes and genomes pathway enrichment analysis

To further analyze the enriched pathways of these DEGs, KEGG pathway enrichment analysis was subsequently conducted. As shown in Table 2, KEGG pathway analysis revealed that the DEGs were mainly enriched in leukocyte transendothelial

### Table 1

| Category | Term | Description                                      | Count | Fold enrichment |
|----------|------|--------------------------------------------------|-------|----------------|
| GO MF    | GO:0030500 | Protein binding, bridging                        |   2   | 33.48245614    |
| GO MF    | GO:0009847 | Protein binding, bridging                        |   4   | 17.74306445    |
| GO MF    | GO:0009848 | Protein binding, bridging                        |   4   | 17.02375102    |
| GO MF    | GO:0009849 | Protein binding, bridging                        |   4   | 14.82067736    |
| GO MF    | GO:0009850 | Protein binding, bridging                        |   3   | 20.95959596    |
| GO MF    | GO:0009851 | Protein binding, bridging                        |   11  | 3.825756266    |
| GO MF    | GO:0009852 | Protein binding, bridging                        |   8   | 5.05283318     |
| GO MF    | GO:0009853 | Protein binding, bridging                        |   7   | 5.18365847     |
| GO MF    | GO:0009854 | Protein binding, bridging                        |   6   | 4.16886169     |
| GO MF    | GO:0009855 | Protein binding, bridging                        |   6   | 5.91474767     |
| GO MF    | GO:0009856 | Protein binding, bridging                        |   6   | 7.85630613     |
| GO MF    | GO:0009857 | Protein binding, bridging                        |   5   | 5.39892665     |
| GO MF    | GO:0009858 | Protein binding, bridging                        |   3   | 5.38982665     |

### Table 2

| KEGG pathway | Term        | Description                                      | Count | Fold enrichment |
|--------------|-------------|--------------------------------------------------|-------|----------------|
| Downregulated | hsa05200    | Pathways in cancer                               |   5   | 4.326923077    |
| Downregulated | hsa04270    | Vascular smooth muscle contraction               |   3   | 8.728448276    |
| Upregulated  | hsa0510     | Focal adhesion                                   |   6   | 5.50062112     |
| Upregulated  | hsa0412     | ECM-receptor interaction                         |   4   | 8.66995074     |
| Upregulated  | hsa0430     | Tight junction                                    |   4   | 8.76233766     |
| Upregulated  | hsa0463     | Leukocyte transendothelial migration             |   4   | 6.60142605     |
| Upregulated  | hsa0514     | Malaria                                           |   3   | 11.80758017    |
| Upregulated  | hsa0411     | Phagosome                                         |   4   | 5.67226890     |
| Upregulated  | hsa00590    | Arachidonic acid metabolism                      |   3   | 9.48477751     |

KEGG = Kyoto encyclopedia of genes and genomes, ECM-receptor = extracellular matrix-receptor.
migration, cellular junction, vascular smooth muscle contraction, focal adhesion, malaria, phagosome, and arachidonic acid metabolism signaling pathways.

3.4. Protein-protein interaction network construction and module analysis

A PPI network of DEGs was constructed (Fig. 2) and the most significant module was obtained using cytoscape (Fig. 3). The functional analyzes of genes involved in this module were performed using DAVID. Results showed that genes in this module were mainly enriched in glycosaminoglycan binding, sulfur compound binding, metalloproteinase activity, heparin binding, extracellular matrix-receptor interaction, vascular smooth muscle contraction, tight junction, and focal adhesion (Table 3).

3.5. Hub gene selection and analysis

A total of 5 genes with a connectivity degree ≥10 were selected as hub genes. The names, abbreviations, and functions for these hub genes are shown in Table 4, and the biological process analysis of the hub genes is shown in Figure 4.

4. Discussion

Endometriosis is a chronic inflammatory hormonal, immune, systemic, and heterogeneous disease defined as the presence of endometrial glands and stroma-like lesions outside of the uterus, often associated with inflammation, severe and chronic pain, and infertility.[3] Diagnosis of endometriosis should be based on patient interviews, examination, and imaging.[21] Lesions identified during laparoscopy are categorized as superficial peritoneal lesions, endometriomas, or deep infiltrating nodules, with a high degree of individual variability in lesion color, size, and morphology.[22] Histopathological analysis requires the presence of at least 2 features - endometrial epithelium, endometrial glands, endometrial stroma, or hemosiderin-filled macrophages - for a diagnosis of endometriosis.[23]
Table 3
GO and KEGG analysis of DEGs in the most significant module (P < .01).

| Pathway ID | Pathway description | Count | FDR    |
|------------|---------------------|-------|--------|
| GO:000539  | Glycosaminoglycan binding | 8     | 0.0675376 |
| GO:1901681 | Sulfur compound binding | 8     | 0.1265086 |
| GO:0004181 | Metallo carboxypeptidase activity | 4     | 0.9614553 |
| GO:0008201 | Heparin binding | 6     | 0.9705039 |
| hsa04512   | ECM-receptor interaction | 6     | 0.8194419 |
| hsa04270   | Vascular smooth muscle contraction | 6     | 3.4249874 |
| hsa04530   | Tight junction | 5     | 6.3134673 |
| hsa04510   | Focal adhesion | 7     | 7.225584 |

GO = gene ontology, KEGG = kyoto encyclopedia of genes and genomes, DEGs = differentially expressed genes, FDR = false discovery rate, ECM-receptor = extracellular matrix-receptor.

The most well-accepted pathophysiological hypothesis for endometriosis is based on retrograde menstruation. Other hypotheses proposed include Müllerian metaplasia, lymphovascular emboli of endometrial cells, and proliferation of endometrial stem cells or bone marrow progenitors.

It is understood that several factors are involved in the pathogenesis and progression of endometriosis, including inflammation, angiogenesis, cytokine/chemokine expression, and endocrine alterations such as steroid and steroid receptor expression.

Angiogenesis is the formation of new blood vessels, and subsequently, is a key process in forming functional blood vessels to ectopic menstrual tissue for the establishment and maintenance of endometriotic lesions. The vascular endothelial growth factor protein family is well known for its roles in angiogenesis. A variety of rodent endometriosis models have shown that vascular endothelial growth factor levels increase in endometriosis-like lesions. Matrix metalloproteinases are proteases required for reorganizing existing blood vessels during budding angiogenesis. They play a known role in endometriosis.

Cytokines and chemokines are emerging as key players in endometriosis pathobiology. Altered levels of a large number of cytokines and chemokines have been found in cyst fluid removed from endometriomas and chocolate cysts.

Endometriosis is intimately associated with steroid metabolism and associated pathways, corresponding to the paramount roles estrogen receptors and progesterone receptors play in uterine biology. Many studies have shown that endometriosis is estrogen dependent and is regulated through alpha and beta estrogen receptors (ESR1 and ESR2).

Although, endometriosis is intimately associated with interaction between inflammation and the endocrine system, which was considered the major mechanism of endometriosis, the exact etiology and pathophysiological mechanisms of endometriosis still remain unclear.

In the present study, a bioinformatics approach was applied to reveal the possible pathways and critical genes related to the development of endometriosis. As some significant biological functions were considered to be common, either in normal endometrium or the development of endometriosis, 3 gene expression datasets (GSE25628, GSE5108, and GSE7305) were analyzed to obtain DEGs. A total of 119 DEGs were identified among the 3 datasets, including 51 downregulated genes and 68 upregulated genes. GO and KEGG enrichment analyzes were performed to explore interactions between the DEGs. The genes were mainly enriched in protein binding, bridging, biccullar tight junction, cell cycle, extracellular region, metallo carboxypeptidase activity, and circulatory and cardiovascular system development.

GO and KEGG enrichment analyzes revealed that changes in the most significant modules were mainly enriched in glycosaminoglycan binding, sulfur compound binding, metallo carboxypeptidase activity, heparin binding, extracellular matrix-receptor interaction, vascular smooth muscle contraction, tight junction, and focal adhesion.

Five genes with a connectivity degree ≥10 were selected as hub genes. Among these hub genes, helicase, lymphoid-specific (HELLS) showed node degrees with 28 of them.

The gene TIMP metalloproteinase inhibitor 2 is a member of the TIMP gene family. The proteins encoded by this gene family are natural inhibitors of the matrix metalloproteinases, a group of peptidases involved in degradation of the extracellular matrix. In addition to an inhibitory role against metalloproteinases, the encoded protein has a unique role among TIMP family members in its ability to directly suppress the proliferation of endothelial cells. As a result, the encoded protein may be critical to the maintenance of tissue homeostasis by suppressing the proliferation of quiescent tissues in response to angiogenic factors, and by inhibiting protease activity in tissues undergoing remodeling of the extracellular matrix. Increased gene TIMP metallopepti-

Table 4
Functional roles of 5 hub genes.

| Number | Gene symbol | Full name                                | Function                                                      |
|--------|-------------|------------------------------------------|---------------------------------------------------------------|
| 1      | HELLs       | Helicase, lymphoid specific              | DNA strand separation, cellular proliferation                |
| 2      | TIMP2       | TIMP metalloproteinase inhibitor 2       | Degradation of the extracellular matrix                       |
| 3      | MYH11       | Myosin heavy chain 11                    | Encode smooth muscle myosin as a major contractile protein    |
| 4      | QSOX1       | Quiescin sulfhydryl oxidase 1            | Exit the proliferative cycle and enter quiescence, growth regulation |
| 5      | LAMA4       | Laminin subunit alpha 4                  | Implicate in a wide variety of biological processes including cell adhesion, differentiation, migration, signaling, neurite outgrowth and metastasis |
dase inhibitor 2 expression has been reported in endometriosis patients.\textsuperscript{[35]}

Literature retrieval results showed that a connection between endometriosis and the hub genes HELLS, myosin heavy chain 11 (MYH11), quiescin sulfhydryl oxidase 1 (QSOX1), and LAMA4 has not been widely reported.

The gene HELLS, a protein coding gene, encodes a lymphoid-specific helicase. Other helicases function in processes involving DNA strand separation, including replication, repair, recombination, and transcription.\textsuperscript{[36]} This protein is thought to be involved in cellular proliferation, associated with the occurrence of cancer. Cheuk-Ting Law found that HELLS, an SWI2/SNF2 chromatin remodeling enzyme, was remarkably overexpressed in hepatocellular carcinoma.\textsuperscript{[37]} A study by Xi Liu revealed that HELLS was significantly upregulated in colorectal cancer.\textsuperscript{[38]}

Aside from cancers, immunodeficiency–centromeric instability–facial anomalies syndrome is caused by ATPase-defective point mutations in HELLS.\textsuperscript{[39]}

MYH11, a protein coding gene that belongs to the myosin heavy chain family, encodes smooth muscle myosin. The gene product is a hexameric protein subunit that consists of 2 heavy chain subunits and 2 pairs of non-identical light chain subunits. It functions as a major contractile protein, converting chemical energy into mechanical energy through adenosine-triphosphate hydrolysis. MYH11 has been traditionally thought of as a specific and exclusive marker for vascular smooth muscle cells and pericytes.\textsuperscript{[40]} Bruce A. Corliss identified MYH11 as a marker of a subset of corneal endothelial cells.\textsuperscript{[41]} Recent studies have identified homozygous or compound heterozygous variants in MYH11 as a candidate gene for megacystis-microcolon-intestinal hypoperistalsis syndrome, a rare and severe disorder characterized by functional obstruction in the urinary and gastrointestinal tract.\textsuperscript{[42,43]}

QSOX1, a protein coding gene that is a member of 2 long-standing gene families, encodes a protein that contains domains of thioredoxin and resolvin E1 receptor.\textsuperscript{[44]} Gene expression is induced as fibroblasts begin to exit the proliferative cycle and enter quiescence, suggesting that this gene plays an important role in growth regulation.\textsuperscript{[45]} Two transcript variants encoding 2 different isoforms have been found for this gene. Amber L Fifield concluded that overexpressed QSOX1 is a potential novel anticancer agent in tumors.\textsuperscript{[46]}

Laminin subunit alpha 4 is a protein-encoding gene. Laminins, a family of extracellular matrix glycoproteins, are the major noncollagenous constituent of basement membranes. They have been implicated in a wide variety of biological processes, including cell adhesion, differentiation, migration, signaling, neurite outgrowth, and metastasis. Down-regulating LAMA4 expression inhibits the proliferation and migration of breast cancer,\textsuperscript{[47]} renal cell carcinoma,\textsuperscript{[48]} gastric cancer,\textsuperscript{[49]} and ovarian cancer.\textsuperscript{[50]}

The biological process analysis the specific signaling pathways involved in the key genes and explore the potential molecular mechanisms by which the key genes influence endometriosis progression. High expression of hub genes mainly enriched maintenance of DNA methylation, protein thiol-disulfide exchange, methylation-dependent chromatin silencing, skeletal myofibril assembly, (skeletal/striated muscle) myosin thick filament assembly, system development, cardiac muscle fiber development, extracellular matrix assembly, centromere complex
assembly, elastic fiber assembly, (centromeric) heterochromatin formation, heterochromatin organization, chromatin remodeling at centromere and other signaling pathways. (Fig. 4). Kuei-Yang Hsiao’s study found that Epigenetic modifications, including DNA methylation, histone modification, and microRNA expression, are involved in the pathogenesis of endometriosis. Mohamed G Ibrahim’s study got the conclusion that the nuclear membrane irregularities are evidence for ultramicro-trauma in adenomyosis. But the connections between endometriosis and the other enriched signaling pathways has not been widely reported. Further studies are needed to confirm the role of these improved signaling pathways in endometriosis.

However, there are still some limitations in this study:

1. A lack of research on detailed molecular mechanisms that hub genes regulate endometriosis progression.
2. elated animal studies are deficient in this study.

5. Conclusion
In conclusion, the present study was designed to identify DEGs that may be involved in the progression of endometriosis. A total of 119 DEGs and 5 hub genes were identified. However, further studies are needed to elucidate the biological function of these genes in endometriosis.

Author contributions
Conceptualization: Ting Wang.
Data curation: Ruoan Jiang, Linhua Qian.
Formal analysis: Ruoan Jiang, Yingsha Yao.
Methodology: Ting Wang, Ruoan Jiang.
Project administration: Xiufeng Huang.
Software: Yingsha Yao, Yu Zhao.
Validation: Ting Wang.
Writing – original draft: Yingsha Yao.
Writing – review & editing: Ruoan Jiang.

References
[1] Shim JY, Laufer MR. Adolescent endometriosis: an update. J Pediatr Adolesc Gynecol 2020;33:112–9.
[2] Greene AD, Lang SA, Kendziorzki JA, et al. Endometriosis: where are we and where are we going? Reproduction 2016;152:R63–78.
[3] Giudice LC, Kao LC. Endometriosis. Lancet 2004;364:1789–99.
[4] Han XT, Guo HY, Kong DL, et al. Analysis of characteristics and overrepresentation of gene ontology categories in biological networks. Bioinformatics 2005;21:3448–9.
[5] Chapron C, Marcellin L, Borghese B, Santulli P. Rethinking mechanisms, diagnosis and management of endometriosis. Nat Rev Endocrinol 2019;15:666–82.
[6] Hsu AL, Khachikyan I, Stratton P. Invasive and noninvasive methods for the diagnosis of endometriosis. Clin Obstet Gynecol 2010;53:413–9.
[7] Czyzyk A, Podfigurna A, Szeliga A, Mezczalski B. Update on endometriosis pathogenesis. Minerva Ginecol 2017;69:447–61.
[8] Vercellini P, Viganò P, Somigliana E, Fedele L. Endometriosis: pathogenesis and treatment. Nat Rev Endocrinol 2014;10:261–75.
[9] Barr RE, Smith RA, Buck LG, et al. Mullerianiosis. Histol Histopathol 2007;22:161–6.
[10] Hufnagel D, Li F, Cosar E, Krikun G, Taylor HS. The role of stem cells in the etiology and pathophysiology of endometriosis. Semin Reprod Med 2015;33:333–40.
[11] Jerman LF, Hey-Cunningham AJ. The role of the lymphatic system in endometriosis: a comprehensive review of the literature. Biol Reprod 2015;92:624.
[12] Szubert M, Suzin J, Ducheler M, Szulawska A, Czyz M, Kowalczyk-Amico K. Evaluation of selected angiogenic and inflammatory markers in endometriosis before and after danazol treatment. Reprod Fertil Dev 2014;26:414–20.
[13] Page-McCaw A, Ewald AJ, Werb Z. Matrix metalloproteinases and the regulation of tissue remodelling. Nat Rev Mol Cell Biol 2007;8:221–33.
[14] Machado DE, Berardo PT, Palmero CY, Nasciuti LE. Higher expression of vascular endothelial growth factor (VEGF) and its receptor VEGFR-2 (Flk-1) and metalloproteinase-9 (MMP-9) in a rat model of peritoneal endometriosis is similar to cancer diseases. J Exp Clin Cancer Res 2010;29:4.
[15] Chen YJ, Wu HH, Liu WT, et al. A tumor necrosis factor-alpha inhibitor reduces the embryotoxic effects of endometriotic peritoneal fluid. Fertil Steril 2013;100:1476–85.
[16] Burns KA, Rodriguez KF, Hewitt SC, et al. Role of estrogen receptor signaling required for endometriosis-like lesion establishment in a mouse model. Endocrinology 2012;153:3960–71.
[17] Han SJ, Jung SY, Wu SP, et al. Estrogen receptor beta modulates apoptosis and the inflammasome to drive the pathogenesis of endometriosis. Cell 2015;163:960–74.
[18] Barisic A, Devic PS, Ostojic S, Perea N, Matrix metalloproteinase and tissue inhibitors of metalloproteinases gene polymorphisms in disorders that influence fertility and pregnancy complications: a systematic review and meta-analysis. Gene 2018;647:48–60.
[19] Liu H, Jiang Y, Jin X, et al. CAPN 7 promotes the migration and invasion of human endometrial stromal cell by regulating matrix metalloproteinase-2 activity. Reprod Biol Endocrinol 2013;11:64.
[20] Unoki M, Funabiki H, Velasco G, et al. CDC47 and HELLIS mutations undermine nonhomologous end joining in centromeric instability syndrome. J Clin Invest 2019;129:78–92.
[21] Eyster KM, Klinsky O, Kennedy V, Hansen KA. Whole genome deoxyribonucleic acid microarray analysis of gene expression in ectopic versus eutopic endometrium. Fertil Steril 2007;88:1505–33.
[22] Hever A, Roth RB, Hevezi P, et al. Human endometriosis is associated with plasma cells and overexpression of B lymphocyte stimulating. Proc Natl Acad Sci U S A 2007;104:12451–6.
[23] Huang DW, Sherman BT, Tan Q, et al. The DAVID Gene Functional Classification Tool: a novel biological module-centric algorithm to functionally analyze large gene lists. Genome Biol 2007;8:R183.
[24] Tilgner M. The KEGG database. Novartis Found Symp 2002;247:91– 101. 103–119, 244–252.
[25] Ashburner M, Ball CA, Blake JA, et al. Gene ontology: tool for the unification of biology. The gene ontology consortium. Nat Genet 2000;25:25–9.
[26] Franceschini A, Szklarczyk D, Frankild S, et al. STRING v9.1: protein–protein interaction networks, with increased coverage and integration. Nucleic Acids Res 2013;41:D808–15.
[27] Smoot ME, Ono K, Ruscheiski J, Wang PL, Ideker T. Cytoscape 2.8: new features for data integration and network visualization. Bioinformatics 2011;27:431–2.
Law CT, Wei L, Tsang FH, et al. HELLS regulates chromatin remodeling and epigenetic silencing of multiple tumor suppressor genes in human hepatocellular carcinoma. Hepatology 2019;69:2013–30.

Liu X, Hou X, Zhou Y, et al. Downregulation of the helicase lymphoid-specific (HELLS) gene impairs cell proliferation and induces cell cycle arrest in colorectal cancer cells. Onco Targets Ther 2019;12:10153–63.

Jenness C, Giunta S, Muller MM, Kimura H, Muir TW, Funabiki H. HELLS and CDCA7 comprise a bipartite nucleosome remodeling complex defective in ICF syndrome. Proc Natl Acad Sci USA 2018;115:E876–85.

Berthiaume AA, Grant RI, McDowell KP, et al. Dynamic remodeling of pericytes in vivo maintains capillary coverage in the adult mouse brain. Cell Rep 2018;22:8–16.

Corliss BA, Ray HC, Mathews C, et al. Myh11 lineage corneal endothelial cells and ASCs populate corneal endothelium. Invest Ophthalmo Vis Sci 2019;60:5095–103.

Kloth K, Renner S, Burmester G, et al. 16p13.11 microdeletion uncovers loss-of-function of a MYH11 missense variant in a patient with megacystis-microcolon-intestinal-hypoperistalsis syndrome. Clin Genet 2019;96:85–90.

Wang Q, Zhang J, Wang H, et al. Compound heterozygous variants in MYH11 underlie autosomal recessive megacystis-microcolon-intestinal hypoperistalsis syndrome in a Chinese family. J Hum Genet 2019;64:1067–73.

Sevier CS. Erv2 and quiescin sulfhydryl oxidases: Erv-domain enzymes associated with the secretory pathway. Antioxid Redox Signal 2012;16:800–8.

Zheng W, Zhang W, Hu W, Zhang C, Yang Y. Exploring the smallest active fragment of HsQSOX1b and finding a highly efficient oxidative engine. Plos One 2012;7:e40935.

Fiehler AL, Hanavan PD, Faigel DO, et al. Molecular inhibitor of QSOX1 suppresses tumor growth in vivo. Mol Cancer Ther 2020;19:112–22.

Yang ZX, Zhang B, Wei J, et al. MiR-539 inhibits proliferation and migration of triple-negative breast cancer cells by down-regulating LAMA4 expression. Cancer Cell Int 2018;18:16.

Li Y, Guan B, Liu J, et al. MicroRNA-200b is downregulated and suppresses metastasis by targeting LAMA4 in renal cell carcinoma. EBioMedicine 2019;44:439–51.

Peng L, Li Y, Wei S, et al. LAMA4 activated by Androgen receptor induces the cisplatin resistance in gastric cancer. Biomed Pharmacother 2020;124:109667.

Liu Y, Xu Y, Ding L, Yu L, Zhang B, Wei D. LncRNA MEG3 suppressed the progression of ovarian cancer via sponging miR-30e-3p and regulating LAMA4 expression. Cancer Cell Int 2020;20:181.

Hsiao KY, Wu MH, Tsai SJ. Epigenetic regulation of the pathological process in endometriosis. Reprod Med Biol 2017;16:314–9.

Ibrahim MG, Chiantera V, Frangini S, et al. Ultramicro-trauma in the endometrial-myometrial junctional zone and pale cell migration in adenomyosis. Fertil Steril 2013;104:1475–83.