



# Molecular mechanisms and signaling pathways involved in immunopathological events of COVID-19

Ali Asghar Peyvandi<sup>1</sup>, Somayeh Niknazar<sup>1\*</sup>, Fatemeh Zare Mehrjardi<sup>2</sup>, Hojjat-Allah Abbaszadeh<sup>3,1,4</sup>, Shahrokh Khoshsirat<sup>1</sup>, Maryam Peyvandi<sup>1</sup>

1. Hearing Disorders Research Center, Loghman Hakim Hospital, Shahid Beheshti University of Medical Sciences, Tehran, Iran

2. Neurobiomedical Research Center, Shahid Sadoughi University of Medical Sciences, Yazd, Iran

3. Laser Application in Medical Sciences Research Center, Shahid Beheshti University of Medical Sciences, Tehran, Iran

4. Department of Anatomical Sciences and Biology, School of Medicine, Shahid Beheshti University of Medical Sciences, Tehran, Iran

## ABSTRACT

**Introduction:** COVID-19, a novel coronavirus that causes severe acute respiratory syndrome (SARS-CoV-2), is currently regarded as the most serious viral disease. During corona infection, viruses bind to host proteins and employ a variety of cellular pathways for their own purposes. Cell signaling is important for the regulation of cellular function. SARS-CoV-2 infection alters multiple signal transduction pathways that are critical for cell survival. The virus causes a severe and prolonged period of hypercytokinemia with misusing of these signaling cascades. Hyperactivation of the host immune system after infection with SARS-CoV-2 is the main cause of death in COVID-19 patients. Thus, to develop effective therapeutic approaches, it is necessary to first understand the problem and the underlying molecular pathways implicated in host immunological function/dysfunction. A number of intracellular signaling cascades have been implicated in infected cell pathways, including MAPK pathway, NF- $\kappa$ B pathway, JAK-STAT signaling pathway, PI3K/AKT/mTOR pathway and TLRI signaling cascades. Here, we have presented the molecular insights on the potential mechanisms involved in immunopathological events of COVID-19.

### Keywords:

SARS-CoV-2 Infection

Signal transduction pathways

Cytokine storm

## Introduction

Infectious diseases such as influenza, acquired immunodeficiency syndrome (AIDS), malaria and meningitis remain the leading causes of death in human populations worldwide (Morens et al., 2004). Humans are infected with a new coronavirus that causes serious pneumonia, which was recognized on 11 2020 by the WHO as coronavirus disease 2019 (COVID-19) (Lai et al., 2020). COVID-19 cause epidemic in all countries and rapidly increasing pandemics move (Gössling et al., 2020). It is

not the first outbreak of severe respiratory disease from coronavirus. Coronaviruses have caused three infectious diseases in only the past two decades, namely Middle East respiratory syndrome (MERS), severe acute respiratory syndrome (SARS) and COVID-19 (Rockx et al., 2020; Mahase, 2020).

Severe acute respiratory coronavirus syndrome 2 (SARS-CoV-2) is genetically related to SARS-CoV, the first pandemic threat of a new and fatal coronavirus that appeared at the end of 2002 and triggered an

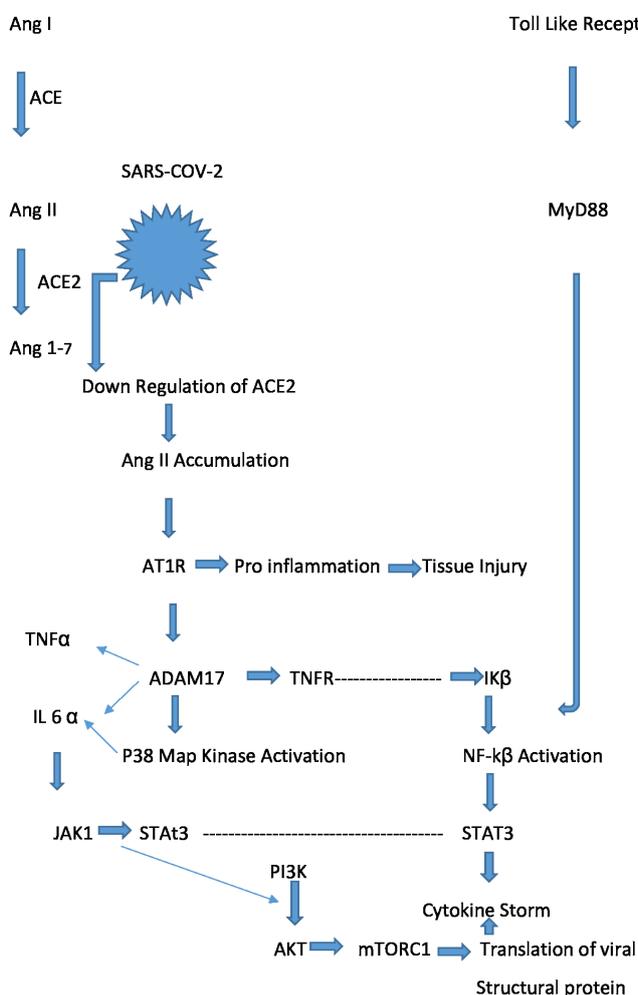
\* Corresponding author: Somayeh Niknazar, E-mail: [niknazar@sbmu.ac.ir](mailto:niknazar@sbmu.ac.ir)

Received 13 April 2021; Revised from 29 June 2021; Accepted 26 July 2021

Citation: Peyvandi AA, Niknazar S, Zare Mehrjardi F, Abbaszadeh H, Khoshsirat S, Peyvandi M. Molecular mechanisms and signaling pathways involved in immunopathological events of COVID-19. *Physiology and Pharmacology* 2021; 25: 193-205. <http://dx.doi.org/10.52547/ppj.25.3.193>

epidemic of SARS. SARS-CoV was extremely lethal but disappeared due to strong public health control (Petersen et al., 2020). According to a recent report, SARS-CoV-2 and SARS-CoV overlap about 80% of their genes (Gralinski and Menachery, 2020; Xu et al., 2020). Another analysis found a 96% similar sequence between SARS-CoV2 and the isolated CoV from *Rhinolophus affinis*, suggesting bats as a virus source (Xu et al., 2020). COVID-19 symptoms involve cough, fever, headache and experiencing shortness of breath. Furthermore, most COVID19 patients developed lymphopenia, with markedly elevated concentration of cytokines such as interleukin (IL)-1b and IL-6 (Prompetchara et al., 2020). COVID-19 uses the angiotensin-converting enzyme II (ACE2) as an entry receptor to infect lung alveolar epithelial cells (Velavan and Meyer, 2020). COVID-19 has the capacity to induce symptoms that range from common cold to acute respiratory distress syndrome (ARDS) (Liu et al., 2020b; Zimmermann and Curtis, 2020). In older COVID19 patients with one or more co-morbidities such as hypertension, diabetes

mellitus, cerebrovascular disease and chronic obstructive pulmonary disease, serious disabilities occur (Barr et al., 2009; Chen et al., 2015). Despite the increasing rate of SARS-CoV-2 transmission and death, no treatment has yet been developed. Studies have shown that viruses have developed a variety of highly sophisticated strategies that affect host cell transcription in purpose to replicate or to survive (Watanabe et al., 2010; Zuniga et al., 2008; Fernandez-Garcia et al., 2009). Extracellular signals regulate cellular homeostasis in multicellular organisms (Krajcsi and Wold, 1998). Several pathways are associated with the COVID-19 pathogenesis and a significant number of proteins are targeted by SARS-CoV-2 (Figure 1). Here, we focus on signaling pathways and molecular mechanisms that are involved in COVID-19 pathogenesis and manipulate host innate immune defenses such as cytokine response pathways. In addition, the study of the mechanisms involved in the pathogenesis of COVID-19 can aid scientists in developing treatments and vaccines that are effective in removing the morbidity and mortality in patients (Table 1).



**FIGURE 1.** Signaling pathways involved in COVID-19 pathophysiology. SARS-CoV-2 down-regulates ACE2 expression and result in production of proinflammatory cytokines and inflammatory mediators. Angiotensin-I is transformed into angiotensin-II by the action of ACE. ACE2 catalyzes Ang-II conversion to Ang (1-7), which promotes anti-inflammatory effects (Gheblawi et al., 2020). Since action of ACE2 is impaired during viral infection, Ang II cause chronic stimulation of AT1R. AT1R signaling induces p38 MAPK activation and inflammatory mediator’s production such as TNF and IL-6. The cytokines IL-6 and TNF bind to specific receptors and promote further NF-κB nuclear translocation and phosphorylation of p38 MAPK, which will result in cytokines storm (Feng et al., 2019; Grimes and Grimes, 2020). IL-6 activates JAK/STAT-3 pathway. Toll-like receptors identify SARS-CoV-2 RNA and trigger the inflammatory response through expression of interferon gene and NF-κB pathway (Battagello et al., 2020). PI3K activation lead to Akt phosphorylation and subsequent activation of mTOR. PI3K-Akt-mTOR pathway was also found to regulate cytokine production in COVID-19 (Ramaiah, 2020). ACE2, Angiotensin-converting enzyme II; Ang, Angiotensin; AT1R, Angiotensin II type 1 receptor; MAPK, Mitogen activated protein kinase; TNF, Tumor necrosis factor; IL, Interleukin; NF-κB, Nuclear factor kappa-B; JAK, Janus kinase; STAT, Signal transducer and activator of transcription; SARS, Severe acute respiratory syndrome; COVID-19, Coronavirus disease 2019; PI3K, Phosphatidylinositol-3-kinase; mTOR, Mammalian target of rapamycin.

**TABLE 1:** Potential treatment against COVID-19 disease.

Virus	Mechanism of modulation	Implications for therapy	Ref
SARS-CoV-2	MAPK Pathway	Silmitasertib (CK2 inhibitor)	(Bouhaddou et al., 2020)
		Ralimetinib (p38 inhibitor)	
		ARRY-797 (p38 inhibitor)	
		Losmapimod (p38 inhibitor)	
	NF-κB pathway	Dilmapimod (p38 inhibitor)	(Grimes and Grimes, 2020a)
		Artesunate (Inhibitor of NF-κB downregulation)	(Uzun and Toptas, 2020)
		Aspirin (Inhibition of ATP-binding to IKKβ)	
	JAK-STAT signaling	Sulfasalazine (Inhibitor of the NF-κB activation)	(Elkhodary, 2020)
		Ruxolitinib (JAK/STAT pathway inhibitor)	(Bagca and Avci, 2020)
		Baricitinib (a selective JAK1 and JAK2 inhibitor)	(Cingolani et al., 2020b)
Toll-like Receptor Signaling Pathway	Tocilizumab (IL6 inhibitor)	(Birra et al., 2020)	
	Anakinra (IL1 inhibitor)		
PI3K/AKT/mTOR pathway	Inhalation of buformin or phenformin	(Lehrer, 2020)	

*Research strategy*

The search for scientific papers was performed by researchers in the electronic databases, including Web of Science, Medline (PubMed) and Scopus. The initial search was carried out in the PubMed database based on the combinations of the following words: SARS-CoV-2, MAPK pathway, NF-κB pathway, JAK-STAT signaling, PI3K/AKT/mTOR pathway and TLRI signaling cascades, cytokine storm and immune defenses.

*Mitogen activated protein kinase (MAPK) pathway*

In response to certain environmental stimuli, MAPK signaling pathways are responsible for controlling several cell functions such as proliferation, differentiation and apoptosis (Cowan and Storey, 2003). The three main MAPK pathways in mammals are MAPK/extracellular signal-regulated kinase (ERK), Jun amino-terminal kinases/stress-activated protein kinases (JNK/SAPK) and p38 MAPK. Pro-inflammatory substances and environmental stimuli primarily activate the p38

MAPK pathway, which has a significant effect on a subset of physiological events such as immune response and inflammatory processes (Deak et al., 1998). Activation of the p38 pathway is required to increase the levels of pro-inflammatory cytokines such as IL-6, tumor necrosis factor (TNF) and IL-1, which appear to play critical roles in the cytokine storm induced by SARS-CoV-2 infection (Catanzaro et al., 2020). Indeed, the excessive immune reaction to COVID-19 infection may be triggered by overly up-regulated p38 activity, as two mechanisms have clarified. Activation of p38 MAPK has been involved in the ACE2 endocytosis (Xiao et al., 2013; Koka et al., 2008; Deshotels et al., 2014). First, the ACE2 activity is lost during SARS-CoV-2 viral entry. ACE2 inhibits related ACE activity by decreasing angiotensin-II and increasing angiotensin 1-7. The stimulation of angiotensin II type 1 receptor (AT1R) by angiotensin II leads to the activation of p38 MAPK and phosphorylation of A disintegrin and metalloprotease 17 (ADAM-17) (Xu and Derynck, 2010; Scott et al., 2011). Phosphorylation increases ADAM17’s catalytic

activity, resulting in increased ACE2 shedding and reduced conversion of angiotensin II into angiotensin 1–7, culminating in renin-angiotensin system (RAS)-mediated adverse consequences in a positive feedback cycle (Patel et al., 2014; Xu et al., 2017). Angiotensin 1-7 is critical for suppressing MAPK cascades and reducing inflammation (Zhang et al., 2014).

Angiotensin II promotes proinflammatory, pro-vasoconstrictive and pro-thrombotic activity through p38 MAPK activation, which is reversed by angiotensin 1-7 down-regulation of p38 activity. When ACE2 is lost during viral infection, it may shift the balance towards harmful p38 signaling via angiotensin II (Grimes and Grimes, 2020). ACE2 activity was found in both the lung and the heart. SARS-CoV-2 binds in the respiratory epithelium and lung alveoli to the same ACE2 receptor (Chen et al., 2020; Li et al., 2020). When a virus gets inside a cell, induce ACE2 shedding. Deficiency of ACE2 is associated with alveoli damage and increases permeability of the pulmonary vascular by angiotensin II (Li and De Clercq, 2020). Angiotensin II levels were directly linked with degree of lung injury and viral load in a study of COVID-19 patients, indicating RAS imbalance in COVID-19 etiology (Liu et al., 2020c).

Second, it has been previously shown that SARS-CoV directly up-regulates p38 activity through a viral protein, identical to several other RNA respiratory viruses that can hijack p38 activation to facilitate reproduction. Because SARS-CoV and SARS-CoV-2 are so similar, the latter may use a similar mechanism. As a result, SARS-CoV-2 might cause widespread inflammation by directly activating p38 and down-regulating a crucial inhibitory pathway, all while exploiting p38 activity to reproduce (Grimes and Grimes, 2020).

According to a report, permissive cell SARS-CoV-infection triggered the p38 MAPK signaling pathway. Up-regulation of the p38 MAPK pathway triggers the activation of IL-6, TNF- $\alpha$  and IL-1 pro-inflammatory cytokines (Zarubin and Jiahuai, 2005). MAPK-activated protein kinase-2 (one of the downstream effectors of p38 MAPK) is triggered in response to SARS-CoV-infection in Vero E6 cells (Foltz et al., 1997; Mizutani et al., 2004). Diffuse alveolar damage, which includes significant infection and viral load in type II pneumocytes, and also pulmonary edema, is the most common finding in COVID-19 postmortem tissue from all vital organs (Bradley et al., 2020; Carsana et al., 2020). CT-

scans with several ground glass opacities are common and have diagnostic value (Parekh et al., 2020). Angiotensin II levels are particularly high in ACE/angiotensin II receptor blocker naive COVID-19 cases and elevated concentrations are related to greater intensity (Liu et al., 2020a). Immune effector cells release massive amounts of pro-inflammatory cytokines and chemokines, lead to lethal uncontrolled systemic inflammation (Cameron et al., 2008; Channappanavar and Perlman, 2017; Huang et al., 2020a). Furthermore, because some COVID-19 patients have endothelial cell apoptosis, these biological effects could be linked to increased MAPK signaling activity (Grimes and Grimes, 2020; Zhou et al., 2020). The over-activation of p38 MAPK in infected cardiomyocytes, which has been demonstrated to cause apoptosis, impair contractility and promote fibrosis, could be part of the reason for cardiac dysfunction in COVID-19 patients (Grimes and Grimes, 2020). Another pathway involved in SARS-CoV infection is the c-jun NH<sub>2</sub>-terminal kinase (JNK) pathway, which may result in a rise in proinflammatory factors, as well as increased lung harm (Mizutani, 2010; Liu et al., 2014). JNK signaling pathway could be a target for SARS-CoV-2 as it includes proteins which are similar in both viruses. This mechanism induces ACE2 receptor binding and opened the way for COVID-19 virus internalization into the respiratory tract's alveolar epithelial cells. JNK signaling is implicated in the extrinsic and intrinsic apoptotic pathway, tissue cytokine production, inflammation and metabolism (Vellingiri et al., 2020).

Although the role of RAS in the pathophysiology of SARS-CoV-2 is still being explored, a recent study indicated that blocking RAS with ACE inhibitors or angiotensin receptor blockers may reduce overall mortality in COVID-19 patients (de Abajo et al., 2021). The specialized viral entrance mechanism of SARS-CoV-2 deactivates a key counterbalancing mechanism that the cell employs to reduce p38 signaling through ACE2 activation, which causes inflammation while also extending the viral lifespan. As a result, SARS-CoV-2 may cause excessive inflammation by directly activating p38 and downregulating a key inhibitory pathway, while also exploiting p38 activity to proliferate. COVID-19 infection could be reduced if p38 is suppressed medically. Losmapimod is the most researched p38 inhibitor in clinical trials and it has a good efficacy. Therefore, p38 MAPK inhibitor could be beneficial in patients with se-

rious COVID-19 health problems (Grimes and Grimes, 2020).

#### *Nuclear factor kappa-B (NF- $\kappa$ B) signaling pathway*

The NF- $\kappa$ B signaling pathway regulates a variety of essential genes in the innate and adaptive immune systems (Hoesel and Schmid, 2013; Liu et al., 2017). NF- $\kappa$ B signaling pathway plays a key role in gene expression involving cytokine/chemokine encoding and anti-apoptotic genes (Tak and Firestein, 2001; Gupta, 2003). NF- $\kappa$ B and its inhibitor (the inhibitory kappa B kinases, I $\kappa$ B) are present as a complex. Release from this complex requires I $\kappa$ B kinase (IKK) activation. The kinase complex of the IKK is the central element of the cascade of the NF- $\kappa$ B. Essentially, it consists of two kinases (IKK $\alpha$  and IKK $\beta$ ) and a regulatory sub-unit, NEMO (NF- $\kappa$ B essential modulator) /IKK $\gamma$  (Bonizzi and Karin, 2004). In most unstimulated cells, NF- $\kappa$ B dimers are kept inactive in the cytosol by interacting with I $\kappa$ B proteins (Oeckinghaus and Ghosh, 2009). After activation, the IKK complex will induce the phosphorylation of the I $\kappa$ B proteins leading to their degradation (Viatour et al., 2005). The degradation of these inhibitors by the IKK complex upon their phosphorylation resulting in the nuclear translocation of NF- $\kappa$ B and the induction of target gene transcription (Magnani et al., 2000). In other types of cell, including mature B cells, macrophages as well as a significant number of tumor cells, NF- $\kappa$ B may also be recognized as a nuclear protein which is constitutively active (Oeckinghaus and Ghosh, 2009).

During a virus infection, the NF- $\kappa$ B signaling pathway is activated and the gene expression of interferon beta (IFN- $\beta$ )/ TNF $\alpha$ / IL8 are increased (Pfeffer, 2011; Liu et al., 2017), suggesting that IKK-mediated NF- $\kappa$ B signaling is necessary for the host's innate immune response (Banoth et al., 2015). In the Vero E6 cells, full-length N protein considerably enhances NF- $\kappa$ B activity. In addition, T helper cells develop proinflammatory cytokines by NF- $\kappa$ B signaling (Liao et al., 2005). NF- $\kappa$ B activation is a characteristic of most infections, including those caused by viruses, which lead to defensive and pathological reactions. Following mice infection with rSARS-CoV-MA15, increased expression of inflammatory cytokine TNF, C-C motif (CC) chemokines [CC chemokine ligand (CCL) 2, CCL5], C-X-C motif (CXC) chemokines [CXC chemokine ligand CXCL1, CXCL2, and CXCL10], and IL-6 were found in neu-

trophils and infected lungs. Elevated levels of IL-6 and chemokines such CCL2 and CXCL10 have also been found in human lungs with fatal SARS (Jiang et al., 2005; Tang et al., 2005; Cameron et al., 2007). Researchers recently investigated the regulatory relation between the protein SARS-CoV-2 mediated pro-inflammatory cytokine/chemokine response and the NF- $\kappa$ B signaling pathway (Huang et al., 2020b; Ingraham et al., 2020; Islam and Fischer, 2020; Neufeldt et al., 2020; Rian et al., 2021). Huang et al. (2020b) showed that a significant transcriptomic transition in infected cells, characterized by a change to an inflammatory phenotype with activation of NF- $\kappa$ B signaling and NF- $\kappa$ B target genes by day 1 post-infection, leads to the loss of the mature alveolar program in a human *in vitro* model that simulates the initial apical infection of alveolar epithelium with SARS-CoV-2, leads to a loss of the mature alveolar program. Differentially expressed genes are enriched for components of pathways related to NF- $\kappa$ B, TNF- $\alpha$  and IL-17 signaling in bronchial epithelial cells infected with SARS-CoV-2 (Enes and Pir, 2020). Elements in the ACE2 gene regulate pirin, a negative regulator of the NF- $\kappa$ B subunit RELA (p65). Pirin expression is thought to be reduced when SARS-CoV-2 disrupts ACE2 (Fadason et al., 2020). Furthermore, in human bronchial epithelial cells, SARS-CoV-2 spike protein subunit 1 (CoV2-S1) caused high rates of NF- $\kappa$ B activation, the development of pro-inflammatory cytokines and chemokines including IL-1, TNF, IL-6 and CCL2, as well as mild epithelial damage. S1 interaction with the human ACE2 receptor, as well as early activation of the endoplasmic reticulum stress, subsequent unfolded protein response and MAP kinase signaling pathways, were all necessary for CoV2-S1-induced NF- $\kappa$ B activation. CoV-2-S1 had a higher NF- $\kappa$ B activation than CoV-S1, which may be attributed to CoV-2-S1's higher affinity for the ACE2 receptor (Hsu et al., 2020). Previous research has shown that an elevated cytokine/chemokine response during extreme SARS infection indicates a dysregulated immune response. *In vivo*, IL-6 is the primary stimulator of signal transducers and activators of transcription (STAT-3), and STAT3 is needed for complete NF- $\kappa$ B pathway activation, particularly during inflammation (Hirano and Murakami, 2020; Murakami et al., 2019). Both NF- $\kappa$ B and STAT-3 are triggered as a result of SARS-CoV-2 infection in the respiratory system, resulting in activation of the IL-6 amplifier, a mech-

anism for STAT-3 hyperactivation of NF- $\kappa$ B, leading to a variety of inflammatory and autoimmune diseases (Murakami et al., 2019). Moreover, previous study reported that thalidomide as an immunomodulatory agent modulates the NF- $\kappa$ B activities in combination with celecoxib (the cyclooxygenase-2 inhibitor) which can restrict the symptoms of inflammation if used to treat severe pneumonia (Hada, 2020). Since immunomodulatory drugs can affect the cytokine storm, these drugs may be effective in treating COVID-19. Immunomodulation of NF- $\kappa$ B activity and inhibitors of NF- $\kappa$ B (I $\kappa$ B) degradation, in combination with TNF- $\alpha$  inhibition may reduce the cytokine storm and lessen the severity of COVID-19. Inhibition of NF- $\kappa$ B pathway may be useful in treating COVID-19 in its most severe form.

Many of the drugs appear to have binds to the NF- $\kappa$ B cascade of immune regulation in COVID-19. Dexamethasone is one of two glucocorticoids (the other being prednisolone) that has an inhibitory effect on the NF- $\kappa$ B pathway (Ye et al., 2020; D'Acquisto et al., 2002). Remdesivir (GS-5734) is a nucleotide analogue that inhibits the RNA dependent RNA polymerase, causing viral replication to be disrupted. It decreases the cytokine storm and severe illness by lowering dsRNA-related activation of the NF- $\kappa$ B pathway. Remdesivir patients had a faster time to recover in the Adaptive COVID-19 Treatment Trial, which compared to a placebo (Beigel et al., 2020). TNF- $\alpha$ , TNF- $1\beta$ , IgG and IFN- $\gamma$  are all reduced by hydroxychloroquine, which suppresses the NF- $\kappa$ B pathway (Liang et al., 2018).

#### *Janus kinase (JAK)–STAT pathway*

The JAK-STAT pathway signaling mechanism, may be a valuable marker of a strong immune response to COVID-19 infections (Bouwman et al., 2020). According to one study, SARS-CoV-2 triggers the biochemical mechanisms mediated by JAK–STAT in the lungs, leading in viral cell proliferation and transmitting (Singh et al., 2020). In another study, inhibiting the JAK-STAT pathway reduced hyperinflammatory conditions while having no effect on viral clearance (Rojas and Sarmiento, 2021). The JAK-STAT pathway is also activated by IL-6 (Billing et al., 2019). The finding demonstrates that induction of the JAK-STAT pathway, particularly through cytokines such IL-6, is associated with the inflammatory response to COVID-19 (Luo et al., 2020b). Angiotensin II binds to the AT1R and activates the JAK-

STAT pathway, leading to the production of IL-6 (Ni et al., 2020). The SARS-CoV-2 S protein inhibits ACE2, causing an increase in angiotensin II expression and, as a result, enhanced IL-6 production. Anti-inflammatory drugs, in particular JAK-STAT inhibitors may be useful against increased cytokine levels and may be effective to prevent viral infection. Ruxolitinib is a JAK1 and JAK2 inhibitor that suppresses STAT activation and nuclear translocation by blocking JAK kinase activity. Ruxolitinib also suppresses the IL6/JAK-STAT3 pathway, decreasing IL-6 levels in the blood (Caocci and La Nasa, 2020; Kusoglu et al., 2020). The role of baricitinib (a specific JAK1 and JAK2 inhibitor) in the treatment of COVID-19 has been proposed, despite its true safety profile has yet to be determined (Cingolani et al., 2020a).

#### *Toll-like receptor (TLR) signaling pathway*

The TLRs are important in the innate immunity by detecting microbes to invade pathogens. TLR signaling pathways are the recruitment of different adaptor molecules resulting in the activation of NF- $\kappa$ B and the IFN regulatory factor transcription factors dictating the outcome of TLR's innate immune responses (Barton and Medzhitov, 2003). While the immune system's effective functioning protects the body from infections, the cytokine storm associated with extreme COVID-19 manifestations is mainly caused by the adaptive immune system's over-expression and exhaustion, rather than an innate immune response (Coperchini et al., 2020). The virus's spread is limited by the host immune response during infection or mild COVID-19 disease, but the innate immune response may also trigger immune-related dysfunction, resulting in extreme pneumonia in cases of high viral load (Soraya and Urmia, 2020). In viral diseases, TLR activators have both defensive and therapeutic effects. The study also discovered that the SARS-CoV-2 spike protein binds to TLR1, 4 and 6 with a higher affinity for TLR4 than the others (Khadke et al., 2020). A recent study offers that TLRs may be involved in both the initial viral clearance failure and the subsequent production of the deadly clinical manifestations of severe COVID-19 primarily ARDS. Lung macrophages can play a critical role in massive release of IL-6 and other cytokines such as IL- $1\beta$ , IL-10, IL-12 and TNF- $\alpha$  via activation of TLRs in patients with severe COVID-19 (Onofrio et al., 2020).

In addition to the development of proinflammato-

ry cytokines, TLRs' interaction with virus particles has immunopathological effects that lead to death in COVID19 patients (Patra et al., 2020). TLR4's pathologic role in patients with an excessive inflammatory response has been documented in other SARS-CoV-2 studies. COVID-19 patients had substantially higher levels of CCL 2, CCL7, CCL8, CCL24, CCL20, CCL13, CCL3, CXCL 2, CXCL10 and IL-1b, and its down-stream inflammatory signaling molecules (IL1R1, Myeloid differentiation primary response [MYD88], interleukin 1 receptor associated kinase 1 [IRAK1], TNF receptor associated factor [TRAF6], NF-KBIA, NF-KB1, RELA). TLR4 and related/down-stream signaling molecules (CD14, MYD88, IRAK1, TRAF6, TIRAP, TICAM) as well as most NF-κB signaling pathway genes (NF-KBIA, NF-KB1, RELA, NF-KB2) were also highly up-regulated, implying that activation of the NF-κB signaling pathway by TLR4 is thought to be responsible for the up-regulation of inflammatory responses in COVID-19 infection patients (Sohn et al., 2020). Furthermore, COVID19 patients have a higher level of neutrophil myeloperoxidase, which triggers oxidized phospholipids and TLR4 pathway activation causes oxidative injury during the pulmonary process of infection (Khadke et al., 2020; Onofrio et al., 2020). Tocilizumab, an anti-IL-6 monoclonal antibody is used to treat rheumatoid arthritis, may be useful in the treatment of critically ill patients with COVID-19 (Kaly and Rosner, 2012). Findings support the use of therapeutic approaches such as dexamethasone that inhibits TLR4-mediated inflammatory signaling through molecular checkpoints (Sohn et al., 2020).

#### *Phosphatidylinositol-3-kinase (PI3K)/ AKT/ mammalian target of rapamycin (mTOR) pathway*

The PI3K/AKT/ mTOR signaling pathways is an important intracellular signaling pathway in the regulation of the cell cycle and cell growth. Therefore, it is specifically associated with cellular proliferation, quiescence and survival. The plasma membrane protein AKT is phosphorylated and activated when PI3K is activated (King et al., 2015). Insulin-like growth factor, epidermal growth factor, sonic hedgehog signaling molecule insulin and CaM can enhance the PI3K / AKT pathway (Man et al., 2003; Peltier et al., 2007; Ojeda et al., 2011; Rafalski and Brunet, 2011). The mTOR signaling pathway modulates protein synthesis in response to stress, hor-

mones and genetic factors. Rapamycin inhibits mTOR by interfering with the PI3K/AKT/mTOR pathway and activating AMP-activated protein kinase (Huang, 2013). MTOR signaling is required for influenza development and regulates the antibody response, resulting in cross-protective immunity against lethal influenza virus infections. Treatment of serious pneumonia caused by H1N1 influenza with rapamycin and steroids has been shown to enhance reporting outcomes in human studies (Chuang et al., 2014; Wang et al., 2014; Lehrer, 2020). The PI3K/AKT/mTOR signaling responses have a key role in MERS-CoV infection which making it a target for therapeutic intervention. Buformin or phenformin (mTOR inhibitor ) inhalation may be an effective novel treatment for coronavirus (Lehrer, 2020). Cytokine storms are the main reason of COVID-19-related serious illness and death. The most significant cause of cytokine storms can be the antibody-dependent enhancement. mTOR inhibitors may suppress antibody-dependent enhancement and decrease the severity of COVID19 by selectively inhibiting memory-B cell activation (Zheng et al., 2020).

The mTOR–PI3K–AKT pathway was identified as a key signaling pathway in SARSCoV2 infection in a recent report. The *in vitro* testing of three mTOR inhibitors showed that they significantly inhibited SARSCoV2 (Garcia Jr et al., 2020). Regarding to recent reports, activation of the PI3K/ AKT/ mTOR pathway appears to be important to promote replication of different viruses and drugs that inhibit PI3K/ AKT/ mTOR signaling pathways may be recommended for SARS-CoV-2 infection. In order to identify potential drug targets, a human protein–protein interaction map for SARSCoV2 was recently developed. The proposed drugs included the mTOR inhibitors rapamycin and sapanisertib, as well as the mTORC1 protein complex modulator metformin. Metformin-treated COVID19 patients have been shown to have a lower mortality rate (Bramante et al., 2020; Cariou et al., 2020; Luo et al., 2020a).

Inflammatory cytokines can be a double-edged sword when it comes to viral infection and disease pathogenesis. To battle viral infection and avoid a cytokine storm, the innate immune system must be fine-tuned (Säemann et al., 2009). As a result, clinical trials should include early and short-term intervention with mTOR inhibitors to reduce the undesirable immunosuppressive effect. Furthermore, IL-6 may play a crucial role in the cyto-

kine storm's substantial negative consequences and IL-6 inhibition has been used to treat severe COVID19 disease with respiratory distress (Zheng et al., 2020). In addition to mTOR inhibitors, combination therapy with an anti-IL6 antibody could be included in the clinical trial for patients suffering SARS-CoV2 pneumonia (Zheng et al., 2020).

## Conclusion

Infection with SARS-CoV-2 changes multiple signal transduction pathways, which contribute to important physiological functions of the cell. The balance of signaling pathway activities is important for cell death, or cell survival determination. The virus takes over mechanisms from the host cell to utilize it for its own benefit. SARS-CoV-2 involved MAPK signaling pathway, NF- $\kappa$ B pathway, PI3K/ AKT/ mTOR pathway, JAK-STAT pathway and toll-like receptors cascades through different mechanisms. In certain infected individuals, SARS-CoV-2 induces excessive and prolonged cytokine/chemokine responses. ARDS, or multi-organ dysfunction, is caused by the cytokine storm and it leads to physiological deterioration and death. The virus manipulates these signaling pathways for inhibiting cytokine antiviral effects.

## Acknowledgment

This work was supported by the Hearing Disorder Research Center of Shahid Beheshti University of Medical Sciences.

## Conflict of interest

The authors declare no conflict of interest.

## References

- Bagca BG, Avci CB. The potential of JAK/STAT pathway inhibition by ruxolitinib in the treatment of COVID-19. *Cytokine Growth Factor Rev* 2020; 54: 51-61. <https://doi.org/10.1016/j.cytogfr.2020.06.013>
- Banoth B, Chatterjee B, Vijayaragavan B, Prasad M, Roy P, Basak S. Stimulus-selective crosstalk via the NF- $\kappa$ B signaling system reinforces innate immune response to alleviate gut infection. *Elife* 2015; 4: e05648. <https://doi.org/10.7554/eLife.05648>
- Barr RG, Celli BR, Mannino DM, Petty T, Rennard SI, Sciurba FC, et al. Comorbidities, patient knowledge, and disease management in a national sample of patients with COPD. *Am J Med* 2009; 122: 348-55. <https://doi.org/10.1016/j.amjmed.2008.09.042>
- Barton GM, Medzhitov R. Toll-like receptor signaling pathways. *Science* 2003; 300: 1524-5. <https://doi.org/10.1126/science.1085536>
- Battagello DS, Dragunas G, Klein MO, Ayub AL, Velloso FJ, Correa RG. Unpuzzling COVID-19: tissue-related signaling pathways associated with SARS-CoV-2 infection and transmission. *Clin Sci* 2020; 134: 2137-60. <https://doi.org/10.1042/CS20200904>
- Beigel JH, Tomashek KM, Dodd LE, Mehta AK, Zingman BS, Kalil AC, et al. Remdesivir for the treatment of Covid-19-preliminary report. *N Engl J Med* 2020.
- Billing U, Jetka T, Nortmann L, Wundrack N, Komorowski M, Waldherr S, et al. Robustness and information transfer within IL-6-induced JAK/STAT signalling. *Commun Biol* 2019; 2: 1-4. <https://doi.org/10.1038/s42003-018-0259-4>
- Birra D, Benucci M, Landolfi L, Merchionda A, Loi G, Amato P, et al. COVID 19: a clue from innate immunity. *Immunol Res* 2020; 161-8. <https://doi.org/10.1007/s12026-020-09137-5>
- Bonizzi G, Karin M. The two NF- $\kappa$ B activation pathways and their role in innate and adaptive immunity. *Trends Immunol* 2004; 25: 280-8. <https://doi.org/10.1016/j.it.2004.03.008>
- Bouhaddou M, Memon D, Meyer B, White KM, Rezelj VV, Marrero MC, et al. The global phosphorylation landscape of SARS-CoV-2 infection. *Cell* 2020; 182: 685-712. <https://doi.org/10.1016/j.cell.2020.06.034>
- Bouwman W, Verhaegh W, Holtzer L, van de Stolpe A. Measurement of cellular immune response to viral infection and vaccination. *Front Immunol* 2020; 11: 575074. <https://doi.org/10.3389/fimmu.2020.575074>
- Bradley BT, Maioli H, Johnston R, Chaudhry I, Fink SL, Xu H, et al. Histopathology and ultrastructural findings of fatal COVID-19 infections in Washington State: a case series. *Lancet* 2020; 396: 320-32. [https://doi.org/10.1016/S0140-6736\(20\)31305-2](https://doi.org/10.1016/S0140-6736(20)31305-2)
- Bramante CT, Ingraham NE, Murray TA, Marmor S, Hoversten S, Gronski J, et al. Observational study of metformin and risk of mortality in patients hospitalized with Covid-19. *MedRxiv* 2020. <https://doi.org/10.1101/2020.06.19.20135095>
- Cameron MJ, Bermejo-Martin JF, Danesh A, Muller MP, Kelvin DJ. Human immunopathogenesis of severe acute respiratory syndrome (SARS). *Virus Res* 2008; 133: 13-9.

- <https://doi.org/10.1016/j.virusres.2007.02.014>
- Cameron MJ, Ran L, Xu L, Danesh A, Bermejo-Martin JF, Cameron CM, et al. Interferon-mediated immunopathological events are associated with atypical innate and adaptive immune responses in patients with severe acute respiratory syndrome. *J Virol* 2007; 81: 8692-706. <https://doi.org/10.1128/JVI.00527-07>
- Caocci G, La Nasa G. Could ruxolitinib be effective in patients with COVID-19 infection at risk of acute respiratory distress syndrome (ARDS)? *Ann Hematol* 2020; 99: 1675-6. <https://doi.org/10.1007/s00277-020-04067-6>
- Cariou B, Hadjadj S, Wargny M, Pichelin M, Al-Salameh A, Allix I, et al. Phenotypic characteristics and prognosis of inpatients with COVID-19 and diabetes: the CORONADO study. *Diabetologia* 2020; 63: 1500-15. <https://doi.org/10.1007/s00125-020-05180-x>
- Carsana L, Sonzogni A, Nasr A, Rossi RS, Pellegrinelli A, Zerbi P, et al. Pulmonary post-mortem findings in a series of COVID-19 cases from northern Italy: a two-centre descriptive study. *Lancet Infect Dis* 2020; 20: 1135-40. [https://doi.org/10.1016/S1473-3099\(20\)30434-5](https://doi.org/10.1016/S1473-3099(20)30434-5)
- Catanzaro M, Fagiani F, Racchi M, Corsini E, Govoni S, Lanni C. Immune response in covid-19: addressing a pharmacological challenge by targeting pathways triggered by SARS-COV-2. *Signal Transduct Target Ther* 2020; 5: 1-10. <https://doi.org/10.1038/s41392-020-0191-1>
- Channappanavar R, Perlman S. Pathogenic human coronavirus infections: causes and consequences of cytokine storm and immunopathology. *Semin Immunopathol* 2017. <https://doi.org/10.1007/s00281-017-0629-x>
- Chen L, Li X, Chen M, Feng Y, Xiong C. The ACE2 expression in human heart indicates new potential mechanism of heart injury among patients infected with SARS-COV-2. *Cardiovasc Res* 2020; 116: 1097-100. <https://doi.org/10.1093/cvr/cvaa078>
- Chen W, Thomas J, Sadatsafavi M, FitzGerald JM. Risk of cardiovascular comorbidity in patients with chronic obstructive pulmonary disease: a systematic review and meta-analysis. *Lancet Respir Med* 2015; 3: 631-9. [https://doi.org/10.1016/S2213-2600\(15\)00241-6](https://doi.org/10.1016/S2213-2600(15)00241-6)
- Chuang YC, Ruan SY, Huang CT. Compelling results of adjuvant therapy with sirolimus for severe H1N1 pneumonia. *Crit Care Med* 2014; 42: 687-8. <https://doi.org/10.1097/CCM.0000000000000489>
- Cingolani A, Tummolo AM, Montemurro G, Gremese E, Larosa L, Cipriani MC, et al. Baricitinib as rescue therapy in a patient with covid-19 with no complete response to sarilumab. *Infection* 2020a; 48: 767-71. <https://doi.org/10.1007/s15010-020-01476-7>
- Cingolani A, Tummolo AM, Montemurro G, Gremese E, Larosa L, Cipriani MC, et al. Baricitinib as rescue therapy in a patient with covid-19 with no complete response to sarilumab. *Infection* 2020b; 48: 767-71. <https://doi.org/10.1007/s15010-020-01476-7>
- Coperchini F, Chiovato L, Croce L, Magri F, Rotondi M. The cytokine storm in covid-19: an overview of the involvement of the chemokine/chemokine-receptor system. *Cytokine Growth Factor Rev* 2020. <https://doi.org/10.1016/j.cytogfr.2020.05.003>
- Cowan KJ, Storey KB. Mitogen-activated protein kinases: new signaling pathways functioning in cellular responses to environmental stress. *J Exp Biol* 2003; 206: 1107-15. <https://doi.org/10.1242/jeb.00220>
- D'Acquisto F, May MJ, Ghosh S. Inhibition of nuclear factor kappa b (NF-B). *Mol Interv* 2002; 2: 22. <https://doi.org/10.1124/mi.2.1.22>
- de Abajo FJ, Rodríguez-Miguel A, Rodríguez-Martín S, Llerma V, García-Lledó A. Impact of in-hospital discontinuation with angiotensin receptor blockers or converting enzyme inhibitors on mortality of covid-19 patients: A retrospective cohort study. *BMC Med* 2021; 19: 1-15. <https://doi.org/10.1186/s12916-021-01992-9>
- Deak M, Clifton AD, Lucocq JM, Alessi DR. Mitogen- and stress-activated protein kinase-1 (MSK1) is directly activated by MAPK and SAPK2/P38, and may mediate activation of CREB. *EMBO J* 1998; 17: 4426-41. <https://doi.org/10.1093/emboj/17.15.4426>
- Deshotel MR, Xia H, Sriramula S, Lazartigues E, Filipeanu CM. Angiotensin II mediates angiotensin converting enzyme type 2 internalization and degradation through an angiotensin II type I receptor-dependent mechanism. *Hypertension* 2014; 64: 1368-75. <https://doi.org/10.1161/HYPERTENSIONAHA.114.03743>
- Elkhodary MS. Treatment of covid-19 by controlling the activity of the nuclear factor-kappa B. *CellBio* 2020; 9: 109-21. <https://doi.org/10.4236/cellbio.2020.92006>
- Enes A, Pir P. Transcriptional response of signaling pathways to SARS-COV-2 infection in normal human bronchial epithelial cells. *bioRxiv* 2020. <https://doi.org/10.1101/2020.06.20.163006>
- Fadason T, Gokuladhas S, Golovina E, Ho D, Farrow S, Nyaga D, et al. A transcription regulatory network within the ACE2 locus may promote a pro-viral environment for SARS-COV-2 by modulating expression of host factors. *bioRxiv*

2020. <https://doi.org/10.1101/2020.04.14.042002>
- Feng Y, Fang Z, Liu B, Zheng X. P38mapk plays a pivotal role in the development of acute respiratory distress syndrome. *Clinics* 2019; 74. <https://doi.org/10.6061/clinics/2019/e509>
- Fernandez-Garcia MD, Mazzon M, Jacobs M, Amara A. Pathogenesis of flavivirus infections: using and abusing the host cell. *Cell Host Microbe* 2009; 5: 318-28. <https://doi.org/10.1016/j.chom.2009.04.001>
- Foltz IN, Lee JC, Young PR, Schrader JW. Hemopoietic growth factors with the exception of interleukin-4 activate the p38 mitogen-activated protein kinase pathway. *J Biol Chem* 1997; 272: 3296-301. <https://doi.org/10.1074/jbc.272.6.3296>
- Garcia Jr G, Sharma A, Ramaiah A, Sen C, Kohn DB, Gomperts BN, et al. Antiviral drug screen of kinase inhibitors identifies cellular signaling pathways critical for SARS-COV-2 replication. Available at SSRN 3682004 2020. <https://doi.org/10.1101/2020.06.24.150326>
- Gheblawi M, Wang K, Viveiros A, Nguyen Q, Zhong JC, Turner AJ, et al. Angiotensin-converting enzyme 2: SARS-COV-2 receptor and regulator of the renin-angiotensin system: celebrating the 20th anniversary of the discovery of ACE2. *Circ Res* 2020; 126: 1456-74. <https://doi.org/10.1161/CIRCRESAHA.120.317015>
- Gössling S, Scott D, Hall CM. Pandemics, tourism and global change: a rapid assessment of covid-19. *J Sustain Tour* 2020;1-20. <https://doi.org/10.1080/09669582.2020.1758708>
- Gralinski LE, Menachery VD. Return of the coronavirus: 2019-nCoV. *Viruses* 2020; 12: 135. <https://doi.org/10.3390/v12020135>
- Grimes JM, Grimes KV. P38 MAPK inhibition: a promising therapeutic approach for covid-19. *J Mol Cell Cardiol* 2020b; 144: 63-5. <https://doi.org/10.1016/j.yjmcc.2020.05.007>
- Gupta S. Molecular signaling in death receptor and mitochondrial pathways of apoptosis. *Int J Oncol* 2003; 22: 15-20. <https://doi.org/10.3892/ijo.22.1.15>
- Hada M. Chemotherapeutic strategy with synbiotics, thalidomide and celecoxib for severe covid-19 pneumonia. Association between Microbiota, Chronic Inflammation and Pneumonia 2020. <https://doi.org/10.22541/au.159188529.93357127>
- Hirano T, Murakami M. Covid-19: a new virus, but a familiar receptor and cytokine release syndrome. *Immunity* 2020. <https://doi.org/10.1016/j.immuni.2020.04.003>
- Hoesel B, Schmid JA. The complexity of NF- $\kappa$ B signaling in inflammation and cancer. *Mol Cancer* 2013; 12: 1-15. <https://doi.org/10.1186/1476-4598-12-86>
- Hsu AC, Wang G, Reid AT, Veerati PC, Pathinayake PS, Daly K, et al. SARS-COV-2 spike protein promotes hyper-inflammatory response that can be ameliorated by spike-antagonistic peptide and FDA-approved ER stress and MAP kinase inhibitors in vitro. *Biorxiv* 2020. <https://doi.org/10.1101/2020.09.30.317818>
- Huang C, Wang Y, Li X, Ren L, Zhao J, Hu Y, et al. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. *Lancet* 2020a; 395: 497-506. [https://doi.org/10.1016/S0140-6736\(20\)30183-5](https://doi.org/10.1016/S0140-6736(20)30183-5)
- Huang J, Hume AJ, Abo KM, Werder RB, Villacorta-Martin C, Alysandratos KD, et al. SARS-COV-2 infection of pluripotent stem cell-derived human lung alveolar type 2 cells elicits a rapid epithelial-intrinsic inflammatory response. *Cell Stem Cell* 2020b; 27: 962-73. <https://doi.org/10.1016/j.stem.2020.09.013>
- Huang S. Inhibition of PI3K/AKT/mTOR signaling by natural products. *Anticancer Agents Med Chem* 2013; 13: 967. <https://doi.org/10.2174/1871520611313070001>
- Ingraham NE, Lotfi-Emran S, Thielen BK, Techar K, Morris RS, Holtan SG, et al. Immunomodulation in covid-19. *Lancet Respir Med* 2020; 8: 544-6. [https://doi.org/10.1016/S2213-2600\(20\)30226-5](https://doi.org/10.1016/S2213-2600(20)30226-5)
- Islam MR, Fischer A. A transcriptome analysis identifies potential preventive and therapeutic approaches towards covid-19. 2020. <https://doi.org/10.20944/preprints202004.0399.v1>
- Jiang Y, Xu J, Zhou C, Wu Z, Zhong S, Liu J, et al. Characterization of cytokine/chemokine profiles of severe acute respiratory syndrome. *Am J Respir Crit Care Med* 2005; 171: 850-7. <https://doi.org/10.1164/rccm.200407-857OC>
- Kaly L, Rosner I. Tocilizumab-a novel therapy for non-organ-specific autoimmune diseases. *Best Pract Res Clin Rheumatol* 2012; 26: 157-65. <https://doi.org/10.1016/j.berh.2012.01.001>
- Khadke S, Ahmed N, Ahmed N, Ratts R, Raju S, Gallogly M, et al. Harnessing the immune system to overcome cytokine storm and reduce viral load in covid-19: a review of the phases of illness and therapeutic agents. *Virol J* 2020; 17: 1-18. <https://doi.org/10.1186/s12985-020-01415-w>
- King D, Yeomanson D, Bryant HE. PI3king the lock: Targeting the PI3K/AKT/mTOR pathway as a novel therapeutic strategy in neuroblastoma. *J Pediatr Hematol Oncol* 2015; 37: 245-51. <https://doi.org/10.1097/>

- MPH.0000000000000329
- Koka V, Huang XR, Chung AC, Wang W, Truong LD, Lan HY. Angiotensin II up-regulates angiotensin I-converting enzyme (ACE), but down-regulates ACE2 via the AT1-ERK/P38 MAP kinase pathway. *A J Pathol* 2008; 172: 1174-83. <https://doi.org/10.2353/ajpath.2008.070762>
- Krajcsi P, Wold WS. Viral proteins that regulate cellular signalling. *J Gen Virol* 1998; 79: 1323-35. <https://doi.org/10.1099/0022-1317-79-6-1323>
- Kusoglu A, Bagca BG, Ay NP, Saydam G, Avci CB. Ruxolitinib regulates the autophagy machinery in multiple myeloma cells. *Anticancer Agents Med Chem* 2020; 20: 2316-23. <https://doi.org/10.2174/1871520620666200218105159>
- Lai CC, Shih TP, Ko WC, Tang HJ, Hsueh PR. Severe acute respiratory syndrome coronavirus 2 (SARS-COV-2) and corona virus disease-2019 (covid-19): The epidemic and the challenges. *Int J Antimicrob Agents* 2020: 105924. <https://doi.org/10.1016/j.ijantimicag.2020.105924>
- Lehrer S. Inhaled biguanides and mTOR inhibition for influenza and coronavirus. *World Acad Sci J* 2020; 2: 1-1. <https://doi.org/10.3892/wasj.2020.68>
- Li G, De Clercq E. Therapeutic options for the 2019 novel coronavirus (2019-nCoV). *Nat Rev Drug Discov* 2020. <https://doi.org/10.1038/d41573-020-00016-0>
- Li G, He X, Zhang L, Ran Q, Wang J, Xiong A, et al. Assessing ACE2 expression patterns in lung tissues in the pathogenesis of covid-19. *J Autoimmun* 2020: 102463. <https://doi.org/10.1016/j.jaut.2020.102463>
- Liang N, Zhong Y, Zhou J, Liu B, Lu R, Guan Y, et al. Immunosuppressive effects of hydroxychloroquine and artemisinin combination therapy via the nuclear factor- $\kappa$ B signaling pathway in lupus nephritis mice. *Exp Ther Med* 2018; 15: 2436-42. <https://doi.org/10.3892/etm.2018.5708>
- Liao QJ, YE LB, Timani KA, ZENG YC, SHE YL, Ye L, et al. Activation of NF- $\kappa$ B by the full-length nucleocapsid protein of the SARS coronavirus. *Acta Biochim Biophys Sin* 2005; 37: 607-12. <https://doi.org/10.1111/j.1745-7270.2005.00082.x>
- Liu DX, Fung TS, Chong KK, Shukla A, Hilgenfeld R. Accessory proteins of SARS-COV and other coronaviruses. *Antivir Res* 2014; 109: 97-109. <https://doi.org/10.1016/j.antiviral.2014.06.013>
- Liu N, Hong Y, Chen RG, Zhu HM. High rate of increased level of plasma angiotensin ii and its gender difference in covid-19: An analysis of 55 hospitalized patients with covid-19 in a single hospital, Wuhan, China. medRxiv 2020a. <https://doi.org/10.1101/2020.04.27.20080432>
- Liu T, Zhang L, Joo D, Sun SC. Nf- $\kappa$ B signaling in inflammation. *Signal Transduct Target Ther* 2017; 2: 1-9. <https://doi.org/10.1038/sigtrans.2017.23>
- Liu Y, Sun W, Li J, Chen L, Wang Y, Zhang L, et al. Clinical features and progression of acute respiratory distress syndrome in coronavirus disease 2019. MedRxiv 2020b. <https://doi.org/10.1101/2020.02.17.20024166>
- Liu Y, Yang Y, Zhang C, Huang F, Wang F, Yuan J, et al. Clinical and biochemical indexes from 2019-nCoV infected patients linked to viral loads and lung injury. *Sci China Life Sci* 2020c; 63: 364-74. <https://doi.org/10.1007/s11427-020-1643-8>
- Luo P, Qiu L, Liu Y, Liu X-l, Zheng JI, Xue Hy, et al. Metformin treatment was associated with decreased mortality in covid-19 patients with diabetes in a retrospective analysis. *Am J Trop Med Hyg* 2020a; 103: 69. <https://doi.org/10.4269/ajtmh.20-0375>
- Luo W, Li YX, Jiang LJ, Chen Q, Wang T, Ye DW. Targeting JAK-STAT signaling to control cytokine release syndrome in covid-19. *Trends Pharmacol Sci* 2020b. <https://doi.org/10.1016/j.tips.2020.06.007>
- Magnani M, Crinelli R, Bianchi M, Antonelli A. The ubiquitin-dependent proteolytic system and other potential targets for the modulation of nuclear factor- $\kappa$ B (NF- $\kappa$ B). *Curr Drug Targets* 2000; 1: 387-99. <https://doi.org/10.2174/1389450003349056>
- Mahase E. Coronavirus: covid-19 has killed more people than SARS and MERS combined, despite lower case fatality rate. *Br Med J* 2020. <https://doi.org/10.1136/bmj.m641>
- Man HY, Wang Q, Lu WY, Ju W, Ahmadian G, Liu L, et al. Activation of PI3-kinase is required for AMPA receptor insertion during LTP of MEPPS in cultured hippocampal neurons. *Neuron* 2003; 38: 611-24. [https://doi.org/10.1016/S0896-6273\(03\)00228-9](https://doi.org/10.1016/S0896-6273(03)00228-9)
- Mizutani T. Signaling pathways of SARS-COV in vitro and in vivo. *Molecular biology of the SARS-coronavirus: Springer*, 2010: 305-322. [https://doi.org/10.1007/978-3-642-03683-5\\_19](https://doi.org/10.1007/978-3-642-03683-5_19)
- Mizutani T, Fukushi S, Saijo M, Kurane I, Morikawa S. Phosphorylation of p38 MAPK and its downstream targets in SARS coronavirus-infected cells. *Biochem Biophys Res Commun* 2004; 319: 1228-34. <https://doi.org/10.1016/j.bbrc.2004.05.107>
- Morens DM, Folkers GK, Fauci AS. The challenge of emerging and re-emerging infectious diseases. *Nature* 2004; 430:

- 242-9. <https://doi.org/10.1038/nature02759>
- Murakami M, Kamimura D, Hirano T. Pleiotropy and specificity: insights from the interleukin 6 family of cytokines. *Immunity* 2019; 50: 812-31. <https://doi.org/10.1016/j.immuni.2019.03.027>
- Neufeldt CJ, Cerikan B, Cortese M, Frankish J, Lee JY, Plociennikowska A, et al. SARS-COV-2 infection induces a pro-inflammatory cytokine response through CGAS-sting and NF- $\kappa$ B. *bioRxiv* 2020. <https://doi.org/10.1101/2020.07.21.212639>
- Ni W, Yang X, Yang D, Bao J, Li R, Xiao Y, et al. Role of angiotensin-converting enzyme 2 (ACE2) in covid-19. *Crit Care* 2020; 24: 1-10. <https://doi.org/10.1186/s13054-020-03120-0>
- Oeckinghaus A, Ghosh S. The NF- $\kappa$ B family of transcription factors and its regulation. *Cold Spring Harb Perspect Biol* 2009; 1: a000034. <https://doi.org/10.1101/cshperspect.a000034>
- Ojeda L, Gao J, Hooten KG, Wang E, Thonhoff JR, Dunn TJ, et al. Critical role of PI3K/AKT/GSK3 $\beta$  in motoneuron specification from human neural stem cells in response to FGF2 and EGF. *PloS One* 2011; 6: e23414. <https://doi.org/10.1371/journal.pone.0023414>
- Onofrio L, Caraglia M, Facchini G, Margherita V, Placido SD, Buonerba C. Toll-like receptors and covid-19: a two-faced story with an exciting ending. 2020. <https://doi.org/10.2144/fsoa-2020-0091>
- Parekh M, Donuru A, Balasubramanya R, Kapur S. Review of the chest CT differential diagnosis of ground-glass opacities in the covid era. *Radiology* 2020; 297: 289-302. <https://doi.org/10.1148/radiol.2020202504>
- Patel VB, Clarke N, Wang Z, Fan D, Parajuli N, Basu R, et al. Angiotensin II induced proteolytic cleavage of myocardial ACE2 is mediated by TACE/ADAM-17: a positive feedback mechanism in the Ras. *J Mol Cell Cardiol* 2014; 66: 167-76. <https://doi.org/10.1016/j.yjmcc.2013.11.017>
- Patra R, Das NC, Mukherjee S. Targeting human TLRs to combat covid-19: a solution? *J Med Virol* 2020. <https://doi.org/10.1002/jmv.26387>
- Peltier J, O'Neill A, Schaffer DV. PI3K/AKT and CREB regulate adult neural hippocampal progenitor proliferation and differentiation. *Dev Neurobiol* 2007; 67: 1348-61. <https://doi.org/10.1002/dneu.20506>
- Petersen E, Koopmans M, Go U, Hamer DH, Petrosillo N, Castelli F, et al. Comparing SARS-COV-2 with SARS-COV and influenza pandemics. *Lancet Infect Dis* 2020. [https://doi.org/10.1016/S1473-3099\(20\)30484-9](https://doi.org/10.1016/S1473-3099(20)30484-9)
- Pfeffer LM. The role of nuclear factor  $\kappa$ B in the interferon response. *J Interferon Cytokine Res* 2011; 31: 553-9. <https://doi.org/10.1089/jir.2011.0028>
- Promptchara E, Ketloy C, Palaga T. Immune responses in covid-19 and potential vaccines: lessons learned from SARS and MERS epidemic. *Asian Pac J Allergy Immunol* 2020; 38: 1-9.
- Rafalski VA, Brunet A. Energy metabolism in adult neural stem cell fate. *Prog Neurobiol* 2011; 93: 182-203. <https://doi.org/10.1016/j.pneurobio.2010.10.007>
- Ramaiah MJ. mTOR inhibition and p53 activation, microRNAs: the possible therapy against pandemic covid-19. *Gene Rep* 2020: 100765. <https://doi.org/10.1016/j.genrep.2020.100765>
- Rian K, Esteban-Medina M, Hidalgo MR, Çubuk C, Falco MM, Loucera C, et al. Mechanistic modeling of the SARS-COV-2 disease map. *BioData Min* 2021; 14: 1-8. <https://doi.org/10.1186/s13040-021-00234-1>
- Rockx B, Kuiken T, Herfst S, Bestebroer T, Lamers MM, Munnink BB, et al. Comparative pathogenesis of covid-19, MERS, and SARS in a nonhuman primate model. *Science* 2020; 368: 1012-5. <https://doi.org/10.1126/science.abb7314>
- Rojas P, Sarmiento M. JAK/STAT pathway inhibition may be a promising therapy for covid-19-related hyperinflammation in hematologic patients. *Acta Haematol* 2021: 144: 312-6. <https://doi.org/10.1159/000510179>
- Säemann M, Haidinger M, Hecking M, Hörl W, Weichhart T. The multifunctional role of mTOR in innate immunity: implications for transplant immunity. *Am J Transplant* 2009; 9: 2655-61. <https://doi.org/10.1111/j.1600-6143.2009.02832.x>
- Scott AJ, O'Dea KP, O'Callaghan D, Williams L, Dokpesi JO, Tatton L, et al. Reactive oxygen species and P38 mitogen-activated protein kinase mediate tumor necrosis factor  $\alpha$ -converting enzyme (TACE/ADAM-17) activation in primary human monocytes. *J Biol Chem* 2011; 286: 35466-76. <https://doi.org/10.1074/jbc.M111.277434>
- Singh Y, Gupta G, Satija S, Pabreja K, Chellappan DK, Dua K. Covid-19 transmission through host cell directed network of GPCR. *Drug Dev Res* 2020. <https://doi.org/10.1002/ddr.21674>
- Sohn KM, Lee SG, Kim HJ, Cheon S, Jeong H, Lee J, et al. Covid-19 patients upregulate toll-like receptor 4-mediated inflammatory signaling that mimics bacterial sepsis. *J Korean Med Sci* 2020; 35. <https://doi.org/10.3346/jkms.2020.35.e343>

- Soraya H, Urmia I. Prophylactic use of chloroquine may impair innate immune system response against SARS-COV-2. *Pharm Sci* 2020. <https://doi.org/10.34172/PS.2020.29>
- Tak PP, Firestein GS. Nf-kb: A key role in inflammatory diseases. *J Clin Invest* 2001; 107: 7-11. <https://doi.org/10.1172/JCI11830>
- Tang NL, Chan PK, Wong CK, To KF, Wu AK, Sung YM, et al. Early enhanced expression of interferon-inducible protein-10 (CXCL-10) and other chemokines predicts adverse outcome in severe acute respiratory syndrome. *Clin Chem* 2005; 51: 2333-40. <https://doi.org/10.1373/clinchem.2005.054460>
- Uzun T, Toptas O. Artesunate: Could be an alternative drug to chloroquine in covid-19 treatment? *Chin Med* 2020; 15: 1-4. <https://doi.org/10.1186/s13020-020-00336-8>
- Velavan TP, Meyer CG. The covid-19 epidemic. *Trop Med Int Health* 2020; 25: 278. <https://doi.org/10.1111/tmi.13383>
- Vellingiri B, Jayaramayya K, Iyer M, Narayanasamy A, Govindasamy V, Giridharan B, et al. Covid-19: a promising cure for the global panic. *Sci Total Environ* 2020: 138277. <https://doi.org/10.1016/j.scitotenv.2020.138277>
- Viatour P, Merville MP, Bours V, Chariot A. Phosphorylation of NF- $\kappa$ B and I $\kappa$ B proteins: implications in cancer and inflammation. *Trends Biochem Sci* 2005; 30: 43-52. <https://doi.org/10.1016/j.tibs.2004.11.009>
- Wang CH, Chung FT, Lin SM, Huang SY, Chou CL, Lee KY, et al. Adjuvant treatment with a mammalian target of rapamycin inhibitor, sirolimus, and steroids improves outcomes in patients with severe H1N1 pneumonia and acute respiratory failure. *Crit Care Med* 2014; 42: 313-21. <https://doi.org/10.1097/CCM.0b013e3182a2727d>
- Watanabe T, Watanabe S, Kawaoka Y. Cellular networks involved in the influenza virus life cycle. *Cell Host Microbe* 2010; 7: 427-39. <https://doi.org/10.1016/j.chom.2010.05.008>
- Xiao L, Haack KK, Zucker IH. Angiotensin II regulates ace and ACE2 in neurons through p38 mitogen-activated protein kinase and extracellular signal-regulated kinase 1/2 signaling. *A J Physiol Cell Physiol* 2013; 304: 1073-9. <https://doi.org/10.1152/ajpcell.00364.2012>
- Xu J, Sriramula S, Xia H, Moreno-Walton L, Culicchia F, Domenig O, et al. Clinical relevance and role of neuronal AT1 receptors in ADAM17-mediated ACE2 shedding in neurogenic hypertension. *Circ Res* 2017; 121: 43-55. <https://doi.org/10.1161/CIRCRESAHA.116.310509>
- Xu J, Zhao S, Teng T, Abdalla AE, Zhu W, Xie L, et al. Systematic comparison of two animal-to-human transmitted human coronaviruses: SARS-COV-2 and SARS-COV. *Virus* 2020; 12: 244. <https://doi.org/10.3390/v12020244>
- Xu P, Derynck R. Direct activation of tace-mediated ectodomain shedding by p38 MAP kinase regulates EGF receptor-dependent cell proliferation. *Mol cell* 2010; 37: 551-66. <https://doi.org/10.1016/j.molcel.2010.01.034>
- Ye Z, Wang Y, Colunga-Lozano LE, Prasad M, Tangamornsuksan W, Rochweg B, et al. Efficacy and safety of corticosteroids in covid-19 based on evidence for covid-19, other coronavirus infections, influenza, community-acquired pneumonia and acute respiratory distress syndrome: a systematic review and meta-analysis. *Cmaj* 2020; 192: 756-67. <https://doi.org/10.1503/cmaj.200645>
- Zarubin T, Jiahuai HA. Activation and signaling of the p38 MAP kinase pathway. *Cell Res* 2005; 15: 11-8. <https://doi.org/10.1038/sj.cr.7290257>
- Zhang Z, Chen L, Zhong J, Gao P, Oudit GY. Ace2/Ang-(1-7) signaling and vascular remodeling. *Sci China Life Sci* 2014; 57: 802-8. <https://doi.org/10.1007/s11427-014-4693-3>
- Zheng Y, Li R, Liu S. Immunoregulation with mTOR inhibitors to prevent covid-19 severity: a novel intervention strategy beyond vaccines and specific antiviral medicines. *J Med Virol* 2020; 92: 1495-500. <https://doi.org/10.1002/jmv.26009>
- Zhou F, Yu T, Du R, Fan G, Liu Y, Liu Z, et al. Clinical course and risk factors for mortality of adult inpatients with covid-19 in Wuhan, China: a retrospective cohort study. *Lancet* 2020; 395: 1054-62. [https://doi.org/10.1016/S0140-6736\(20\)30566-3](https://doi.org/10.1016/S0140-6736(20)30566-3)
- Zimmermann P, Curtis N. Coronavirus infections in children including covid-19: an overview of the epidemiology, clinical features, diagnosis, treatment and prevention options in children. *Pediatr Infect Dis J* 2020; 39: 355. <https://doi.org/10.1097/INF.0000000000002660>
- Zuniga EI, Liou LY, Mack L, Mendoza M, Oldstone MB. Persistent virus infection inhibits type I interferon production by plasmacytoid dendritic cells to facilitate opportunistic infections. *Cell Host Microbe* 2008; 4: 374-86. <https://doi.org/10.1016/j.chom.2008.08.016>