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Original Article

Transcription factor-mediated signaling pathways' contribution to the pathology of acute lung injury and acute respiratory distress syndrome

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Abstract: The 2019 novel coronavirus (2019-nCoV) is still spreading rapidly around the world, and one cause of lethality for patients infected with 2019-nCoV is acute respiratory distress syndrome (ARDS). ARDS is a severe syndrome of acute lung injury (ALI) that is predominantly triggered by inflammation and results in a sudden loss of, or damage to, kidney function. Emerging studies reveal that multiple transcription factor-associated signaling pathways are activated in the pathology of ALI/ARDS. Of these pathways, the activation of NF-κB (nuclear factor kappa-light-chain-enhancer of activated B cells), AP-1 (activator protein 1), IRFs (interferon regulatory factors), STATs (signal transducer and activator of transcription), Wnt/β-catenin-TCF/LEF (T-cell factor/lymphoid enhancer-binding factor), and CtBP2 (C-Terminal binding protein 2)-associated transcriptional complex contributes to ALI/ARDS pathology through diverse mechanisms, such as inducing proinflammatory cytokine levels and mediating macrophage polarization. In this review, we present an updated summary of the mechanisms underlying these signaling activations and regulations, as well as their contribution to the pathogenesis of ALI/ARDS. We aim to develop a better understanding of how ALI/ARDS occurs and improve ALI/ARDS therapy.

Keywords: Inflammation, ALI, NF-κB, AP-1, Wnt/β-catenin, transcriptional regulation

Introduction

The lungs are the major pathological target of many bacteria and viruses (e.g., SARS coronavirus [SARS-CoV] and the 2019 novel coronavirus [2019-nCoV]) [1, 2]. After infection, such bacteria and viruses often cause respiratory syndrome due to acute lung injury (ALI) or its severe form-acute respiratory distress syndrome (ARDS) [1, 2]. ALI and ARDS are disorders characterized by rapid dysfunction and damage of the lung within a short period (several hours to days) [3, 4]. In the past several decades, scientists have made great efforts to investigate the pathological mechanisms of ALI/ARDS [5-9]. Experimental and clinical evidence has revealed that inflammation is an important trigger in the occurrence of ALI/ARDS [5-9].

An inflammation response is a process in which innate immune cells respond to external infection or tissue injury [10, 11]. After infection or lung injury, multiple cell types-including resident neutrophils, macrophages, and dendritic cells-as well as circulating monocytes can initiate an immune response [5-11]. The local lung injury, or a systemic injury in the extrapulmonary site, can affect the functions of the vascular endothelium (VE), bronchial epithelium (BE), and alveolar macrophages (AMs), resulting in an accumulation of edema fluid in the alveoli that can, in turn, cause hypoxemia, a gas barrier disorder [12, 13]. Of these cell types, resident AMs are dominant in mediating inflammation response and the resolution of ALI/ARDS [12, 13]. Based on the polarization of macrophages, they can be classified into two major types: M1 (the classically activated phenotype) and M2 (an alternatively activated phenotype) [14]. When ALI/ARDS occurs, resident AMs are activated, becoming M1-type to initiate a broad range of signaling pathways and perpetuate an

inflammation response that results in the up-regulation of proinflammatory cytokines such as interleukin 1 beta (IL-1β), IL-6, IL-8, and tumor necrosis factor-alpha (TNF-α) [15-17]. In late-phase ALI/ARDS, the activated AMs shift to M2-type macrophages, which can eliminate apoptotic cells and contribute to lung fibrosis [15-17].

Transcription factors' contribution to ALI/ARDS pathogenesis

Immune cells such as macrophages and helper T cells secrete proinflammatory cytokines, which play a dominant role in the pathogenesis of inflammatory diseases including ALI/ARDS [18, 19]. Proinflammatory cytokines mainly include IL-1β, IL-6, IL-8, IL-12, IL-18, TNF-α, interferon gamma (IFN-γ), and granulocyte-macrophage colony-stimulating factor (GMCSF) [20]. Their expression is mainly controlled by transcription factors such as NF-κB (nuclear factor kappa-light-chain-enhancer of activated B cells), AP-1 (activator protein 1), IRFs (interferon regulatory factors), and TCF/LEF (T-cell factor/lymphoid enhancer-binding factor) [21]. Mechanically, PAMPs (pathogen-associated molecular patterns) (e.g., lipopolysaccharides [LPS], endotoxins), and DAMPs (damage-associated molecular patterns) (e.g., heat-shock proteins and HMGB1 [high-mobility group box 1]) are recognized by PRRs (pattern recognition receptors) (e.g., TLRs [Toll-like receptors], mannose receptors, and nucleotide-binding oligomerization domain-like receptors) [22]. PRRs further trigger intracellular signaling cascades and, ultimately, activate transcription factors in order to translocate into the nucleus and induce the expression of a variety of proinflammatory cytokines [22]. The extracellular secretion of matured proinflammatory cytokines causes inflammation, eventually leading to the pathogenesis inflammatory diseases [21, 22]. Similarly, the published results suggest that the induction of proinflammatory cytokines in ALI/ARDS pathogenesis is predominantly controlled by transcription factors. In addition to the direct activation of proinflammatory cytokines, transcription factors also contribute to regulate the expression of some important genes and miRNAs in ALI/ARDS pathogenesis. We summarize their contributions in the following sections of this review.

NF-κB signaling and ALI/ARDS

NF-κB family transcription factors include p65 (also known as "RelA"), RelB, c-Rel, p50 (also known as "NF-κB1"), and p52 (also known as "NF-κB2"), which can assemble as different heterodimers and homodimers and bind to a specific DNA element (5'-GGGRNYYYCC-3'; R represents a purine, Y represents a pyrimidine, and N represents any nucleotide) of their target genes [23]. NF-κB activation is mainly controlled by two pathways: canonical and non-canonical [23-25]. In the canonical pathway, the receptors (e.g., TLR4, IL1 receptor [IL-1R] and TNF receptor [TNFR]) on the cellular membrane sense different stimuli and recruit the intracellular adaptor proteins TIRAP (TIR domain-containing adaptor protein) and TRAM (translocation-associated membrane protein) [23-25]. They can further activate a cascade to, in turn, activate the IKK (IκB kinase) complex. The activation of IKKs phosphorylates IκB results in the degradation of phosphorylated-IκB by proteases [23-25]. The degraded IκB eliminates its inhibition of NF-κB and causes the translocation of NF-κB (especially the p50/p65 heterodimer) from the cytoplasm to the nucleus, where it induces the expression of genes including proinflammatory cytokines [23-25]. In the non-canonical pathway, receptors such as BAFFR (B-cell activating factor), LTβR (lymphotoxin-β receptor), and RANK (receptor activator for nuclear factor-kappa B) initiate signaling cascades and cause the activation of NIK (NF-κB-inducing kinase) [23-25]. The activated NIK promotes the phosphorylation of IKKα, inducing the processing of p100 into the active RelB-p52 isoform, which translocates into the nucleus to regulate gene expression [23-25].

In ARDS subjects, the activation of TLR4-dependent canonical signaling has been observed to contribute to increased levels of proinflammatory cytokines such as IL-1β, TNF-α, IL-6, IL-8, and IL-18. Elevated levels of these cytokines have been observed in both bronchoalveolar lavage fluid and circulating plasma [26-28]. Mechanically, TLR4 activation recruits MyD88 (myeloid differentiation primary response 88) and TIRAP, IRAKs (IL-1 receptor-associated kinases), and TRAF6 (TNF receptor-associated factor 6) to activate IKKs, causing IκB to dissociate from the IκB-NF-κB complex

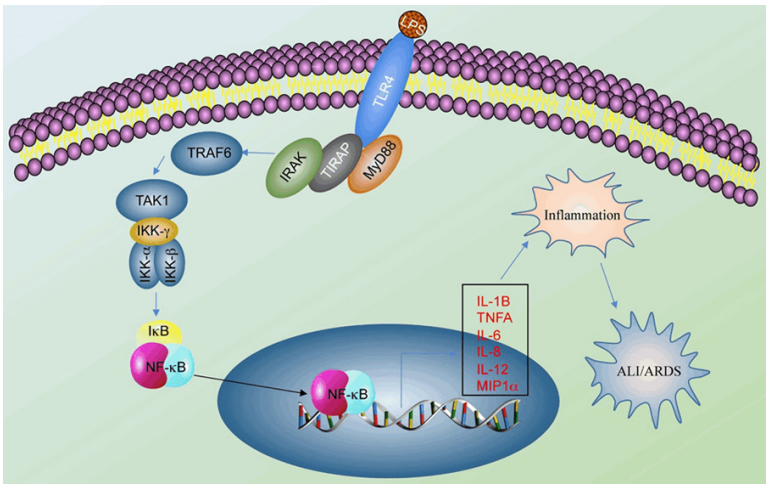

Figure 1. TLR4/NF-κB signaling's contribution to ALI/ARDS pathogenesis. The stimulation of pathologic stimuli such as LPS activates canonical TLR4/NF-κB signaling. TLR4 recruits MyD88, TIRAP, IRAK, and TRAF6 to initiate a kinase cascade, including IKKs and IκB, causing the release of NF-κB and its subsequent translocation to the nucleus, where it induces the expression of genes including proinflammatory cytokines such as *IL-1B*, *IL-6*, *IL-8*, *IL-12*, *TNFA*, and *MIP-1α*. The elevated proinflammatory cytokines cause inflammation, eventually leading to ALI/ARDS pathology.

[26-28]. The released NF-κB subunits-including p50, p65, and c-Rel-translocate from the cytoplasm to the nucleus in order to activate the expression of proinflammatory cytokines (**Figure 1**) [26-28]. Interestingly, several studies have shown that the p50/p50 homodimer is dominant in the later phase of inflammation, and that it plays an inhibitory role in regulating proinflammatory cytokines but enhances the expression of anti-inflammatory cytokines such as IL-10 [29-31]. In an LPS-induced ARDS rat model, Resolvin D1 (an endogenous lipid mediator) can inhibit inflammation by activating p50/p50-mediated *COX-2* (Cyclooxygenase-2) and *PGD2* (Prostaglandin D2) expression [29]. In LPS-treated cells and in an LPS-injected ARDS mouse model, Pooladanda et al. also found that TLR4 activation can promote the cytoplasmic-to-nuclear translocation of NF-κB and HDAC3 (histone deacetylase 3), which coordinate to increase proinflammatory cytokines (including IL-1β, IL-6, IL-12, TNF-α, and TGF-β [transforming growth factor-beta]) as well as chemokines (including MIP-1α [macrophage inflammatory protein 1-alpha] and MIP-1β) but decrease levels of anti-inflammatory cytokines (including IL-4, IL-10, and IL-13) [32]. Nimbolide, a limonoid tetranortriterpenoid, exhibits HDAC-inhibitory activity by repressing the nuclear translocation of HDAC3 and NF-κB, abrogating LPS-induced proinflammatory cytokines and alleviating lung injury [32]. In addition, the NLRP3 (NLR family, pyrin domain containing 3) inflammasome has also been reported to play a pivotal role in causing sterile inflammation in ALI [33, 24]. The NLRP3 inflammasome comprises three components-NLRP3, ASC (apoptosis-associated speck-like protein containing a CARD), and procaspase-1, which can cleave pro-IL-1β and pro-IL-18 to generate the matured IL-1β and IL-18 [33, 34].

AP1-associated transcriptional regulation in ALI/ARDS

The AP1 transcription factor family proteins comprise seven members: three Jun proteins (JunB, c-Jun, and JunD) and four Fos proteins (FosB, c-Fos, Fra1, and Fra2) [35, 36]. These proteins bind to their target genes through two consensus nucleotide sequences, comprising the cAMP-response element (CRE, 5'-TGACGTCA-3') and the tetradecanoylphorbol acetate (TPA)-response element (TRE, 5'-TGAG/CTCA-3') [35, 36]. Similar to the activation of NF-κB signaling, AP1 is also activated by stress inducers, pathogens (bacteria and virus), and inflammatory cytokines [35, 36]. Upon different stimulations, TLR4 also triggers and recruits intracellular adaptor proteins to initiate a signaling cascade in which the TLR4/TRAF6 axis signaling molecules are the same as in NF-κB signaling [35-38]. TRAF6 activates TAK1 (transforming growth factor-activated kinase 1), which further triggers a MAPK (mitogen-activated protein kinase) cascade, including ERK (extracellular signal-regulated kinases), JNK (c-Jun N-terminal kinases), and p38, causing the activation and nuclear translocation of AP1 [35-38]. The activated AP1 binds to the promoters of proinflammatory cytokines to increase their expression [35-38]. The overlapped upstream signaling molecules of AP1 and NF-κB suggest that these two pathways can cooperate to regulate the expression of some proinflammatory cytokines [35-38].

AP1 activation is also involved in ALI/ARDS pathogenesis through transactivating proinflammatory cytokines and other genes that

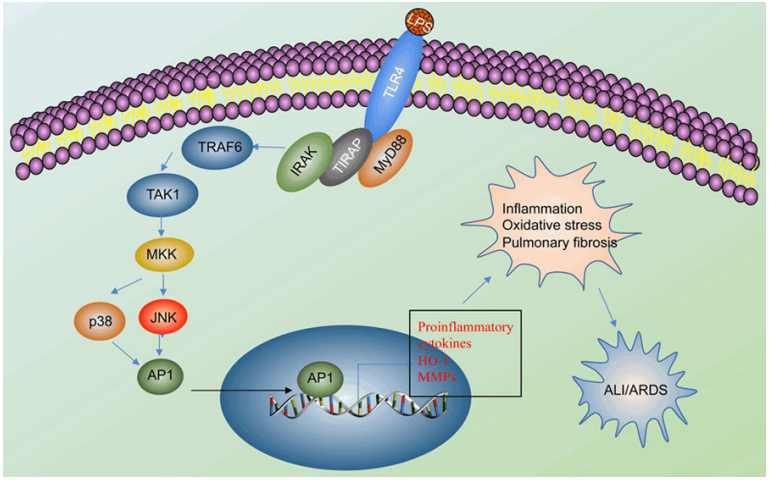

Figure 2. JNK/AP1 signaling's contribution to ALI/ARDS pathogenesis. Upon LPS stimulation, TLR4 recruits MyD88, TIRAP, IRAK, and TRAF6 to initiate a kinase cascade including TAK1, MKK, JNK (or p38) to activate AP1. The activated AP1 subsequently translocates into the nucleus, where it induces the expression of genes including proinflammatory cytokines, *HO-1*, and *MMPs*. The dysregulation of these genes results in inflammation, oxidative stress, and pulmonary fibrosis, eventually leading to ALI/ARDS pathology.

contribute to lung injury (**Figure 2**). In nuclear extracts from AMs and whole-lung tissues from ALI patients, AP1 subunits-including JunB, c-Jun, JunD, and c-FOS-are significantly abundant [39]. Depletion of the lung macrophage or treatment with a TNF-α inhibitor sharply decreases AP1 activation [39]. In a study evaluating AP1's role in LPS-induced ALI, Vaz et al. found that Fra-1 knockout (Fra-1$^{-/-}$) mice were much more tolerant of LPS administration than wild-type mice [40]. Fra-1$^{-/-}$ mice experienced decreased mortality, lung injury, and proinflammatory cytokine levels [40]. Heme oxygenase 1 (HO-1), a cytoprotective enzyme required for the lungs' defense against oxidative and inflammatory responses, is significantly upregulated in ARDS patients and rodent animal models [41]. The promoter of the *HO-1* gene contains an AP-1 consensus motif, which is required for *HO-1* activation in the treatment of LPS (**Figure 2**) [41]. Many patients with ALI/ARDS-and especially patients with COVID-2019 (coronavirus disease 2019)-have significant pulmonary fibrosis [2]. A group of genes known as "MMPs" (matrix metalloproteases) have been shown to contribute to the pathology of pulmonary fibrosis [42]. AP1 plays a critical role in regulating MMPs through directly binding to their promoters [42]. Fra-1$^{-/-}$ mice are much more susceptible to bleomycin-induced pulmonary fibrosis than wild-type mice, and AP1 is believed to function in pulmonary fibrosis through regulating MMPs (**Figure 2**) [43].

Interferon regulatory factors (IRFs) and signal transducer and activator of transcription (STATs)-mediated macrophage polarization in ALI/ARDS

As we mentioned earlier, macrophages can be polarized into M1 and M2 types, and the mechanisms of macrophage polarization have been well-characterized in inflammatory diseases [15-17]. Several transcription factors-such as IRFs, STATs (signal transducer and activator of transcription), NF-κB, and AP1-are involved in the regulation of macrophage polarization [44, 45]. In the M1 macrophage, different stimuli and signals from the microenvironment-such as IFN-LPS, and inflammatory cytokines-can activate STAT1, IRF5, and NF-κB as well as AP1, respectively, which induces proinflammatory cytokines [44, 45]. Specifically, STAT1 activation induces the expression of *IL-12*, *NOS2* (nitric oxide synthase 2), and *MHC II* (major histocompatibility complex II) [44-46]. IRF5 activation upregulates *TNF-α*, *IL-12*, and *IL-23* while the activation of both NF-κB and AP1 causes an increase in *TNF-α*, *IL-6*, and *NOS-2* [44-46]. IL-4 and IL-13–induced M2 macrophage polarization activates STAT6 and IRF4, leading to the induction of *Arg1* (Arginase 1), *Ym1* (also known as "*CHIA*", chitinase acidic), and *IL-10* [44-46].

Emerging evidence has suggested that the polarization of macrophages, including resident AMs and circulating macrophages, plays a critical role in ALI/ARDS pathogenesis [47]. Similarly, IRFs and STATs also contribute to the polarization of AMs in ALI/ARDS [47]. AMs' polarization is activated mainly via JAK (Janus kinase)/STAT1 signaling [47]. Once extracellular cytokines bind to their receptors (e.g., IFN-γ binds to its receptor, INFGR), they trigger the activation of JAK1 and JAK2, causing the phosphorylation of STAT1 [47, 48]. The activated STAT1 translocates to the nucleus, where it regulates the expression of *SOCS* (suppressor

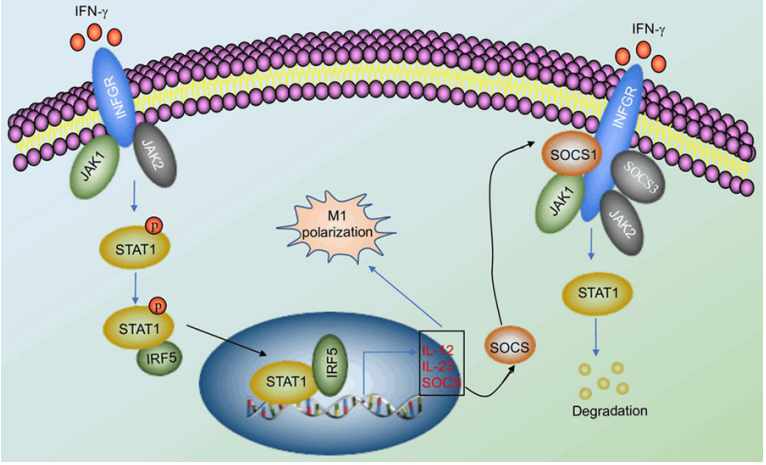

Figure 3. STAT1/IRF5 signaling's contribution to ALI/ARDS pathogenesis. The binding of IFN-γ to its receptor, INFGR, activates two kinases-JAK1 and JAK2-which phosphorylate STAT1. The phosphorylated STAT1 interacts with IRF5, and the complex further translocates into the nucleus, where it activates *IL-12*, *IL-23*, and *SOCS*. The upregulation of IL-12 and IL-23 promotes the M1 polarization of the macrophage. The increased SOCS1 and SOCS3 proteins bind to the phosphotyrosine residues of INFGR, causing the inactivation of JAKs, preventing the phosphorylation of STAT1, and inhibiting M1 macrophage polarization.

of cytokine signaling) genes (**Figure 3**) [49]. On the other hand, SOCSs also negatively mediate JAK/STAT1 signaling [49]. The cytoplasmic SOCS1 and SOCS3 proteins bind to the phosphotyrosine residues of intracellular cytokine receptors, blocking STAT1 from its docking sites (**Figure 3**) [49]. SOCSs' binding to the cytokine receptors inhibits JAKs' kinase activities, preventing the phosphorylation of STAT1 (**Figure 3**) [49]. As a result, SOCSs inhibit M1 macrophage polarization [49]. In the absence of SOCS3, IRF5 expression is increased, and it enhances M1 polarization by directly regulating IL-12 and IL-23 expression (**Figure 3**) [50]. Circulating leukocytes and monocytes can be recruited to the inflammation sites where they shift into the M1 macrophage [50]. In the later stage of ALI/ARDS, the M1 phenotype shifts to the M2 phenotype when pathogenic factors are eliminated, and this process is controlled by IRF4 and STAT6 transcription factors, which induce the expression of anti-inflammatory cytokines [47, 48].

Wnt/β-catenin-TCF/LEF signaling and ALI/ARDS

Wnt/β-catenin signaling is a conserved pathway to function in a variety of biological processes, such as embryonic development, axis patterning, stem cell pluripotency, cell fate specification, cell proliferation and migration, and inflammation [51, 52]. In the absence of Wnt, β-catenin is modified and degraded by a destruction complex, which comprises Axin, APC (adenomatosis polyposis coli), GSK3 (glycogen synthase kinase 3), PP2A (protein phosphatase 2A), and CK1α (casein kinase 1α). Once Wnt binds to its receptor, Frizzled, its coreceptors LRP5/6 (low-density lipoprotein receptor-related proteins 5 and 6) recruit the destruction complex to the membrane, where CK-1α and GSK3 phosphorylate LRP5/6, causing the accumulation of β-catenin in the cytoplasm [51, 52]. The accumulated β-catenin then translocates into the nucleus, where it interacts with TCF/LEF (T cell factor/lymphoid enhancer factor) to initiate gene transcription [51, 52]. The Wnt/β-catenin signaling cross-talks with the NF-κB pathway in inflammation response regulation [53, 54]. Current evidence has suggested that Wnt/β-catenin signaling plays a dominant anti-inflammatory role by repressing NF-κB activity [53, 54]. For example, in primary human MRC-5 lung fibroblast, β-catenin has been revealed to be able to affect RelA acetylation and, thus, repress the expression of NF-κB target genes such as *IL-1B* and *IL-6* [55]. In bacteria-colonized intestinal epithelial cells, overexpression of β-catenin causes a significant reduction in NF-κB levels as well as the expression of its target genes, *IL-6, IL-8*, and *TNFA* [56].

The activation of Wnt/β-Catenin signaling and a variety of its target genes has been observed in different pulmonary fibrosis models. In an idiopathic pulmonary fibrosis (IPF) mouse model and IPF patient, Wnt/β-Catenin signaling and its target gene, *WISP1* (WNT-inducible signaling protein 1), were activated [57]. Treatment with recombinant WISP1 in mouse alveolar epithelial type II cells can increase cell proliferation and epithelial-mesenchymal transition (EMT) [57]. Villar et al. found that Wnt/β-Catenin

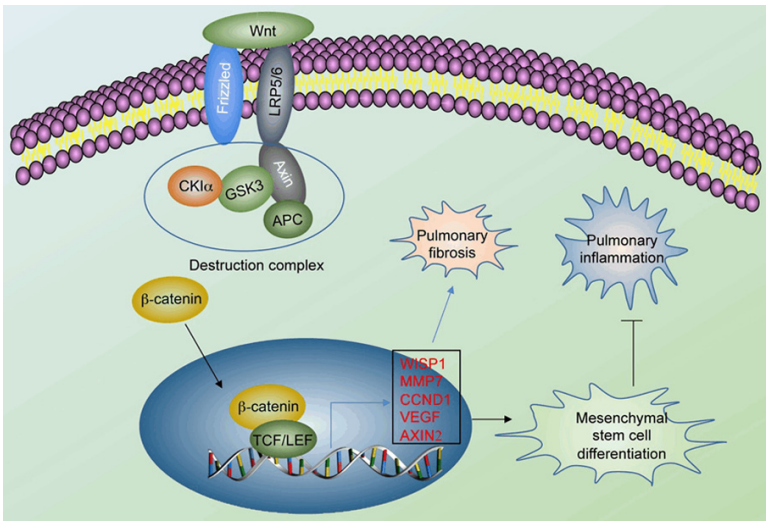

Figure 4. Wnt/β-catenin signaling's involvement in ALI/ARDS pathogenesis. The binding of Wnt to its receptor, Frizzled, and coreceptors LRP5/6 recruits the destruction complex, which comprises Axin, APC, GSK3, and CK1α. This recruitment causes the release of β-catenin from the destruction complex, and the accumulated β-catenin translocates into the nucleus to assemble a complex with TCF/LEF, which further activates the expression of *WISP1*, *MMP7*, *CCND1*, *VEGF*, and *AXIN2*. The increase in *WISP1* can induce pulmonary fibrosis. The upregulation of *MMP7*, *CCND1*, *VEGF*, and *AXIN2* contributes to a differentiation of MSCs, which inhibits pulmonary inflammation.

signaling was controlled by mechanical ventilation (MV) in the lungs, without preexistent lung disease [58]. This process significantly increases WNT5A and non-phosphorylated β-Catenin, as well as several Wnt target genes-including *MMP-7*, *CCND1* (Cyclin D1), *VEGF* (vascular endothelial growth factor A), and AXIN2 (Axin 2) (**Figure 4**) [58]. Mesenchymal stem cells (MSCs) are required for re-epithelization in ARDS [59]. Overexpression of β-catenin in MSCs promotes their differentiation into alveolar epithelial cells, improves LPS-induced lung permeability, attenuates pulmonary inflammation, and inhibits lung fibrosis [59]. In an LPS-induced mouse model, Cheng et al. found that Wnt/β-catenin promoted the occurrence of ALI through triggering a Th117 (T helper cell 17) response [60]. The activated Th17 response promoted neutrophils infiltration and proinflammatory cytokines production [60]. The overexpression of a circular RNA known as "circ0001434" was revealed to reduce LPS-induced lung inflammation through upregulation of Wnt/β-catenin, but with downregulation of NF-κB [61]. On the contrary, miRNA-1246 overexpression promotes an inflammation response through activating NF-κB signaling but repressing Wnt/β-catenin signaling [62].

Hypoxia-inducible factors (HIFs)-associated transcriptional regulation in ALI/ARDS

The lungs are oxygenated organs, and the activation of hypoxia signaling pathways is involved in ALI pathology [63]. Mammals have evolved precise mechanisms regulated by HIFs in response to hypoxia. The HIF family of transcription factors comprises three HIF-a subunits (HIF-1α, HIF-2α, and HIF-3α) induced by hypoxia and one constitutively expressed HIF-1β subunit [64]. In the hypoxia condition, HIF-1α is accumulated because the activities of prolyl-4-hydroxylases (PHDs) and factor inhibiting HIF-1 (FIH) are decreased [65]. The accumulated HIF-1α subunits translocate into the nucleus, where they dimerize with HIF-1β [64, 65]. The heterodimers bind to the hypoxia responsive-elements (HREs) of their target genes and initiate transcription [64, 65].

A few studies have found that the stabilization of HIFs can benefit lung protection in ALI/ARDS pathology [66, 67]. In LPS or SEB (staphylococcal enterotoxin B)-induced mouse models, high concentrations of oxygen cause a decreased survival rate within 48-60 hours after the injection of toxins because hyperoxia can inhibit the activation of the anti-inflammatory response mediated by the adenosine A2A receptor [68]. Several studies have endeavored to identify HIF targets during ALI, finding that *NT5E* (5'-Nucleotidase, also known as "CD73") [69], *Adora2a* (Adenosine A2a receptor), and *Adora2b* are direct HIF target genes (**Figure 5**) [70]. A deficiency of the Adora2a and Adora2b receptors has been shown to decrease survival but also increase inflammation in different experimental ALI models [68, 70]. The deletion of the Adora2b receptor increased proinflammatory cytokine levels and caused the activation of NF-κB in the hearts of septic mice, which implies that HIF-mediated signaling may crosstalk with other pathways involved in ALI/ARDS pathogenesis [71]. In addition, several studies have also indicated that hypoxic HIFs

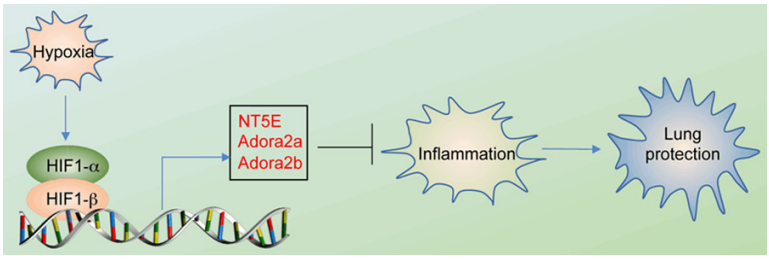

Figure 5. Hypoxia activates HIFs to protect against lung injury. Hypoxia activates the heterodimer of HIF1-α and HIF1-β, which activates the expression of *NT5E*, *Adora2a*, and *Adora2b*, whose induction inhibits the inflammation response, causing lung protection.

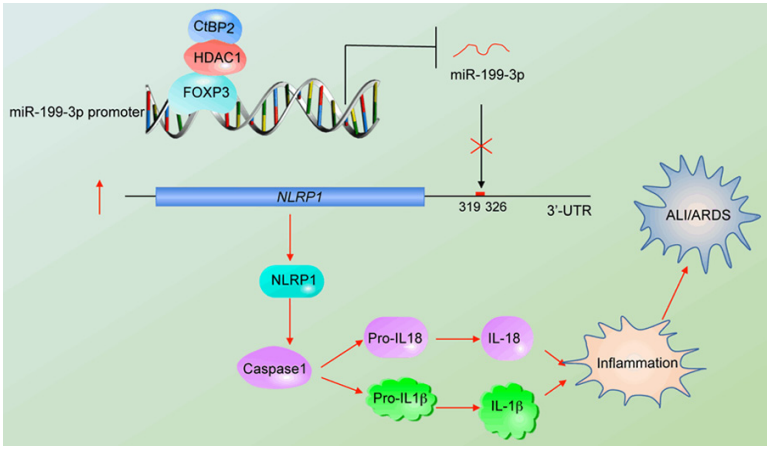

Figure 6. The CtBP2-HDAC1-FOXP3 complex's involvement in ALI/ARDS. The CtBP2-HDAC1-FOXP3 complex binds to the promoter of miR-199a-3p and represses its expression. The downregulated miR-199a-3p eliminates the inhibition of *NLRP1*. The functional NLRP1 inflammasome cleaves pro-IL-1β and pro-IL-18 to release their mature proteins, leading to an inflammation response and causing ALI/ARDS pathology.

activation acts as a critical regulator in the acute inflammatory response. In an HIF-1α conditional knockout mouse model, Suresh et al. identified hypoxic activation of HIF-1α in type-2 alveolar epithelial cells as a major regulator to drive inflammation following lung contusion (LC), and they found that the downregulation of HIF-1α could reverse lung injury and inflammation [72]. Several proinflammatory cytokines-including IL-1, IL-6, and MCP-5 (monocyte chemotactic protein 5)-significantly decreased following HIF-1α knockdown [72]. In a bleomycin-induced ALI model, HIF-1α has been shown to promote NLRP3 inflammasome activation [73].

C-terminal-binding proteins (CtBPs)-associated transcriptional regulation in ALI/ARDS

CtBPs are important transcription regulators in multiple biological processes, such as apoptosis, tumorigenesis, EMT, and cell cycle progression [74]. The mammalian genome contains two CtBP proteins: CtBP1 and CtBP2. They share a highly conserved protein sequence identity (nearly 80%) [74]. Mechanically, CtBPs function as both corepressors and coactivators to associate with histone enzymes (e.g., histone acetyltransferases p300/CBP, and histone deacetylases [HDACs]) and transcription factors (e.g., KLF3 [Krüppel-like factor 3], KLF8, KLF12, FOXP2 [Forkhead box protein P2], and FOXP3) to control gene expression [74, 75].

Using the lung tissues from ALI patients, we recently discovered that CtBP2 is overexpressed, and that it assembles a transcriptional complex with HDAC1 and FOXP3 to repress the expression of miR-199a-3p, which can target the 3'-UTR of *NLRP1* (**Figure 6**) [75]. The downregulation of miR-199a-3p causes the overexpression of *NLRP1*, which further assembles an inflammasome with Caspase-1 to cleave two immature proinflammatory cytokines, pro-IL-1β and pro-IL-18 (**Figure 6**) [75]. The secretion of mature IL-1β and IL-18 aggravates the inflammation response and causes ALI pathology (**Figure 6**) [75]. Recently, Li and colleagues found that CtBP1 and CtBP2 are involved in the induction of the inflammation response during traumatic brain injury (TBI) [76]. CtBPs can transactivate a series of proinflammatory cytokine genes, such as *IL1B*, *IL6*, and *TNFA* [76]. Although they did not reveal how CtBPs couple with other transcription regulators and transcription factors to transactivate these proinflammatory cytokine genes, we speculate that NF-κB is involved in this process because the CtBP-activated genes are NF-κB targets. These findings suggest that CtBPs may have direct and indirect roles in regulating proinflammatory genes and, thus, causing inflammation.

Summary and perspective

ALI/ARDS are seriously life-threatening syndromes with high mortality-especially currently, while COVID-2019 is rapidly spreading globally. A better understanding of ALI/ARDS pathology can help scientists and doctors develop therapeutic strategies to treat patients. Currently, our understanding of the mechanisms of ALI/ARDS has significantly improved. Widespread inflammation in the lungs is a major cause of ALI/ARDS. A variety of signaling pathways-especially transcription factor-dependent pathways-are activated to directly or indirectly induce the inflammation response in this process.

In this review, we have mainly discussed the contributions of six transcription factor-dependent pathways-comprising TLR4/NF-κB axis signaling, TLR4/AP1 axis signaling, IRFs and STATs-mediated signaling, Wnt/β-catenin-TCF/LEF axis signaling, HIFs-mediated signaling, and CtBPs-associated signaling-in the pathology of ALI/ARDS. Among these pathways, the activation of NF-κB and AP1 plays a dominant role in inducting an inflammation response through upregulating proinflammatory cytokine genes at the transcriptional levels [32, 40]. IRFs and STATs-mediated signaling pathways mainly function in promoting macrophage polarization [47-49]. Wnt/β-catenin-TCF/LEF signaling functions in the regulation of genes involved in pulmonary fibrosis [57-59]. Hypoxia induces HIFs stabilization, which can benefit lung protection. The CtBP2-HDAC1-FOXP3 transcriptional complex contributes to ALI pathogenesis through indirectly activating NLRP1 and promoting the cleavage of pro-IL-1β and pro-IL-18 [76]. Some of these pathways have crosslinks. For instance, the upstream signaling of both NF-κB and AP1 depend on the activation of a TLR4-initiating kinase cascade. In regulating the inflammation response, Wnt/β-catenin signaling plays an anti-inflammatory role by repressing NF-κB activity. Although significant progress has been achieved regarding the molecular mechanisms of ALI/ARDS in past decades, several key questions have not been answered well. First, the transcription regulator proteins that associate with transcription factors have not been investigated during ALI/ARDS pathology. The transcription machinery is assembled by transcription factors and other transcription regulators, such as corepressors and corepressors [76]; however, little is known about how transcription factors coordinate with other transcriptional regulators to regulate the expression of genes involved in ALI/ARDS pathology. Second, more efforts are required to investigate the crosstalk among different pathways. Proinflammatory cytokines are controlled by several transcription factors, such as NF-κB, AP1, and the CtBP2-associated transcription complex [32, 40, 76]. The inhibition of a single signaling pathway may not be sufficient to completely suppress inflammation. A better understanding of the crosstalk among these pathways would promote the establishment of effective therapeutic strategies. Third, investigating whether the other five types of transcription-mediated signaling are controlled by hypoxia is also necessary. The lungs are oxygenated organs, and their dysfunction causes ALI/ARDS. The cells in lung tissues are exposed in hypoxia conditions; thus, we speculate that hypoxia may also affect other transcription factors, in addition to HIFs.

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Disclosure of conflict of interest

None.

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