



Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Elsevier hereby grants permission to make all its COVID-19-related research that is available on the COVID-19 resource centre - including this research content - immediately available in PubMed Central and other publicly funded repositories, such as the WHO COVID database with rights for unrestricted research re-use and analyses in any form or by any means with acknowledgement of the original source. These permissions are granted for free by Elsevier for as long as the COVID-19 resource centre remains active.



Role of CCL2/CCR2 axis in the pathogenesis of COVID-19 and possible Treatments: All options on the Table

Mitra Ranjbar^a, Ali Rahimi^b, Zeinab Baghernejadan^b, Atousa Ghorbani^c, Hossein Khorramdelazad^{b,d,*}

^a Department of Infectious Disease, School of Medicine, Iran University of Medical Sciences, Tehran, Iran

^b Department of Immunology, School of Medicine, Iran University of Medical Sciences, Tehran, Iran

^c Department of Biology, East Tehran Branch, Islamic Azad University, Tehran, Iran

^d Department of Immunology, School of Medicine, Rafsanjan University of Medical Sciences, Rafsanjan, Iran

ARTICLE INFO

Keywords:

CCL2
CCR2
COVID-19
CXCL17
MDSCs
Immunotherapy

ABSTRACT

Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) is cause of the novel coronavirus disease (COVID-19). In the last two years, SARS-CoV-2 has infected millions of people worldwide with different waves, resulting in the death of many individuals. The evidence disclosed that the host immune responses to SARS-CoV-2 play a pivotal role in COVID-19 pathogenesis and clinical manifestations. In addition to inducing antiviral immune responses, SARS-CoV-2 can also cause dysregulated inflammatory responses characterized by the noticeable release of proinflammatory mediators in COVID-19 patients. Among these proinflammatory mediators, chemokines are considered a subset of cytokines that participate in the chemotaxis process to recruit immune and non-immune cells to the site of inflammation and infection. Researchers have demonstrated that monocyte chemoattractant protein-1 (MCP-1/CCL2) and its receptor (CCR2) are involved in the recruitment of monocytes and infiltration of these cells into the lungs of patients suffering from COVID-19. Moreover, elevated levels of CCL2 have been reported in the bronchoalveolar lavage fluid (BALF) obtained from patients with severe COVID-19, initiating cytokine storm and promoting CD163⁺ myeloid cells infiltration in the airways and further alveolar damage. Therefore, CCL2/CCR axis plays a key role in the immunopathogenesis of COVID-19 and targeted therapy of involved molecules in this axis can be a potential therapeutic approach for these patients. This review discusses the biology of the CCL2/CCR2 axis as well as the role of this axis in COVID-19 immunopathogenesis, along with therapeutic options aimed at inhibiting CCL2/CCR2 and modulating dysregulated inflammatory responses in patients with severe SARS-CoV-2 infection.

1. Introduction

In December 2019, the first cases of pneumonia due to an unknown infection were recorded in Wuhan, China [1]. Extensive research has shown that the cause of this infection is a new beta-coronavirus with many phylogenetic similarities to the severe acute respiratory syndrome coronavirus (SARS-CoV) that caused the 2003 outbreak in China [2]. The new SARS-CoV-2 infected millions of people worldwide, leading to an epidemic and a public health emergency of international concern by the World Health Organization (WHO) [3]. The latest data shows 614,561,191 confirmed COVID-19 cases, and 6,518,801 deaths had been reported to WHO (until 14 September 2022) [4].

The main clinical manifestations of SARS-CoV-2 infection are

pneumonia associated with lung injury with pulmonary edema and diffuse alveolar injury, leading to acute respiratory distress syndrome (ARDS), decreased blood O₂ saturation, and lethal hypoxemia [5]. Other complications in various body organs may occur in the form of thrombotic disorders, neurological complications, as well as gastrointestinal and renal disorders, which are often consequences of uncontrolled systemic inflammation and cytokine release syndrome (CRS) [6–10]. Based on available knowledge, the immune system and its components, especially inflammatory mediators, are involved in the pathogenesis of COVID-19 [11]. Chemokines are a group of these mediators that can be classified into four categories, CC, CXC, CX3C, and C, depending on the position of the conserved cysteine amino acid residues [12]. CCL2 belongs to the group of CC chemokines and is also known as monocyte

* Corresponding author at: Department of Immunology, School of Medicine, Rafsanjan University of Medical Sciences, Rafsanjan, Iran.
E-mail address: Khorramdelazad@gmail.com (H. Khorramdelazad).

chemoattractant protein-1 (MCP-1) due to its participation in monocyte recruitment and can bind to CC chemokine receptor type 2 (CCR2, CD192), triggering various downstream signaling pathways [13–16].

It has been revealed that CD163⁺ myeloid cells can be detected in damaged alveolar tissue of patients with COVID-19 [17–19]. Moreover, in a time-and-severity manner, lymphocyte frequency in blood circulation was reduced in patients with severe SARS-CoV-2 infection [20]. In contrast, the frequency of inflammatory HLA-DR^{low}CD163^{high} monocytes increased within the blood circulation, resulting in infiltration of these monocytes into the airways upon release of CCL2 by activated airway tissue-resident memory T cells [20]. Furthermore, to confirm the importance of monocytes in the immunopathogenesis of COVID-19, high plasma levels of several inflammatory mediators, including CCL2, granulocyte-macrophage colony-stimulating factor (GM-CSF), CXCL8 (Interleukin-8), interferon gamma-induced protein 10 (IP-10), and osteopontin were detected in patients with SARS-CoV-2 infection [21]. Besides, studies have proposed that hyper-inflammation is associated with the severity of COVID-19, characterized by upregulation of inflammatory cytokines and chemokines including IL-6, IL-10, tumor necrosis factor-alpha (TNF- α), IP-10, CCL2, and CCL3, leading to CRS [22–24]. Interestingly, an investigation demonstrated that human pluripotent stem cell-derived cardiomyocytes and adult cardiomyocytes could be productively infected by SARS-CoV-2, resulting in secretion of the CCL2 and following monocyte recruitment. Augmented CCL2 expression and monocyte infiltration was also detected in the hearts of SARS-CoV-2 infected hamsters [25].

Considering the importance of the CCL2/CCR2 axis in monocyte infiltration into the lung and further tissue damage, it seems that the CCL2/CCR2 axis can be a potential therapeutic target for the treatment and immunotherapy of patients with severe COVID-19 [26]. Although CCL2 has not yet been directly targeted for the treatment of COVID-19, evaluating existing options and learning from other CCL2-mediated inflammatory diseases could lead to the emergence of novel therapeutic approaches [27]. Moreover, despite multiple booster vaccinations, due to numerous virus mutations, new waves of the SARS-CoV-2 infection, such as Omicron, still infect people [28]. As a result, efforts to manage uncontrolled inflammation are still essential.

This review summarized the biology of CCL2 and its role in the immunopathogenesis of COVID-19 and discussed available treatment options targeting the CCL2/CCR2 axis.

2. CCL2 biology

2.1. Biostructure and biofunction

CCL2, also known as the small inducible cytokine A2, is a prominent member of the CC chemokine superfamily. CCL2 exerts its effect as an inflammatory chemokine, and it is essential for the recruitment of leukocytes, including monocytes, dendritic cells (DCs), and memory T cells during inflammation [29,30]. Furthermore, CCL2 regulates cancer progression through inflammatory responses [31]. The survival of different species through intense selective pressure is probably one of the reasons that CCL2 has developed rapidly in humans [29]. Like other chemokines, the crystal structure analysis of CCL2 reveals stable multimerization through its function. Upon the dimerization of CCL2, tetramers of this chemokine are generated by dimer-dimers interaction [32,33]. CCL2 was the first identified CC chemokine among other CCL family members, and it is well characterized in humans and other species. The gene of *CCL2* is located on chromosome 17 (chr.17, q11.2), which encodes a 13 kDa protein with 76 amino acids [34]. In mice, the *JE* gene is remarkably similar to the *CCL2* gene in humans, and the platelet-derived growth factor (PDGF) is known as a stimulator of the *JE* expression in fibroblasts [35]. According to sequence alignment assessments, CCL2 exhibits a high-level amino acid sequence homology among various species, with an extra 21 amino acids in rats and mice, compared to humans. However, CCL2 has a conserved sequence with

minimal sequence identity than other CCL family members. The human CCL2 amino acid sequence contains different sections, including the signal peptide, pyrrolidone carboxylic acid, and N-linked glycosylation asparagine, linked with two disulfide bonds [36]. The chemoattract potency of CCL2 can be slightly reduced by extreme glycosylation in its structure through inflammatory responses [37]. According to the protein modeling analysis, several α -helices and β -sheets confirmations are involved in the CCL2 structure formation [38].

A wide range of immune and non-immune cells can produce and release CCL2 spontaneously or by inducing other stimuli such as oxidative stress, cytokines, and growth factors. Mutational research revealed that the basic structure of CCL2 comprised two essential domains related to its biological function [39]. The primary domain includes the sequence from Thr-10 to Tyr-13, while the second location exists at amino acid residues 34 and 35. It is reported that permanent mutation at the position of residue 10 or 13 can hamper CCL2 function [40]. On the other hand, the necessity of the second location was demonstrated by the outcome of two mutations. There were two mutations, the first affecting a proline between Ser-34 and Lys-35 and the second affecting the Gly-Pro-His sequence. In addition, it has been mentioned that mutations of residues 28 and 30 can alter cell-type specificity [39].

The formation of the secondary structure of CCL2 illustrates four β -sheets sections, which contain residues 9–11 (β_0), 27–31 (β_1), 40–45 (β_2), and 51–54 (β_3). There are also two helical domains and a long helix that extends from residue 58 to 69 [37]. Furthermore, the position of amino acid residues from 6 to 16 through the protein structure plays a crucial role in the CCL2 dimerization [41]. The CCL2 interface is generated by the residues Glu50, Ile51, and Cys52, whereas particularly amino acid residues, such as Asn6, Ala7, Val9, Tyr13, Asn14, Phe15, and Thr16, are important for the formation of *N*-terminus which are necessary for its conformational structure and proper activity [32].

Evidence shows that inflammatory chemokines such as CCL2 contribute to defense against viral infections by recruiting innate and adaptive immune cells to the infection site, enhancing their cytotoxicity and capacity to release antiviral mediators. On the other hand, their overwhelming secretion following infection is the major reason for hyper-inflammation [42,43]. In this regard, CCL2 may be one of the main reasons for ARDS and even death in patients with severe SARS-CoV-2 infection [44].

2.2. Signaling

Chemokines exert biological effects via interactions with G-protein coupled receptors (GPCRs) and related signaling pathways [45] (Fig. 1). Several studies have indicated that five different inflammatory chemokines can activate CCR2, and CCL2 is one of the most important ligands for initiating the CCR2 signaling pathway [46–49]. CCR2 is a GPCR composed of a single polypeptide chain with seven hydrophobic transmembrane domains attributed to some intracellular and extracellular loops, an extracellular acidic domain from *N*-terminal with an intracellular serine/threonine C-terminal domain [50]. The extracellular region from the *N*-terminal of the receptor is the most essential for the high-affinity binding to specific ligands. The other three extracellular sites of the receptor are critical for activating subsequent intracellular signal transduction [51,52]. CCR2A and CCR2B are the two alternatively spliced variants of CCR2 with a different number of amino acids in their structure found in humans [53]. These two variants of CCR2 can activate various signaling pathways and participate in various functions. However, CCR2B is the main variant representing the most CCR2 expressed on the surface of different cells [34]. The binding and oligomerization of some compounds, such as glycosaminoglycan (GAG), are necessary for CCL2 and other chemokines to exhibit their function *in vivo* [54]. However, the affinity of the final binding is lower than the first attachment between CCL2 and the *N*-terminus of CCR2 [55].

There are two well-established CCL2 signaling pathways.

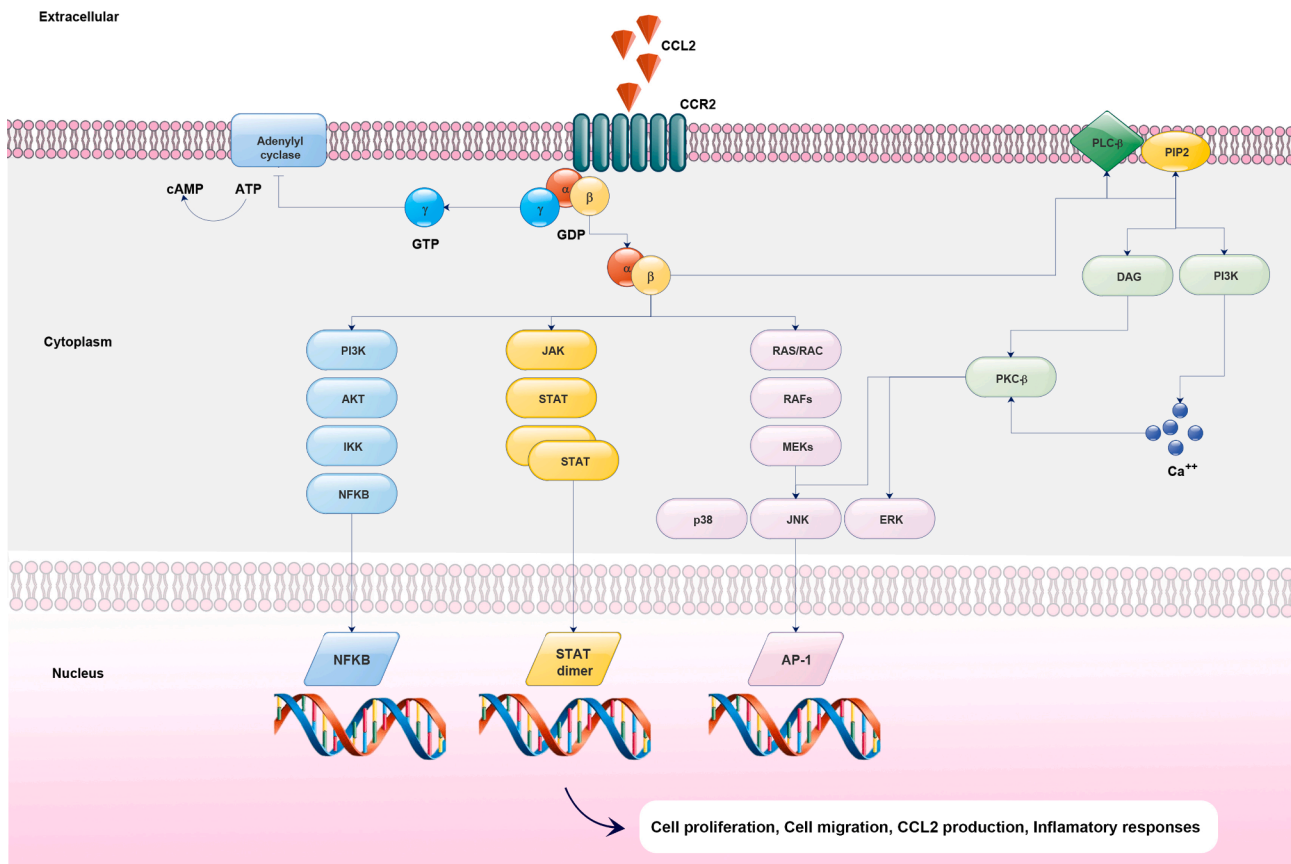


Fig. 1. CCL2/CCR2 signaling pathway. The ligation of CCL2 to CCR2 can activate various signaling pathways such as JAK/STAT, MAPK and NFκB, which finally induces the expression of various genes by activating transcription factors, leading to cell proliferation, migration of immune cells such as monocytes and macrophages, expression of inflammatory mediators, and hyperinflammation.

Inflammatory cytokines like tumor necrosis factor (TNF)- α can bind to their receptor and activate the nuclear factor- κ B (NF κ B) inhibitor (IKK) through the canonical pathway. IKK activation subsequently promotes the phosphorylation of NF κ B-bound inhibitor NF- κ B (IB), leading to IB degradation. Consequently, the translocation of activated NF- κ B homodimers through the nucleus can stimulate the transcription of the inflammation-related genes, including CCL2, TNF- α , and IL-6 [56,57]. Additionally, CCL2 signaling can be initiated via a noncanonical pathway through the expression of NF- κ B-independent CCL2, activated by PDGF or insulin [35]. It has been elucidated that normal insulin concentrations remarkably decrease both NF- κ B and CCL2 expression in human aortic endothelial cells [58]. It has been revealed that a noncanonical pathway can promote phosphatidylinositol 3-kinase (PI3K)-Akt signaling by augmenting Ras homolog enriched in the brain (RHEB) as the inhibitor of mTORC1-repressor. Subsequently, mTORC1 dephosphorylates forkhead box K1 (FOXK1) by protein phosphatase 2A (PP2A), resulting in CCL2 expression [59].

Interaction between the N-terminus site of CCL2 and CCR2 causes GDP to dissociate from $G_{\alpha i}$ and be replaced by intracellular GTP [50], and following these occurrences; the $G_{\alpha i}$ -GTP complex dissociates from the receptor and the generation of the $G_{\beta\gamma}$ heterodimer. As a result, $G_{\alpha i}$ blocks adenylate cyclase, while $G_{\beta\gamma}$ stimulates phospholipase C, leading to diacylglycerol and inositol 1,4,5-trisphosphate (IP3) formation. These events eventually result in the production of calcium from intracellular sources and the initiation of signaling cascades, such as the calmodulin-dependent protein kinase II (CaMKII), protein kinase C (PKC), PI3K, Akt, and ERK, involved in cell mobility, survival, gene transcription, and the activation of pro-nociceptive molecules [60,61]. $G_{\beta\gamma}$ components also stimulate PI3K and Akt, activate the polymerization of actin and pseudopod formation, and promote monocyte migration [62]. Furthermore,

it has been demonstrated that CCL2 significantly increases the p38 MAPK phosphorylation, which subsequently enhances transcription factor phosphorylation of some mediators, i.e., cAMP response element-binding protein (CREB). The CCL2/CCR2 axis influences Ca^{2+} influx and synaptic network function within hippocampal neurons [63]. Arachidonic acid (AA) is an essential second messenger through the GPCR signaling pathway during CCL2-induced chemotaxis. AA is synthesized from phospholipids through phospholipase A2 (cPLA2) enzyme function and plays an essential role as an inflammatory eicosanoid precursor. It has been reported that antisense cPLA2 deletion significantly inhibits CCL2-induced chemotaxis, suggesting that AA production is essential for monocyte locomotion and recruitment [64]. During chemotaxis, activated CCR2 interfaces directly with FROUNT, a specific clathrin heavy-chain protein, contributing to the development of clusters along the cell front [65]. Notably, abundant serine and threonine residues within the CCR2 C-terminal can be phosphorylated after interacting with the receptor, leading to receptor desensitization and eventually interrupting signal transduction [34].

3. Role of CCL2 in the immune system

Inflammatory responses could initiate upon the ligation of CCL2 to CCR2 on the surface of various cells, such as monocytes, resulting in leukocyte infiltration and T cell proliferation in inflammatory diseases, including atherosclerotic cardiovascular disease, related vasculopathy, and multiple sclerosis [66–70]. Moreover, CCL2 has a crucial role in inflammation and immune response, and binding to CCR2 initiates various signaling pathways to regulate the activation and migration of target cells.

3.1. Innate immunity

CCL2 is the first purified and best characterized human CC chemokine, identified because of its monocyte chemotactic property *in-vitro* in human cell lines [37]. CCL2 is believed to be identical to *JE*, a gene whose expression is induced in mouse fibroblasts by PDGF [35]. However, the human homolog, characterized as *CCL2*, was first purified from human cell lines based on its monocyte chemoattractant properties. Many cell types, including monocytes, macrophages, fibroblasts, endothelial, epithelial, smooth muscle cells (SMCs), mesangial, astrocytic, and microglial cells, produce CCL2 constitutively or after induction by oxidative stress, cytokines, or growth factors [71–74]. These cells are essential for antiviral immune responses in the peripheral circulation and tissues. However, monocyte/macrophages are the major sources of CCL2 [75,76]. The main inducers of CCL2 expression include pro-inflammatory cytokines [e.g., interleukin (IL)-1, IL-4, IL-6, TNF- α , interferon-gamma (IFN- γ)], growth factors [e.g., macrophage colony-stimulating factor (M-CSF), granulocyte-macrophage colony-stimulating factor (GM-CSF), PDGF, vascular endothelial growth factor (VEGF)], lipopolysaccharides, reactive oxygen species (ROS), oxidized low-density lipoprotein (oxLDL) and immune complexes [77] (Fig. 2). Conversely, transforming growth factor-beta (TGF)- β and retinoic acid may down-regulate CCL2 expression [78]. CCL2 regulates the migration and infiltration of monocytes, DCs, memory T lymphocytes, and NK cells

[30,79]. CCL2 mediates its effects through its receptor CCR2, and, unlike CCL2, CCR2 expression is relatively restricted to certain types of cells, including monocytes, T lymphocytes and vascular SMCs [37]. The interaction of CCL2 with CCR2 is crucial in inflammation and inflammatory-associated diseases and is involved in the innate immune response by recruiting monocytes into the site of inflammation [80]. There are two alternatively spliced forms of CCR2, namely, CCR2A and CCR2B, which differ only in their C-terminal tails [53]. CCR2A is the major isoform expressed by mononuclear cells and vascular smooth muscle cells [81], whereas monocytes and activated NK cells predominantly express the CCR2B isoform. CCR2A and CCR2B may activate different signaling pathways and exert different actions. For example, CCL2/CCR2A-mediated migration occurs without Ca²⁺ mobilization, but Ca²⁺ flux is induced in the CCR2B-positive cells [82,83]. Furthermore, CCL2 promotes the concurrent initiation of different sign falls; their effect on monocyte chemotaxis could be extraordinary. Some studies demonstrated that the p38 MAPK pathway is involved in the CCL2-mediated relocation of monocytes [84].

3.2. Adaptive immunity

Besides recruiting and directing leukocyte movement, several lines of evidence indicate that CCL2 participates in adaptive immunity, controlling the preferential differentiation of T helper (Th) lymphocytes

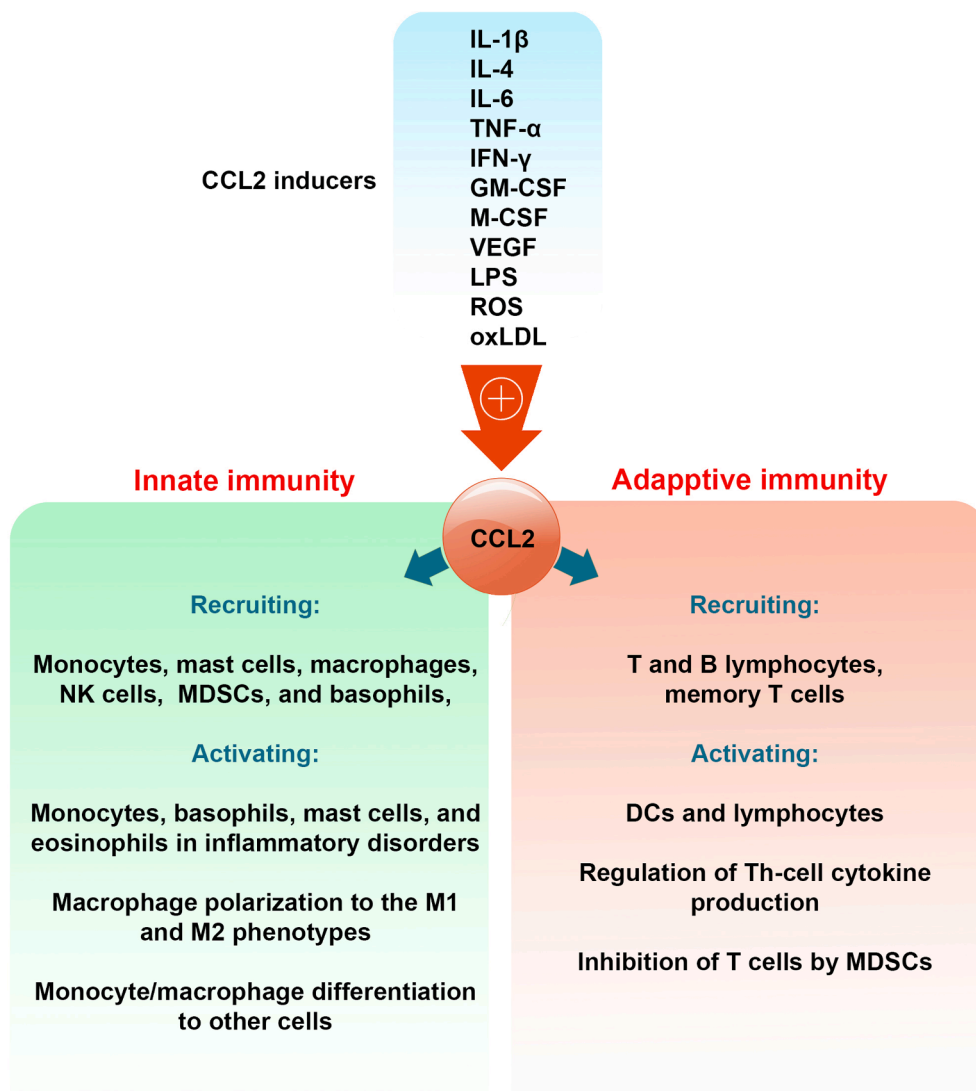


Fig. 2. CCL2 inducers and role of CCL2 in innate and adaptive immunity. Various mediators such as IL-1 β , IL-4, IL-6, TNF- α , IFN- γ , GM-CSF, M-CSF, VEGF, LPS, oxLDL, and ROS can lead to increased expression of CCL2. Following the increased CCL2 level, this chemokine can affect the innate and adaptive arms of the immune system. In general, CCL2 causes the recruitment and activation of various immune cells. It can also affect the function of monocytes and macrophages and cause their differentiation and phenotype alteration in different physiologic and pathologic states.

toward the Th1 or Th2 phenotype. Since IL-4 production is increased in cells that are given a primary T cell receptor (TCR) stimulus in the presence of CCL2, CCL2 may activate the IL-4 promoter [85]. Therefore, CCL2 expression is more associated with developing Th2 responses [32,86]. Unlike other chemokines of the C–C family, which trigger the Th1 phenotype upon their interaction with CCR5 on T-helper cells [34], CCL2 acts as a potent factor in the polarization of Th0 cells into the Th2 phenotype [87]. However, Th1 and Th2 responses can be promoted by CCL2 *in-vivo*, depending on other reported factors, such as CCL2 induction timing, target tissue, and type of pathogen [88]. CCL2 regulates the differentiation of monocytes into DCs and modulates Th1 immune response by selectively suppressing naïve T cells differentiation into the Th1 effector cells via regulating the release of DC-derived IL-12 [89]. Additionally, CCL2 is involved in cytokine production by naïve T cells [85]. It has been reported that the released CCL2 in the draining lymph nodes of the skin can be presented on the surface of high endothelial venules (HEVs) to recruit lymphocytes [90]. In addition, CCL2 participates in recruiting memory T-cells [30,91].

Neutrophils-derived CCL2 in a Th1 microenvironment has been suggested to be involved in Th1-associated adaptive responses [92]. These findings provide an important clue as to why there is a switch from Th1 to Th2 cytokine response in viral infections such as SARS-CoV-2 and the human immunodeficiency virus (HIV). The reciprocal inhibition between Th1 and Th2 cytokines is the major factor that induces Th2 differentiation and inhibits the development of IFN- γ -secreting cells [93]. In other Th2 immune-mediated diseases, such as asthma, CCL2 is expressed at high levels, and its neutralization in animal models ameliorated the disease [94]. Interestingly, CCR2 could have an anti-inflammatory action. As mentioned above, the proinflammatory role of CCR2 is dependent on antigen-presenting cells (APCs) and T cells, whereas the anti-inflammatory role of CCR2 is dependent on CCR2 expression on the surface of regulatory T cells (Tregs) [37].

4. CCL2/CCR2 axis in viral infections

Evidence reveals that CCL2 contributes to the pathogenesis of bacterial, fungal, and viral infection diseases, such as salmonella, *C. elegans*, severe acute respiratory syndrome (SARS), HIV, respiratory syncytial virus (RSV), human cytomegalovirus (HCMV), human rhinovirus (HRV), and simian-human immunodeficiency virus (SHIV) [95–97]. Like many inflammatory cytokines and chemokines, CCL2 expression is increased following infection to facilitate locomotion and leukocyte activation [98]. Following the migration of effector cells such as neutrophils and monocytes to the site of infection, inflammatory innate immune responses against pathogenic microorganisms begin to clear the infection. The regulated immune responses induced by chemokines prevent tissue damage due to hyperinflammation and microorganisms from escaping the immune system [97]. As mentioned earlier, CCL2 induces monocyte migration to the site of infection, which has also been reported in viral infections such as influenza, West Nile virus (WNV), and mouse cytomegalovirus (MCMV). In some infections, such as HIV, the role of CCL2 is more harmful than protective because recruited and infiltrated leukocytes become new targets of the virus for infection, resulting in more replication of HIV [99]. In addition, it has been shown that sometimes CCL2 can directly affect the replication of viruses and increase viral load [100]. In middle east respiratory syndrome (MERS) and SARS infections, CCL2 and CXCL10 are responsible for the recruitment of monocytes, macrophages, and T cells to the lungs [101–104].

In contrast, it has been revealed that CCL2 and CXCL10 could suppress the multiplication of lymphoid progenitor cells leading to lymphopenia in MERS and SARS infections [104–107]. Evaluation of inflammatory monocyte-macrophages (IMMs)-derived chemokines demonstrate that CCL2 dominantly supports the activation of these CCR2⁺ cells [108]. In patients with SARS infection, the bronchoalveolar lavage fluid (BALF) analysis showed that levels of CCL2 were significantly increased and associated with alveolar macrophage infiltration

[109–111]. Furthermore, CCR1, CCR5, and CCR2 could have a protective role in infected DCs with SARS-CoV. Therefore, impaired expression or function of these receptors might be associated with disease severity and mortality [112]. Previous studies also disclosed that CXCR3 and CCR2 were upregulated in the RSV, associated with disease severity [113]. Additionally, an experimental study used a CCR2 antagonist in influenza H1N1 infected mice, and findings showed that antagonizing CCR2 could induce the activity of nucleoprotein-specific cytotoxic T cells [114].

It seems that the CCL2/CCR2 axis could have a dual role in the pathogenesis of viral infections, and targeting this axis might be a potential therapeutic approach to reduce hyperinflammation (Fig. 3).

5. The CCL2/CCR2 axis in COVID-19

SARS-CoV-2 appears the same biological and viral characterization as SARS-CoV and MERS-CoV; therefore, the chemokine signature of COVID-19 patients is assumed to have similar inflammatory mediators; however, the transmission rate of SARS-CoV-2 is higher, along with lower mortality rate compared to MERS and SARS [115]. Furthermore, hyperinflammation triggered by other viral pathogens, including influenza H1N1 and avian H5N1, may assist in identifying the effect of chemokines implicated in forming inflammatory responses against SARS-CoV-2 infection [116–118]. Therefore, recognizing the SARS-CoV-2 chemokine signature and distinguishing it from non-COVID-19 pathogenic microbial ARDS would minimize complications and decrease mortality by developing interventional techniques [119]. Although resident alveolar macrophages are beneficial in the early stages of the disease, infiltrating monocytes and macrophages are essential in the progression of COVID-19 [115]. Infiltrated monocytes as the major leukocytes migrated into the infected lungs, and excessive secretion of cytokines and chemokines by these immune cells can lead to remarkable severe lung inflammation in SARS-CoV-2 patients [109,120,121]. According to the comparison of the SARS-CoV-2 chemokine profile with SARS-CoV and MERS-CoV, CCL2 has a critical contribution to the pathogenesis of pulmonary disease in all coronaviruses (Fig. 4). Nevertheless, different behaviors of SARS-CoV-2 and SARS-CoV in isolated human lung tissues is notably associated with increased capacity of replication and infection, suggesting the dissimilar pattern of cytokine and chemokine production in these viruses [122].

Although various chemoattractant mediators induce monocyte migration, CCL2 and CCL7 are often expressed quickly by stromal and immune cells upon activation of pattern recognition receptors (PRR) or cytokine secretion [123]. As mentioned above, CCL2 is the predominant chemokine related to COVID-19 severity, and it is increased during the primary infection and is significantly enhanced during the late stages in expired cases [124,125]. CCL2 is primarily produced in the lungs by alveolar macrophages, T cells, and endothelial cells, while CCR2 is mainly upregulated on the surface of monocytes, macrophages, and T cells in an inflammatory condition [126]. Following the expression of CCR2 in extracellular matrix (ECM) glycosaminoglycans (GAGs), CCL2 can stimulate monocyte recruitment into the infected lungs, where they trigger calcium influx, produce oxygen radicals, and express integrin in COVID-19 patients [123,127,128]. The CCL2/CCR2 axis also participated in mast cell progenitors migration during inflammatory responses *in vitro* and *in vivo* [129]. Histamine and leukotrienes produced by mast cells interact to promote T helper 2 (Th2) polarization [130]. Correspondingly, CCR2⁺ blood monocytes significantly increase neutrophil accumulation, demonstrating the interaction between monocytes and neutrophils through leukocyte efflux during respiratory inflammation [131].

Moreover, CCL2 has been stimulating fibroblast procollagen production [132]. These effects of CCL2 may result in fibroproliferative disorders in ARDS [130]. According to animal model studies, the administration of CCL2 antagonists could minimize pulmonary hyperinflammation and improve the longevity of infected mice [114].

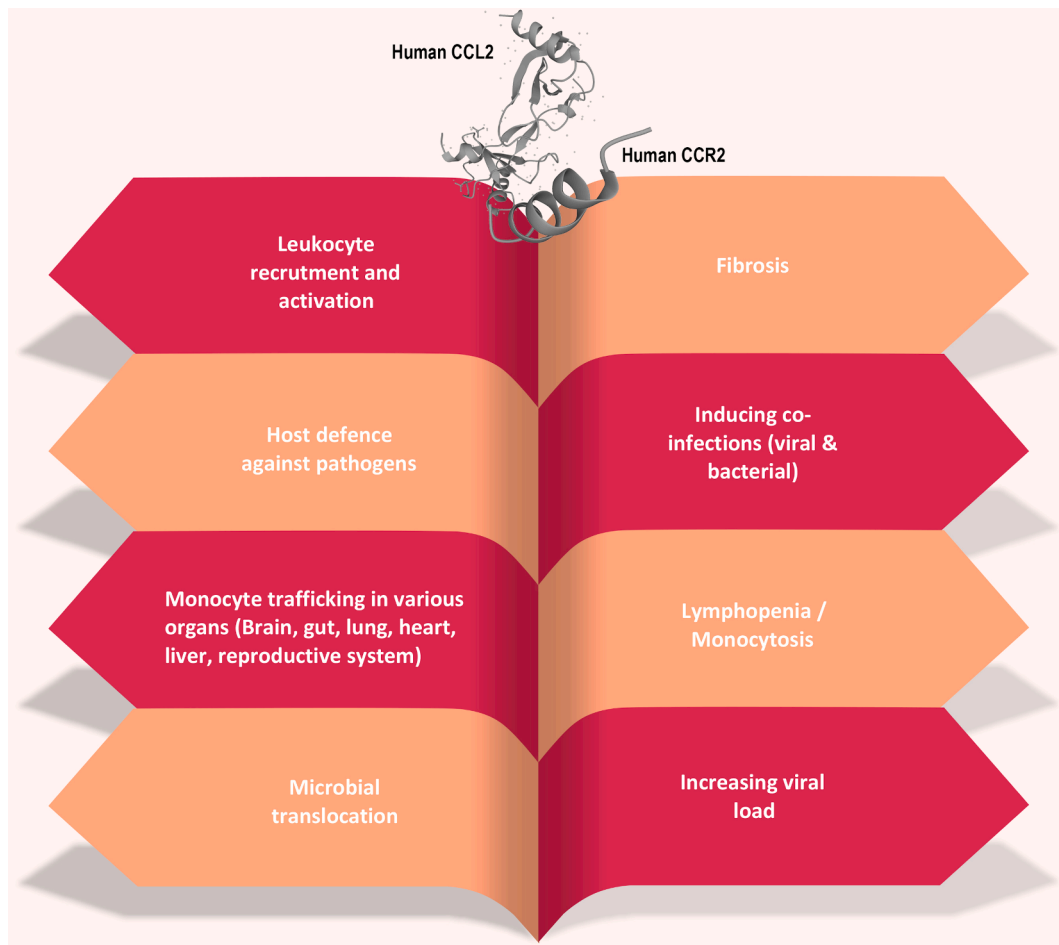


Fig. 3. Consequences of the CCL2/CCR2 axis activation in viral infections. When different pathogenic viruses enter the body, host inflammatory processes are activated. Virus-infected cells in various organs can produce CCL2, leading to the recruiting and activation of leukocytes, mainly monocytes and macrophages, into the brain, lungs, liver, gut, heart, and reproductive system. With the infiltration of these inflammatory cells, severe inflammatory responses are formed at the site of infection. Increased CCL2 levels can also lead to tissue fibrosis. Monocytosis and lymphopenia, along with the induction of bacterial or viral co-infections, are other outcomes of increased expression of CCL2 following viral infections.

Although recruited macrophages initially express a high level of CCL2, CXCL10 and CCL3 are augmented in advanced stages of the disease with high severity [133]. Compared to convalescent individuals, symptomatic patients revealed a higher level of CCL2, leading to monocytes and macrophages locomotion into the lungs [134]. It has been reported that among the chemokines, which were considerably increased in fatal COVID-19 cases, CCL2 was increased equally in moderate to severe patients during the primary infection and maintained at steady levels after that in moderate cases. Additionally, CCL2 levels were elevated in the late stages of the disease [124]. Furthermore, transcriptional investigations on post-mortem lung tissue patients with COVID-19 revealed a substantial upregulation from the CCL2 coding gene [135]. COVID-19 patients admitted to ICU exhibited higher levels of CCL2 than patients with mild SARS-CoV-2 infection [24,136].

In contrast with the exacerbating effect of the CCL2/CCR2 axis during the pulmonary inflammation in patients with various severity of COVID-19, an investigation demonstrated that during the mouse-adapted SARS-CoV-2 infection, CCR2 reduced the viral load and the weight of animals through inflammatory responses. In this study, *Ccr2*^{-/-} mice represented a higher level of neutrophils and inflammatory cytokines than wild-type mice, probably due to the excessive viral load in their lungs. Therefore, the CCL2/CCR2 axis in the mouse-adapted SARS-CoV-2 can promote cytokine secretion and neutrophil recruitment into the lung, leading to viral load restriction [137].

Another crucial issue in the pathogenesis and vaccination of COVID-

19, which is related to CCL2, is the increase in the frequency of monocytic myeloid-derived suppressor cells (M-MDSCs) and the recruitment of these cells, which disrupt the host's antiviral responses in a chronic inflammation condition. On the other hand, how MDSCs influence immunization following COVID-19 vaccination remains to be determined [138]. An investigation reported that the number of M-MDSCs was significantly higher in the blood of patients with severe COVID-19 than in control subjects. These immunosuppressive cells can suppress effector T cells and the release of IFN γ by increasing the expression of arginase-1 (Arg-1) and IL-6 [139]. Regarding the role of CCL2 in the recruitment of M-MDSCs, it can be argued that inhibiting this chemokine or its receptor may help reduce the migration of M-MDSCs and increase the effectiveness of vaccination and antiviral responses of the immune system [140–142].

Altogether, it seems that understanding the dual effect of the CCR2/CCL2 axis on disease severity and immune responses priming is essential to designing different therapeutic strategies and vaccines for patients with COVID-19.

6. The CCL2/CCR2 axis in the different variants of SARS-CoV-2

In addition to Alpha (B.1.1.7), Beta (B.1.351), Gamma (P.1), and Delta (B.1.617.2) SARS-CoV-2 variants of concern (VOCs), Omicron (B.1.1.529) has been added to the list [143]. The lineages B.1.1.7, recognized first in the United Kingdom, and B.1.351, recognized first in

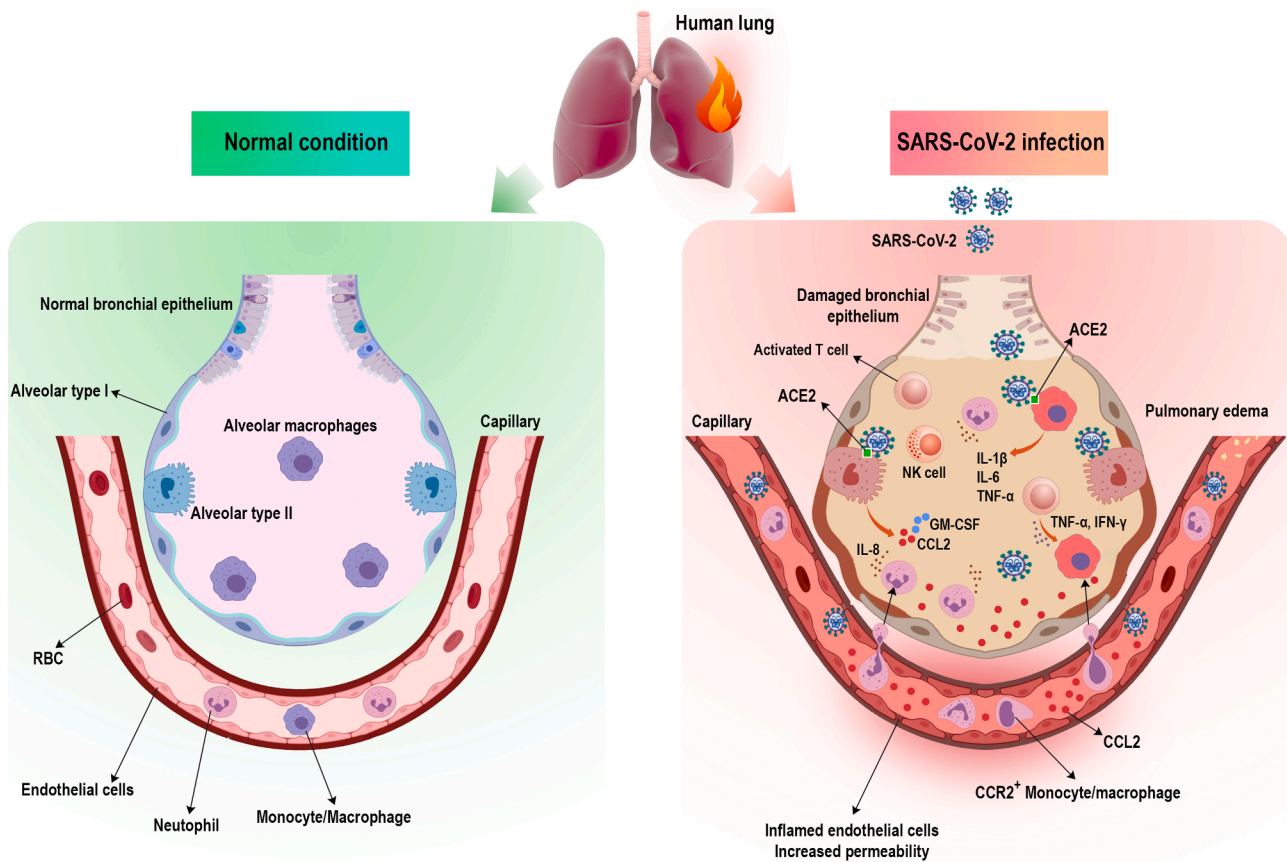


Fig. 4. Normal and inflamed lung before and after SARS-CoV-2 infection. After entering the SARS-CoV-2, it usually enters the lungs and infects immune and non-immune cells such as macrophages and type 2 alveolar cells that express the ACE2 receptor. One of the critical events in infection control is the migration of inflammatory cells to the site of infection. In the case of viral infections such as SARS-CoV-2, neutrophils and monocytes are the first cells that migrate to the damaged lung tissue. Moreover, CCR2⁺ cells, such as monocytes and macrophages, migrate from circulation to the lung alveoli, where CCL2 is overexpressed by resident cells, leading to activation of infiltrated leukocytes and lung inflammation. Neutrophils, macrophages, and T cells also aggravate this situation by releasing inflammatory mediators such as IL-1 β , IL-6, IFN- γ and TNF- α . Alveolar macrophages have a protective and anti-inflammatory role in normal conditions as well as in the initial stages of SARS-CoV-2 infection; nonetheless, with the severity of the disease, the phenotype of infiltrated macrophages and monocytes changed to inflammatory.

South Africa, have been categorized as VOCs according to the transmissibility, disease severity, and immune escape capability [144,145]. Mutations in the spike protein receptor-binding domain (S-RBD) may also contribute to their adaptation to new hosts [146,147].

Past lessons from coronaviruses show that the level of IL-6, CCL2, CXCL1, CXCL5, and CXCL10 is higher in SARS-CoV-2 than in SARS-CoV-1 [148]. According to animal and clinical studies, infection with SARS-CoV-2 variants stimulated the production of a wide range of pro-inflammatory cytokines, including IL-1 α , IL-1 β , IL-2, IL-4, IL-7, IL-17, TNF- α , IFN- γ , CCL2, CCL3, CCL4, and CXCL1 [149–153]. Moreover, researchers found that levels of CCL2, IL-8, and CXCL10 were significantly increased in patients infected with the ancestral Wuhan strain than in Alpha, Delta and Omicron variants [154].

Investigating the lung tissue levels of CCL2 showed that the concentrations of this CC chemokine were elevated following the infection of mice with B.1.1.7 and B.1.351 SARS-CoV-2 variants [155]. Another preclinical study reported that serum levels of CCL2 and CXCL10 were lower in Beta variant-infected mice at the beginning of infection because these animals produced significantly lower levels of CCL2 and CXCL10 than wild-type mice at two days post-infection. However, CCL2 and CXCL10 levels gradually increased over time in the studied mice [156]. According to an early study in K18-hACE2 mice, infection by 614D variant enhanced pulmonary levels of CCL2 and CCL5, infiltrating myeloid and activated CD8⁺ T cells in the lungs [150]. In this regard, examining the pattern of cytokines secreted in the lungs and livers of K18-hACE2 mice disclosed that the inflammatory profile in mice infected by

B.1.1.7 and B.1.351 variants was entirely different from those infected with early SARS-CoV-2 strains bearing 614D or 614G because the expression of CCL2 and CCL5 were significantly upregulated in liver and lung of the studied animals. Therefore, CCL2 and CCL5 could be the tissue-specific chemokine signatures for mice infected with 614D and 614G variants [157].

In patients with moderate SARS-CoV-2 infection, it has been reported that CCL2 levels in the Wuhan variant are significantly higher than in Alpha, Delta, and Omicron variants [154]. The comparison of CCL2 levels in the same patients shows that after the Wuhan variant, patients with Delta and Alpha variants have higher concentrations than the Omicron variant, respectively. Interestingly, in the severe form of COVID-19, the levels of CCL2 in patients with Wuhan variants, followed by Delta and Omicron, respectively, show a higher expression than the Alpha variant, although this difference is not significant. These findings indicated that although the rate of CCL2-mediated inflammatory responses in the Omicron variant is lower in mild and moderate forms of the disease, with the severity of the disease, these responses can increase at the level of the Delta variant and lead to severe symptoms in COVID-19 patients [154].

It was also discovered that deletion variants of open reading frame 6 (ORF6) could significantly induce the host's inflammatory response without affecting virus replication. In this variant, the gene expression of CCL2 was upregulated, inducing hyperinflammation. Moreover, the expression of CCL2 in the 26-nucleotide deletion variant (D26) was higher than in the 34-nucleotide deletion (D34) variant [158].

Collectively, CCL2 concentrations can vary significantly depending on SARS-CoV-2 variants, which implies that viral protein mutations affect immune responses at the cellular and molecular levels.

7. Therapeutic approaches based on CCL2/CCR2 axis targeting

As discussed before, the role of this chemokine is critical in inflammation because following the ligation of CCL2 to CCR2 and activation of signaling pathways, migration of monocytes initiated that play a protective role in the clearance of pathogen microorganisms [92,159,160]. This section summarized the outcomes of the CCL2/CCR2 axis inhibition using various available compounds and drugs, as well as the possibility of employing these inhibitors in patients with COVID-19 (Table 1).

7.1. CCL2/CCR2 inhibitors in Non-viral pathological conditions

Anti-CCL2 monoclonal antibodies, including carlumab and ABN912, are a group of CCL2 inhibitors that bind and neutralize human CCL2 [161]. In patients with rheumatoid arthritis (RA), using ABN912 worsens the disease because the CCL2 serum levels increase in these patients following ABN912 therapy [162]. This may occur in a wide range of antibody-targeted therapies. Carlumab is another anti-CCL2 antibody. The effectiveness of this inhibitor is also debatable because it could not bring satisfactory results in several inflammatory diseases, such as idiopathic pulmonary fibrosis [163], castration-resistant metastatic prostate cancer [164], and inflammatory-induced solid tumors [165]. Carlumab and ABN912 have not been used in patients with COVID-19, which can be due to the lack of effectiveness of this drug in other inflammatory-based diseases.

Ingramon is a synthetic peptide antagonist of CCL2. This peptide can suppress the chemotactic effects of CCL2 in a glioma-conditioned medium, reducing monocyte's locomotion [166]. Another study showed that treatment with Ingramon was associated with a less noticeable rise of high-sensitivity C-reactive protein (hsCRP) and reduced CCL2 and fibrinogen plasma levels in patients with coronary disorders. This study claimed that Ingramon did not affect CCL2 dimerization or interaction of cell receptors with CCL2. However, this synthetic peptide suppressed CCL2 binding to heparin. Therefore, the anti-inflammatory action of Ingramon may be due to impairing the interaction of CCL2 with glycosaminoglycans [167]. A study on patients with osteoarthritis (OA) showed that the CCL2/CCR2 axis mediates monocyte-derived inflammation and cartilage destruction [168]. In this regard, Bindarit, another synthetic CCL2 inhibitor, was used to treat mice with OA. The outcome showed that blockade of CCL2 could significantly decrease macrophage infiltration in the synovium and further cartilage damage in OA mice [168].

Since CCL2 is overexpressed in breast cancer tissues, and patients with high expression of CCL2 have early recurrence or poor prognosis in breast cancer [169–172], a study evaluated the impact of propagermanium (PG), a CCL2 inhibitor, in perioperative patients with primary breast cancer [173]. They found that IL-6 levels were downregulated in treated patients with PG dose-dependently. Consistent with these data, a mouse model of metastatic breast cancer showed that IL-6 levels were downregulated by anti-CCL2 neutralizing antibodies, resulting in inhibition of metastasis [174]. Concerning the critical pathologic role of IL-6 in hyperinflammation and the formation of cytokine storm COVID-19, CCL2 blockade by PG and other agents might be beneficial for the COVID-19 treatment by inhibiting monocyte/macrophage recruitment and downregulating IL-6 levels [11].

TNF- α inhibitors are another group of agents that might be effective in COVID-19 management by reducing inflammation [175]. Interestingly, TNF- α inhibitors such as can etanercept and adalimumab inhibit CCL2 production through epigenetic regulation in THP-1 monocytes [176]. Mechanistically, TNF- α inhibitors decrease NFKB-associated acetyltransferases, including p300, CBP, and PCAF, reducing the acetylation of H3 and H4 histones in the CCL2 promoter. Moreover, the

expression of the WDR5 and SMYD2 methyltransferases down-regulated by TNF- α inhibitors, inducing trimethylation of H3K4, H3K27, H3K36 and H3K79 in the mentioned promoter [176]. Diallyl disulfide (DADS), the organo-sulfur compound found in garlic, is another CCL2 inhibitor that can be effective in inflammatory disorders. An investigation reported that DADS decreased TNF- α -induced CCL2 production in MDAMB-231 cells [177]. Another study explored the mechanism by which DADS attenuates TNF- α -induced CCL2 production and found that this compound inhibits TNF- α -induced CCL2 production primarily by reducing the expression of IKK ϵ and pERK, impairing MAPK/ERK, and NFKB pathway signals [178]. Accordingly, the DADS mechanism of action is similar to TNF-inhibitors and influences CCL2 production by creating a disturbance in upstream signaling pathways involved in CCL2 expression [179]. Since anti-TNFs have been effective in reducing the severity of the disease in patients with COVID-19, perhaps these compounds can be used together with other CCL2/CCR2 inhibitors to make a synergistic therapeutic effect [180].

Bindarit (AF 2838) or 2-methyl-2-[[1-(phenylmethyl)-1Hindazol-3-yl]methoxy] propanoic acid is a small anti-inflammatory molecule that exhibits an inhibitory effect on CCL2, CCL3, and CCL8 production by down-regulating NF- κ B pathway [181,182]. Efficacy of Bindarit has been revealed in various experimental and clinical inflammatory conditions, such as Alzheimer's disease (AD) [183], experimental autoimmune encephalomyelitis (EAE) [184], diabetes-associated periodontitis [185], diabetic nephropathy [186], and lupus nephritis [186]. Accordingly, Bindarit might have the same effect in COVID-19, in which hyperinflammation plays a critical role.

Blockade of CCL2 using L-RNA aptamer (Spiegelmer) is another potential therapeutic approach to manage inflammatory states such as COVID-19. L-RNA aptamers are nuclease-resistant RNA-like molecules constructed from L-ribose units that neutralize the biologic functions of target molecules [187]. Because of their high biostability without any further chemical modifications, L-RNA aptamers are considered potential drugs in various diseases [188,189]. The effect of CCL2 blockade was examined in autoimmune-prone MRL ^{lpr/lpr} mice using pegylated mNOX-E36, a Spiegelmer that binds murine CCL2 with high affinity and neutralizes its action *in vitro* and *in vivo* [190]. This study found that mNOX-E36-3'PEG effectively improves the mice's lupus nephritis, autoimmune peribronchitis, and lupus-like skin disease.

A small heterodimer partner (SHP, NR0B2) is a transcriptional repressor that inhibits the transcription of its downstream target genes, such as CCL2 [191–194]. A recent study discovered 5-(diethylsulfamoyl)-3-hydroxynaphthalene-2-carboxylic acid (DSHN), a novel agonist for SHP that acts as a transcriptional activator for SHP [191]. It was reported that activation of SHP by DSHN inhibited hepatocellular carcinoma (HCC) cell migration and invasion by suppressing CCL2 expression. According to this data, it is expected that SHP and its agonists like DSHN be effective in inflammatory disorders such as COVID-19. However, further studies are required in patients with SARS-CoV-2 infection.

Because of the critical role of CCR2 in the recruitment of inflammatory monocytes, the therapeutic potential of CCR2 antagonists for preventing and treating inflammatory, infectious, and autoimmune diseases has always been of interest to researchers. Although CCR2 antagonists could not succeed in clinical trials [179], using CCR2 inhibitors achieved promising outcomes in cancer settings.

Experimental and clinical studies on solid tumors and peritonitis demonstrated that a group of small molecules, such as BMS-813160 and BMS-687681, can inhibit CCR2 and CCR5 in a dual-function manner [195,196]. These CCR2/CCR5 antagonists suppress the migration and infiltration of monocytes and macrophages, reducing inflammatory responses. Since CD14⁺, CD16⁺, and CCR5⁺ monocytes are involved in the pathogenesis of COVID-19, using these dual-antagonists could be beneficial in managing hyperinflammation in SARS-CoV-2 infected patients. In this regard, another study reported that by inhibiting the CCR5 receptor with leronlimab, a CCR5-specific antibody, plasma levels of IL-

Table 1

List of available therapeutic agents for targeting the CCL2/CCR2 axis.

	Drug or compound	Type of study and disease	Mechanism of action and consequences	Ref
<i>CCL2 inhibitors</i>	ABN912	Human / RA	<ul style="list-style-type: none"> o Inhibiting CCL2 o Worsening the disease because the CCL2 serum levels increase following the therapy 	[162]
	Carlumab	Human / idiopathic pulmonary fibrosis, castration-resistant metastatic prostate cancer, and inflammatory-induced solid tumors	<ul style="list-style-type: none"> o Inhibiting CCL2 o It was not effective in the mentioned disorders 	[163-165]
	Ingramon	<i>In vitro</i> / GliomaHuman/ Coronary disorder	<ul style="list-style-type: none"> o Inhibiting CCL2 o Suppressing the chemotactic effects of CCL2 in a glioma-conditioned medium, reducing monocyte's locomotion o Less noticeable rise of hsCRP and reduced CCL2 and fibrinogen plasma levels D o Cannot affect CCL2 dimerization or interaction of cell receptors with CCL2 o Suppressing CCL2 binding to heparin o Impairing the interaction of CCL2 with glycosaminoglycans 	[166,167]
	Bindarit	Animal/ OA	<ul style="list-style-type: none"> o Decreasing macrophage infiltration in the synovium and further cartilage damage 	[168]
	Propagermanium (PG)	Human/Metastatic breast cancer	<ul style="list-style-type: none"> o A CCL2 inhibitor o Decreasing IL-6 levels dose-dependently 	[173]
	Etanercept and adalimumab	<i>In vitro</i> / Inflammatory condition	<ul style="list-style-type: none"> o Anti-TNF o Inhibiting CCL2 production through epigenetic regulation in THP-1 monocytes o Decrease NFkB-associated acetyltransferases, including p300, CBP, and PCAF, reducing the acetylation of H3 and H4 histones in the CCL2 promoter o Down-regulating the expression of WDR5 and SMYD2 methyltransferases o Inducing trimethylation of H3K4, H3K27, H3K36 and H3K79 in the CCL2 promoter 	[176]
	Diallyl disulfide (DADS)	<i>In vitro</i> / Inflammatory condition	<ul style="list-style-type: none"> o A CCL2 inhibitor o Decreasing TNF-α-induced CCL2 production in MDAMB-231 cells o Inhibiting TNF-α-induced CCL2 production primarily by reducing the expression of IKKϵ and pERK o Impairing MAPK/ERK and NFkB pathway 	[177,178]
	Bindarit (AF 2838)	<i>In vitro</i> / <i>in vivo</i> /AD, EAE, diabetes-associated periodontitis, diabetic nephropathy, and lupus nephritis	<ul style="list-style-type: none"> o Inhibiting CCL2, CCL3, and CCL8 production o Down-regulating NFkB pathway 	[181-186]
	L-RNA aptamer (Spiegelmer)	<i>In vivo</i> / Autoimmune-prone MRL ^{lpr/lpr} mice	<ul style="list-style-type: none"> o Blocking CCL2 blockade o Neutralizing murine CCL2 o Improving the mice's lupus nephritis, autoimmune peribronchitis, and lupus-like skin disease 	[190]
	DSHN	<i>In vitro</i> / HCC	<ul style="list-style-type: none"> o A novel agonist for SHP that acts as a transcriptional activator for SHP o Activation of SHP by DSHN inhibited HCC cell migration and invasion by suppressing CCL2 expression 	[191-194]
Anti-CCL2 antibody	Animal study18-hACE2 mouse model	<ul style="list-style-type: none"> o Inhibiting CCL2 delayed virus-induced death in mice infected with B.1.351 SARS-CoV-2 variant 	[156]	
<i>CCR2 inhibitors</i>	BMS-813160 and BMS-687681	Human/AnimalSolid tumors, peritonitis	<ul style="list-style-type: none"> o CCR2/CCR5 antagonists o Inhibiting CCR2 and CCR5 in a dual function manner o Suppressing the migration and infiltration of monocytes and macrophages o Reducing inflammatory responses. 	[195,196]
	PF-04178903	Animal/Influenza-infected mice	<ul style="list-style-type: none"> o A CCR2 antagonist o Reducing mortality and improving the frequency of cytotoxic CD8⁺ T cells, hypothermia and weight loss o Reducing the levels of total protein, albumin, and lactose dehydrogenase activity in bronchoalveolar lavage fluid o Cannot influence the viral titers and <i>Streptococcus pneumoniae</i>-induced secondary infection o Cannot influence the anti-influenza-neutralizing antibodies 	[114]
	RS504393	Animal/ mice model of lipoatrophic diabetes <i>In silico</i>	<ul style="list-style-type: none"> o A CCR2 antagonist o Reducing CCL2/CCR2-mediated and adipose tissue-independent inflammation o Improving metabolic processes o Binding to the S-RBD of SARS-CoV-2 and ACE2 receptors o Reducing monocyte and macrophage infiltration o Reducing chronic inflammation o Inhibiting the virus and its specific receptor 	[198,202]
	CCX140-B	Human/ patients with type 2 diabetes with nephropathy	<ul style="list-style-type: none"> o A CCR2 antagonist o Inhibiting CCL2-dependent monocyte activation and migration 	[203]

(continued on next page)

Table 1 (continued)

Drug or compound	Type of study and disease	Mechanism of action and consequences	Ref
Extracellular loop 1 inverso (ECL1i)	Animal/ murine models of peritonitis and multiple sclerosis	<ul style="list-style-type: none"> o Reduce CCR2⁺ monocyte-induced inflammation and had a reno-protective impact o A potent, allosteric, non-competitive, and selective CCR2 inhibitor o Decreasing inflammatory responses in murine models of peritonitis and multiple sclerosis 	[215]
Genicriviroc (CVC)	Human/COVID-19	<ul style="list-style-type: none"> o A dual CCR2/CCR5 antagonist with a potent antiviral activity o Attenuating or repressing dysregulated inflammatory responses as well as fibrosis 	[216]
CAS445479-97-0	<i>In vitro</i> /lung adenocarcinoma A549 cells	<ul style="list-style-type: none"> o A CCR2 antagonist o significantly suppressed motility and invasion ability of CCR2⁺ tumor cells 	[204]
Natural compounds Progesterone (P4)	Human/ COVID-19	<ul style="list-style-type: none"> o Inhibiting the production of pro-inflammatory cytokines and chemokines such as IL-1β and TNF-α, and CCL2 o Progesterone therapy might effectively treat male patients with moderate to severe COVID-19 	[214]
Curcumin	<i>In vitro/in vivo</i> /inflammatory diseases	<ul style="list-style-type: none"> o Down-regulating of some critical elements involved in cellular and molecular pathways, such as NFκB o Inhibiting the production of CCL2 by osteoblastic cells, blood monocytes, and alveolar macrophages o Antiviral effect o ACE2 inhibition in SARS-CoV2 infection o Inhibiting CCL2 production in LPS-stimulated THP-1 cells 	[224-227,229,230]
<i>Lippia sidoides</i> and <i>Terminalia glabrescens</i> extracts	<i>In vitro</i> / Inflammation	<ul style="list-style-type: none"> o ACE2 inhibition in SARS-CoV2 infection o Inhibiting CCL2 production in LPS-stimulated THP-1 cells 	[217]
<i>Shufeng Jiedu</i> capsule (SFJDC)	Human/ upper respiratory tract infections	<ul style="list-style-type: none"> o Antiviral, anti-inflammatory, antipyretic, and immune regulatory activity o Its most potent compounds are quercetin, kaempferol, luteolin, wogonin, 7-Methoxy-2-methyl isoflavone, naringenin, beta-sitosterol, stigmasterol, physovenine, and formononetin o CCL2 is one of the main targets of SFJDC 	[218-221]

6 were remarkably reduced, and the CD4/CD8 ratio was restored. Furthermore, SARS-CoV2 plasma viremia decreased, and CD8⁺ cytotoxic T cells' frequency significantly increased following the treatment [197].

It has been reported that the CCL2/CCR2 axis is involved in hepatic macrophage infiltration and insulin resistance by inducing a chronic inflammatory condition [198]. Recent studies showed that patients with COVID-19 and diabetes had a higher absolute neutrophil count and lower lymphocyte count. Moreover, the levels of ferritin, erythrocyte sedimentation rate (ESR), lactate dehydrogenase (LDH), C-reactive protein, CCL2, IL-6, IL-8 and interferon beta 1 (IFN β 1) were higher in diabetic patients with COVID-19 [199–201]. RS504393 is another CCR2 antagonist, and it has been revealed that administration of this antagonist could reduce CCL2/CCR2-mediated and adipose tissue-independent inflammation, improving metabolic processes in mice models of lipotrophic diabetes [198]. Surprisingly, an *in silico* study shows that RS504393 can also bind to the S-RBD of SARS-CoV-2 and angiotensin-converting enzyme 2 (ACE2) receptors [202]. Therefore, it may be possible to use this antagonist in treating diabetic patients with COVID-19 to reduce monocyte and macrophage infiltration, reduce chronic inflammation, and inhibit the virus and its specific receptor. CCX140-B is another small molecule CCR2 antagonist that inhibits CCL2-dependent monocyte activation and migration [203]. In a randomized clinical trial, the clinical efficacy of CCX140-B was evaluated in patients with type 2 diabetes with nephropathy; outcomes indicated that CCX140-B could reduce CCR2⁺ monocyte-induced inflammation and had a reno-protective impact [203]. It has been reported that treating lung adenocarcinoma A549 cells with (CAS445479-97-0), a CCR2 antagonist significantly suppressed their motility and invasion ability [204]. This study suggested the potential therapeutic role of the CCR2 inhibition of CCR2⁺ tumor cells in patients with non-small cell lung cancer (NSCLC).

7.2. CCL2/CCR2 inhibitors in viral infections

Studies on viral infections have shown that neutralizing CCL2 can help improve the condition by reducing inflammation. For example, in acquired immunodeficiency syndrome (AIDS), it was shown that CCL2 neutralization hinders human immunodeficiency viruses (HIV)-1 replication in monocyte-derived macrophages [205]. Experimental studies demonstrated that administering anti-CCL2 in mice with Theiler's murine encephalomyelitis virus-induced demyelinating disease (TMEV-IDD) could reduce mononuclear cell infiltration and significantly decrease disease progression through modulating central nervous system (CNS) inflammation [206].

An investigation reported that administration of PF-04178903, a CCR2 antagonist small molecule in influenza-infected mice, significantly reduced mortality and improved the frequency of cytotoxic CD8⁺ T cells, hypothermia and weight loss in the studied animals. Moreover, the levels of total protein, albumin, and lactose dehydrogenase activity in bronchoalveolar lavage fluid of the treated mice were meaningfully reduced. However, PF-04178903 could not influence the viral titers and *Streptococcus pneumoniae*-induced secondary infection, as well as anti-influenza-neutralizing antibodies [114]. Since inflammatory CCR⁺ monocytes and their migration to the lung is similar in the pathogenesis of influenza and COVID-19, the use of PF-04178903 may be effective in managing inflammation in patients with COVID-19 [137,207].

Evidence strongly designates that COVID-19 severity and mortality are consistently lower in women than in men [208–213]. It is theorized that female biological factors such as 17 β -estradiol (E2) and progesterone (P4) confer protection against death. Since studies have demonstrated that E2 exerts its anti-inflammatory effect by inhibiting the production of pro-inflammatory cytokines and chemokines such as IL-1 β and TNF- α , and CCL2, two clinical trials were conducted to evaluate the clinical benefits of E2 (NCT04359329) and P4 (NCT04365127) in COVID-19 patients. Outcomes of these studies demonstrated that progesterone therapy might effectively treat male patients with moderate to

severe COVID-19 [214].

Researchers have established that CCL2 is the first and most potent chemokine released during SARS-CoV-2 infection, and inhibiting CCL2 with an anti-CCL2 antibody could delay virus-induced death in mice infected with SARS-CoV-2 beta variant (B.1.351) [156].

Extracellular loop 1 inverso (ECL1i) is also a potent, allosteric, non-competitive, and selective CCR2 inhibitor [215]. It has been revealed that the administration of ECL1i significantly decreased inflammatory responses in murine models of peritonitis and multiple sclerosis [215]. It has been reported that in both early and late stages of COVID-19, Ceniciviroc (CVC) as a dual CCR2/CCR5 antagonist with a potent antiviral activity can attenuate or repress dysregulated inflammatory responses as well as fibrosis [216]. However, the only clinical trial (NCT04500418) designed to evaluate CVC in patients with COVID-19 was terminated due to the non-cooperation of the patients in this study.

There are many plant extracts and natural compounds with anti-inflammatory properties; however, the anti-inflammatory mechanisms of most of these herbal compounds are not elucidated. In this context, it was reported that *two Brazilian plants, Lippia sidoides and Terminalia glabrescens* extracts inhibit CCL2 production in LPS-stimulated THP-1 cells [217]. *Shufeng Jiedu* capsule (SFJDC) is another traditional Chinese medicine containing eight types of herbal medicines with antiviral, anti-inflammatory, antipyretic, and immune regulatory activity [218], used to treat upper respiratory tract infections such as influenza [219,220] and recommended for COVID-19 treatment in China. According to network pharmacology analysis, its most potent compounds are quercetin, kaempferol, luteolin, wogonin, 7-Methoxy-2-methyl isoflavone, naringenin, beta-sitosterol, stigmaterol, physovenine, and formononetin. CCL2 is one of the main targets of SFJDC; however, examining the inhibition of CCL2 and the anti-inflammatory effects of each SFJDC component in patients with COVID-19 requires further studies [221].

Curcumin is a polyphenolic compound of *Curcuma longa L. rhizome* with anti-oxidant and anti-inflammatory effects [222,223]. This compound exerts its effects by down-regulation of some critical elements involved in cellular and molecular pathways, such as NF κ B, which plays a critical role in inflammation. Evidence revealed that curcumin inhibits the production of CCL2 by osteoblastic cells, blood monocytes, and alveolar macrophages [224–227]. In addition, the inhibitory effect of curcumin on LPS-induced CCL2 production has been reported in rat astrocytoma cell C6 via the JNK pathway [228]. Correspondingly, the antiviral effect of curcumin has been reported through ACE2 inhibition in SARS-CoV2 [229,230]. These findings suggest that curcumin might modulate CCL2-mediated inflammatory responses in COVID-19 [231,232].

8. Concluding remarks and future direction

According to the results of numerous studies conducted in the field of COVID-19 pathogenesis mechanisms, it can be concluded that the infiltration of inflammatory monocytes and macrophages and the dysregulated inflammation caused by the function of these cells and the released inflammatory mediators are one of the most important pathological events of severe SARS-CoV-2 infection. On the other hand, the CCL2/CCR2 chemokine axis plays a vital role in the recruitment and migration of monocytes and macrophages to the lung tissue. Therefore, inhibiting this axis using different compounds may reduce the disease's severity and manage hyperinflammation. However, few studies have been conducted in this field, the reasons for which are still unclear [233]. The critical point in this therapeutic approach is to pay attention to other mechanisms involved in migrating monocytes and macrophages to the lung.

Interestingly, in addition to CCL2, the CXCL17/GPR35 axis can also participate in the migration of alveolar macrophages or the precursors of these cells from circulation to the lungs [234]. Perhaps this parallel mechanism is one of the reasons for the lack of efficacy required by CCL2

or CCR2 inhibition. Therefore, combination therapies aimed at inhibiting CXCL17/GPR35 and CCL2/CCR2 axes may be more effective than monotherapy in inhibiting the migration of monocytes and macrophages to the lung. On the other hand, using CCL2 or CCR2 inhibitors can effectively increase vaccine immunization by inhibiting the function of MDSCs in vaccinated people.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

No data was used for the research described in the article.

Acknowledgments

Rafsanjan University of Medical Sciences supported this study.

References

- [1] H. Khorramdelazad, et al., Immunopathological similarities between COVID-19 and influenza: investigating the consequences of Co-infection, *Microb. Pathog.* 152 (2021), 104554.
- [2] L.M. Khosroshahi, et al., Immunology, immunopathogenesis and immunotherapeutics of COVID-19; an overview, *Int. Immunopharmacol.* 93 (2021), 107364.
- [3] H.R. Güner, I. Hasanoğlu, F. Aktaş, COVID-19: prevention and control measures in community, *Turkish J. Medical Sci.* 50 (SI-1) (2020) 571–577.
- [4] WHO Coronavirus (COVID-19) Dashboard. [cited 2022 11.2.2022]; Available from: <https://covid19.who.int/>.
- [5] Z. Xu, et al., Pathological findings of COVID-19 associated with acute respiratory distress syndrome, *Lancet Respir. Med.* 8 (4) (2020) 420–422.
- [6] C. Wu, et al., Risk factors associated with acute respiratory distress syndrome and death in patients with coronavirus disease 2019 pneumonia in Wuhan, China, *JAMA Internal Med.* 180 (7) (2020) 934–943.
- [7] Y.-Y. Zheng, et al., COVID-19 and the cardiovascular system, *Nat. Rev. Cardiol.* 17 (5) (2020) 259–260.
- [8] L. Lu, et al., The potential neurological effect of the COVID-19 vaccines: a review, *Acta Neurol. Scand.* 144 (1) (2021) 3–12.
- [9] Z. Li et al., Caution on kidney dysfunctions of COVID-19 patients. 2020.
- [10] G. Farnoosh, et al., Are Iranian sulfur mustard gas-exposed survivors more vulnerable to SARS-CoV-2? Some similarity in their pathogenesis, *Disaster Medicine Public Health Preparedness* 14 (6) (2020) 826–832.
- [11] M. Abbasifard, H. Khorramdelazad, The bio-mission of interleukin-6 in the pathogenesis of COVID-19: A brief look at potential therapeutic tactics, *Life Sci.* 257 (2020), 118097.
- [12] V. Bagheri, et al., CXC chemokine CXCL12 tissue expression and circulating levels in peptic ulcer patients with *Helicobacter pylori* infection, *Cytokine* 85 (2016) 1–4.
- [13] A. Vakilian, et al., CCL2/CCR2 signaling pathway in glioblastoma multiforme, *Neurochem. Int.* 103 (2017) 1–7.
- [14] F. Moadab, H. Khorramdelazad, M. Abbasifard, Role of CCL2/CCR2 axis in the immunopathogenesis of rheumatoid arthritis: Latest evidence and therapeutic approaches, *Life Sci.* 269 (2021), 119034.
- [15] Y. Taghavi, et al., Monocyte chemoattractant protein-1 (MCP-1/CCL2) in diabetic retinopathy: latest evidence and clinical considerations, *J. Cell Commun. Signaling* 13 (4) (2019) 451–462.
- [16] S. Behfar, et al., A brief look at the role of monocyte chemoattractant protein-1 (CCL2) in the pathophysiology of psoriasis, *Cytokine* 110 (2018) 226–231.
- [17] M. Merad, J.C. Martin, Pathological inflammation in patients with COVID-19: a key role for monocytes and macrophages, *Nat. Rev. Immunol.* 20 (6) (2020) 355–362.
- [18] Z. Zhou et al., Overly exuberant innate immune response to SARS-CoV-2 infection. 2020.
- [19] P.A. Szabo, et al., Longitudinal profiling of respiratory and systemic immune responses reveals myeloid cell-driven lung inflammation in severe COVID-19, *Immunity* 54 (4) (2021), pp. 797–814. e6.
- [20] R.J. Dress, F. Ginhoux, Monocytes and macrophages in severe COVID-19—friend, foe or both? *Immunol. Cell Biol.* (2021).
- [21] L. Gibellini, et al., Altered bioenergetics and mitochondrial dysfunction of monocytes in patients with COVID-19 pneumonia, *EMBO Mol. Med.* 12 (12) (2020) e13001.
- [22] M.Z. Tay, et al., The trinity of COVID-19: immunity, inflammation and intervention, *Nat. Rev. Immunol.* 20 (6) (2020) 363–374.
- [23] J.B. Moore, C.H. June, Cytokine release syndrome in severe COVID-19, *Science* 368 (6490) (2020) 473–474.

- [24] C. Huang, et al., Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China, *The Lancet* 395 (10223) (2020) 497–506.
- [25] L. Yang, et al., Cardiomyocytes recruit monocytes upon SARS-CoV-2 infection by secreting CCL2, *Stem Cell Rep.* 16 (9) (2021) 2274–2288.
- [26] M. Rahmati, M.A. Moosavi, Cytokine-targeted therapy in severely ill COVID-19 patients: options and cautions, *Ejmo* 4 (2) (2020) 179–181.
- [27] N. Morgulchik, et al., Potential therapeutic approaches for targeted inhibition of inflammatory cytokines following COVID-19 infection-induced cytokine storm, *Interface focus* 12 (1) (2021) 20210006.
- [28] T.K. Burki, *OMICRON variant and booster COVID-19 vaccines*. *The Lancet, Respir. Med.* (2021).
- [29] A. Zlotnik, O. Yoshie, The chemokine superfamily revisited, *Immunity* 36 (5) (2012) 705–716.
- [30] M.W. Carr, et al., Monocyte chemoattractant protein 1 acts as a T-lymphocyte chemoattractant, *Proc. Natl. Acad. Sci.* 91 (9) (1994) 3652–3656.
- [31] G. Soria, A. Ben-Baruch, The inflammatory chemokines CCL2 and CCL5 in breast cancer, *Cancer Lett.* 267 (2) (2008) 271–285.
- [32] T.M. Handel, P.J. Domaille, Heteronuclear (1H, 13C, 15N) NMR assignments and solution structure of the monocyte chemoattractant protein-1 (MCP-1) dimer, *Biochemistry* 35 (21) (1996) 6569–6584.
- [33] J. Lubkowski, et al., The structure of MCP-1 in two crystal forms provides a rare example of variable quaternary interactions, *Nat. Struct. Biol.* 4 (1) (1997) 64–69.
- [34] E. Van Coillie, J. Van Damme, G. Opdenakker, The MCP/eotaxin subfamily of CC chemokines, *Cytokine Growth Factor Rev.* 10 (1) (1999) 61–86.
- [35] B.H. Cochran, A.C. Reffel, C.D. Stiles, Molecular cloning of gene sequences regulated by platelet-derived growth factor, *Cell* 33 (3) (1983) 939–947.
- [36] S. Zhu, et al., The molecular structure and role of CCL2 (MCP-1) and C-C chemokine receptor CCR2 in skeletal biology and diseases, *J. Cell. Physiol.* (2021).
- [37] S.L. Deshmane, et al., Monocyte chemoattractant protein-1 (MCP-1): an overview, *J. Interferon Cytokine Res.* 29 (6) (2009) 313–326.
- [38] L.A. Kelley, et al., The Phyre2 web portal for protein modeling, prediction and analysis, *Nat. Protoc.* 10 (6) (2015) 845–858.
- [39] C.J. Beall, et al., Site-directed mutagenesis of monocyte chemoattractant protein-1 identifies two regions of the polypeptide essential for biological activity, *Biochem. J.* 313 (2) (1996) 633–640.
- [40] M. Ebisawa, et al., Eosinophil transendothelial migration induced by cytokines. III. Effect of the chemokine RANTES, *J. Immunol.* 153 (5) (1994) 2153–2160.
- [41] Y. Zhang, B.J. Rollins, A dominant negative inhibitor indicates that monocyte chemoattractant protein 1 functions as a dimer, *Mol. Cell. Biol.* 15 (9) (1995) 4851–4855.
- [42] J. Melchjorsen, L.N. Sørensen, S.R. Paludan, Expression and function of chemokines during viral infections: from molecular mechanisms to in vivo function, *J. Leukoc. Biol.* 74 (3) (2003) 331–343.
- [43] S. Mahalingam, et al., Chemokines and viruses: friends or foes? *Trends Microbiol.* 11 (8) (2003) 383–391.
- [44] B.A. Khalil, N.M. Elemam, A.A. Maghazachi, Chemokines and chemokine receptors during COVID-19 infection, *Comput. Struct. Biotechnol. J.* (2021).
- [45] R. Ransohoff, The chemokine system in neuroinflammation: an update, *J. Infect. Dis.* 186 (Supplement 2) (2002) S152–S156.
- [46] J.-H. Gong, et al., An antagonist of monocyte chemoattractant protein 1 (MCP-1) inhibits arthritis in the MRL-*lpr* mouse model, *J. Exp. Med.* 186 (1) (1997) 131–137.
- [47] J. Wain, J. Kirby, S. Ali, Leucocyte chemotaxis: Examination of mitogen-activated protein kinase and phosphoinositide 3-kinase activation by Monocyte Chemoattractant Proteins-1,-2,-3 and-4, *Clin. Exp. Immunol.* 127 (3) (2002) 436–444.
- [48] M. Gouwy, et al., Synergy between proinflammatory ligands of G protein-coupled receptors in neutrophil activation and migration, *J. Leukoc. Biol.* 76 (1) (2004) 185–194.
- [49] S. Sozzani, et al., Receptors and transduction pathways for monocyte chemotactic protein-2 and monocyte chemotactic protein-3. Similarities and differences with MCP-1, *J. Immunol.* 152 (7) (1994) 3615–3622.
- [50] S. Bose, J. Cho, Role of chemokine CCL2 and its receptor CCR2 in neurodegenerative diseases, *Arch. Pharmacol. Res.* 36 (9) (2013) 1039–1050.
- [51] F.S. Monteclaro, I.F. Charo, The amino-terminal extracellular domain of the MCP-1 receptor, but not the RANTES/MIP-1 α receptor, confers chemokine selectivity: evidence for a two-step mechanism for MCP-1 receptor activation, *J. Biol. Chem.* 271 (32) (1996) 19084–19092.
- [52] F.S. Monteclaro, I.F. Charo, The amino-terminal domain of CCR2 is both necessary and sufficient for high affinity binding of monocyte chemoattractant protein 1: receptor activation by a pseudo-tethered ligand, *J. Biol. Chem.* 272 (37) (1997) 23186–23190.
- [53] I.F. Charo, et al., Molecular cloning and functional expression of two monocyte chemoattractant protein 1 receptors reveals alternative splicing of the carboxyl-terminal tails, *Proc. Natl. Acad. Sci.* 91 (7) (1994) 2752–2756.
- [54] E.K. Lau, et al., Identification of the glycosaminoglycan binding site of the CC chemokine, MCP-1: implications for structure and function in vivo, *J. Biol. Chem.* 279 (21) (2004) 22294–22305.
- [55] R. Yamasaki, et al., Role of CCR2 in immunobiology and neurobiology, *Clin. Experimental Neuroimmunol.* 3 (1) (2012) 16–29.
- [56] M.S. Hayden, S. Ghosh, Regulation of NF- κ B by TNF family cytokines. *Seminars in Immunology*, Elsevier, 2014.
- [57] S. Dommel, M. Blüher, Does CC Motif Chemokine Ligand 2 (CCL2) Link Obesity to a Pro-Inflammatory State? *Int. J. Mol. Sci.* 22 (3) (2021) 1500.
- [58] A. Aljada, et al., Insulin inhibits NF κ B and MCP-1 expression in human aortic endothelial cells, *J. Clin. Endocrinol. Metab.* 86 (1) (2001) 450–453.
- [59] H. Nakatsumi, M. Matsumoto, K.I. Nakayama, Noncanonical pathway for regulation of CCL2 expression by an mTORC1-FOXK1 axis promotes recruitment of tumor-associated macrophages, *Cell Rep.* 21 (9) (2017) 2471–2486.
- [60] B.J. Rollins, Chemokines, *Blood, J. Am. Soc. Hematol.* 90 (3) (1997) 909–928.
- [61] E.A. Old, M. Malcangio, Chemokine mediated neuron–glia communication and aberrant signalling in neuropathic pain states, *Curr. Opin. Pharmacol.* 12 (1) (2012) 67–73.
- [62] J. Dawson, et al., Targeting monocyte chemoattractant protein-1 signalling in disease, *Expert opinion on therapeutic targets* 7 (1) (2003) 35–48.
- [63] J. Cho, D.L. Gruol, The chemokine CCL2 activates p38 mitogen-activated protein kinase pathway in cultured rat hippocampal cells, *J. Neuroimmunol.* 199 (1–2) (2008) 94–103.
- [64] M. Locati, et al., Inhibition of Monocyte Chemotaxis to CC Chemokines by Antisense Oligonucleotide for Cytosolic Phospholipase A2 (*), *J. Biol. Chem.* 271 (11) (1996) 6010–6016.
- [65] Y. Terashima, et al., Pivotal function for cytoplasmic protein FROUNT in CCR2-mediated monocyte chemotaxis, *Nat. Immunol.* 6 (8) (2005) 827–835.
- [66] J. Gosling, S. Slaymaker, L. Gu, S. Tseng, C.H. Zlot, S.G. Young, B.J. Rollins, I. F. Charo, MCP-1 deficiency reduces susceptibility to atherosclerosis in mice that overexpress human apolipoprotein B, *J Clin Invest* 103 (1999) 773–778.
- [67] L. Gu, et al., Absence of monocyte chemoattractant protein-1 reduces atherosclerosis in low density lipoprotein receptor–deficient mice, *Mol. Cell* 2 (2) (1998) 275–281.
- [68] C. McManus, et al., MCP-1, MCP-2 and MCP-3 expression in multiple sclerosis lesions: an immunohistochemical and in situ hybridization study, *J. Neuroimmunol.* 86 (1) (1998) 20–29.
- [69] R.M. Ransohoff, et al., Astrocyte expression of mRNA encoding cytokines IP-10 and JE/MCP-1 in experimental autoimmune encephalomyelitis, *FASEB J.* 7 (6) (1993) 592–600.
- [70] J. Simpson, et al., Expression of monocyte chemoattractant protein-1 and other β -chemokines by resident glia and inflammatory cells in multiple sclerosis lesions, *J. Neuroimmunol.* 84 (2) (1998) 238–249.
- [71] B.P. Barna, et al., Regulation of monocyte chemoattractant protein-1 expression in adult human non-neoplastic astrocytes is sensitive to tumor necrosis factor (TNF) or antibody to the 55-kDa TNF receptor, *J. Neuroimmunol.* 50 (1) (1994) 101–107.
- [72] Z. Brown, et al., IL-1 receptor antagonist inhibits monocyte chemotactic peptide 1 generation by human mesangial cells, *Kidney Int.* 42 (1) (1992) 95–101.
- [73] S.D. Cushing, et al., Minimally modified low density lipoprotein induces monocyte chemotactic protein 1 in human endothelial cells and smooth muscle cells, *Proc. Natl. Acad. Sci.* 87 (13) (1990) 5134–5138.
- [74] T.J. Standiford, et al., Alveolar macrophage-derived cytokines induce monocyte chemoattractant protein-1 expression from human pulmonary type II-like epithelial cells, *J. Biol. Chem.* 266 (15) (1991) 9912–9918.
- [75] T. Yoshimura, et al., Purification and amino acid analysis of two human monocyte chemoattractants produced by phytohemagglutinin-stimulated human blood mononuclear leukocytes, *J. Immunol.* 142 (6) (1989) 1956–1962.
- [76] T. Yoshimura, et al., Human monocyte chemoattractant protein-1 (MCP-1) Full-length cDNA cloning, expression in mitogen-stimulated blood mononuclear leukocytes, and sequence similarity to mouse competence gene JE, *FEBS Lett.* 244 (2) (1989) 487–493.
- [77] V. Bianconi, et al., The regulation and importance of monocyte chemoattractant protein-1, *Curr. Opin. Hematol.* 25 (1) (2018) 44–51.
- [78] M.W. Feinberg, et al., Essential role for Smad3 in regulating MCP-1 expression and vascular inflammation, *Circ. Res.* 94 (5) (2004) 601–608.
- [79] L.L. Xu, et al., Human recombinant monocyte chemotactic protein and other C-C chemokines bind and induce directional migration of dendritic cells in vitro, *J. Leukoc. Biol.* 60 (3) (1996) 365–371.
- [80] C. Daly, B.J. Rollins, Monocyte chemoattractant protein-1 (CCL2) in inflammatory disease and adaptive immunity: therapeutic opportunities and controversies, *Microcirculation* 10 (3–4) (2003) 247–257.
- [81] C. Bartoli, et al., CCR2A and CCR2B, the two isoforms of the monocyte chemoattractant protein-1 receptor are up-regulated and expressed by different cell subsets in idiopathic inflammatory myopathies, *Acta Neuropathol.* 102 (4) (2001) 385–392.
- [82] M.-L. Cho, et al., Expression of CCR2A, an isoform of MCP-1 receptor, is increased by MCP-1, CD40 ligand and TGF- β in fibroblast like synoviocytes of patients with RA, *Exp. Mol. Med.* 39 (4) (2007) 499–507.
- [83] S.K. Sanders, et al., Functional differences between monocyte chemotactic protein-1 receptor A and monocyte chemotactic protein-1 receptor B expressed in a Jurkat T cell, *J. Immunol.* 165 (9) (2000) 4877–4883.
- [84] T.I. Arefieva, et al., MCP-1-stimulated chemotaxis of monocyte and endothelial cells is dependent on activation of different signaling cascades, *Cytokine* 31 (6) (2005) 439–446.
- [85] W.J. Karpus, et al., Differential CC chemokine-induced enhancement of T helper cell cytokine production, *J. Immunol.* 158 (9) (1997) 4129–4136.
- [86] S.W. Chensue, et al., Monocyte chemotactic protein expression during schistosome egg granuloma formation. Sequence of production, localization, contribution, and regulation, *Am. J. Pathol.* 146 (1) (1995) 130.
- [87] L. Gu, et al., Control of TH2 polarization by the chemokine monocyte chemoattractant protein-1, *Nature* 404 (6776) (2000) 407–411.
- [88] T.R. Traynor, et al., Differential roles of CC chemokine ligand 2/monocyte chemotactic protein-1 and CCR2 in the development of T1 immunity, *J. Immunol.* 168 (9) (2002) 4659–4666.

- [89] N. Omata, et al., Monocyte chemoattractant protein-1 selectively inhibits the acquisition of CD40 ligand-dependent IL-12-producing capacity of monocyte-derived dendritic cells and modulates Th1 immune response, *J. Immunol.* 169 (9) (2002) 4861–4866.
- [90] R.T. Palframan, et al., Inflammatory chemokine transport and presentation in heV a remote control mechanism for monocyte recruitment to lymph nodes in inflamed tissues, *J. Exp. Med.* 194 (9) (2001) 1361–1374.
- [91] A.A. Maghazachi, A. Al-Aoukaty, T.J. Schall, CC chemokines induce the chemotaxis of NK and IL-2-activated NK cells. Role for G proteins, *J. Immunol.* 153 (11) (1994) 4969–4977.
- [92] T. Yoshimura, M. Takahashi, IFN- γ -mediated survival enables human neutrophils to produce MCP-1/CCL2 in response to activation by TLR ligands, *J. Immunol.* 179 (3) (2007) 1942–1949.
- [93] M.A. Brown, J. Hural, Functions of IL-4 and control of its expression, *Critical Rev.™ Immunol.* 17 (1) (1997).
- [94] J.-A. Gonzalo, et al., The coordinated action of CC chemokines in the lung orchestrates allergic inflammation and airway hyperresponsiveness, *J. Exp. Med.* 188 (1) (1998) 157–167.
- [95] R. Badia, et al., β -Galactomannan and Saccharomyces cerevisiae var. boulardii modulate the immune response against Salmonella enterica serovar Typhimurium in porcine intestinal epithelial and dendritic cells, *Clin. Vaccine Immunol.* 19 (3) (2012) 368–376.
- [96] K. Stutz, et al., Disruption of the C. elegans intestinal brush border by the fungal lectin CCL2 phenocopies dietary lectin toxicity in mammals, *PLoS ONE* 10 (6) (2015) e0129381.
- [97] D. Angela Covino, M. Sabbatucci, L. Fantuzzi, The CCL2/CCR2 axis in the pathogenesis of HIV-1 infection: a new cellular target for therapy? *Curr. Drug Targets* 17 (1) (2016) 76–110.
- [98] C.D. Conrady, et al., IFN- α -driven CCL2 production recruits inflammatory monocytes to infection site in mice, *Mucosal Immunol.* 6 (1) (2013) 45–55.
- [99] C. Shi, E.G. Pamer, Monocyte recruitment during infection and inflammation, *Nat. Rev. Immunol.* 11 (11) (2011) 762–774.
- [100] A.W. Ansari, et al., CCL2: A potential prognostic marker and target of anti-inflammatory strategy in HIV/AIDS pathogenesis, *Eur. J. Immunol.* 41 (12) (2011) 3412–3418.
- [101] Y.L. Lau, J.M. Peiris, Pathogenesis of severe acute respiratory syndrome, *Curr. Opin. Immunol.* 17 (4) (2005) 404–410.
- [102] L.F. Ng, et al., A human in vitro model system for investigating genome-wide host responses to SARS coronavirus infection, *BMC Infect. Dis.* 4 (1) (2004) 1–11.
- [103] J. Chen, et al., Cellular immune responses to severe acute respiratory syndrome coronavirus (SARS-CoV) infection in senescent BALB/c mice: CD4+ T cells are important in control of SARS-CoV infection, *J. Virol.* 84 (3) (2010) 1289–1301.
- [104] M. Yilla, et al., SARS-coronavirus replication in human peripheral monocytes/macrophages, *Virus Res.* 107 (1) (2005) 93–101.
- [105] J.S. Peiris, Y. Guan, K. Yuen, Severe acute respiratory syndrome, *Nat. Med.* 10 (12) (2004) S88–S97.
- [106] J. Zhou, et al., Active replication of Middle East respiratory syndrome coronavirus and aberrant induction of inflammatory cytokines and chemokines in human macrophages: implications for pathogenesis, *J. Infect. Dis.* 209 (9) (2014) 1331–1342.
- [107] T. Ziegler, et al., Severe acute respiratory syndrome coronavirus fails to activate cytokine-mediated innate immune responses in cultured human monocyte-derived dendritic cells, *J. Virol.* 79 (21) (2005) 13800–13805.
- [108] R. Channappanavar, et al., Dysregulated type I interferon and inflammatory monocyte-macrophage responses cause lethal pneumonia in SARS-CoV-infected mice, *Cell Host Microbe* 19 (2) (2016) 181–193.
- [109] J.M. Nicholls, et al., Lung pathology of fatal severe acute respiratory syndrome, *The Lancet* 361 (9371) (2003) 1773–1778.
- [110] S. Rosseau, et al., Phenotypic characterization of alveolar monocyte recruitment in acute respiratory distress syndrome, *Am. J. Physiol.-Lung Cellular Mol. Physiol.* 279 (1) (2000) L25–L35.
- [111] K.W. Tsang, et al., A cluster of cases of severe acute respiratory syndrome in Hong Kong, *N. Engl. J. Med.* 348 (20) (2003) 1977–1985.
- [112] H.R. Jonsdottir et al., Molnupiravir combined with different repurposed drugs further inhibits SARS-CoV-2 infection in human nasal epithelium in vitro. *bioRxiv*, 2022.
- [113] P.T. Morrison, et al., Chemokine-receptor upregulation and disease severity in respiratory syncytial virus infection, *Clin. Immunol.* 128 (1) (2008) 85–93.
- [114] K.L. Lin, et al., CCR2-antagonist prophylaxis reduces pulmonary immune pathology and markedly improves survival during influenza infection, *J. Immunol.* 186 (1) (2011) 508–515.
- [115] A. Jafarzadeh, et al., Contribution of monocytes and macrophages to the local tissue inflammation and cytokine storm in COVID-19: Lessons from SARS and MERS, and potential therapeutic interventions, *Life Sci.* 257 (2020), 118102.
- [116] M.S. Hayney, et al., Serum IFN- γ -induced protein 10 (IP-10) as a biomarker for severity of acute respiratory infection in healthy adults, *J. Clin. Virol.* 90 (2017) 32–37.
- [117] S.L. Cole, et al., M1-like monocytes are a major immunological determinant of severity in previously healthy adults with life-threatening influenza, *JCI insight* 7 (7) (2017).
- [118] P.S. McNamara, et al., Production of chemokines in the lungs of infants with severe respiratory syncytial virus bronchiolitis, *J. Infect. Dis.* 191 (8) (2005) 1225–1232.
- [119] B.A. Khalil, N.M. Elemam, A.A. Maghazachi, Chemokines and chemokine receptors during COVID-19 infection, *Comput. Struct. Biotechnol. J.* 19 (2021) 976–988.
- [120] T.J. Franks, et al., Lung pathology of severe acute respiratory syndrome (SARS): a study of 8 autopsy cases from Singapore, *Hum. Pathol.* 34 (8) (2003) 743–748.
- [121] M. Swiecki, M. Colonna, Type I interferons: diversity of sources, production pathways and effects on immune responses, *Curr. Opin. Virol.* 1 (6) (2011) 463–475.
- [122] F. Coperchini, et al., The cytokine storm in COVID-19: an overview of the involvement of the chemokine/chemokine-receptor system, *Cytokine Growth Factor Rev.* 53 (2020) 25–32.
- [123] O. Yoshie, T. Imai, H. Nomiya, Chemokines in immunity, *Adv. Immunol.* 78 (2001) 57–110.
- [124] Z.-S. Xu, et al., Temporal profiling of plasma cytokines, chemokines and growth factors from mild, severe and fatal COVID-19 patients, *Signal Transduction Targeted Ther.* 5 (1) (2020) 1–3.
- [125] R.B. Goodman, et al., Inflammatory cytokines in patients with persistence of the acute respiratory distress syndrome, *Am. J. Respir. Crit. Care Med.* 154 (3) (1996) 602–611.
- [126] P. Henrot, et al., Chemokines in COPD: from implication to therapeutic use, *Int. J. Mol. Sci.* 20 (11) (2019) 2785.
- [127] B.J. Rollins, A. Walz, M. Baggiolini, Recombinant human MCP-1/JE induces chemotaxis, calcium flux, and the respiratory burst in human monocytes. 1991.
- [128] A.E. Proudfoot, et al., Glycosaminoglycan binding and oligomerization are essential for the in vivo activity of certain chemokines, *Proc. Natl. Acad. Sci.* 100 (4) (2003) 1885–1890.
- [129] S.J. Collington, et al., The role of the CCL2/CCR2 axis in mouse mast cell migration in vitro and in vivo, *J. Immunol.* 184 (11) (2010) 6114–6123.
- [130] C.E. Rose Jr, S.S.J. Sung, S.M. Fu, Significant involvement of CCL2 (MCP-1) in inflammatory disorders of the lung, *Microcirculation* 10 (3–4) (2003) 273–288.
- [131] U.A. Maus, et al., Monocytes are potent facilitators of alveolar neutrophil emigration during lung inflammation: role of the CCL2-CCR2 axis, *J. Immunol.* 170 (6) (2003) 3273–3278.
- [132] C.M. Hogaboam, et al., Differential monocyte chemoattractant protein-1 and chemokine receptor 2 expression by murine lung fibroblasts derived from Th1- and Th2-type pulmonary granuloma models, *J. Immunol.* 163 (4) (1999) 2193–2201.
- [133] M. Buszko et al., The dynamic changes in cytokine responses in COVID-19: a snapshot of the current state of knowledge. 2020, *Nature Publishing Group*.
- [134] Y. Chi, et al., Serum cytokine and chemokine profile in relation to the severity of coronavirus disease 2019 in China, *J. Infect. Dis.* 222 (5) (2020) 746–754.
- [135] D. Blanco-Melo, et al., Imbalanced host response to SARS-CoV-2 drives development of COVID-19, *Cell* 181 (5) (2020) 1036–1045, e9.
- [136] Y. Yang, et al., Plasma IP-10 and MCP-3 levels are highly associated with disease severity and predict the progression of COVID-19, *J. Allergy Clin. Immunol.* 146 (1) (2020) 119–127, e4.
- [137] A. Vanderheiden, et al., CCR2 Signaling Restricts SARS-CoV-2 Infection, *MBio* 12 (6) (2021) e02749–e2821.
- [138] G. Grassi, et al., Myeloid-Derived Suppressor Cells in COVID-19: The Paradox of Good, *Front. Immunol.* 13 (2022).
- [139] S. Falck-Jones et al., Functional myeloid-derived suppressor cells expand in blood but not airways of COVID-19 patients and predict disease severity. *medRxiv*, 2020.
- [140] X. Guan, et al., Myeloid-derived suppressor cell accumulation in renal cell carcinoma is correlated with CCL2, IL-17 and IL-18 expression in blood and tumors, *Adv. Clin. Experimental Medicine* 27 (7) (2018) 947–953.
- [141] H. Gu, et al., CCL2 produced by pancreatic ductal adenocarcinoma is essential for the accumulation and activation of monocytic myeloid-derived suppressor cells, *Inflammation Dis.* 9 (4) (2021) 1686–1695.
- [142] Y.V. Perflyeva, et al., Myeloid-derived suppressor cells in COVID-19: a review, *Clin. Immunol.* (2022), 109024.
- [143] A. Chugh, et al., Changing dynamics of SARS-CoV-2: a global challenge, *Appl. Sci.* 12 (11) (2022) 5546.
- [144] R.P. Walensky, H.T. Walke, A.S. Fauci, SARS-CoV-2 variants of concern in the United States—challenges and opportunities, *JAMA* 325 (11) (2021) 1037–1038.
- [145] *SARS-CoV-2 Variant Classifications and Definitions*. 2022 [cited 2022 26.4.2022]; Available from: https://www.cdc.gov/coronavirus/2019-ncov/variants/variant-classifications.html?CDC_AA_refVal=https%3A%2F%2Fwww.cdc.gov%2Fcoronavirus%2F2019-ncov%2Fvariants%2Fvariant-info.html.
- [146] M. Ramanathan et al., SARS-CoV-2 B. 1.1. 7 and B. 1.351 spike variants bind human ACE2 with increased affinity. *Lancet Infect. Dis.* 21(8) (2021) 1070.
- [147] A. Khan, et al., Higher infectivity of the SARS-CoV-2 new variants is associated with K417N/T, E484K, and N501Y mutants: an insight from structural data, *J. Cell. Physiol.* 236 (10) (2021) 7045–7057.
- [148] X. Li, et al., Molecular immune pathogenesis and diagnosis of COVID-19, *J. Pharm. Anal.* 10 (2) (2020) 102–108.
- [149] A. Pum, et al., Cytokines and chemokines in SARS-CoV-2 infections—therapeutic strategies targeting cytokine storm, *Biomolecules* 11 (1) (2021) 91.
- [150] E.S. Winkler, et al., SARS-CoV-2 infection of human ACE2-transgenic mice causes severe lung inflammation and impaired function, *Nat. Immunol.* 21 (11) (2020) 1327–1335.
- [151] C.K. Yinda, et al., K18-hACE2 mice develop respiratory disease resembling severe COVID-19, *PLoS Pathog.* 17 (1) (2021) e1009195.
- [152] M. Olbei, et al., SARS-CoV-2 causes a different cytokine response compared to other cytokine storm-causing respiratory viruses in severely ill patients, *Front. Immunol.* 12 (2021), 629193.
- [153] B. Stolp, et al., SARS-CoV-2 variants of concern display enhanced intrinsic pathogenic properties and expanded organ tropism in mouse models, *Cell Rep.* 38 (7) (2022), 110387.

- [154] Z.R. Korobova, et al., A Comparative Study of the Plasma Chemokine Profile in COVID-19 Patients Infected with Different SARS-CoV-2 Variants, *Int. J. Mol. Sci.* 23 (16) (2022) 9058.
- [155] S. Stone, et al., SARS-CoV-2 variants of concern infect the respiratory tract and induce inflammatory response in wild-type laboratory mice, *Viruses* 14 (1) (2021) 27.
- [156] S. Lee, et al., Immunological and Pathological Peculiarity of Severe Acute Respiratory Syndrome Coronavirus 2 Beta Variant, *Microbiol. Spectrum* (2022) e02371–e10422.
- [157] P. Radvak et al., B. 1.1. 7 and B. 1.351 variants are highly virulent in K18-ACE2 transgenic mice and show different pathogenic patterns from early SARS-CoV-2 strains. *bioRxiv*, 2021.
- [158] G. Quéromès, et al., Characterization of SARS-CoV-2 ORF6 deletion variants detected in a nosocomial cluster during routine genomic surveillance, Lyon, France, *Emerging Microbes Infections* 10 (1) (2021) 167–177.
- [159] M.C. Dessing, et al., Monocyte chemoattractant protein 1 contributes to an adequate immune response in influenza pneumonia, *Clin. Immunol.* 125 (3) (2007) 328–336.
- [160] T. Yoshimura, et al., Non-myeloid cells are major contributors to innate immune responses via production of monocyte chemoattractant protein-1/CCL2, *Front. Immunol.* (2014) 482.
- [161] M. Majety, et al., A drug development perspective on targeting tumor-associated myeloid cells, *FEBS J.* 285 (4) (2018) 763–776.
- [162] J.J. Haringman, et al., A randomized controlled trial with an anti-CCL2 (anti-monocyte chemoattractant protein 1) monoclonal antibody in patients with rheumatoid arthritis, *Arthritis & Rheumatism: Official J. Am. College Rheumatol.* 54 (8) (2006) 2387–2392.
- [163] G. Raghui, et al., CC-chemokine ligand 2 inhibition in idiopathic pulmonary fibrosis: a phase 2 trial of carlumab, *Eur. Respir. J.* 46 (6) (2015) 1740–1750.
- [164] K.J. Pienta, et al., Phase 2 study of carlumab (CNTO 888), a human monoclonal antibody against CC-chemokine ligand 2 (CCL2), in metastatic castration-resistant prostate cancer, *Invest. New Drugs* 31 (3) (2013) 760–768.
- [165] S.K. Sandhu, et al., A first-in-human, first-in-class, phase I study of carlumab (CNTO 888), a human monoclonal antibody against CC-chemokine ligand 2 in patients with solid tumors, *Cancer Chemother. Pharmacol.* 71 (4) (2013) 1041–1050.
- [166] T. Krasnikova, et al., Ingramon, a peptide inhibitor of MCP-1 chemokine, reduces migration of blood monocytes stimulated by glioma-conditioned medium, *Bull. Exp. Biol. Med.* 160 (4) (2016) 480–482.
- [167] T. Arefieva, et al., Synthetic peptide fragment (65–76) of monocyte chemoattractant protein-1 (MCP-1) inhibits MCP-1 binding to heparin and possesses anti-inflammatory activity in stable angina patients after coronary stenting, *Inflamm. Res.* 60 (10) (2011) 955–964.
- [168] H. Raghui, et al., CCL2/CCR2, but not CCL5/CCR5, mediates monocyte recruitment, inflammation and cartilage destruction in osteoarthritis, *Ann. Rheum. Dis.* 76 (5) (2017) 914–922.
- [169] T. Ueno, et al., Significance of macrophage chemoattractant protein-1 in macrophage recruitment, angiogenesis, and survival in human breast cancer, *Clin. Cancer Res.* 6 (8) (2000) 3282–3289.
- [170] H. Fujimoto, et al., Stromal MCP-1 in mammary tumors induces tumor-associated macrophage infiltration and contributes to tumor progression, *Int. J. Cancer* 125 (6) (2009) 1276–1284.
- [171] M. Heiskala, et al., High expression of CCL2 in tumor cells and abundant infiltration with CD14 positive macrophages predict early relapse in breast cancer, *Virchows Arch.* 474 (1) (2019) 3–12.
- [172] H. Saji, et al., Significant correlation of monocyte chemoattractant protein-1 expression with neovascularization and progression of breast carcinoma, *Cancer: Interdisciplinary Int. J. Am. Cancer Soc.* 92 (5) (2001) 1085–1091.
- [173] T. Masuda, et al., Phase I dose-escalation trial to repurpose propagermanium, an oral CCL2 inhibitor, in patients with breast cancer, *Cancer Sci.* 111 (3) (2020) 924–931.
- [174] L. Bonapace, et al., Cessation of CCL2 inhibition accelerates breast cancer metastasis by promoting angiogenesis, *Nature* 515 (7525) (2014) 130–133.
- [175] A. Hariharan, et al., The role and therapeutic potential of NF-kappa-B pathway in severe COVID-19 patients, *Inflammopharmacology* 29 (1) (2021) 91–100.
- [176] Y.-C. Lin, et al., Tumor necrosis factor-alpha inhibitors suppress CCL2 chemokine in monocytes via epigenetic modification, *Mol. Immunol.* 83 (2017) 82–91.
- [177] D. Bauer, et al., Diallyl disulfide inhibits TNF α -induced CCL2 release by MDA-MB-231 cells, *Anticancer Res.* 34 (6) (2014) 2763–2770.
- [178] D. Bauer, et al., Diallyl disulfide inhibits TNF α induced CCL2 release through MAPK/ERK and NF-Kappa-B signaling, *Cytokine* 75 (1) (2015) 117–126.
- [179] K.-J. Kanga, et al., Effect of Diallyl Trisulfide on TNF- α -induced CCL2/MCP-1 Release in Genetically Different Triple-negative Breast Cancer Cells, *Anticancer Res.* 41 (12) (2021) 5919–5933.
- [180] P.C. Robinson, et al., The Potential for Repurposing Anti-TNF as a Therapy for the Treatment of COVID-19, *Med* 1 (1) (2020) 90–102.
- [181] E. Mora, et al., Bindarit: an anti-inflammatory small molecule that modulates the NFkB pathway, *Cell Cycle* 11 (1) (2012) 159–169.
- [182] M. Mirolo, et al., Impact of the anti-inflammatory agent bindarit on the chemokine: selective inhibition of the monocyte chemotactic proteins, *Eur. Cytokine Netw.* 19 (3) (2008) 119–122.
- [183] C. Severini, et al., Bindarit, inhibitor of CCL2 synthesis, protects neurons against amyloid- β -induced toxicity, *Journal of Alzheimer's Disease* 38 (2) (2014) 281–293.
- [184] S. Ge, et al., The CCL2 synthesis inhibitor bindarit targets cells of the neurovascular unit, and suppresses experimental autoimmune encephalomyelitis, *Journal of neuroinflammation* 9 (1) (2012) 1–13.
- [185] Z. Shen, et al., Inhibition of CCL2 by bindarit alleviates diabetes-associated periodontitis by suppressing inflammatory monocyte infiltration and altering macrophage properties, *Cell. Mol. Immunol.* 18 (9) (2021) 2224–2235.
- [186] A. Ble, et al., Antiproteinuric effect of chemokine CC motif ligand 2 inhibition in subjects with acute proliferative lupus nephritis, *Am. J. Nephrol.* 34 (4) (2011) 367–372.
- [187] S. Klussmann, et al., Mirror-image RNA that binds D-adenosine, *Nat. Biotechnol.* 14 (9) (1996) 1112–1115.
- [188] D. Oberthür, et al., Crystal structure of a mirror-image L-RNA aptamer (Spiegelmer) in complex with the natural L-protein target CCL2, *Nat. Commun.* 6 (1) (2015) 1–11.
- [189] J. Menne, et al., CC motif-ligand 2 inhibition with emaptic pegol (NOX-E36) in type 2 diabetic patients with albuminuria, *Nephrol. Dial. Transplant.* 32 (2) (2017) 307–315.
- [190] O. Kulkarni, et al., Spiegelmer inhibition of CCL2/MCP-1 ameliorates lupus nephritis in MRL-(Fas) lpr mice, *J. Am. Soc. Nephrol.* 18 (8) (2007) 2350–2358.
- [191] Z. Yang, A.N. Koehler, L. Wang, A novel small molecule activator of nuclear receptor SHP inhibits HCC cell migration via suppressing Ccl2, *Mol. Cancer Ther.* 15 (10) (2016) 2294–2301.
- [192] Y. Zhang, C.H. Hagedorn, L. Wang, Role of nuclear receptor SHP in metabolism and cancer, *Biochimica et Biophysica Acta (BBA)-Molecular Basis of Disease* 1812 (8) (2011) 893–908.
- [193] S. Rudraiah, X. Zhang, L. Wang, Nuclear receptors as therapeutic targets in liver disease: are we there yet? *Annu. Rev. Pharmacol. Toxicol.* 56 (2016) 605–626.
- [194] N.S. Magee, et al., A novel hepatocyte SHP/CCL2 axis controls liver inflammation in NAFL to NASH transition, *FASEB J.* 31 (2017), p. 328.4-328.4.
- [195] Q. Zhao et al., Preclinical antitumor activity of a CC chemokine receptor (CCR) 2/5 dual antagonist as monotherapy and in combination with immune checkpoint blockade. 2018, AACR.
- [196] R.J. Cherney, et al., BMS-813160: a potent CCR2 and CCR5 dual antagonist selected as a clinical candidate, *ACS Med. Chem. Lett.* 12 (11) (2021) 1753–1758.
- [197] B.K. Patterson, et al., CCR5 inhibition in critical COVID-19 patients decreases inflammatory cytokines, increases CD8 T-cells, and decreases SARS-CoV2 RNA in plasma by day 14, *Int. J. Infect. Dis.* 103 (2021) 25–32.
- [198] S. Yang, et al., Inhibition of the chemokine (C-C motif) ligand 2/chemokine (C-C motif) receptor 2 pathway attenuates hyperglycaemia and inflammation in a mouse model of hepatic steatosis and lipodystrophy, *Diabetologia* 52 (5) (2009) 972–981.
- [199] W. Guo, et al., Diabetes is a risk factor for the progression and prognosis of COVID-19, *Diabetes/metabolism Res. Rev.* 36 (7) (2020) e3319.
- [200] L. Zhu, et al., Association of blood glucose control and outcomes in patients with COVID-19 and pre-existing type 2 diabetes, *Cell Metab.* 31 (6) (2020), pp. 1068–1077. e3.
- [201] F. Alzaid, et al., Monocytopenia, monocyte morphological anomalies and hyperinflammation characterise severe COVID-19 in type 2 diabetes, *EMBO Mol. Med.* 12 (10) (2020) e13038.
- [202] C. Fong, Covid-19: Predicting inhibition of SARS-CoV-2 in Caco-2 cells: viral cellular entry. 2020, Eigenenergy, Adelaide, Australia.
- [203] D. de Zeeuw, et al., The effect of CCR2 inhibitor CCX140-B on residual albuminuria in patients with type 2 diabetes and nephropathy: a randomised trial, *Lancet Diabetes Endocrinol.* 3 (9) (2015) 687–696.
- [204] J. An, et al., Targeting CCR2 with its antagonist suppresses viability, motility and invasion by downregulating MMP-9 expression in non-small cell lung cancer cells, *Oncotarget* 8 (24) (2017) 39230.
- [205] M. Sabbatucci, et al., Endogenous CCL2 neutralization restricts HIV-1 replication in primary human macrophages by inhibiting viral DNA accumulation, *Retrovirology* 12 (1) (2015) 1–22.
- [206] W.J. Karpus, et al., Anti-CCL2 treatment inhibits Theiler's murine encephalomyelitis virus-induced demyelinating disease, *J. Neurovirol.* 12 (4) (2006) 251–261.
- [207] K.L. Lin, et al., CCR2+ monocyte-derived dendritic cells and exudate macrophages produce influenza-induced pulmonary immune pathology and mortality, *J. Immunol.* 180 (4) (2008) 2562–2572.
- [208] W.-J. Guan, et al., Clinical characteristics of coronavirus disease 2019 in China, *N. Engl. J. Med.* 382 (18) (2020) 1708–1720.
- [209] S.L. Klein, et al., Biological sex impacts COVID-19 outcomes, *PLoS Pathog.* 16 (6) (2020) e1008570.
- [210] F. Mauvais-Jarvis, et al., Sex and gender: modifiers of health, disease, and medicine, *The Lancet* 396 (10250) (2020) 565–582.
- [211] G. Onder, G. Rezza, S. Brusaferro, Case-fatality rate and characteristics of patients dying in relation to COVID-19 in Italy, *JAMA* 323 (18) (2020) 1775–1776.
- [212] Y.J. Park, et al., COVID-19 national emergency response center, epidemiology and case management team. Contact tracing during coronavirus disease outbreak, South Korea 2020, *Emerg. Infect. Dis.* 26 (10) (2020) 2465–2468.
- [213] S. Richardson, et al., Presenting characteristics, comorbidities, and outcomes among 5700 patients hospitalized with COVID-19 in the New York City area, *JAMA* 323 (20) (2020) 2052–2059.
- [214] S. Ghandehari, et al., Progesterone in addition to standard of care vs standard of care alone in the treatment of men hospitalized with moderate to severe COVID-19: a randomized, controlled pilot trial, *Chest* 160 (1) (2021) 74–84.
- [215] C. Auvynet, et al., ECL1i, d (LGTFLLK), a novel, small peptide that specifically inhibits CCL2-dependent migration, *FASEB J.* 30 (6) (2016) 2370–2381.

- [216] D.C. Files, et al., Rationale of using the dual chemokine receptor CCR2/CCR5 inhibitor cenicriviroc for the treatment of COVID-19, *PLoS Pathog.* 18 (6) (2022) e1010547.
- [217] G.S. Gusman, et al., Evaluation of the effects of some Brazilian medicinal plants on the production of TNF- α and CCL2 by THP-1 cells, *Evidence-Based Complementary Alternative Med.* 2015 (2015).
- [218] Y. Yuan, et al., Shufeng Jiedu capsules alleviate lipopolysaccharide-induced acute lung inflammatory injury via activation of GPR18 by verbenalin, *Cell. Physiol. Biochem.* 50 (2) (2018) 629–639.
- [219] Z. Tao, et al., Systematic analyses on the potential immune and anti-inflammatory mechanisms of Shufeng Jiedu Capsule against Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2)-caused pneumonia, *J. Funct. Foods* 75 (2020), 104243.
- [220] Z. Tao, et al., Complementary and alternative medicine is expected to make greater contribution in controlling the prevalence of influenza, *Biosci. Trends* 7 (5) (2013) 253–256.
- [221] Y. Liu et al. Study on mechanism of Shufeng Jiedu granules in treating novel coronavirus pneumonia based on network pharmacology, in *AIP Conference Proceedings*. 2020. AIP Publishing LLC.
- [222] S.K. Sandur, et al., Curcumin, demethoxycurcumin, bisdemethoxycurcumin, tetrahydrocurcumin and turmerones differentially regulate anti-inflammatory and anti-proliferative responses through a ROS-independent mechanism, *Carcinogenesis* 28 (8) (2007) 1765–1773.
- [223] H.P. Ammon, M.A. Wahl, *Pharmacology of Curcuma longa*, *Planta Med.* 57 (01) (1991) 1–7.
- [224] Y. Abe, S. Hashimoto, T. Horie, Curcumin inhibition of inflammatory cytokine production by human peripheral blood monocytes and alveolar macrophages, *Pharmacol. Res.* 39 (1) (1999) 41–47.
- [225] S. Hanazawa, et al., Tumor necrosis factor- α induces expression of monocyte chemoattractant JE via fos and jun genes in clonal osteoblastic MC3T3-E1 cells, *J. Biol. Chem.* 268 (13) (1993) 9526–9532.
- [226] A. Goel, A.B. Kunnumakkara, B.B. Aggarwal, Curcumin as “Curecumin”: from kitchen to clinic, *Biochem. Pharmacol.* 75 (4) (2008) 787–809.
- [227] S. Shishodia, T. Singh, M.M. Chaturvedi, Modulation of transcription factors by curcumin, The molecular targets and therapeutic uses of curcumin in health and disease (2007) 127–148.
- [228] Z.-J. Zhang, et al., Curcumin inhibits LPS-induced CCL2 expression via JNK pathway in C6 rat astrocytoma cells, *Cell. Mol. Neurobiol.* 32 (6) (2012) 1003–1010.
- [229] T.-Y. Chen, et al., Inhibition of enveloped viruses infectivity by curcumin, *PLoS ONE* 8 (5) (2013) e62482.
- [230] R.Y. Utomo, M. Ikawati, E. Meiyanto, Revealing the potency of citrus and galangal constituents to halt SARS-CoV-2 infection. 2020.
- [231] F. Zahedipour, et al., Potential effects of curcumin in the treatment of COVID-19 infection, *Phytother. Res.* 34 (11) (2020) 2911–2920.
- [232] M. Nemati, et al., Curcumin, an inhibitor of PAK1, potential treatment for COVID-19, *J. Infectiol. Epidemiol.* 3 (2) (2020).
- [233] M. Xia, Z. Sui, Recent developments in CCR2 antagonists, *Expert Opin. Ther. Pat.* 19 (3) (2009) 295–303.
- [234] A.M. Burkhardt, et al., CXCL17 is a major chemotactic factor for lung macrophages, *J. Immunol.* 193 (3) (2014) 1468–1474.