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Sepsis-Induced Endothelial Dysfunction: Permeability and Regulated Cell Death

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Abstract: Sepsis is a life-threatening organ dysfunction caused by a dysregulated host response to infection. Endothelial cells (ECs) are an important cell type typically affected in sepsis, resulting in compromised barrier function and various forms of regulated cell death (RCD). However, the precise mechanisms underlying sepsis-induced EC damage remain unclear. This review summarizes the recent research progress on factors and mechanisms that may affect the permeability and RCD of ECs under septic conditions, including glycocalyx, damage-associated molecular patterns, and various forms of RCD in ECs, such as apoptosis, pyroptosis, ferroptosis, and autophagy. This review offers important insights into the underlying mechanisms of endothelial dysfunction in sepsis, aiming to contribute to developing small-molecule targeted clinical therapies.

Keywords: damage-associated protein, endothelial cell, glycocalyx, permeability, regulated cell death, sepsis

Introduction

As outlined in the most recent international consensus, sepsis is a severe organ dysfunction that continues to pose a significant threat to public health.[1] In 2017, an estimated 48.9 million cases and 11 million sepsis-related fatalities worldwide were documented, constituting roughly 20% of the annual global mortality.[2] The damage and dysfunction of vascular endothelial cells (ECs) are the shared characteristics in systemic reactions caused by sepsis.

Under physiological conditions, the extensive microvessels in the alveoli facilitate effective gas exchange. Meanwhile, ECs establish a semi-permeable membrane barrier that regulates the passage of fluids, proteins, and cells into tissues, thus maintaining the microvascular bed in an anti-thrombotic and anti-inflammatory state.[3] However, during infections, including bacterial, fungal, or viral infections, both exogenous pathogen-associated molecular patterns (PAMPs) and endogenous damage-associated molecular patterns (DAMPs) can induce overactivation of ECs, impair vascular integrity and barrier function, leading to increased permeability, interstitial edema, and other serious disruptions to gas exchange.[3] Impaired EC function in vital organs can be particularly fatal. The lungs are the most susceptible target organ in cases of sepsis. This can result in the development of acute lung injury (ALI) and even acute respiratory distress syndrome (ARDS) finally.[4] Similarly, EC damage and dysfunction can lead to damage to other vital organs, such as acute liver injury,[5] acute kidney injury (AKI),[6] encephalopathy,[7] and disseminated intravascular coagulation (DIC).[8] Thus, it is particularly important to analyze the specific mechanisms by which EC dysfunction occurs under septic conditions.

This review summarizes the recent research progress on factors and mechanisms that may affect the permeability and regulated cell death (RCD) of ECs under septic conditions, including glycocalyx, DMAPs, and various forms of RCD in ECs, such as apoptosis, pyroptosis, ferroptosis, and autophagy. This review offers important insights into the underlying mechanisms of endothelial dysfunction in sepsis, aiming to contribute to developing small-molecule targeted clinical therapies.

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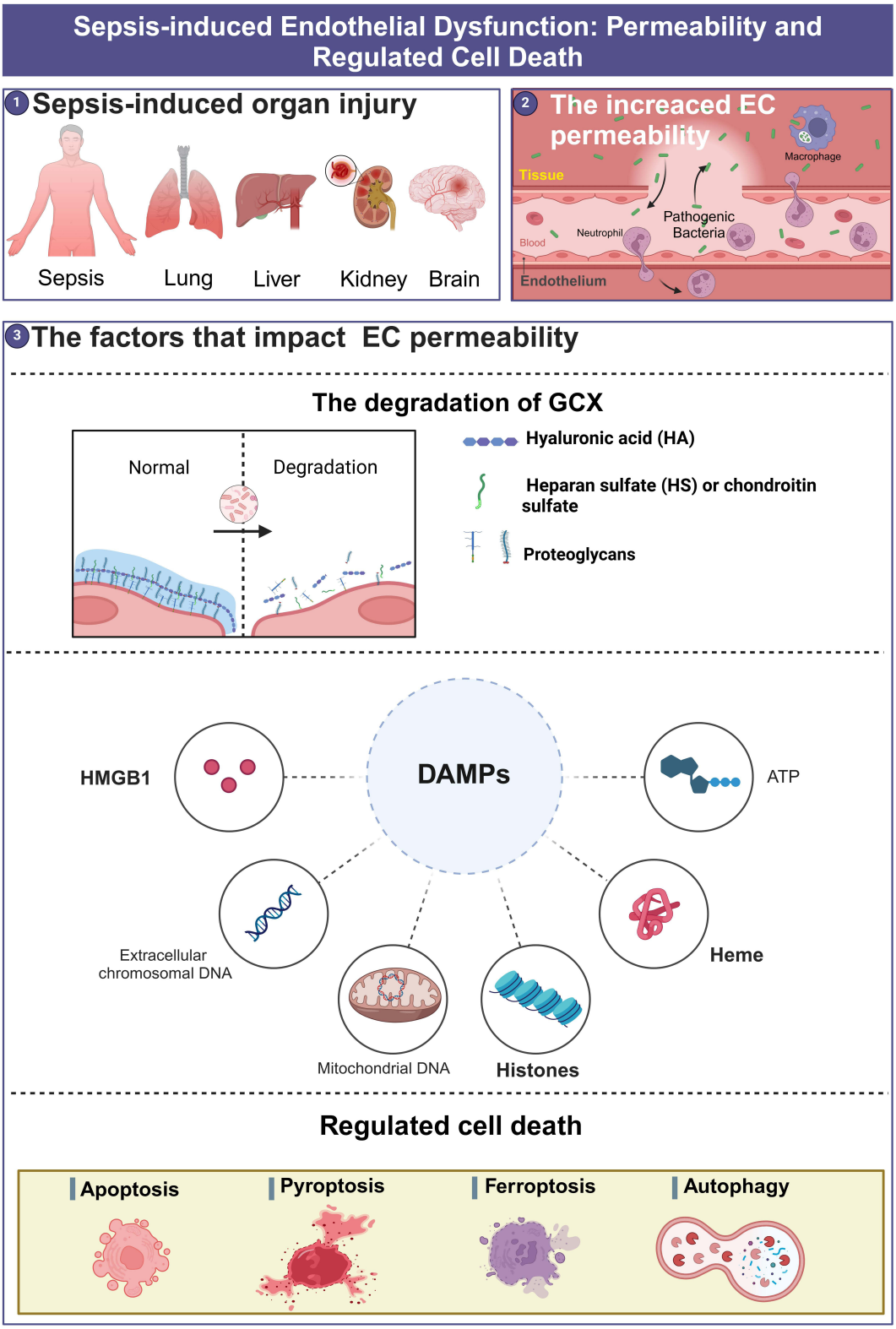
Graphical Abstract



GCX

The GCX is a large molecular substance that attaches to the luminal surface of ECs. Initially identified in 1966 via ruthenium red staining by Luft and his team,[9] the GCX comprises sulfated proteoglycans (including syndecans and glypicans), glycosaminoglycans (such as hyaluronic acid [HA], heparan sulfate [HS], and chondroitin sulfate), and sialic acid proteins.[10,11] In addition to the well-researched cell–cell junction, the GCX plays a crucial role in maintaining barrier function.[12] It can respond to shear stress sensed in the bloodstream by modulating vasoconstriction via the regulation of nitric oxide (NO), a biologically important inorganic compound that facilitates vasodilation and controls vascular tone.[13,14] Initially, the GCX was perceived as a physical barrier that separates the blood from ECs, and its impact on EC water permeability was the primary focus.[15] Recently, clinical studies have revealed that GCX degradation and shedding are prevalent in patients with sepsis or severe trauma, particularly in individuals with low plasma colloid osmolality.[16,17]

The mechanisms associated with GCX degradation are complex. The classical theory suggests that pro-inflammatory factors, such as tumor necrosis factor-α(TNF-α) and interleukin-1β (IL-1β), activate specific enzymes, such as metalloproteinase, heparanase (HPSE), and hyaluronidase, resulting in GCX degradation.[16,18] The pathophysiology of GCX damage may be attributed to the involvement of a disintegrin and metalloproteinase 15 (ADAM15), a membrane-bound metzincin metalloproteinase implicated in GCX shedding and vascular penetration in sepsis.[19] This process may involve the degradation of the GCX protein, CD44, on the EC membrane, resulting in the disruption of the VE-cadherin–β-catenin complex and compromised cell–cell adhesion.[19] Additionally, ADAM15 facilitates neutrophil transmigration across cell membranes. Thus, ECs may be further placed in an inflammatory microenvironment.[19] Recent in vitro experiments have further supported the association between the GCX and vascular permeability.[20–22] For example, during sepsis, GCX shedding and degradation are closely linked to thrombin depletion. This phenomenon might be caused by the loss of HA and syndecan-1 (SDC-1) and the heightened EC permeability resulting from concurrent catecholamine responses.[20,21] Moreover, increases in the plasma levels of free SDC-1, which is released upon GCX damage, predict adverse outcomes.[23,24] In the clinical setting, sepsis induces significant changes in polysaccharide metabolism, and different EC subtypes exhibit similar metabolic characteristics with a significant upregulation of glycan and fatty acid metabolisms. GCX shedding is also more evident in patients with reduced colloid osmotic pressure.[25,26]

The analysis of publicly available serum metabolomic data from patients with septic shock revealed significant changes in glycan metabolism among non-survivors.[25,26] One clinical trial reported an increase in the translation of HPSE and the expression of its active form during sepsis, which was associated with sepsis-related mortality.[27] HPSE is the only known endoglycosidase in mammals that can degrade HS, which is a major component of the GCX.[28] Heparin inhibits the cytosolic delivery of lipopolysaccharide (LPS) via the prevention of HPSE-mediated GCX degradation. Therefore, using heparin in the treatment of sepsis can improve the prognosis through anticoagulation and protect ECs by inhibiting HPSE.[29,30] However, the mechanisms that affect the structural integrity of the GCX on ECs require further investigation.

The specific manifestations of the effect of GCX damage on the EC barrier function have also been investigated. Reliable experimental data have indicated that the GCX increases vascular permeability and promotes leukocyte adhesion following injury.[22] However, distinguishing between EC connectivity and the GCX in terms of vascular permeability is challenging.[31] Under septic conditions, GCX degradation forms a vicious cycle. For example, the GCX plays an important role in EC homeostasis, while GCX degradation reduces the bioavailability of NO, which is closely related to the increase in reactive oxygen species (ROS) (Figure 1).[32,33] Consequently, increased ROS activates various specific enzymes (such as metalloproteinase, HPSE, and hyaluronidase) that destroy the GCX structure[34] and cause ECs to transform into myofibroblasts.[35] Damage to the GCX structure further affects the expressions of tight junction (TJ) proteins, thereby impairing barrier function.[36] HPSE can selectively strip HS from GCX to form the HPSE–HS complex, interfering with the synthesis of the TJ protein, zona occludens-1 (ZO-1).[36] Additionally, HPSE can impact the integrity of the GCX and interfere with the expression of *ZO-1* mRNA, subsequently affecting the assembly and function of TJ proteins. These effects can be counteracted by *N-desulfated/re*-N-*acetylated* heparin, which may be effective in protecting the GCX.[36,37]

Neutrophils participate in intrinsic immunity and typically play a role in the phagocytosis of fine microorganisms in infectious diseases such as sepsis. Their numbers are commonly clinically indicative of the degree of infection or inflammation. Golden and his team proposed a mechanism of HS-dependent neutrophil enrichment in the liver.[38] In a mouse model of *Staphylococcus aureus*-induced septicemia, rapid remodeling of the protein composition on the surface

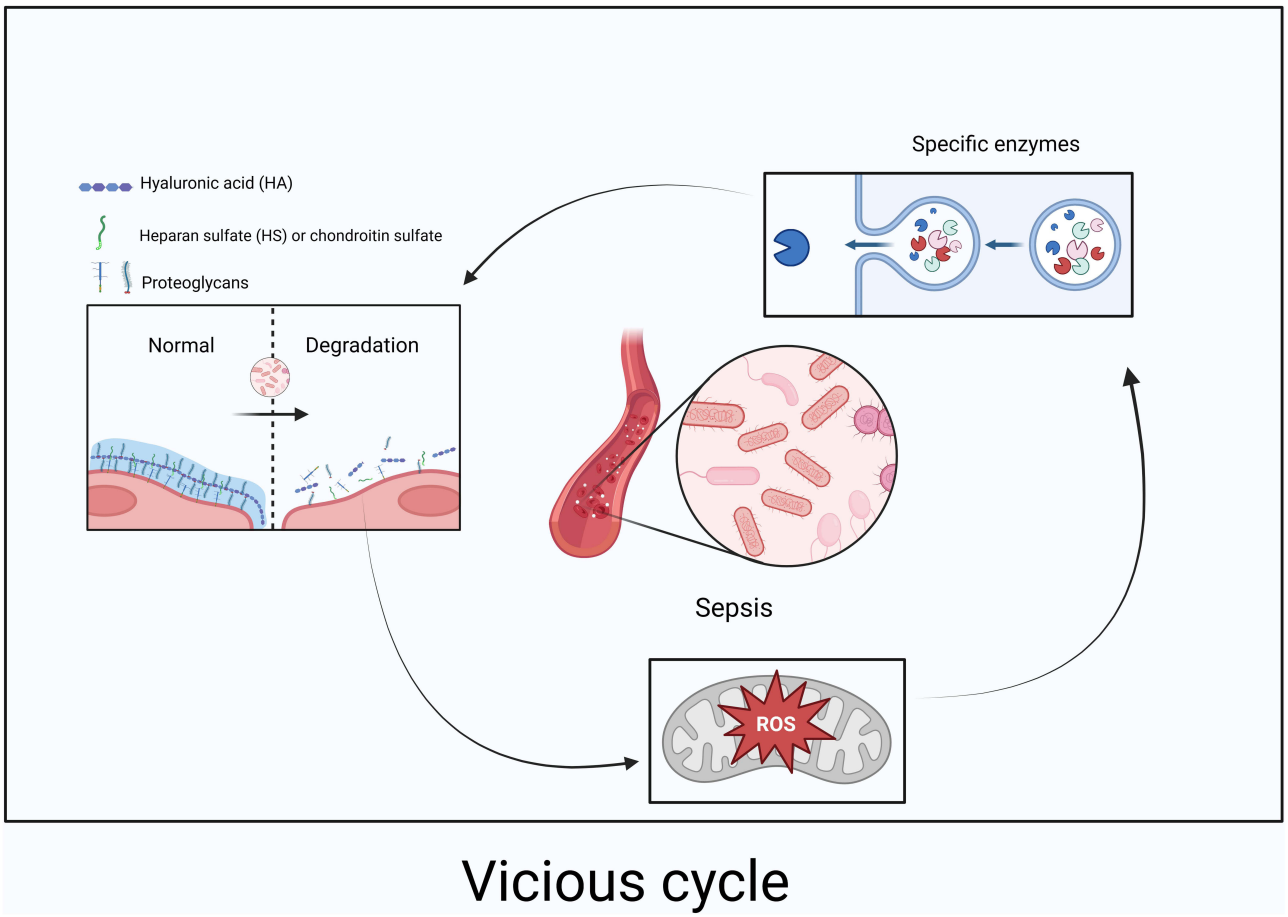


Figure 1 Schematic illustration of the vicious cycle that occurs when the GCX breaks down. First, sepsis functions as an initiating factor to destroy the GCX. Subsequently, ROS levels increase. Finally, specific enzymes are activated to decompose the GCX, completing the closed loop. Created in BioRender. Jiang, L. (2024) http://BioRender.com/w57v515.

of hepatic blood vessels was observed. These changes included the deposition of proteins from neutrophils (such as myeloperoxidase, neutrophil elastase, matrix metallopeptidase 9, and neutrophil gelatinase-associated lipocalin) and alterations in the levels of HS-binding factors that occurred before the onset of liver injury.[38] Their study on sepsis-related liver failure has provided experimental evidence that the genetic modification of fine HS structures can reduce the transport of neutrophils to the liver[39,40] and that the modulation of vascular GCX can reduce neutrophil infiltration in aseptic liver injury.[38] Although further research is required, elevated levels of plasma-derived GCX fragments (HS or HA) can be used to predict adverse outcomes in sepsis, potentially involving HS-dependent neutrophil enrichment.[23,24,38] Empagliflozin reduces the inflammatory state of the endothelium caused by GCX degradation; however, this mechanism may be independent of HS. Empagliflozin can reduce the activation of nucleotide-binding domain, leucine-rich repeat-containing family, pyrin domain-containing 3 (NLRP3) by downregulating pro-apoptotic signals and the thioredoxin-interacting protein, thereby reducing oxidative damage from endoplasmic reticulum (ER) stress.[41,42] Thus, protecting the GCX from being shed or removing GCX breakdown products may help to relieve EC dysfunction and protect blood vessels during sepsis.

DAMPs

In the mid-1990s, Polly Matzinger introduced the concept of DAMPs, which are endogenous molecules extracellularly released following tissue injury that activate pro-inflammatory cascade responses and play a crucial role in sepsis.[43] Various potential DAMPs, such as ATP, high mobility group box 1 (HMGB1), extracellular chromosomal DNA, mitochondrial DNA, histones, and heme, participate in the development of sepsis injury. Among them, HMGB1, histones, and heme have been more widely studied;[44–47] therefore, we summarize the related research progress as follows.

HMGB1

As a nuclear factor and secreted protein, HMGB1 exhibits a high degree of conservation, with 99% homology between human and rodent species.[48] HMGB1 can exist in the nucleus and extracellular matrix. Within the nucleus, HMGB1 normally functions as a chromatin-binding factor, aiding in the assembly of proteins on specific DNA targets and inducing DNA bending.[44] However, HMGB1 exhibits a weak binding affinity to chromatin and is rapidly released into the extracellular environment upon cell damage.[49]

HMGB1 has multiple deleterious on endothelial cell function. During sepsis, HMGB1 is passively released owing to intense inflammation or cytolytic necrosis, particularly in cells undergoing mitosis or those in interphase. Type 1 interferon-mediate signaling can induce the release of HMGB1 in macrophages and monocytes.[50] Moreover, lactylation and acetylation modifications of HMGB1 facilitate its secretion from macrophages through exosomes.[51] Extracellular HMGB1 interacts with LPS, a critical constituent of the outer membrane of gram-negative bacteria, to facilitate LPS internalization into ECs via endocytosis, thereby triggering caspase-11-dependent pyroptosis.[52] HMGB1 can also bind to LPS, mediating its transport to macrophages during sepsis. This process activates caspase-11 and caspase-1 signaling, which triggers apoptosis, autophagy, and pyroptosis (Figure 2).[49] Moreover, HMGB1 increases the procoagulant activity of tissue factors and promotes the exposure of procoagulant phosphatidylserine on the cell surface. This contributes to the assembly of prothrombin complexes and ultimately results in widespread DIC.[50]

At the same time, HMGB1 has multiple beneficial effects on endothelial cell function. HMGB1 can promote mitochondrial fusion in ECs via the C-X-C-chemokine receptor 4 (CXCR4) /20S proteasome subunit beta 5 pathway without significantly affecting their inflammatory phenotype.[53] The current view is that proper mitochondrial fusion facilitates the maintenance of normal mitochondrial and cellular functions and supports increased energy metabolism during cell proliferation.[54,55] HMGB1 may also play a regulatory role in angiogenesis. The internalization of HMGB1 via the receptor for advanced glycation end-products (RAGE) located on the cellular membrane could induce the secretion of vascular endothelial growth factor-A and endoglin.[56] This process is facilitated by the release of fragments from a crucial functional domain of HMGB1, which occurs owing to cytokine activity. Notably, in vivo experiments conducted on mice demonstrated that this phenomenon promotes angiogenesis in ligated femoral arteries.[56] Thus, these findings provided new insights into the mechanism underlying HMGB1-mediated cytoprotective effects and revealed that HMGB1 may exhibit a specific double-edged sword effect on ECs.

Histones

Histones are evolutionarily conserved nuclear proteins that interact with DNA to create nucleosomes. In particular, the histone subunits H1B, H2A, H2B, H3, and H4 play crucial roles in chromatin compaction and chromosomal structure formation.[57] In the instances of severe trauma or infections such as sepsis, extracellular DNA-binding proteins, such as histones, can trigger immune responses and vascular dysfunction in the host.[47,58] Histones are released either passively as DAMPs during cell necrosis or actively by neutrophils as part of neutrophil extracellular traps, which contain histones, myeloperoxidase, and extracellular DNA.[43,59] At present, circulating histones are considered as the primary mediators of cell death and multiple organ dysfunction in sepsis.[60–62] Furthermore, the administration of anti-histone antibodies in different animal models of sepsis resulted in reduced mortality rates.[58,63] EC function impairment has since been confirmed to be primarily mediated by Toll-like receptor 4 (TLR4) signaling.[64,65] A recent experimental study revealed that the levels of impact of the histone subunits H3 and H4 on EC permeability differed from those of the H2A and H2B subunits. H3 and H4 are responsible for damaging ECs, and an increase in ECs permeability can be observed even at low histone concentrations.[47] Once the histone concentration surpasses a certain threshold, the expression of EC adhesion molecules, such as intercellular adhesion molecule 1 (ICAM1) and vascular cell adhesion molecule 1 (VCAM1), is significantly increased.[47] Notably, both ICAM1 and VCAM1 belong to the immunoglobulin superfamily of cell adhesion molecules and are essential in facilitating the robust adhesion of leukocytes to ECs. Histones disrupt cellular junctions by phosphorylating the tight junction molecule, VE-cadherin, resulting in EC barrier dysfunction, increased permeability, and the upregulation of inflammatory responses (Figure 2).[47]

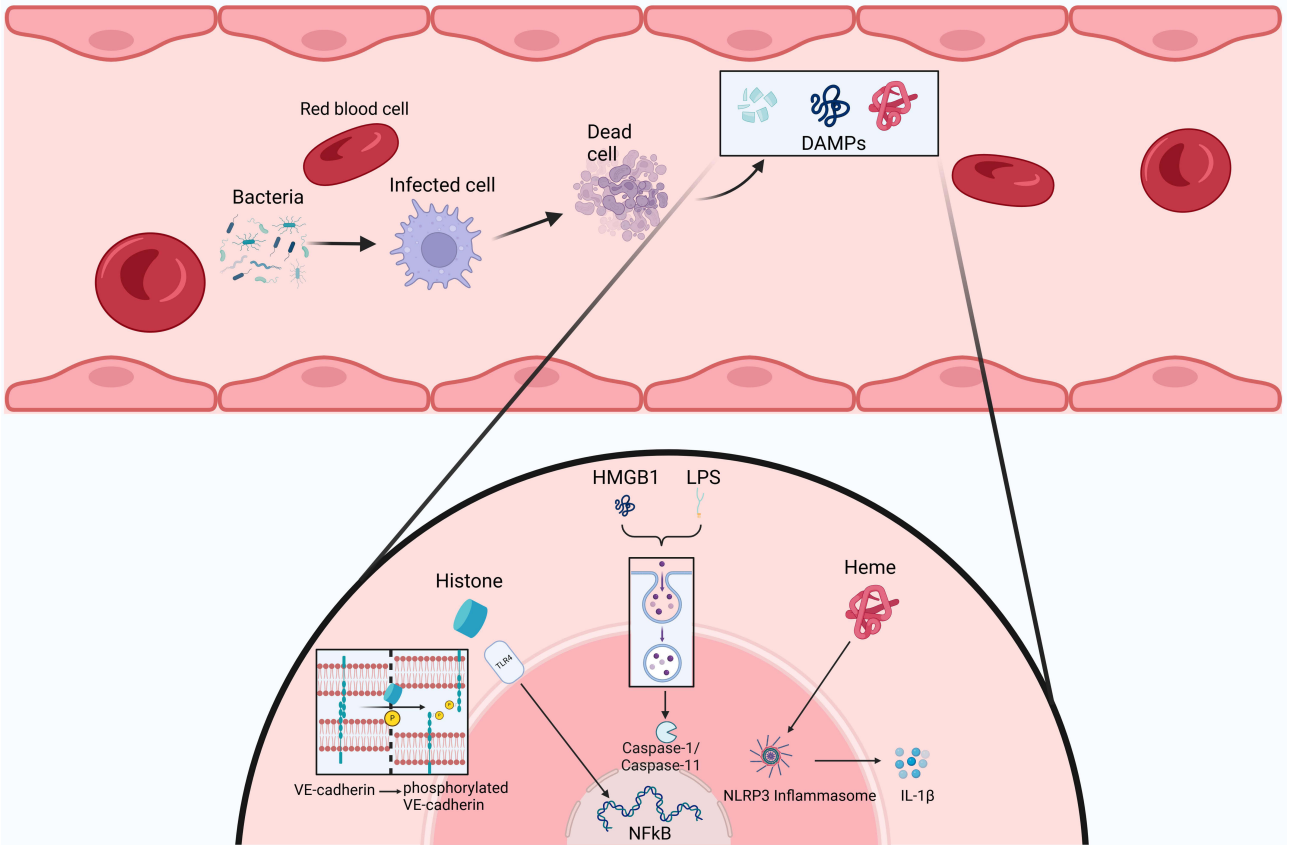


Figure 2 Effect of DAMPs on vascular ECs during sepsis or severe bacterial infections. Right: Histones primarily function via the activation of related signaling pathways by TLR4 and can phosphorylate VE-cadherin to further weaken the tight junctions between ECs. Center: HMGB1 and LPS combine and enter cells via endocytosis and activate the caspase1/11-related signaling pathway. Left: In ECs, heme promotes IL-1β release by activating the NLRP3 inflammasome, resulting in an inflammatory response. Created in BioRender. Jiang, L. (2024) http://BioRender.com/v55n286.

Heme

Heme (iron protoporphyrin-IX) is a naturally occurring iron chelate synthesized by the mitochondria that is primarily involved in oxygen transport and storage and cellular metabolic enzyme activity.[66–68] Increased circulating free heme levels have been linked to a higher risk of hemolytic disorders, such as sickle cell anemia, and regulated EC death in patients with severe sepsis.[66,69] Moreover, the excessive release of free heme under pathological conditions is associated with EC barrier dysfunction. In macrophages, heme or the oxidized form of hemoglobin stimulates NLRP3, resulting in the production of the inflammatory factor IL-1β.[70,71] In ECs, heme, rather than hemoglobin, activates the NLRP3 inflammasome, resulting in the release of IL-1β (Figure 2).[45] Heme oxygenase-1 (HO-1) can effectively degrade heme, converting it to carbon monoxide, iron, and biliverdin, and is beneficial and therapeutic for maintaining EC barrier function.[72,73] Recently, MitoQ (a mitochondria-targeted antioxidant) was found to effectively reduce EC damage and inhibit endothelial apoptosis by suppressing the degradation of nuclear factor erythroid 2-related factor 2 (NRF2) and upregulating downstream HO-1.[74] Regarding the molecular mechanism by which heme affects EC barrier function, one study reported that following heme exposure, the levels of the TJ proteins, ZO-1, claudin-1, and claudin-5, were primarily reduced, while the p38 mitogen-activated protein kinase (MAPK)/heat shock protein 27 pathway, which mediates endothelial cytoskeleton changes, underwent significant alterations.[75] Notably, the MAPK family is activated by various environmental stress and inflammatory cytokines and the heat shock protein 27 is one of the downstream targets of p38 MAPK.[75] These conditions improved following the knockout of a key regulator, MKK3, suggesting that targeting this pathway may be a therapeutic strategy to mitigate sepsis and sepsis-related disorders.[75]

The DAMPs discussed above are not independent entities; they interact with one another and collectively contribute to severe infections such as sepsis. As previously discussed, HMGB1 is a pivotal pro-inflammatory factor in sepsis; it can bind to DNA, RNA, nucleosomes, and histones, thereby participating in the inflammatory regulation response.[76] HMGB1 may also interact with histones, affecting their acetylation state.[77,78] Similarly, HMGB1 can bind to the toll-like receptor 4 (TLR4) receptor to activate the innate immune response.[79] Heme, an important iron-containing compound in the human body, can induce the release of HMGB1 and produce a synergistic effect with it, both participating in the pathogenic mechanism of sepsis.[69] Notably, HMGB1 can affect enzymes involved in heme metabolism, such as HO-1, promoting ferroptosis in ECs.[80] These DAMPs, as part of the DAMP family, influence each other through various pathways and play a pro-inflammatory synergistic role in sepsis.

Oxidative Stress

Normally, cells themselves can produce oxidants such as ROS. The high oxidative stress environment of sepsis can impair the EC antioxidant function due to excessive consumption of antioxidants, such as glutathione, superoxide dismutase, and catalase, thus disrupting the balance between ROS, reactive nitrogen species, and antioxidants.[32] Oxidative stress, a pathway associated with damage, can induce cell damage and even EC necrosis, which directly leads to the release of DAMPs.[81,82] ROS relies on its strong oxidizing properties, oxidizing the lipid membrane of the cell, as well as the proteins, DNA and organelles within the cell.[32] The ER exposed to oxidants could lead to ER stress and the release of vesicles containing HMGB1, thereby facilitating inflammation development.[42] Similarly, DAMPs can be oxidatively modified, leading to inactivation of physiological functions and promoting pathology.[83]

Meanwhile, the release of DAMPs could further aggravate tissue damage and inflammatory responses, exacerbates oxidative stress, leading to ROS production.[84] Upon increased ROS levels within the cell, HMGB1 shifts from its reduced form, which exhibits chemotactic activity, to its oxidized form with disulfide bonds, known as oxidized HMGB1 (Ox-HMGB1).[85] Extracellularly, Ox-HMGB1 with intramolecular disulfide bonds may have a stronger pro-inflammatory effect. Ox-HMGB1 binds more to TLR4, which activates immune cells more strongly and releases more inflammatory factors, exacerbating the inflammatory response and ROS production.[86] Notably, when intracellular levels of ROS exceed a certain threshold, two Ox-HMGB1 molecules can form disulfide bonds between cysteine residues, resulting in the production of an anti-parallel oxidative self-dimerized HMGB1 that confers protection to DNA.[85] In addition, free heme is detrimental and exacerbates oxidative stress in ECs, leading to cellular damage.[87] Heme can catalyze ROS production and oxidize proteins and lipids, leading to DNA damage.[88] Heme has a high affinity for biological membranes, rendering them susceptible to ROS and resulting in oxidative damage to membrane lipids.[89] It also activates the unfolded protein response pathway, leading to ER stress.[90] These results suggest the interaction between DAMPs and oxidative stress, and the regulation of DAMPs or oxidative stress level may contribute to the improvement of the inflammatory environment in sepsis.

RCD

Distinct from instantaneous and catastrophic cell death under physical or mechanical conditions, RCD is a form of cell death caused by intracellular events. It relies on precise molecular regulatory mechanisms and can be modulated by drugs or genetic intervention.[91,92] In 2018, the Nomenclature Committee on Cell Death (NCCD) updated the classification of RCD by dividing it into 13 categories.[93] Through an extensive review of the literature, we found that increasing evidence indicates that EC apoptosis, pyroptosis, ferroptosis, and autophagy triggered by pathogenic components, such as LPS, inflammatory cytokines, and oxidative stress, are involved in multi-organ dysfunction and septic shock caused by sepsis. Here, we briefly discuss the specific mechanisms and targets of apoptosis, pyroptosis, ferroptosis, and autophagy in ECs during sepsis.

Apoptosis of ECs in Sepsis

Apoptosis is an active and orderly cell death process regulated by gene expression that aims to maintain cellular homeostasis under physiological or pathological conditions. Both in vitro and in vivo experiments have demonstrated that sepsis can induce EC injury and apoptosis, which is characterized by the formation of apoptotic bodies.[94–96] We

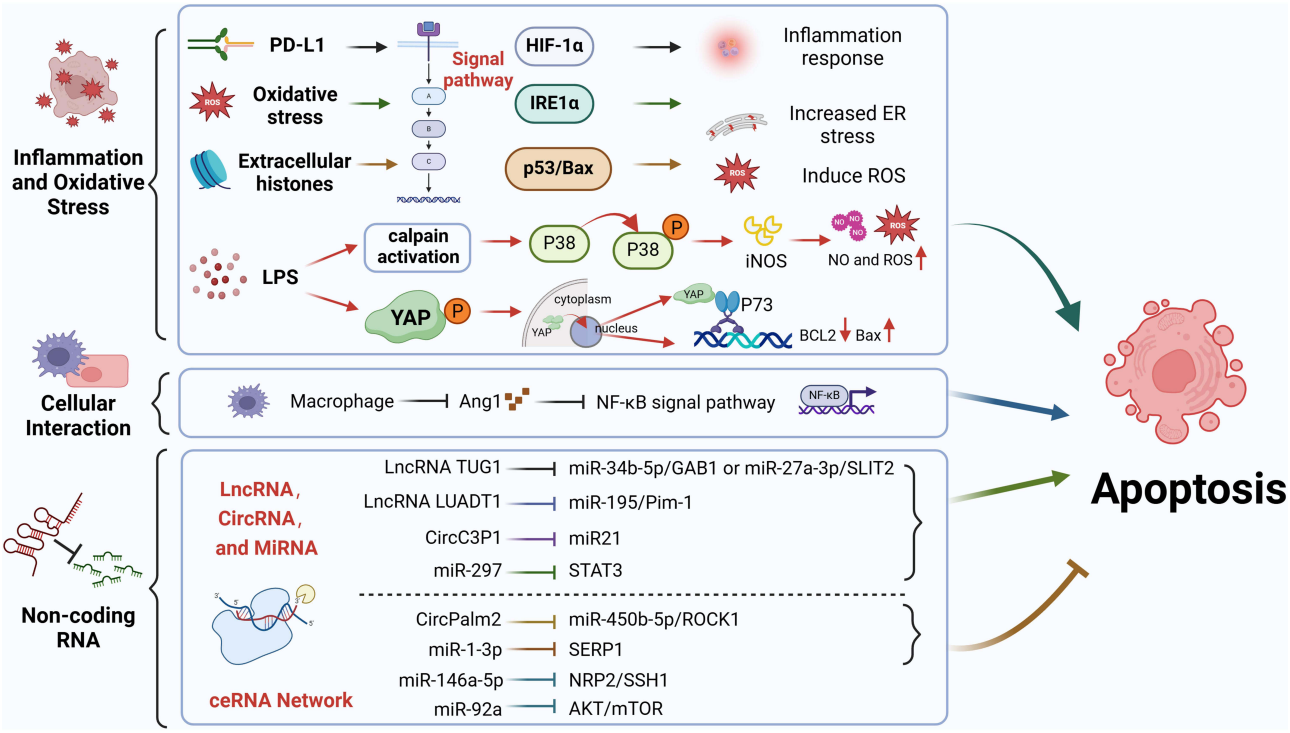


Figure 3 Specific pathophysiological mechanisms of EC apoptosis in sepsis. Increased inflammation and oxidative stress and the activation of the NF-kB signal pathway due to macrophage could aggravate EC apoptosis. Moreover, the ceRNA network comprising non-coding RNA could also regulate EC apoptosis. Created in BioRender. Jiang, L. (2024) http://BioRender.com/k50n929.

present the specific pathophysiological mechanisms of EC apoptosis in sepsis from three aspects: inflammation and oxidative stress, cellular communication, and non-coding RNA (Figure 3).

A highly inflammatory environment and elevated oxidative stress levels under septic conditions are the primary factors resulting in EC apoptosis, which involves complex molecular mechanisms. PD-L1 can activate the inflammatory response of pulmonary vascular ECs via the hypoxia-inducible factor-1α (HIF-1α) signaling pathway, thereby increasing apoptosis.[97] Inhibiting inflammation is considered a reliable way to protect against apoptosis. Research has indicated that interleukin-35, an anti-inflammatory cytokine, exerts anti-inflammatory and anti-apoptotic effects on LPS-induced ECs by activating the signal transducer and activator of transcription (STAT) 1 and 4 signaling pathways.[98] Another study used genome-wide CRISPR/Cas9 knockout screening technology to explore potential targets in the TNF-α signaling pathway using TNF-α-stimulated human umbilical vein endothelial cells (HUVECs). This study identified and confirmed NLR family member X1 (NLRX1) as a new regulatory factor involved in TNF-α-induced EC apoptosis.[99] Moreover, the regulatory role of Yes-associated protein (YAP) in EC apoptosis has been reported. As a key effector molecule of the Hippo signaling pathway, YAP participates in regulating various biological processes of cells, including proliferation, differentiation, transformation, and apoptosis. In human pulmonary microvascular endothelial cells (HPMECs), LPS can promote the phosphorylation of YAP on Y357, leading to its translocation from the cytoplasm to the nucleus, where it interacts with the transcription factor P73 to induce EC apoptosis. This suggests that YAP may serve as a potential therapeutic strategy for sepsis-induced lung injury.[100] Moreover, calpain and its signaling pathway also contribute to sepsis-induced EC injury. In LPS-induced AKI, the activation of endothelial calpain can promote p38 phosphorylation, increase the expression of inducible nitric oxide synthase (iNOS), and further enhance the excessive expression of NO and ROS, ultimately inducing EC apoptosis.[101]

ER stress is also involved in the apoptosis of ECs during sepsis, primarily through the oxidative stress-activated inositol requiring enzyme-1α (IRE1α) signaling pathway. This can be inhibited by adipocyte-secreted adiponectin, thereby reducing apoptosis in ECs.[96] Extracellular histones can function as DAMPs, inducing oxidative stress and dose-

dependently inducing apoptosis of primary HUVEC in a p53-Bax-dependent manner at concentrations of 50 μg/mL or higher.[102] Moreover, in LPS-induced ECs, calpain activation and p38 MAPK phosphorylation are enhanced, resulting in apoptosis, whereas heat stress protects ECs by blocking these pathways.[103]

Interactions between cells also play crucial roles in EC dysfunction and apoptosis. As a key immune cell type involved in septic conditions, macrophages exacerbate LPS-induced EC dysfunction and promote apoptosis by reducing angiopoietin-1 (Ang1) expression and activating the nuclear factor kappa-B (NF-κB) pathway.[104]

Recent studies have demonstrated that non-coding RNAs, including long non-coding RNAs (lncRNAs), circular RNAs, and microRNAs (miRNAs), participate in regulating apoptosis in ECs under septic conditions, offering potential targets for sepsis treatment through competing endogenous RNA (ceRNA) regulatory networks. The lncRNA taurine-upregulated gene 1 (TUG1) can inhibit sepsis-induced inflammation and EC apoptosis by targeting miR-34b-5p and its downstream target gene *GRB2 associated binding protein 1 (GAB1)*[105] or miR-27a-3p and its downstream target gene *slit guidance ligand 2* (*SLIT2)*.[95] Another study demonstrated the downregulation of lncRNA LUADT1 in plasma samples collected from patients with sepsis. This downregulation of LUADT1 has also been confirmed in LPS-stimulated human primary coronary artery ECs (HPCAECs). Further bioinformatics analysis, combined with experimental verification, indicated that LUADT1 can inhibit LPS-induced apoptosis of HPCAECs by sponging miR-195 and increasing the expression of the target gene *Pim-1*.[106] CircC3P1 exerts anti-inflammatory and anti-apoptotic effects on LPS-treated primary mouse lung microvascular ECs by targeting miR-21.[107] CircPALM2 inhibition regulates the miR-450b-5p/Rho-associated coiled-coil containing protein kinase 1 (ROCK1) axis to alleviate sepsis-induced apoptosis and inflammation in primary mouse lung microvascular ECs.[108] Individual miRNAs intrinsically regulate apoptosis in ECs. For example, miR-1-3p can target *stress-associated ER protein 1* (*SERP1*), inhibit EC proliferation, and promote apoptosis, thereby exacerbating EC dysfunction and promoting ALI development.[109] miR-297 reduces STAT3 expression, thereby alleviating LPS-induced inflammation and protecting ECs from apoptosis.[110] The downregulation of miR-146a-5p can upregulate *neuropilin 2* (*NRP2*) and *slingshot protein phosphatase 1* (*SSH1*), inhibit the expression of pro-inflammatory factors, and reduce LPS-induced EC apoptosis.[111] MiR-92a plays an important role in sepsis-induced ARDS. It promotes apoptosis in LPS-induced HPMECs by inhibiting the protein kinase B (AKT)/mammalian target of rapamycin (mTOR) signaling pathway, thereby adversely affecting ARDS.[112] These studies provide effective targets against the apoptosis of ECs and the alleviation of EC dysfunction in sepsis, offering the potential for clinical translation.

Pyroptosis of ECs in Sepsis

Pyroptosis is one of the most important types of RCD associated with septic shock and tissue damage and is triggered by PAMPs, DAMPs, and inflammatory cytokines. The primary distinction between pyroptosis and apoptosis lies in the disruption of the cell plasma membrane integrity, resulting in the release of cell contents. This phenomenon amplifies local inflammatory responses and the early release of IL-1β and IL-18, which results in an imbalanced immune response and exacerbation of tissue damage.[113,114] The specific pathophysiological mechanisms of EC pyroptosis in sepsis are illustrated from three aspects: GSDMD-NT dependent pathway, NLRP3 dependent pathway, and cellular communication (Figure 4).

Pyroptosis is an inflammatory form of cell death. First, LPS binds to the TLR4 co-receptor myeloid differentiation protein-2 (MD2) complex on the cell membrane. The LPS then activates caspase-11, which cleaves gasdermin D (GSDMD) to release its N-terminal fragment (GSDMD-NT), forming a transmembrane pore and inducing GSDMD-dependent pyroptosis.[115–118] Furthermore, the GSDMD-dependent pyroptotic pathway participates in a novel mitochondrial mechanism. The LPS-induced GSDMD-NT can bind to the mitochondrial membrane and form mitochondrial pores, causing mitochondrial damage and subsequent mitochondrial DNA release into the cytosol. This is accompanied by the activation of cyclic GMP-AMP synthase (cGAS)-stimulator of interferon genes (STING) pathway and the suppression of YAP-mediated EC proliferation and vascular regeneration.[119] A recent study also discovered a calcium-dependent mechanism in GSDMD-mediated pyroptosis, in which cytosolic calcium signaling promoted the translocation of the NH_2-terminal fragment of GSDMD-NT to the plasma membrane, enhancing endothelial pyroptosis. Phospholipase Cγ1 can also induce endothelial barrier dysfunction via this pathway, exacerbating perfusion impairment and increasing mortality in mice with sepsis.[120] In addition, PTMs of GSDMD may offer a potential approach for treating sepsis-

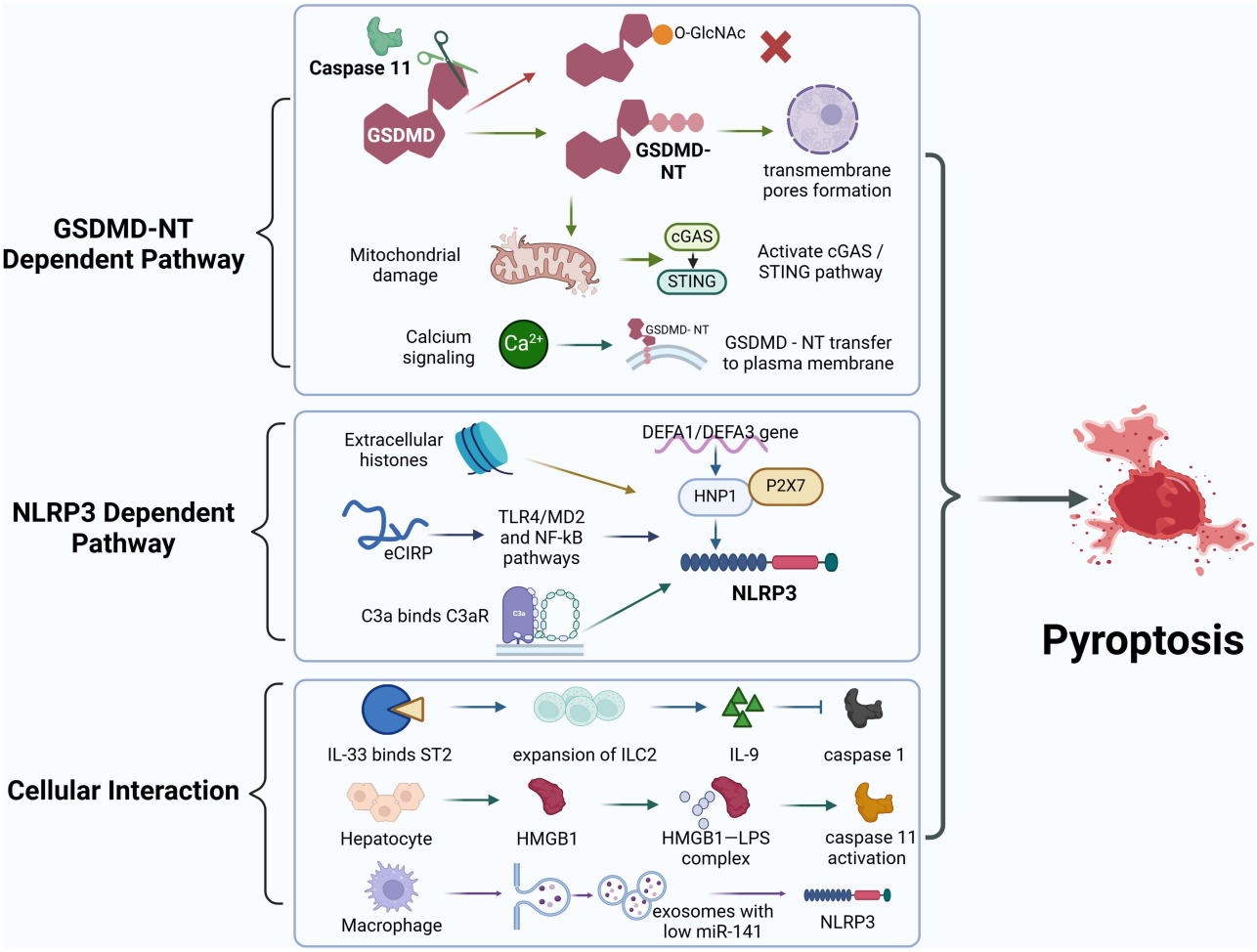


Figure 4 Potential mechanisms of EC pyroptosis in sepsis. The GSDMD-NT-dependent pathway participates in activating pyroptosis by inducing the formation of transmembrane pores and mitochondrial damage, which further promotes pyroptosis. The NLRP3-dependent pathway is important in pyroptosis occurrence, and several DAMPs, such as extracellular histones and eCIRP, and complement C3a can increase NLRP3 levels. Recent research has demonstrated that Group 2 innate lymphoid cells (ILC2), hepatocytes, and macrophage can regulate pyroptosis by influencing caspase family proteins or NLRP3. Created in BioRender. Jiang, L. (2024) http://BioRender.com/b96h492.

induced EC injury. Notably, O-linked β-N-acetylglucosamine (O-GlcNAc) modification of GSDMD can reduce LPS-induced EC pyroptosis by inhibiting the interaction between LPS and caspase-11.[121]

The NLRP3 inflammasome plays a critical role in the initiation and development of endothelial pyroptosis. Extracellular histones function as DAMPs, activating the NLRP3 inflammasome and mediating endothelial pyroptosis. The high acetylation of extracellular histones or the use of antioxidants can significantly reduce pyroptosis, suggesting that blocking histone-mediated pyroptosis is a feasible therapeutic strategy against sepsis.[122] Extracellular cold-inducible RNA-binding protein (eCIRP) is a DAMP with a significant impact on the pathophysiology of inflammatory diseases. In ECs, eCIRP can induce NLRP3 inflammasome activation via the TLR4/MD2 and NF-kB-dependent pathways, increase ICAM1 expression, activate the inflammatory response, and subsequently induce caspase-1, IL-1β, and IL-18 expressions, leading to pyroptosis.[123,124] Blocking NLRP3-mediated inflammation through drug intervention is an effective approach for inhibiting pyroptosis. Astragaloside IV can alleviate LPS-induced endothelial pyroptosis by inhibiting the ROS/NLRP3-mediated inflammatory response, providing a scientific basis for its clinical application.[125] Notably, some studies have suggested that genetic factors can affect the severity of sepsis. For example, in the Chinese Han population, individuals with a high genetic copy number of *DEFA1/DEFA3* genes, which encode human neutrophil peptides (HNP) 1–3, are more likely to develop severe sepsis.[126] Another study indicated that HNP-1 can mediate the activation of the canonical NLRP3/caspase-1 inflammasome through its interaction with the purinergic receptor P2X ligand-gated ion

channel 7 (P2X7), subsequently inducing pyroptosis of ECs and playing an important role in sepsis-induced EC injury and organ dysfunction.[127]

The complement system is an essential part of host defense mechanisms and promotes inflammation by activating leukocytes and ECs. The complement pathway also plays a crucial role in promoting pyroptosis during sepsis.[128,129] Complement C3 is at the core of the complement activation pathway. C3a levels in organs affected by sepsis are significantly elevated and are closely correlated with sepsis-related organ dysfunction and poor prognosis. Further investigations into the role and mechanism of C3a in sepsis-induced ALI have revealed that C3a binds to the C3a receptor, subsequently activating the NLRP3/caspase-1 and caspase-11 signaling pathways, resulting in pyroptosis in pulmonary vascular ECs.[130]

Various cells are also widely involved in the regulation of EC pyroptosis. Group 2 innate lymphoid cells (ILC2) are one of the three subgroups of innate lymphoid cells (ILC1, ILC2, and ILC3) and are the primary ILC population in the lungs. The release of IL-33 during sepsis can induce the expansion of ILC2 in the lungs via its receptor suppression of tumorigenicity 2 (ST2), and the expanded ILC2 can secrete IL-9, which inhibits caspase-1 activation, thereby preventing pyroptosis in pulmonary ECs.[131] Hepatocytes can sense PAMPs in circulation and release HMGB1. HMGB1 then forms complexes with LPS and enters EC lysosomes via the RAGE, causing lysosomal instability and enabling LPS to enter the cytoplasm. Subsequently, caspase-11 is activated, resulting in EC pyroptosis.[52] Heparin or non-anticoagulant-modified heparin can inhibit the interaction between HMGB1 and LPS, minimizing the entry of LPS into the cytoplasm, and blocking caspase-11 activation while reducing pyroptosis.[132] This may represent a potential therapy to alleviate ALI. In addition, exosomes with low miR-141 expression derived from macrophages can activate the downstream target NLRP3 and mediate pyroptosis of ECs, thereby promoting the progression of sepsis;[133] this represents a potentially valuable target for the treatment of sepsis.

Ferroptosis of ECs in Sepsis

Ferroptosis, a newly recognized form of unique cell death, is driven by iron-dependent lipid peroxidation and regulated by various cellular metabolic pathways and signaling cascades.[134,135] Notably, ferroptosis is prevalent in sepsis and may contribute to the development of multi-organ dysfunction syndrome.[136] In this section, the relevant progress of ferroptosis of ECs under septic conditions will be comprehensively discussed (Figure 5).

Metabolomic analysis has revealed that sepsis induces ferroptosis in vascular ECs, leading to elevated lipid peroxidation.[137] Recent studies have reported that glutathione peroxidase 4 (GPX4) and NRF2 are the primary factors involved in ferroptosis regulation, with GPX4 serving as an antioxidant enzyme that reduces lipid peroxidation and negatively modulates ferroptosis.[138,139] NRF2 is a crucial transcription factor that plays a pivotal role in regulating various cytoprotective genes, including *GPX4*.[140] Therefore, interventions targeting GPX4 and NRF2 may serve as effective approaches to ameliorate the ferroptosis of ECs in sepsis. In addition, research teams have confirmed that dexmedetomidine enhances EC resilience against ferroptosis in sepsis by upregulating the NRF2 signaling pathway, suppressing mitochondrial fission, and promoting metabolic reprogramming. Dex treatment reinforces cell–cell connections and improves barrier function, offering protective effects against sepsis-induced organ dysfunction.[137] The membrane protein six transmembrane epithelial antigen of the prostate 1 (STEAP1), which plays a pivotal role in cell adhesion and communication, has been implicated in ferroptosis. A sepsis-induced ALI model was used to demonstrate that STEAP1 expression was upregulated under septic conditions, and its inhibition reduced ROS production and inflammatory levels. Additionally, STEAP1 inhibition upregulated NRF2 expression, thereby attenuating EC ferroptosis, which may be mediated by the solute carrier family 7 member 11/GPX4 axis.[141]

In addition, the mutual regulation between cells is crucial for EC ferroptosis. Extracellular vesicles secreted by adipose-derived stem cells (ADSCs) can attenuate ferroptosis in sepsis-induced pulmonary microvascular ECs. Mechanistic experiments have confirmed that ADSC-derived extracellular vesicles specifically delivered miR-125b-5p to inhibit the target gene *kelch-like ECH-associated protein-1* (*KEAP1)*, resulting in increased expression of *NRF2* and *GPX4*. This process reduces oxidative stress levels in lung tissues and mitigates EC ferroptosis, thereby alleviating tissue damage.[142] Neutrophil extracellular traps (NETs) secreted by neutrophils play an important role in ALI. The component neutrophil elastase is significantly increased in the plasma of patients with sepsis. Moreover, NETs can induce damage in

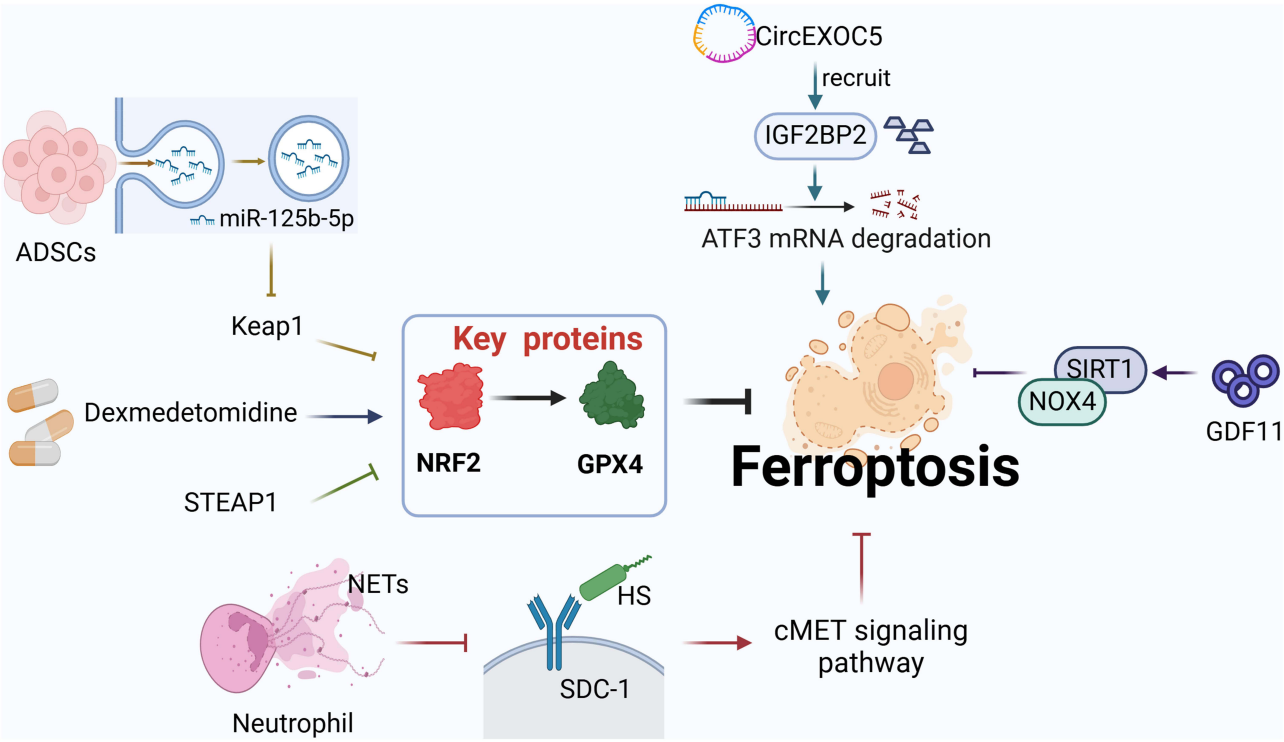


Figure 5 Overview of the mechanisms related to ferroptosis of ECs in sepsis. NRF2 and GPX4 act as the key proteins in ferroptosis. Exosomes derived from ADSCs, rich in miR-125-5p, can influence the expression of NRF2, alleviating ferroptosis. Dexmedetomidine and STEAP1 can also regulate ferroptosis via NRF2. NETs may promote ferroptosis by blocking the formation of the SDC-1/HS complex and interfering with the cMET signaling pathway. Moreover, circEXOC5 can recruit IGF2BP2 to induce the degradation of *ATF3* mRNA, inducing EC ferroptosis. GDF11 can inhibit ferroptosis by activating the SIRT1/ NOX4 pathway. Created in BioRender. Jiang, L. (2024) http://BioRender.com/p37f510.

the glycocalyx of ECs, disrupting the formation of the SDC-1/HS complex. This disruption inhibits the normal conduction of the mesenchymal-epithelial transition factor (cMET) signaling pathway, ultimately leading to EC ferroptosis and aggravation of the severity of ALI.[143]

Circular RNAs can also participate in the regulation of ferroptosis of ECs in sepsis. For example, CircEXOC5 is highly expressed in patients with sepsis and cecal ligation and puncture (CLP) mice. Its upregulation promotes ferroptosis of primary mouse pulmonary microvascular endothelial cells (MPMECs) by recruiting IGF2BP2 to degrade *activating transcription factor 3* (*Atf3*) mRNA, exacerbating sepsis-induced ALI.[144]

Inhibition of ferroptosis can also improve the barrier function of ECs. Overexpression of growth differentiation factor 11 (GDF11) can inhibit ferroptosis and reduce sepsis-induced pulmonary microvascular endothelial barrier injury by activating the sirtuin 1 (SIRT1)/NADPH oxidase 4 (NOX4) signaling pathway, providing a new molecular target for the clinical treatment of ALI.[145] Thus, the alleviation of EC ferroptosis represents a promising protective strategy against sepsis.

Autophagy of ECs in Sepsis

Autophagy is a form of RCD that mechanistically depends on the autophagic machinery and its components, according to the NCCD.[93] As a highly conserved catabolic process, autophagy can eliminate cellular metabolic waste and maintain overall balance and homeostasis. In addition, the autophagy pathway can actively remove intracellular microorganisms and prevent microbial invasion by regulating inflammation and immunity, especially during sepsis.[146] Thus, autophagy, which serves as an important self-protection mechanism for cell survival, may be a promising therapeutic target for reversing sepsis-induced immunosuppression.[147] Recent studies related to sepsis have increasingly focused on the autophagy of ECs. Notably, these studies have demonstrated that the role of autophagy in sepsis may be contradictory.[148,149] Therefore, this section mainly summarizes the autophagy and functional effects of ECs in the sepsis environment based on recent high-quality studies to further explain the complex mechanism of autophagy in sepsis (Figure 6).

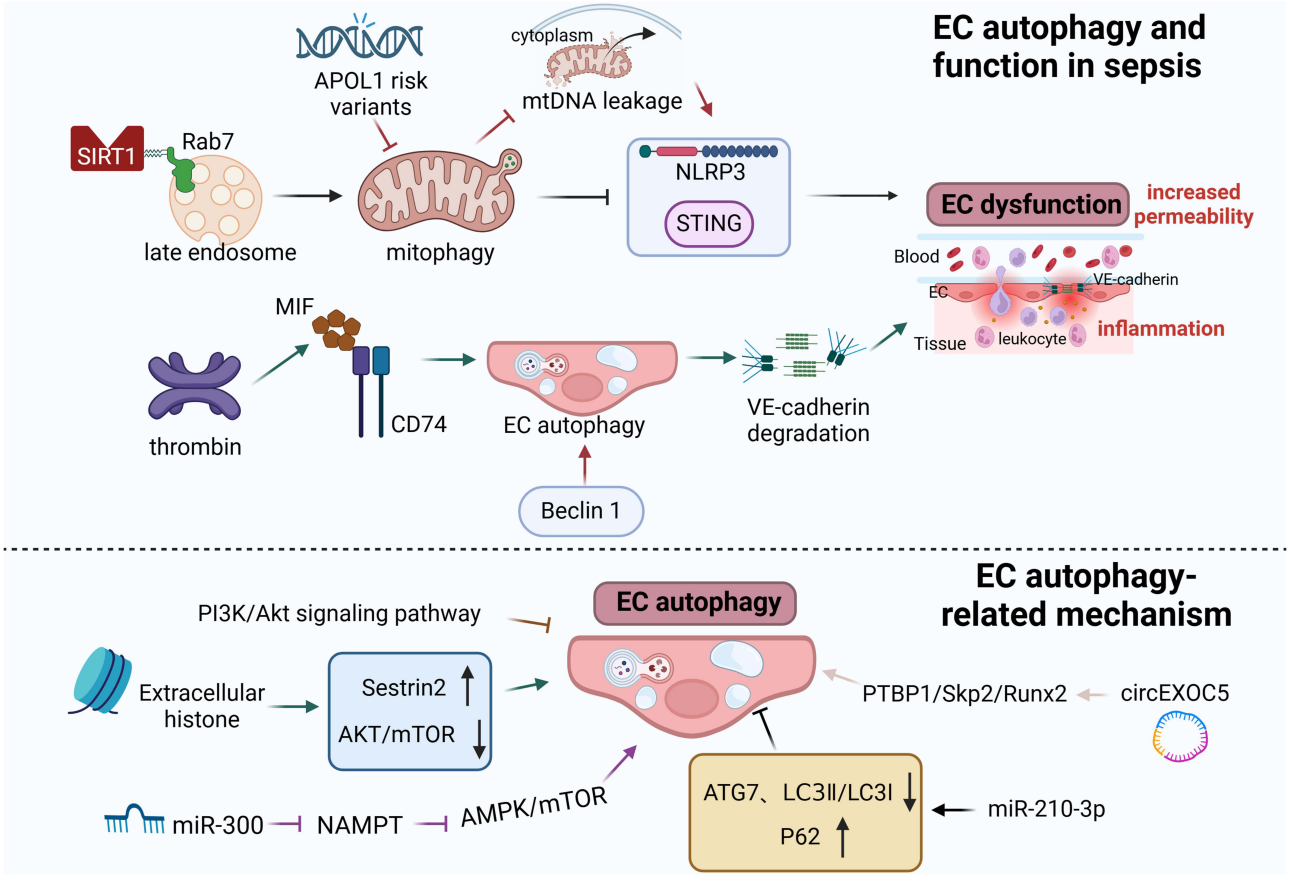


Figure 6 The relationship of EC autophagy and function and EC autophagy-related mechanisms in sepsis. The impact of EC autophagy on its function is controversial. Late endosomal-dependent mitophagy induced by the SIRT1-Rab7 axis can attenuate NLRP3 and STING activation, thereby improving EC dysfunction, including inflammation and barrier function. *APOL1* risk variants can inhibit mitophagy, leading to the leakage of mtDNA into the cytoplasm, activation of NLRP3 and STING, and ultimately resulting in EC dysfunction. However, EC autophagy activated by thrombin or beclin 1 increases the degradation of VE-cadherin, which is important in maintaining EC barrier function. Moreover, EC autophagy-related mechanisms are mainly involved in the mTOR signaling pathway. Non-coding RNAs, such as circEXOC5, miR-210-3p, and miR-300, can also regulate EC autophagy. Created in BioRender. Jiang, L. (2024) http://BioRender.com/r00x283.

Autophagy deficiency will increase the susceptibility of the body to microbial infection, while enhanced autophagy may play an important role in improving inflammation and EC function. LC3B is a key autophagy protein involved in the formation and maturation of autophagosomes. Compared to wild-type normal mice, *Lc3b*-deficient mice exhibit higher mortality and more severe tissue and organ damage after CLP modeling. Moreover, these mice also exhibited increased bacterial counts in various organs, indicating that autophagy deficiency can lead to impaired bacterial clearance and resistance to polymicrobial sepsis.[148] In the study of sepsis-associated encephalopathy, promoting mitochondrial autophagy of cerebral microvascular ECs can block the activation of NLRP3 inflammasome, inhibit the secretion of IL-1β to the central nervous system, reduce neuroinflammation, and improve cognitive dysfunction.[150] In sepsis-induced ALI, mitochondrial dysfunction is considered an important pathogenesis. One study identified a late endosome-dependent mitochondrial autophagy. SIRT1, a histone deacetylase, can interact with Rab7 on late endosomes to promote mitochondrial autophagy and limit the excessive activation of STING and NLRP3 in lung ECs, thereby playing a protective role.[151] Moreover, treatment with the drug hyperoside can reduce LPS-induced damage to human lung microvascular ECs (HLMVECs) by activating autophagy-related (Atg) 13-mediated autophagy.[152] In AKI caused by sepsis, the autophagy activator rapamycin can further enhance sepsis-induced autophagy in renal ECs and reduce renal endothelial injury and AKI. This finding supports the hypothesis that upregulation of autophagy in renal ECs may provide therapeutic benefits in sepsis-induced AKI.[153] Notably, the occurrence and severity of sepsis are closely associated with race. For example, the incidence and severity of sepsis in individuals of African ancestry are higher than those of European ancestry, and only individuals of African ancestry have genetic risk variants (RVs) in the *trypanolytic factor*

apolipoprotein L1 (*APOL1*). Moreover, RV *APOL1* can mediate mitochondrial autophagy defects in ECs, leading to the leakage of mitochondrial DNA (mtDNA) into the cytoplasm and activation of the NLRP3 and STING, resulting in an inflammatory and pro-adhesive endothelial phenotype that adversely affects sepsis.[154]

In addition, numerous studies support that autophagy activation can aggravate sepsis and EC dysfunction. Autophagy activation can participate in impairing the endothelial barrier. In a study using the human microvascular endothelial cell line-1 and HUVEC, thrombin-induced macrophage migration inhibitory factor (MIF) secretion was found to promote autophagy by binding to CD74. This interaction leads to the disorganization and degradation of EC VE-cadherin, resulting in high permeability.[155] In contrast, the autophagy inhibitor hydroxychloroquine can significantly reduce LPS-induced endothelial hyperpermeability.[156] Beclin1, a key regulator of autophagy, has recently been found to serve as a new mechanistic link between autophagy and EC dysfunction, including inflammation and permeability. Silencing of *Beclin1* can downregulate the expression of pro-inflammatory genes, such as *NF-κB*, in ECs in response to thrombin, while also preventing the loss of VE-cadherin, thereby protecting the EC barrier function.[157]

Based on current research, there is considerable controversy regarding the role of autophagy in sepsis. A comprehensive consideration of the occurrence and development process suggests that, in the early stage of infection, autophagy may be beneficial as it assists in clearance, resistance, and cell survival.[158] However, in the later stage of infection, continuous activation of autophagy may lead to uncontrolled vascular leakage and excessive inflammation.[159] From this perspective, the timing of autophagy regulation is crucial for the effective treatment of sepsis.

Finally, the review summarizes the important autophagy-related signaling pathways. mTOR is an important signaling pathway involved in the autophagy process. Dl-3-n-butylphthalide, a drug that can improve multi-organ ischemic diseases, can alleviate the autophagy of microvascular ECs in CLP rats and improve their intestinal microcirculation disorders. This effect is achieved through the activation of the PI3K/Akt signaling pathway, which usually serves as the upstream regulator of mTOR.[160] Extracellular histones can induce autophagy in HUVECs by upregulating Sestrin2 and inhibiting the AKT/mTOR signaling pathway.[102] There is also evidence indicating that in a sepsis mouse model, the overexpression of miR-300 can enhance autophagy by targeting nicotinamide phosphoribosyltransferase (NAMPT) and activating the AMP-activated protein kinase (AMPK)/mTOR signaling pathway, thereby inhibiting apoptosis and mitigating inflammatory responses.[161] Non-coding RNAs can also participate in the regulation of autophagy. For example, circEXOC5 is implicated in the ferroptosis process of ECs during sepsis-induced ALI.[134] In addition, the upregulation of circEXOC5 is associated with increased inflammation and activated autophagy in ECs. This upregulation can promote inflammation and autophagy in MPMECs through the polypyrimidine tract binding protein 1 (PTBP1)/ S-Phase kinase associated protein 2 (Skp2)/ RUNX family transcription factor 2 (Runx2) signaling cascade and aggravate sepsis-induced ALI.[149] Conversely, miR-210-3p can decrease the expression of ATG7 and LC3II/LC3I and increase the expression of P62 in HLMVECs, thereby inhibiting autophagy.[162]

Understanding the pathogenesis of sepsis, particularly RCD, poses a significant challenge in critical care medicine. RCD is a dynamic response of the body to internal and external environmental changes. The specific type of cell death depends on the microenvironment and the molecular mechanisms activated in response to stress. In addition to the RCD forms discussed above—including apoptosis, pyroptosis, ferroptosis, and autophagy—other forms of RCD, such as necroptosis, also participate in the development of sepsis.[163,164] However, owing to the limitations in existing literature, we did not conduct a further review of these RCD forms. Different forms of cell death involve distinct pathways; however, they are also interconnected,[93] co-occurring and synergistically contributing to sepsis progression.[102,165,166] Thus, recognizing that the different forms of RCD can counterbalance each other is essential. For instance, in an in vitro sepsis model using LPS-stimulated human umbilical vein ECs, an intervention with rapamycin to enhance autophagy was found to reduce the expressions of pyroptosis-related proteins, such as GSDMD and caspase-1, and decrease the secretions of IL-1β and IL-18, thereby improving the inflammatory microenvironment of sepsis and providing a potential therapeutic approach for sepsis treatment.[167] Furthermore, it must be acknowledged that RCD is a double-edged sword. Moderate cell death can remove damaged cells and facilitate tissue renewal, thus exerting a protective effect on the body. However, excessive apoptosis, pyroptosis, ferroptosis, or autophagy can significantly impair EC function, thereby compromising cell barrier integrity and vitality and contributing to the development of various diseases.

At present, the accumulated evidence enhances our understanding of the role of RCD in EC biology and sepsis-induced dysfunction. However, this evidence is predominately based on in vitro cell culture studies, primarily involving various ECs stimulated by LPS. In vivo studies have mainly focused on CLP or LPS-stimulated sepsis models, which still vary from real clinical scenarios of sepsis. Therefore, the current treatment strategies targeting RCD of ECs during sepsis remain largely hypothetical. Although the regulation of RCD by many commonly used RCD regulators, such as ferrostatin-1, rapamycin, and chloroquine, has been elucidated, their broad-spectrum effects in vivo limit their clinical applicability. Nevertheless, emerging drugs and interventions for regulating RCD in sepsis are also being developed and tested. For example, QiShenYiQi pills have been proven to improve pulmonary diseases clinically; however, their role in sepsis remains unknown. One study suggested that QiShen YiQi pills may inhibit ferroptosis in ECs by inhibiting the RAGE pathway and the target cyclooxygenase-2, thereby maintaining the integrity of the pulmonary vascular barrier in CLP mice. Therefore, these pills could be a promising option for treating sepsis-induced ALI.[168] Another study has indicated that hepatocyte growth factor (HGF) can partially improve pyroptosis in ECs and alleviate vascular endothelial injury in septic animals by activating the mTOR signaling pathway, thereby protecting mitochondrial physiological functions and alleviating ALI.[169] Finally, new treatment methods should be further verified in various preclinical models. Future research and discoveries of sepsis-related pathways will aid in identifying other therapeutic targets.

Conclusion

EC dysfunction plays a critical role in sepsis development and leads to organ dysfunction. The primary manifestations of endothelial damage include barrier dysfunction and various forms of RCD. Barrier dysfunction can promote white blood cell migration and enhance inflammatory responses. A heightened inflammatory response can increase the risk of EC death through complex mechanisms, thereby hindering the repair of sepsis-induced damage. Through a systematic literature review, we identified multiple factors contributing to barrier dysfunction in ECs, including GCX degradation and shedding and the release of various DAMPs. RCD in ECs, such as apoptosis, pyroptosis, ferroptosis, and autophagy, is closely linked to the high levels of inflammation and oxidative stress induced by sepsis. Thus, investigating the interconnections and regulatory relationships between various modes of cell death is crucial. With the recent advancements in single-cell sequencing and multi-omics technologies, further exploration of the pathological molecular mechanisms underlying endothelial dysfunction in sepsis, as well as identifying effective treatment targets and developing specific small-molecule drugs, remain a focal point for future research.

Abbreviations

ADAM15, a disintegrin and metalloproteinase 15; ADSC, adipose-derived stem cell; AKI, acute kidney injury; AKT, protein kinase B; ALI:acute lung injury; AMPK, AMP-activated protein kinase; Ang1, angiopoietin-1; APOL1, apolipoprotein L1; ARDS, acute respiratory distress syndrome; ATF3, activating transcription factor 3; ATG7, autophagy-related 7; ceRNA, competing endogenous RNA; cGAS, cyclic GMP-AMP synthase; CLP, cecal ligation and puncture; cMET, the mesenchymal-epithelial transition factor; DAMPs, damage-associated molecular patterns; DIC, disseminated intravascular coagulation; eCIRP, extracellular cold-inducible RNA-binding protein; ECs, endothelial cells; ER, endoplasmic reticulum; GAB1, GRB2 associated binding protein 1; GCX, glycocalyx; GDF11, growth differentiation factor 11; GPX4, glutathione peroxidase 4; GSDMD, gasdermin D; GSDMD-NT, gasdermin D N-terminal fragment; HA, hyaluronic acid; HIF-1α, hypoxia-inducible factor-1α; HLMVEC, human lung microvascular ECs; HMGB1, high mobility group box 1; HNP, human neutrophil peptides; HO-1, heme oxygenase-1; HPCAECs, human primary coronary artery ECs; HPMECs, human pulmonary microvascular endothelial cells; HPSE, heparanase; HS, heparan sulfate; HUVECs, human umbilical vein endothelial cells; ICAM1, intercellular adhesion molecule 1; IFN, interferon; IL-1β, interleukin-1β; ILC2, group 2 innate lymphoid cells; iNOS, inducible nitric oxide synthase; IRE1α, inositol requiring enzyme-1α; KEAP1, kelch-like ECH-associated protein-1; lncRNAs, long non-coding RNAs; LPS, lipopolysaccharide; MAPK, mitogen-activated protein kinase; MD2, myeloid differentiation protein-2; MIF, macrophage migration inhibitory factor; miRNAs, microRNAs; MPMECs, mouse pulmonary microvascular endothelial cells; mtDNA, mitochondrial DNA; mTOR, mammalian target of rapamycin; NAMPT, nicotinamide phosphoribosyltransferase; NCCD, Nomenclature Committee on Cell Death; NET, neutrophil extracellular traps; NF-κB, nuclear factor kappa-B; NLRP3, nucleotide-

binding domain, leucine-rich repeat-containing family, pyrin domain-containing 3; NLRX1, NLR family member X1; NO, nitric oxide; NOX4, NADPH oxidase 4; NRF2, nuclear factor erythroid 2-related factor 2; NRP2, neuropilin 2; O-GlcNAc, O-linked β-N-acetylglucosamine; Ox-HMGB1, oxidized HMGB1; P2X7, P2X ligand-gated ion channel 7; PAMPs, pathogen-associated molecular patterns; PECAM-1, platelet endothelial cell adhesion molecule-1; PI3K, phosphoinositide-3-kinase; PTBP1, polypyrimidine tract binding protein 1; PTMs, post-translational modifications; RAGE, receptor for advanced glycation end-products; RCD, regulated cell death; ROCK1, Rho-associated coiled-coil containing protein kinase 1; ROS, reactive oxygen species; Runx2, RUNX family transcription factor 2; SDC-1, syndecan-1; SERP1, stress-associated endoplasmic reticulum protein 1; SIRT1, sirtuin 1; Skp2, S-Phase kinase associated protein 2; SLIT2, slit guidance ligand 2; SSH1, slingshot protein phosphatase 1; ST2, suppression of tumorigenicity 2; STAT, signal transducer and activator of transcription; STEAP1, six transmembrane epithelial antigen of the prostate 1; STING, stimulator of interferon genes; TJ, tight junction; TLR4, toll-like receptor 4; TNF-α, tumor necrosis factor-alpha; TUG1, taurine-upregulated gene 1; TXNIP, thioredoxin-interacting protein; VCAM1, vascular cell adhesion molecule 1; VE-cadherin, vascular endothelial cadherin; YAP, yes-associated protein; ZO-1, zona occludens-1.

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Author Contributions

All authors made a significant contribution to the work reported, including the conception, study design, execution, acquisition of data, analysis, and interpretation. They all participated in drafting, revising, or critically reviewing the article; provided final approval of the version to be published; and agreed to be accountable for all aspects of the work.

Disclosure

The authors report no conflicts of interest in this work.

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