Ferroptosis in female reproductive diseases: from potential pathogenesis to therapy

Jieru Zhang[1], Guanru Chen[2], Mengqing Zhang[1], Juan Wang[3], Qiong Xing[3], Wenxiang Zhang[3], Xiaojin He[4], Yuping Xu[3], Yunxia Cao[3], and Tianjuan Wang[3]

[1]Anhui Medical University
[2]Anhui Provincial Children's Hospital
[3]The First Affiliated Hospital of Anhui Medical University
[4]Affiliation not available

January 14, 2023

Abstract

Ferroptosis, a cohering network integrating iron, amino acids, lipids, and redox chemicals together,is a unique regulated cell death (RCD). Iron overload, excessive reactive oxygen species (ROS), and lipid peroxidation all contribute to the onset of ferroptosis. In recent years, a growing body of evidence suggests that ferroptosis is associated with some female reproductive diseases. The purpose of our review is to give a brief description of ferroptosis activation mechanism and relationship to female reproductive diseases including infertility, pregnancy associated diseases and ovarian cancer.

Ferroptosis in female reproductive diseases: from potential pathogenesis to therapy

Jie-Ru Zhang[a#], Guan-Ru Chen[b#], Meng-Qing Zhang[a], Juan Wang[c], Qiong Xing[c], Wen-Xiang Zhang[c], Xiao-Jin He[c], Yu-Ping Xu[c], Yun-Xia Cao[b, *], Tian-Juan Wang[b, *]

[a] Anhui Medical University, No 81 Meishan Road, Hefei 230022, Anhui, China;

[b] Department of Clinical Pharmacy, Anhui Provincial Children's Hospital, Hefei, China;

[c] Department of Obstetrics and Gynecology, the First Affiliated Hospital of Anhui Medical University, No 218 Jixi Road, Hefei 230022, Anhui, China.

[#] Jie-Ru Zhang and Guan-Ru Chen have contributed equally to this paper;

[*] Dr. Yun-Xia Cao and Dr. Tian-Juan Wang contributed equally to this paper.

Corresponding author:

Tian-Juan Wang, Department of Obstetrics and Gynecology, the First Affiliated Hospital of Anhui Medical University, No 218 Jixi Road, Hefei 230022, Anhui, China, Email:*wangtj6@126.com*;

Yun-Xia Cao, Department of Obstetrics and Gynecology, the First Affiliated Hospital of Anhui Medical University, No 218 Jixi Road, Hefei 230022, Anhui, China, Email:*caoyunxia6@126.com*.

Running head : Ferroptosis in female reproduction

Abstract:

Ferroptosis, a cohering network integrating iron, amino acids, lipids, and redox chemicals together,is a unique regulated cell death (RCD). Iron overload, excessive reactive oxygen species (ROS), and lipid peroxidation

all contribute to the onset of ferroptosis. In recent years, a growing body of evidence suggests that ferroptosis is associated with some female reproductive diseases. The purpose of our review is to give a brief description of ferroptosis activation mechanism and relationship to female reproductive diseases including infertility, pregnancy associated diseases and ovarian cancer.

Keywords: ferroptosis, regulated cell death, female reproductive diseases

Introduction

Recently, ferroptosis as one of regulated cell death (RCD) types was discovered and quickly emerged as a new area of focus for characterizing the pathophysiology and developing novel therapeutic and preventative strategies for associated illnesses.[1] It is iron-dependent and visibly distinct from apoptosis, necroptosis, and pyroptosis.[2-4] Numerous studies have demonstrated recently that ferroptosis has a role in the regulation of various disorders including tumorigenesis, ischemia–reperfusion injury, hematological diseases and reproductive diseases.[5, 6]In mice models of ischemia injury, ferrostatins and numerous other inhibitors of ferroptosis have been reported to perform a protective effect in the liver, kidney, brain, and heart.[7-9]

Reproductive diseases are common in female, and threaten large numbers of female health. Notably, mounting data suggests that ferroptosis is a key contributor to the ovarian and placental malfunction that underlies the majority of severe reproductive illnesses in recent years.[10, 11] In this review, we have provided a comprehensive knowledge of ferroptosis and the connections between it and diseases of female reproductive system, including infertility, pregnancy associated diseases and ovarian cancer, such as endometriosis (EMS), polycystic ovarian syndrome (PCOS), primary ovarian insufficiency (POI), preeclampsia (PE) and gestational diabetes mellitus (GDM).

Ferroptosis and its activation mechanism

In contrast to other RCD, ferroptosis is an iron-dependent modality that produces deadly levels of lipid peroxidation to induce oxidative damage to cell membranes. Cells experiencing ferroptosis exhibit the morphological characteristics of packed and tightly packed mitochondria lacking cristae and intact plasma membrane, normal nuclei without the chromatin aggregation and display severe iron-dependent lipid peroxidation biochemically.[2, 4, 12]

Ferroptosis has a complicated activation mechanism. It is a comprehensive network with amounts of elements that can be divided into three aspects: imbalance iron level, GSH homeostasis and redox regulation, and excessive lipid peroxidation (**Figure 1**). Iron is a vital trace for the maintenance of life. One of the important cues to initiate membrane oxidation is the accumulation of free iron. The Fenton reaction takes place as free iron levels rise inside of cells, causing considerable amounts of ROS and lipid peroxidation as well as the breakdown of cell membrane structure and ferroptosis.[2] Ferroptosis sensitive cells usually show higher transferrin receptor (TfR1) to increase iron intake while lower ferritin (FTHL1/FTL) to reduce iron storage, which eventually lead to iron overload and induce ferroptosis.[13]

GPX4, a powerful antioxidant, is required for the regulation of ferroptosis, which transforms hazardous lipid hydroperoxides to non-toxic ones.[14] For GPX4 to function, glutathione production (GSH) is an essential cofactor. A cystine/glutamate antiporter called System XC- trades extracellular cystine (Cys2) required for GSH for intracellular glutamate. Of note, it consists of two subunits, SLC7A11 and SLC3A2. SLC7A11 is in charge of exchanging glutamate for cysteine, and SLC3A2 acts as a chaperone.[15] Both the direct inhibition of GPX4 and the inhibition of GSH can lead to the accumulation of lipid peroxide in the cell membrane and the initiation of ferroptosis.

Additionally, polyunsaturated fatty acids (PUFAs) are a crucial component in the buildup of lipid peroxidation. Lysophosphatidylcholineacyl-transferase 3 (LPCAT3) and Acyl-CoA synthetase long-chain family member 4 (ACSL4) are essential mediators of ferroptosis that are involved in the manufacture and modification of PUFA-phosphatidyl-ethanolamine phospholipids in cell membranes.[16, 17] Large amounts of free lipid base are produced by PUFAs, which causes the cell membrane and plasma membrane to weaken, create protein holes, and lose their barrier function, disrupting intracellular homeostasis.[2, 3, 18]Furthermore, the

decomposition products of lipid peroxidation activate a more serious cascade reaction and eventually lead to ferroptosis.[4, 19]

Other regulated cell deaths

Apart from ferroptosis, RCD also includes apoptosis, necroptosis, and pyroptosis, each of them with their own characteristics and functions from morphologically and biochemically. Caspase activation is a key component of apoptosis, which exhibits distinctive morphological characteristics such as cell shrinkage, nuclear fragmentation, chromosomal DNA fragmentation, and plasma-membrane blebbing.[20-23] It is essential for preserving homeostasis and eliminating undesirable cells to prevent harming nearby cells throughout growth and aging.[24] Moreover, it plays essential role for the smooth working of the female reproductive tract. Apoptosis resistance is present in a few reproductive tract malignancies. What's more, it has been thought that apoptosis is a significant kind of cell death in oocyte loss.[25, 26]Necroptosis is activated by pro-inflammatory signal transduction as well as ischemic injury and viral infection displaying Plasma membrane rupture and cell enlargement. The activation and assembly of necrosome complexes containing receptor-interacting serine/threonine protein kinase 1 (RIPK1) and RIPK3 will lead to phosphorylation and oligomerization of MLKL, which will then lead to lipid peroxidation, cation influx, and ultimately cell death.[27] It has been identified is effective for suppressing ovarian cancer growth.[28, 29] Moreover, Induction of necroptosis in granulosa cell and oocyte may lead to follicular atresia.[30] It also has been identified contribute to placental pathophysiology that underlies serious pregnancy complications such as PE and fetal growth restriction (FGR).[31]Pyroptosis, in contrast to other types of cell death, is brought on by pathogen invasion and is reliant on caspase activation. It is regarded as an inflammatory type of cell death that is brought on by intracellular sensors like NLRP3 that identify DAMPs, PAMPs, membrane disruptions, etc. It displays distinctive physical and morphological traits, such as chromatin condensation, intact nuclei, cellular enlargement, and plasma membrane rupture.[32] It is a critical inflammatory pathway in the ovary and placenta linked with PE, PCOS, POI.[33-35](**Table 1**)

Ferroptosis and female reproductive diseases

Ferroptosis has different effects on different diseases. In OC, ferroptosis is imperative for the treatment, but for PCOS, pregnancy diseases, like PE and GDM, is responsible for the occurrence of them. Interestingly, in EMS, ferroptosis-resistance seems to be responsible for the form of early ectopic lesion, while, ferroptosis is also an important contributor in subsequent disease progression. It is meaningful for illuminating a variety of ferroptosis pathways in female reproductive diseases, such as infertility, pregnancy illnesses, and ovarian cancer (Figure 2). Ferroptosis could be a therapeutic target for these diseases (Table 2).

Ferroptosis and infertility

Infertility affects between 8% and 12% of reproductive-aged couples worldwide.[36] The pathology is not clear probably result from an immune system failure in the pelvic and uterine cavities, which affects ovulation, fallopian tube function, and endometrial receptivity, among other things. Notably, it is common to see that iron overload and oxidant stress microenvironment which have been found hinder preimplantation embryo development via ferroptosis in infertility patients. What's more, many infertility-related diseases such as EMs, POI, and PCOS are found linked with ferroptosis.

5.1 Endometriosis

Endometriosis (EMs) is, a chronic inflammatory disorder characterized mainly affecting pelvic tissues, including ovaries and fallopian tubes.[37, 38] It affects 11% of women worldwide who are of reproductive age, with up to 50% of them reporting pelvic pain or infertility.[37-41]

Based on previous research, the microenvironment in pelvic cavity of endometriosis patients contains excessive free iron and ROS, which is more conducive to the occurrence of ferroptosis [42]. Ferroptosis is an important contributor to the pathogenesis of endometriosis. First, endometrial cells can survive and develop endometriotic lesions during retrograde menstruation due to the resistance to ferroptosis, the pathophysiological alterations that follow in EMs are largely due to ferroptosis.

5.1.1 The two sides effects of ferroptosis in EMs

On one hand, it is believed that ferroptosis is the process by which endometrial cells dispersed during retrograde menstruation are expelled from the peritoneal cavity. Additionally, a prior study found that ectopic endometrial stromal cells (EESCs) were more susceptible to the effects of erastin therapy than normal endometrial stromal cells (NESCs), with the help of unique microenvironment and reduced ferroportin expression.[43] However, the endometrial cells enable to survive, and implant in vivo of EMs patients.[44] It seems that EESCs are resistant to ferroptosis displaying a general tendency to block the ferroptosis gene pathway, according to the most recent meta-analysis study.[45] Endometriotic tissues appear to be able to benefit from intracellular high iron loads for energy metabolism and cell proliferation by the resistance to ferroptosis while safeguarding themselves from the ferroptosis.[46-48]

Several ferroptosis regulators are found to alter for getting rid of ferroptosis (Figure 3). Ferritin can be transported to autophagosomes by nuclear receptor coactivator 4 (NCOA4). and then fuse with lysosomes to degrade ferritin into active iron.[49] In endometrium, it was found to be diminished. Additionally, it was discovered that the ectopic endometrium had increased expression of FTL, a crucial component of the iron ion storage protein.[50]These modifications enhance the insensitivity to ferroptosis and lower the intracellular amounts of free iron ions.[45]

Interestingly, GSH is inhibited in ectopic endometrium.[51] The expression of the lncRNA ADAMTS9-AS, on the other hand, was improved in the ectopic endometrium. By sponging miR-6516-5p to increase the expression of GPX4, this promoted ESCs to grow and migrate and prevented ectopic endometrium from ferroptosis.[52] In addition, repressing lipid oxidation plays important role in ferroptosis-resistance in EMs. Through lower expression of reticulocyte-type 15-lipooxygenase-1 (ALOX15) and spermine N1-acetyltransferase 1(SAT1), coding for a crucial enzyme in polyamine metabolism and also promotes p53-dependent ferroptosis,[53] lipid oxidation is hindered in eutopic endometrial tissue of individuals with endometriosis. Of note, this inhibition increased in ectopic endometrium, due to ACSL5 and LPCAT3 downregulation.[45]

Moreover, Fibulin-1 (FBLN1), FBLN1 expression was enhanced in eutopic and ectopic endometrial tissues with EMS, increasing ESC viability and migration while decreasing ESC ferroptosis via enhancing the stability of EFEMP1 protein.[54] VDAC2 serving as the main pathway for metabolite diffusion through the outer mitochondrial membrane and crucial for intracellular redox response expression was significantly downregulated in eutopic endometrium.[55] Overall, ferroptosis-resistance is a key contribute to the establishment and maintain of EMs with complex regulatory pathways, and ferroptosis-inducers may be a therapeutic option for it.

On the other hand, ferroptosis contributes to the subsequent process of EMs, including infertility. Study showed that some EECS that are undergoing ferroptosis have paracrine effects that promote the development of new vascular systems in the surrounding tissues, which may promote the growth of benign cells and hasten the progression of this illness.[56]Furthermore, endometriosis, which causes uterine dysfunction and interferes with embryo implantation, is closely associated with infertility. Moreover, almost half of the infertility patients are accompanied with EMs.[57] Ferroptosis may contribute to the infertility brought on by EMs.[56] Follicle dysplasia and decreased oocyte quality are the primary reasons of infertility associated with EMs. It has been established that iron overload is a common feature of the peritoneal fluid (PF) and follicular fluid (FF) in EMs patients, causing ferroptosis, damaging oocytes and embryos, and eventually leading to infertility.[50, 58, 59]

Oocyte maturation requires granulosa cells (GCs), the biggest cell group and the primary functional cells in follicles. The normal development of oocyte is reliant on the paracrine and nutritional activities of the surrounding GCs.[60] However, high levels of iron in FF of EMs patients cause GCs to ferroptosis and produce exosomes after ferroptosis, which further impair oocyte maturation.[61]Exosomes are extracellular vesicles that carry proteins, mRNAs, and miRNAs to target cells.[62] In a setting of iron overload, granulosa cells' exosomal miRNAs can control the expression of several signal pathways, which ultimately reduces ovarian

reserve.[61] Furthermore, ferroptosis is a reaction to higher iron damage at the blastocyst stage, may cause to developmental stoppage. Additionally, endometriosis PF's iron excess prevents blastocyst development and damages developing embryos by causing ferroptosis and mitochondrial malfunction.[59, 63] The consequences of iron excess on embryo development have been the subject of numerous studies, although the precise connection between ferroptosis and embryotoxicity has not yet been established. According to a study, HMOX1 is elevated during embryonic ferroptosis and inhibits it by maintaining mitochondrial activity and protect them from oxidative stress.[63] When considered collectively, we cannot ignore the impact of ferroptosis for the subsequent pathologic processes of EMs.

In conclusion, endometriosis exists ferroptosis and ferroptosis-resistance simultaneously. Ferroptosis-resistance is essential for the form of endometriotic lesions, while ferroptosis plays important role in the subsequent lesions. Recent studies have proposed to promote ferroptosis in endometriotic lesions in order to treat EMs, but iron overload microenvironment and ferroptosis impairing oocyte function should be considered.[64] Vitamin E and iron chelators can significantly alleviate the symptoms of EMs-related fertility, by increasing GPX4 and reducing iron overload.[61] Therefore, treatment methods related to ferroptosis need to be considered comprehensively.

5.2 PCOS

About 15%–21% of women of reproductive age have PCOS, which is the most frequent cause of anovulatory infertility and is characterized by hyperandrogenism, insulin resistance, and irregular ovulation or anovulation.[65] Previous research discovered that the primary factor behind the onset and progression of PCOS is aberrant ovarian folliculogenesis.[66] There are growing evidence showing that ferroptosis may be associated with it. Therefore, it is meaningful for us to understanding the mechanism of ferroptosis in PCOS and discovery novel treatment for POCS-related infertility.

Several studies have identified that ferroptosis may have a key role in the pathogenesis of PCOS. MiRNA and circ RNA can regulate the progress of PCOS via ferroptosis. Circ RHBG has been shown to compete with SLC7A11 for the ability to bind to miR-515-5p, upregulate the expression of SLC7A11, and ultimately prevent GC ferroptosis.[67]However, Silencing MiR-93-5p guards against GCs dysfunction through controlling the NF-B signaling pathway and encouraging GCs apoptosis and ferroptosis.[68]

Moreover, the unique microenvironment in PCOS patients vivo causes ferroptosis in gravid uterine and the placenta. The results of one study showed that co-exposure to 5 dihydrotestosterone and insulin resulted in decreased levels of glutathione and GPX4, altered expression of genes related to ferroptosis such as ACSL4, SLC7A11, increased levels of the oxidative stress marker malondialdehyde (MDA) and iron deposition, increased levels of the ERK/p38/JNK pathway and mitochondrial morphology.[69] But the specific relationship between ferroptosis and PCOS has not been illustrated. A previous study showed that gravid uterine and placental ferroptosis is modulated by hyperandrogenism and insulin resistance in PCOS.[69]Proteomic analysis of CD4+ T cells in infertile patients with PCOS showed that three key proteins, which namely phosphatidylethanolaminebinding protein 1, proteasome activator complex subunit 1 and triosephosphate isomerase 1 are overexpressed are involved in the ferroptosis pathway.[70] We hypothesize that ferroptosis may have an impact on PCOS-related infertility. Of note, it was revealed by differential gene expression analysis that the interaction of autophagy, apoptosis, and ferroptosis contributed to the development of porcine ovarian atresia.[10]

In addition, these ferroptosis inhibitors, Ferrostatin-1,[71] N-acetylcysteine,[72]Cryptotanshinone,[73] have shown that they can relieve symptoms in PCOS patients. Ferroptosis is generally considered to be intimately associated with the pathogenic alterations of PCOS, while the precise mechanism is still currently unclear. Overall, the present studies have explored the role of ferroptosis in the pathological process of PCOS, targeting the inhibition of ferroptosis may be an effective treatment for PCOS.

5.3 POI

POI, characterized as early exhaustion of primordial follicles that may cause low fertility, affects about 1%

of women under the age of 40.[74] This condition typically results from flaws in the development of follicular atresia. Of note, previous researches have confirmed the strong connection between ferroptosis and follicular atresia and oocyte loss.[10, 75] A latest study found that BNC1 loss causes excessive follicular atresia and early follicular activation by inducing oocyte ferroptosis via the NF2-YAP pathway, in accordance with earlier findings that BNC1 mutation is related to ovarian dysfunction through genome whole exon sequencing and transgenic mouse animal models.[76, 77] The levels of ferroptosis-associated markers were aberrant in Bnc1 truncated mutant mouse oocytes, and the corresponding phenotype was aggravated by the ferroptosis agonist RLS3 and reversed by fer-1. This study also discovered the downstream target gene Nf2 of BNC1 and the mechanism governing the HiPO-YEP-TFRC/ACSL4 pathway, which controls oocyte ferroptosis and is mediated by Nf2.

Ferroptosis and pregnancy diseases

Normally, during pregnancy, iron levels in pregnant women will increase to meet the requirements of maternal and infant hematopoiesis. In addition, hepcidin (hepc), an important regulator of iron homeostasis, has been identified that the expression level decreased, and thus both dietary iron absorption and stored iron release rise during normal pregnancy. These findings imply the capacity to react to ambient iron exposure may be diminished in healthy pregnant women, which may easily lead to clinical diseases due to excessive iron.[78]Recent years, several pregnancy diseases have been uncovered their pathogenesis is linked with ferroptosis.[79, 80]

6.1 PE

PE is a common disease of pregnancy, about 3–10% of all pregnant women suffer from preeclampsia worldwide, and together with eclampsia,[81] It is a significant contributor to poor pregnancy outcomes and maternal mortality; over 50,000 mothers die from them each year across the globe.[82] The exact reason for preeclampsia is unclear, a key pathophysiologic feature though is the development of an abnormal placenta which may be linked with ferroptosis.

6.1.1 Abnormal ferroptosis to Hypoxia/Reperfusion during pregnancy

Before 10 weeks into the pregnancy, blood clots and congealed endothelial cells fully block the maternal spiral arteries. The embryo's environment is hypoxic and hypoglycemia during 8–10 weeks. The spiral arteries don't fully open until 10 to 12 weeks of pregnancy, when maternal blood can enter the placental gap and initially expose the fetal villi to an environment rich in glucose, oxygen, and iron. Similar to the hypoxia/reperfusion event, which causes excessive cell membrane lipid peroxidation and ferroptosis at the maternal-fetal interface, especially in trophoblast cells, this mechanism causes excessive cell membrane lipid peroxidation. The pathologic characteristics of PE are caused by superficial endovascular invasion of extravillous cytotrophoblast cells and inadequate remodeling of the maternal spiral arteries. Placental ischemia and oxidative stress are caused by inadequately modified spiral arteries, which also cause insufficient placental perfusion, high-speed blood flow, and turbulence,[83] thus damaging placental villi and causing abnormal levels of angiogenic proteins in maternal blood.[84]

6.1.2 The mechanism of ferroptosis in PE

Hepc is an iron metabolism regulator. Hepc inhibits iron ion transport by binding to the membrane iron transporter (FPN) on the basolateral surface of intestinal epithelial cells and the plasma membrane of reticuloendothelial cells (macrophages). Finally, Hepc degrades the transporters in lysosomes, inhibits FPN, and prevents the output of iron ions from cells. Its levels have been observed to decrease in PE patients in some research [85], while being raised during pregnancy in other studies.[86] According to reports, the occurrence of PE and early pregnancy's high serum hepc level.[86] The study on the change in hepc level in PE patients is still debatable at this time, and more research is required to understand its mechanism. In PE, free iron, ferritin, and transferrin saturation levels rise in contrast to normal pregnancy, but transferrin receptor levels fall.[87-89] Additionally, the mother's plasma blood volume increases during pregnancy, this has little impact on pregnancy-related issues. However, the increased plasma volume has the ability to change other

parameters, such as the iron concentration, which have differing impacts on pregnancies with normal and PE.[90]

In addition, in the PE model, mir-30b-5p can inhibit the cys2 / glutamate reverse transporter and Pax3, which decreases the expression of transferrin 1, increasing the amount of unstable Fe^{2+} and encouraging ferroptosis in PE patients.[79] In addition, the expression of mir-210 in placenta has been found increased, leading to iron accumulation and autophagosome formation in trophoblasts, as well as hemosiderin deposition in placental stromal trophoblasts.[91]Therefore, mounting evidence demonstrated iron overload in PE can lead to ferroptosis, especially in trophoblasts which are sensitive to ferroptosis due to the high expression of two ferroptosis- inducer genes Sat1, Lpcat3,[53] and further lead to placental dysfunction and trophoblast damage.

Studies have revealed that patients with PE and eclampsia had considerably higher serum levels of MDA, an oxidation byproduct that probably leads to ferroptosis.[92-94] Moreover, it has been discovered that PE's hypoxia activates the nuclear factor erythroid-2-related factor 2 (Nrf2), a key antioxidant regulator. It can relieve the symptoms of PE by the activation of the Nrf2/HO-1 signaling pathway and the expression of SLC7A11, GPX4, and FPN1 to inhibit oxidation stress and ferroptosis.[95] In addition, DJ-1, essential for activating and stabilizing Nrf2 to carry out further function, has also been found elevated to play a protective role in PE.[96] Currently, the primary focus of PE treatment is a placental hypoxia cure. Although various PE-related parameters have been linked to placental oxidative stress, it is unclear how these factors specifically affect ferroptosis regulation.

6.2 GDM

GDM is one of the most prevalent metabolic problems of pregnancy, associated with abnormal placental functioning.[97, 98]A recent study found that greater blood ferritin levels, which in turn enhance the oxidative stress, can be caused by higher pre-pregnancy BMI. In GDM patients, ferritin and oxidative stress can raise blood sugar levels, possibly via causing pancreatic beta-cell ferroptosis in vivo.[99]

Additionally, the diabetes placenta typically exhibits abnormal trophoblast proliferation, autophagy, and cell cycle regulation, which promotes the progression of GDM.[100-102] High glucose concentration induces trophoblasts to produce an excessive amount of ROS, which may starts the pathogenesis of GDM.[103, 104]Moreover, in trophoblastic cells of GMD patients, high glucose exposure causes an increase in SIRT3 protein level, iron buildup, and lipid peroxidation. Eventually, this causes ferroptosis.[105]It is worth noting that in GDM, abnormalities in lipid metabolism, which is a key factor in ferroptosis, occur in addition to glucose metabolism. Adiponectin, a lipid metabolism regulator, can reduce placental injury in GDM by restoring CPT-1 activity to inhibit ferroptosis.[80]

Ferroptosis and OC

OC is one of the most fatal malignant tumors with a five-year relative survival below 50%.[106, 107] Mounting evidence have been revealed that ferroptosis has a closely relationship with OC, although the potent therapeutic strategies and the pathogenesis of OC have yet to been fully uncovered. Accumulation of iron promotes the development of OC. Iron excess intracellular iron and an increased reliance on iron for proliferation are typical features of high-grade ovarian cancer. A forced reduction in intracellular iron inhibits tumor growth as well as tumor cell intraperitoneal dissemination.[108] Therefore, the drugs which can reduce intracellular iron, such as binding iron chelators and cytotoxic drugs to TfR1 are effective on the treatment of OC.[108]

Moreover, ferroptosis provides a new solution for the problems of drugs resistance in OC. For example, erastin, a ferroptosis reducer, was proved to inhibit System Xc- and result in depletion of GSH.[109] According to a study, erastin and cisplatin jointly limit the proliferation of OC cells, which may be affected by a ROS-mediated mechanism that improves cisplatin therapy. This suggests a unique method for overcoming cisplatin therapy resistance.[110] Since the role of ferroptosis in ovarian cancer has been reviewed in previous reviews, this article will not repeat it again. However, the ability of ferroptosis inducers to kill tumor cells in vitro has been demonstrated, but in experimental animal models using immunocompromised mice, they have

not been particularly successful. Fortunately, a recent study showed that ferroptosis promotes iron death in PMN MDSCs in the tumor microenvironment as a distinct and specific immunosuppressive mechanism.[111] Overall, ferroptosis is an effective target for tumor treatment according to the current researches.

C onclusion and perspective

An aberrant humoral microenvironment, iron overload, excessive ROS, causes normal female reproductive cells such as GCs and trophoblasts, to ferroptosis. This causes the ovary, placenta, and embryo to become dysfunctional, which ultimately results in a number of illnesses associated to reproduction. Ferroptosis connects iron, amino acids, lipids, and redox chemistry into a single, coherent network. Today, it is understood that this network plays a role in a wide range of biological activities in the female reproductive system, including both normal physiology and various diseases. As more information is revealed about the intricate regulatory mechanism and consequences of ferroptosis that controls various diseases, the field will undoubtedly become more and more exciting.

We realize that there are still some challenges in the research of ferroptosis in female reproductive system related diseases. Firstly, it is worth to note that several studies have simply demonstrated the coexistence of ferroptosis and a pathological state of sickness or the special vivo microenvironment of patients which tends to promote ferroptosis in reproductive function related cells, such as GCs, Trophoblasts. We yet not to know the direct molecular events are responsible for the eventual pathological changes of these diseases and this will be a hotspot in future research.

Furthermore, ferroptosis plays a crucial role in the treatment of illnesses of the female reproductive system. However, more attention should be attached on that ferroptosis may play a dual role in some disease such as EMs. Furthermore, we cannot ignore that ferroptosis occurs throughout normal physiological processes and is crucial for the normal development of tissues and organs. Thus, one area of future research will focus on tailored ferroptosis therapy for various tissues. Exosomes have gained increasing notoriety recently as highly effective targeted drug delivery systems in tumor therapy. Targeting and biocompatibility of preparations based on ferroptosis-related exosomes may offer a cutting-edge and potent delivery platform for the treatment of anti-reproductive disorders. In addition, there are numerous factors that have been found to cause and prevent ferroptosis, but it is unknown whether one can be the optimal therapeutic index, be employed in both patients and animal models, and be chemically managed.

Of note, A new type of death known as "Cuproptosis" is induced by an overabundance of copper. Interestingly, ferroptosis and cuproptsis, these two distinct types of cell death, both closely associated with mitochondria. It must be exciting to find out whether there is any connection between these two pathways that might be significant and lead to further treatment possibilities for female reproductive diseases.

In summary, there is a wealth of foreseeable opportunities to elucidate both the and the effects and execution mechanisms of ferroptosis in female reproduction. Such studies will not only provide new targets for the therapy for diseases but also provide new ideas for illuminating the breadth of physiological and pathological roles of ferroptosis in other disease linked with iron overload and oxidant stress.

Disclosures of interests

There are no conflicts of interest reported by the authors.

Author contributions

ZJ and CG researched the data and drafted the manuscript. ZM helped collect the data and edited the manuscript. WJ, XQ, ZW, HX and XY participated in the conception and design of the study and interpretation of the data. CY and WT reviewed the data, made substantial contributions to the discussion of the content, and edited the article before submission. All authors read and approved the final manuscript.

Details of ethics approval

Due to the nature of this review ethical approvals were not required.

Funding

This study was supported by Clinical Cooperative Research and Promotion Program of Anhui Medical University (2021xkjT031)

Acknowledgement s

Not applicable

References

1. Li J, Cao F, Yin HL, Huang ZJ, Lin ZT, Mao N, et al. Ferroptosis: past, present and future. Cell Death Dis. 2020 Feb 3;11(2):88.

2. Xie Y, Hou W, Song X, Yu Y, Huang J, Sun X, et al. Ferroptosis: process and function. Cell Death Differ. 2016 Mar;23(3):369-79.

3. Stockwell BR, Friedmann Angeli JP, Bayir H, Bush AI, Conrad M, Dixon SJ, et al. Ferroptosis: A Regulated Cell Death Nexus Linking Metabolism, Redox Biology, and Disease. Cell. 2017 Oct 5;171(2):273-85.

4. Chen X, Comish PB, Tang D, Kang R. Characteristics and Biomarkers of Ferroptosis. Front Cell Dev Biol. 2021;9:637162.

5. Gao M, Deng J, Liu F, Fan A, Wang Y, Wu H, et al. Triggered ferroptotic polymer micelles for reversing multidrug resistance to chemotherapy. Biomaterials. 2019 Dec;223:119486.

6. Ma LL, Liang L, Zhou D, Wang SW. Tumor suppressor miR-424-5p abrogates ferroptosis in ovarian cancer through targeting ACSL4. Neoplasma. 2020 Oct 7.

7. Linkermann A, Skouta R, Himmerkus N, Mulay SR, Dewitz C, De Zen F, et al. Synchronized renal tubular cell death involves ferroptosis. Proc Natl Acad Sci U S A. 2014 Nov 25;111(47):16836-41.

8. Tuo QZ, Lei P, Jackman KA, Li XL, Xiong H, Li XL, et al. Tau-mediated iron export prevents ferroptotic damage after ischemic stroke. Mol Psychiatry. 2017 Nov;22(11):1520-30.

9. Gao M, Monian P, Quadri N, Ramasamy R, Jiang X. Glutaminolysis and Transferrin Regulate Ferroptosis. Mol Cell. 2015 Jul 16;59(2):298-308.

10. Zhang J, Liu Y, Yao W, Li Q, Liu H, Pan Z. Initiation of follicular atresia: gene networks during early atresia in pig ovaries. Reproduction. 2018 Jul;156(1):23-33.

11. Ng SW, Norwitz SG, Norwitz ER. The Impact of Iron Overload and Ferroptosis on Reproductive Disorders in Humans: Implications for Preeclampsia. Int J Mol Sci. 2019 Jul 4;20(13).

12. Dolma S, Lessnick SL, Hahn WC, Stockwell BR. Identification of genotype-selective antitumor agents using synthetic lethal chemical screening in engineered human tumor cells. Cancer Cell. 2003 Mar;3(3):285-96.

13. McCullough K, Bolisetty S. Ferritins in Kidney Disease. Semin Nephrol. 2020 Mar;40(2):160-72.

14. Ursini F, Maiorino M, Valente M, Ferri L, Gregolin C. Purification from pig liver of a protein which protects liposomes and biomembranes from peroxidative degradation and exhibits glutathione peroxidase activity on phosphatidylcholine hydroperoxides. Biochim Biophys Acta. 1982 Feb 15;710(2):197-211.

15. Lewerenz J, Hewett SJ, Huang Y, Lambros M, Gout PW, Kalivas PW, et al. The cystine/glutamate antiporter system x(c)(-) in health and disease: from molecular mechanisms to novel therapeutic opportunities. Antioxid Redox Signal. 2013 Feb 10;18(5):522-55.

16. Kagan VE, Mao G, Qu F, Angeli JP, Doll S, Croix CS, et al. Oxidized arachidonic and adrenic PEs navigate cells to ferroptosis. Nat Chem Biol. 2017 Jan;13(1):81-90.

17. Dixon SJ, Winter GE, Musavi LS, Lee ED, Snijder B, Rebsamen M, et al. Human Haploid Cell Genetics Reveals Roles for Lipid Metabolism Genes in Nonapoptotic Cell Death. ACS Chem Biol. 2015 Jul 17;10(7):1604-9.

18. Feng H, Stockwell BR. Unsolved mysteries: How does lipid peroxidation cause ferroptosis? PLoS Biol. 2018 May;16(5):e2006203.

19. Dixon SJ, Lemberg KM, Lamprecht MR, Skouta R, Zaitsev EM, Gleason CE, et al. Ferroptosis: an iron-dependent form of nonapoptotic cell death. Cell. 2012 May 25;149(5):1060-72.

20. Slee EA, Adrain C, Martin SJ. Executioner caspase-3, -6, and -7 perform distinct, non-redundant roles during the demolition phase of apoptosis. J Biol Chem. 2001 Mar 9;276(10):7320-6.

21. Susin SA, Lorenzo HK, Zamzami N, Marzo I, Snow BE, Brothers GM, et al. Molecular characterization of mitochondrial apoptosis-inducing factor. Nature. 1999 Feb 4;397(6718):441-6.

22. Li P, Nijhawan D, Budihardjo I, Srinivasula SM, Ahmad M, Alnemri ES, et al. Cytochrome c and dATP-dependent formation of Apaf-1/caspase-9 complex initiates an apoptotic protease cascade. Cell. 1997 Nov 14;91(4):479-89.

23. Joza N, Susin SA, Daugas E, Stanford WL, Cho SK, Li CY, et al. Essential role of the mitochondrial apoptosis-inducing factor in programmed cell death. Nature. 2001 Mar 29;410(6828):549-54.

24. Chinnaiyan AM. The apoptosome: heart and soul of the cell death machine. Neoplasia. 1999 Apr;1(1):5-15.

25. Kong QQ, Wang GL, An JS, Wang J, Cheng H, Liu T, et al. Effects of postovulatory oviduct changes and female restraint stress on aging of mouse oocytes. Reproduction. 2021 Jun 16;162(1):95-105.

26. Singla S, Iwamoto-Stohl LK, Zhu M, Zernicka-Goetz M. Autophagy-mediated apoptosis eliminates aneuploid cells in a mouse model of chromosome mosaicism. Nat Commun. 2020 Jun 11;11(1):2958.

27. Sun L, Wang H, Wang Z, He S, Chen S, Liao D, et al. Mixed lineage kinase domain-like protein mediates necrosis signaling downstream of RIP3 kinase. Cell. 2012 Jan 20;148(1-2):213-27.

28. Li S, Li H, Zhang YL, Xin QL, Guan ZQ, Chen X, et al. SFTSV Infection Induces BAK/BAX-Dependent Mitochondrial DNA Release to Trigger NLRP3 Inflammasome Activation. Cell Rep. 2020 Mar 31;30(13):4370-85.e7.

29. McCabe KE, Bacos K, Lu D, Delaney JR, Axelrod J, Potter MD, et al. Triggering necroptosis in cisplatin and IAP antagonist-resistant ovarian carcinoma. Cell Death Dis. 2014 Oct 30;5(10):e1496.

30. Chaudhary GR, Yadav PK, Yadav AK, Tiwari M, Gupta A, Sharma A, et al. Necroptosis in stressed ovary. J Biomed Sci. 2019 Jan 21;26(1):11.

31. Hannan NJ, Beard S, Binder NK, Onda K, Kaitu'u-Lino TJ, Chen Q, et al. Key players of the necroptosis pathway RIPK1 and SIRT2 are altered in placenta from preeclampsia and fetal growth restriction. Placenta. 2017 Mar;51:1-9.

32. Bergsbaken T, Fink SL, Cookson BT. Pyroptosis: host cell death and inflammation. Nat Rev Microbiol. 2009 Feb;7(2):99-109.

33. Cheng SB, Nakashima A, Huber WJ, Davis S, Banerjee S, Huang Z, et al. Pyroptosis is a critical inflammatory pathway in the placenta from early onset preeclampsia and in human trophoblasts exposed to hypoxia and endoplasmic reticulum stressors. Cell Death Dis. 2019 Dec 5;10(12):927.

34. Ala M, Ala M. Metformin for Cardiovascular Protection, Inflammatory Bowel Disease, Osteoporosis, Periodontitis, Polycystic Ovarian Syndrome, Neurodegeneration, Cancer, Inflammation and Senescence: What Is Next? ACS Pharmacol Transl Sci. 2021 Dec 10;4(6):1747-70.

35. Zhang CR, Zhu WN, Tao W, Lin WQ, Cheng CC, Deng H, et al. Moxibustion against Cyclophosphamide-Induced Premature Ovarian Failure in Rats through Inhibiting NLRP3-/Caspase-1-/GSDMD-Dependent Pyroptosis. Evid Based Complement Alternat Med. 2021;2021:8874757.

36. Vander Borght M, Wyns C. Fertility and infertility: Definition and epidemiology. Clin Biochem. 2018 Dec;62:2-10.

37. Laganà AS, Vitale SG, Salmeri FM, Triolo O, Ban Frangež H, Vrtačnik-Bokal E, et al. Unus pro omnibus, omnes pro uno: A novel, evidence-based, unifying theory for the pathogenesis of endometriosis. Med Hypotheses. 2017 Jun;103:10-20.

38. Bulun SE, Yilmaz BD, Sison C, Miyazaki K, Bernardi L, Liu S, et al. Endometriosis. Endocr Rev. 2019 Aug 1;40(4):1048-79.

39. Bulun SE. Endometriosis. N Engl J Med. 2009 Jan 15;360(3):268-79.

40. Nnoaham KE, Hummelshoj L, Webster P, d'Hooghe T, de Cicco Nardone F, de Cicco Nardone C, et al. Impact of endometriosis on quality of life and work productivity: a multicenter study across ten countries. Fertil Steril. 2011 Aug;96(2):366-73.e8.

41. Simoens S, Dunselman G, Dirksen C, Hummelshoj L, Bokor A, Brandes I, et al. The burden of endometriosis: costs and quality of life of women with endometriosis and treated in referral centres. Hum Reprod. 2012 May;27(5):1292-9.

42. Lousse JC, Defrère S, Van Langendonckt A, Gras J, González-Ramos R, Colette S, et al. Iron storage is significantly increased in peritoneal macrophages of endometriosis patients and correlates with iron overload in peritoneal fluid. Fertil Steril. 2009 May;91(5):1668-75.

43. Li Y, Zeng X, Lu D, Yin M, Shan M, Gao Y. Erastin induces ferroptosis via ferroportin-mediated iron accumulation in endometriosis. Hum Reprod. 2021 Mar 18;36(4):951-64.

44. Ng SW, Norwitz SG, Taylor HS, Norwitz ER. Endometriosis: The Role of Iron Overload and Ferroptosis. Reprod Sci 3060. 2020 Jul;27(7):1383-90.

45. Li B, Duan H, Wang S, Li Y. Ferroptosis resistance mechanisms in endometriosis for diagnostic model establishment. Reprod Biomed Online. 2021 Jul;43(1):127-38.

46. Murphy AA, Santanam N, Morales AJ, Parthasarathy S. Lysophosphatidyl choline, a chemotactic factor for monocytes/T-lymphocytes is elevated in endometriosis. J Clin Endocrinol Metab. 1998 Jun;83(6):2110-3.

47. Defrère S, Lousse JC, González-Ramos R, Colette S, Donnez J, Van Langendonckt A. Potential involvement of iron in the pathogenesis of peritoneal endometriosis. Mol Hum Reprod. 2008 Jul;14(7):377-85.

48. Murphy AA, Santanam N, Parthasarathy S. Endometriosis: a disease of oxidative stress? Semin Reprod Endocrinol. 1998;16(4):263-73.

49. Goodall M, Thorburn A. Identifying specific receptors for cargo-mediated autophagy. Cell Research. 2014 2014/07/01;24(7):783-4.

50. Woo JH, Choi YS, Choi JH. Iron-Storage Protein Ferritin Is Upregulated in Endometriosis and Iron Overload Contributes to a Migratory Phenotype. Biomedicines. 2020 Oct 27;8(11).

51. Andrisani A, Donà G, Brunati AM, Clari G, Armanini D, Ragazzi E, et al. Increased oxidation-related glutathionylation and carbonic anhydrase activity in endometriosis. Reprod Biomed Online. 2014 Jun;28(6):773-9.

52. Wan Y, Gu C, Kong J, Sui J, Zuo L, Song Y, et al. Long noncoding RNA ADAMTS9-AS1 represses ferroptosis of endometrial stromal cells by regulating the miR-6516-5p/GPX4 axis in endometriosis. Sci Rep. 2022 Feb 16;12(1):2618.

53. Ou Y, Wang SJ, Li D, Chu B, Gu W. Activation of SAT1 engages polyamine metabolism with p53-mediated ferroptotic responses. Proc Natl Acad Sci U S A. 2016 Nov 1;113(44):E6806-e12.

54. Wan Y, Song Y, Chen J, Kong J, Gu C, Huang J, et al. Upregulated Fibulin-1 Increased Endometrial Stromal Cell Viability and Migration by Repressing EFEMP1-Dependent Ferroptosis in Endometriosis. Biomed Res Int. 2022;2022:4809415.

55. Srivastava SR, Zadafiya P, Mahalakshmi R. Hydrophobic Mismatch Modulates Stability and Plasticity of Human Mitochondrial VDAC2. Biophys J. 2018 Dec 18;115(12):2386-94.

56. Li G, Lin Y, Zhang Y, Gu N, Yang B, Shan S, et al. Endometrial stromal cell ferroptosis promotes angiogenesis in endometriosis. Cell Death Discov. 2022 Jan 17;8(1):29.

57. Meuleman C, Vandenabeele B, Fieuws S, Spiessens C, Timmerman D, D'Hooghe T. High prevalence of endometriosis in infertile women with normal ovulation and normospermic partners. Fertil Steril. 2009 Jul;92(1):68-74.

58. Li A, Ni Z, Zhang J, Cai Z, Kuang Y, Yu C. Transferrin Insufficiency and Iron Overload in Follicular Fluid Contribute to Oocyte Dysmaturity in Infertile Women With Advanced Endometriosis. Front Endocrinol (Lausanne). 2020;11:391.

59. Chen X, Zhou Y, Wu D, Shu C, Wu R, Li S, et al. Iron overload compromises preimplantation mouse embryo development. Reprod Toxicol. 2021 Oct;105:156-65.

60. Almeida CP, Ferreira MCF, Silveira CO, Campos JR, Borges IT, Baeta PG, et al. Clinical correlation of apoptosis in human granulosa cells-A review. Cell Biol Int. 2018 Sep;42(10):1276-81.

61. Ni Z, Li Y, Song D, Ding J, Mei S, Sun S, et al. Iron-overloaded follicular fluid increases the risk of endometriosis-related infertility by triggering granulosa cell ferroptosis and oocyte dysmaturity. Cell Death Dis. 2022 Jul 4;13(7):579.

62. Tkach M, Théry C. Communication by Extracellular Vesicles: Where We Are and Where We Need to Go. Cell. 2016 Mar 10;164(6):1226-32.

63. Li S, Zhou Y, Huang Q, Fu X, Zhang L, Gao F, et al. Iron overload in endometriosis peritoneal fluid induces early embryo ferroptosis mediated by HMOX1. Cell Death Discov. 2021 Nov 15;7(1):355.

64. Hu W, Zhang Y, Wang D, Yang T, Qi J, Zhang Y, et al. Iron Overload-Induced Ferroptosis Impairs Porcine Oocyte Maturation and Subsequent Embryonic Developmental Competence in vitro. Front Cell Dev Biol. 2021;9:673291.

65. Sirmans SM, Pate KA. Epidemiology, diagnosis, and management of polycystic ovary syndrome. Clin Epidemiol. 2013 Dec 18;6:1-13.

66. Dewailly D, Robin G, Peigne M, Decanter C, Pigny P, Catteau-Jonard S. Interactions between androgens, FSH, anti-Müllerian hormone and estradiol during folliculogenesis in the human normal and polycystic ovary. Hum Reprod Update. 2016 Nov;22(6):709-24.

67. Zhang D, Yi S, Cai B, Wang Z, Chen M, Zheng Z, et al. Involvement of ferroptosis in the granulosa cells proliferation of PCOS through the circRHBG/miR-515/SLC7A11 axis. Ann Transl Med. 2021 Aug;9(16):1348.

68. Tan W, Dai F, Yang D, Deng Z, Gu R, Zhao X, et al. MiR-93-5p promotes granulosa cell apoptosis and ferroptosis by the NF-kB signaling pathway in polycystic ovary syndrome. Front Immunol. 2022;13:967151.

69. Zhang Y, Hu M, Jia W, Liu G, Zhang J, Wang B, et al. Hyperandrogenism and insulin resistance modulate gravid uterine and placental ferroptosis in PCOS-like rats. J Endocrinol. 2020 Sep;246(3):247-63.

70. Nasri F, Zare M, Doroudchi M, Gharesi-Fard B. Proteome Analysis of CD4(+) T Cells Reveals Differentially Expressed Proteins in Infertile Polycystic Ovary Syndrome Patients. Endocr Metab Immune Disord Drug Targets. 2021;21(11):1998-2004.

71. Shi Q, Liu R, Chen L. Ferroptosis inhibitor ferrostatin-1 alleviates homocysteine-induced ovarian granulosa cell injury by regulating TET activity and DNA methylation. Mol Med Rep. 2022 Apr;25(4).

72. Hu M, Zhang Y, Ma S, Li J, Wang X, Liang M, et al. Suppression of uterine and placental ferroptosis by N-acetylcysteine in a rat model of polycystic ovary syndrome. Mol Hum Reprod. 2021 Nov 27;27(12).

73. Liu H, Xie J, Fan L, Xia Y, Peng X, Zhou J, et al. Cryptotanshinone Protects against PCOS-Induced Damage of Ovarian Tissue via Regulating Oxidative Stress, Mitochondrial Membrane Potential, Inflammation, and Apoptosis via Regulating Ferroptosis. Oxid Med Cell Longev. 2022;2022:8011850.

74. Xu Y, Qin Z, Ma J, Cao W, Zhang P. Recent progress in nanotechnology based ferroptotic therapies for clinical applications. Eur J Pharmacol. 2020 Aug 5;880:173198.

75. Wang JJ, Ge W, Zhai QY, Liu JC, Sun XW, Liu WX, et al. Single-cell transcriptome landscape of ovarian cells during primordial follicle assembly in mice. PLoS Biol. 2020 Dec;18(12):e3001025.

76. Wang F, Liu Y, Ni F, Jin J, Wu Y, Huang Y, et al. BNC1 deficiency-triggered ferroptosis through the NF2-YAP pathway induces primary ovarian insufficiency. Nat Commun. 2022 Oct 5;13(1):5871.

77. Zhang D, Liu Y, Zhang Z, Lv P, Liu Y, Li J, et al. Basonuclin 1 deficiency is a cause of primary ovarian insufficiency. Hum Mol Genet. 2018 Nov 1;27(21):3787-800.

78. Young MF, Griffin I, Pressman E, McIntyre AW, Cooper E, McNanley T, et al. Maternal hepcidin is associated with placental transfer of iron derived from dietary heme and nonheme sources. J Nutr. 2012 Jan;142(1):33-9.

79. Zhang H, He Y, Wang JX, Chen MH, Xu JJ, Jiang MH, et al. miR-30-5p-mediated ferroptosis of trophoblasts is implicated in the pathogenesis of preeclampsia. Redox Biol. 2020 Jan;29:101402.

80. Zheng Y, Hu Q, Wu J. Adiponectin ameliorates placental injury in gestational diabetes mice by correcting fatty acid oxidation/peroxide imbalance-induced ferroptosis via restoration of CPT-1 activity. Endocrine. 2022 Mar;75(3):781-93.

81. Wallis AB, Saftlas AF, Hsia J, Atrash HK. Secular trends in the rates of preeclampsia, eclampsia, and gestational hypertension, United States, 1987-2004. Am J Hypertens. 2008 May;21(5):521-6.

82. Ghulmiyyah L, Sibai B. Maternal mortality from preeclampsia/eclampsia. Semin Perinatol. 2012 Feb;36(1):56-9.

83. Zur RL, Kingdom JC, Parks WT, Hobson SR. The Placental Basis of Fetal Growth Restriction. Obstet Gynecol Clin North Am. 2020 Mar;47(1):81-98.

84. Leanos-Miranda A, Campos-Galicia I, Berumen-Lechuga MG, Molina-Perez CJ, Garcia-Paleta Y, Isordia-Salas I, et al. Circulating Angiogenic Factors and the Risk of Preeclampsia in Systemic Lupus Erythematosus Pregnancies. J Rheumatol. 2015 Jul;42(7):1141-9.

85. Cardaropoli S, Todros T, Nuzzo AM, Rolfo A. Maternal serum levels and placental expression of hepcidin in preeclampsia. Pregnancy Hypertens. 2018 Jan;11:47-53.

86. Aires Rodrigues de Freitas M, Vieira da Costa A, Alves de Medeiros L, da Silva Garrote Filho M, Lemos Debs Diniz A, Penha-Silva N. Are There Differences in the Anthropometric, Hemodynamic, Hematologic, and Biochemical Profiles between Late- and Early-Onset Preeclampsia? Obstet Gynecol Int. 2018;2018:9628726.

87. Shaji Geetha N, Bobby Z, Dorairajan G, Jacob SE. Increased hepcidin levels in preeclampsia: a protective mechanism against iron overload mediated oxidative stress? J Matern Fetal Neonatal Med. 2022 Feb;35(4):636-41.

88. Toldi G, Stenczer B, Molvarec A, Takats Z, Beko G, Rigo J, Jr., et al. Hepcidin concentrations and iron homeostasis in preeclampsia. Clin Chem Lab Med. 2010 Oct;48(10):1423-6.

89. Brunacci F, Rocha VS, De Carli E, Esposito BP, Ruano R, Colli C. Increased serum iron in preeclamptic women is likely due to low hepcidin levels. Nutr Res. 2018 May;53:32-9.

90. Kajiwara K, Beharier O, Chng CP, Goff JP, Ouyang Y, St Croix CM, et al. Ferroptosis induces membrane blebbing in placental trophoblasts. J Cell Sci. 2022 Mar 1;135(5).

91. Yang N, Wang Q, Ding B, Gong Y, Wu Y, Sun J, et al. Expression profiles and functions of ferroptosis-related genes in the placental tissue samples of early- and late-onset preeclampsia patients. BMC Pregnancy Childbirth. 2022 Jan 31;22(1):87.

92. Patil SB, Kodliwadmath MV, Kodliwadmath M. Lipid peroxidation and antioxidant activity in complicated pregnancies. Clin Exp Obstet Gynecol. 2009;36(2):110-2.

93. Ahmadi R, Rahimi Z, Vaisi-Raygani A, Kiani A, Jalilian N, Rahimi Z. Apolipoprotein E genotypes, lipid peroxidation, and antioxidant status among mild and severe preeclamptic women from western Iran: protective role of apolipoprotein ε2 allele in severe preeclampsia. Hypertens Pregnancy. 2012;31(4):405-18.

94. Sarandöl E, Safak O, Dirican M, Uncu G. Oxidizability of apolipoprotein B-containing lipoproteins and serum paraoxonase/arylesterase activities in preeclampsia. Clin Biochem. 2004 Nov;37(11):990-6.

95. Wang Y, Zhang L, Zhou X. Activation of Nrf2 signaling protects hypoxia-induced HTR-8/SVneo cells against ferroptosis. J Obstet Gynaecol Res. 2021 Nov;47(11):3797-806.

96. Liao T, Xu X, Ye X, Yan J. DJ-1 upregulates the Nrf2/GPX4 signal pathway to inhibit trophoblast ferroptosis in the pathogenesis of preeclampsia. Sci Rep. 2022 Feb 21;12(1):2934.

97. Dipla K, Triantafyllou A, Grigoriadou I, Kintiraki E, Triantafyllou GA, Poulios P, et al. Impairments in microvascular function and skeletal muscle oxygenation in women with gestational diabetes mellitus: links to cardiovascular disease risk factors. Diabetologia. 2017 Jan;60(1):192-201.

98. Murthi P, Vaillancourt C. RETRACTED: Placental serotonin systems in pregnancy metabolic complications associated with maternal obesity and gestational diabetes mellitus. Biochim Biophys Acta Mol Basis Dis. 2020 Feb 1;1866(2):165391.

99. Gautam S, Alam F, Moin S, Noor N, Arif SH. Role of ferritin and oxidative stress index in gestational diabetes mellitus. J Diabetes Metab Disord. 2021 Dec;20(2):1615-9.

100. Filomeni G, De Zio D, Cecconi F. Oxidative stress and autophagy: the clash between damage and metabolic needs. Cell Death Differ. 2015 Mar;22(3):377-88.

101. Gauster M, Desoye G, Tötsch M, Hiden U. The placenta and gestational diabetes mellitus. Curr Diab Rep. 2012 Feb;12(1):16-23.

102. Hernandez TL, Brand-Miller JC. Nutrition Therapy in Gestational Diabetes Mellitus: Time to Move Forward. Diabetes Care. 2018 Jul;41(7):1343-5.

103. Peng HY, Li MQ, Li HP. High glucose suppresses the viability and proliferation of HTR-8/SVneo cells through regulation of the miR-137/PRKAA1/IL-6 axis. Int J Mol Med. 2018 Aug;42(2):799-810.

104. Yung HW, Alnaes-Katjavivi P, Jones CJ, El-Bacha T, Golic M, Staff AC, et al. Placental endoplasmic reticulum stress in gestational diabetes: the potential for therapeutic intervention with chemical chaperones and antioxidants. Diabetologia. 2016 Oct;59(10):2240-50.

105. Han D, Jiang L, Gu X, Huang S, Pang J, Wu Y, et al. SIRT3 deficiency is resistant to autophagy-dependent ferroptosis by inhibiting the AMPK/mTOR pathway and promoting GPX4 levels. J Cell Physiol. 2020 Nov;235(11):8839-51.

106. Torre LA, Trabert B, DeSantis CE, Miller KD, Samimi G, Runowicz CD, et al. Ovarian cancer statistics, 2018. CA Cancer J Clin. 2018 Jul;68(4):284-96.

107. Cannistra SA. Cancer of the ovary. N Engl J Med. 2004 Dec 9;351(24):2519-29.

108. Basuli D, Tesfay L, Deng Z, Paul B, Yamamoto Y, Ning G, et al. Iron addiction: a novel therapeutic target in ovarian cancer. Oncogene. 2017 Jul 20;36(29):4089-99.

109. Liu N, Lin X, Huang C. Activation of the reverse transsulfuration pathway through NRF2/CBS confers erastin-induced ferroptosis resistance. British Journal of Cancer. 2020 2020/01/01;122(2):279-92.

110. Cheng Q, Bao L, Li M, Chang K, Yi X. Erastin synergizes with cisplatin via ferroptosis to inhibit ovarian cancer growth in vitro and in vivo. J Obstet Gynaecol Res 173. 2021 Jul;47(7):2481-91.

111. Kim R, Hashimoto A, Markosyan N, Tyurin VA, Tyurina YY, Kar G, et al. Ferroptosis of tumour neutrophils causes immune suppression in cancer. Nature. 2022 Nov 16.

112. Maiorino M, Conrad M, Ursini F. GPx4, Lipid Peroxidation, and Cell Death: Discoveries, Rediscoveries, and Open Issues. Antioxid Redox Signal. 2018 Jul 1;29(1):61-74.

113. Yan HF, Zou T, Tuo QZ, Xu S, Li H, Belaidi AA, et al. Ferroptosis: mechanisms and links with diseases. Signal Transduct Target Ther. 2021 Feb 3;6(1):49.

Table 1. The comparison of main cell death

Types	Morphologic hallmarks	Biochemical hallmarks	Protein and Genetic hallmarks	Related female reproductive diseases	References
Ferroptosis	Cell swelling, plasma membrane rupture, smaller mitochondria, increased mitochondrial, membrane densities	Iron accumulation, lipid peroxidation increase, Antioxidant defense decrease	GPX4, GSH, ACSL4, TFRC, PTGS2, CHAC1	EMs, PCOS, POI, GMD, PE, OC	[2, 4, 12]
Apoptosis	nuclear fragment, chromatin agglutination, cell membrane rupture	Activation of caspase proteins, DNA fragmentation Cytochrome C release	caspase-8 caspase-3	POI, PE PCOS	[20-23]
Necroptosis	Cell swelling, plasma-membrane rupture	ATP deplete, Activate inflammatory reaction	Phospho-MLKL, phospho-RIP, RIP, phospho-RIP3, RIP3	PE, OC, FGR	[27-31]

Types	**Morphologic hallmarks**	**Biochemical hallmarks**	**Protein and Genetic hallmarks**	**Related female reproductive diseases**	**References**
Pyroptosis	chromatin condensation, intact nuclei, cellular swelling, and plasma membrane rupture	caspase-dependent,	caspase-1,IL1β , IL18, Gasdermin D	PE, PCOS, POI	32-35

Table 2 the application of ferroptosis in female reproductive diseases.

Diseases	Drugs	Mechanism	Effects	References
EMs	Vitamin E	Inhibit the peroxidation chain and lipoxygenase expression	Inhibiting ferroptosis and improve the fertility	61, 112, 113
	deferoxamine mesylate	Decrease iron level		
PCOS	Ferrostatin-1	decrease ROS, MDA and LDH improve GPX4	Reduce ferroptosis and protect ovarian granulosa cells	71-73
	N-acetylcysteine	Increase GPX4	Attenuate gravid uterine and placental ferroptosis, reduce fetal loss	
	Cryptotanshinone	inhibit MMP and NF-ϰB, activate MAPK/ERK signaling	Regulate ferroptosis and protects against PCOS-induced damage	
PE	Nrf2,DJ-1	inhibit the levels of GSH, MDA, ROSs, and Fe^{2+} and improve the SLC7A11, GPX4, and FPN1,	Inhibit ferroptosis and relieve hypoxia	95, 96
GMD	Adiponectin	restoration of CPT-1 activity to reduce oxidation stress	ameliorate placental injury	80
OC	erastin	inhibit System Xc- and result in depletion of GSH	inhibit tumor growth	109

Figure Legends

Figure 1. The brief activation mechanism of ferroptosis.

Excessive ROS, free iron, and lipid peroxidation all contribute to ferroptosis. Iron overload can result from increasing TfR1 or reducing ferritin expression (FTL and FTH1). In cells, the Fenton reaction results in a significant amount of ROS. PUFAs produce PUFA-PE and ROS under the influence of ASCL4 and LPAT3, ultimately cause ferroptosis. GPX4, a vital antioxidant enzyme in the body, has the ability to successfully anti-peroxide. Cys2, which is necessary for the production of GSH and is delivered by System Xc-, is essential for the operation of GPX 4.

Figure 2. The potential mechanism of ferroptosis in reproductive diseases.

In EMs,EECs are resistant to ferroptosis which allows them proliferation, migration and form ectopic tissue eventually, but ferroptosis can also promote the progress of EMs such as angiogenesis in surrounding tissues and leading to infertility, etc. In PCOS, Ferroptosis related proteins are highly expressed in infertile PCOS patients, and ferroptosis inhibitors can reduce the symptoms of polycystic ovary. In POI, Ferroptosis leads to premature apoptosis of ovarian cells, thus reducing ovarian reserve. In PE, iron overload, excessive ROS in vivo and other ferroptosis inducers lead trophoblast cells ferroptosis, and then lead placental injury. In GDM, higher BMI have a higher risk of β Cells ferroptosis, causing further increase of blood sugar. At the same time, high fat induced ferroptosis of trophoblasts, resulting in placental damage. In OC, ferroptosis inducers like erastin are thought as an effective drug to kill tumor cells and treat drug resistance.

Figure 3. The mechanism of ferroptosis-resistance in EMs.

Green represents the molecules that change in ectopic endometrium: the genes inducing ferroptosis ACSL4, LPCAT3 are downregulated. Moreover, ferritin and lnc RNA ADAMTS9-AS are upregulated, preventing ferroptosis. Blue represents the molecules that change in eutopic endometrium: the genes inducing ferroptosis including STA1, ALOX15, NCOA4, VDAC2 are downregulated in eutopic endometrium. FBLN1 which can increase ESC viability and migration by prohibiting ferroptosis are upregulated.

Hosted file

`Table.docx` available at https://authorea.com/users/575995/articles/619014-ferroptosis-in-female-reproductive-diseases-from-potential-pathogenesis-to-therapy

Hosted file

`figure.pptx` available at https://authorea.com/users/575995/articles/619014-ferroptosis-in-female-reproductive-diseases-from-potential-pathogenesis-to-therapy