EDITORIAL

Contribution of mast cells in SARS-CoV-2-induced inflammation

G. Ronconi[1], C.E. Gallenga[2], Al. Caraffa[3], S.K. Kritas[4], I. Frydas[5], P. Conti[6], V. Calvisi[7] and M. Trimarchi[8]

[1]Clinica dei Pazienti del Territorio, Fondazione Policlinico Gemelli, Rome, Italy; [2]Department of Biomedical Sciences and Specialist Surgery, Section of Ophthalmology, University of Ferrara, Ferrara, Italy; [3]School of Pharmacy, University of Camerino, Camerino, Italy; [4]Department of Microbiology and Infectious Diseases, School of Veterinary Medicine, Aristotle University of Thessaloniki, Macedonia, Greece; [5]Aristotelian University, Thessaloniki, Greece; [6]Immunology Division, Postgraduate Medical School, University of Chieti, Pescara 65100, Italy [7]Department of Orthopedics, School of Medicine, L'Aquila, Italy; [8]Department of Medicine and Surgery, Centre of Neuroscience of Milan, University of Milan, Italy

COVID-19, which first appeared in China, has so far caused an unexpected number of deaths, as our immune system has not been able to annihilate the SARS-CoV-2 virus. SARS-CoV-2 reacts to both innate and acquired immunity. In the first instance, when the virus enters our organism, it is attacked by innate immune cells, including macrophages and mast cells (MCs), which produce defensive cytokines such as IL-1, IL-6, IL-33 and TNF; but the overproduction of these cytokines is very harmful to the patient. Here, in this editorial, we report that the inflammatory cytokine network established in COVID-19, in the most serious cases, can lead to the death of the patient. Therefore, it is pertinent to think that by blocking the pro-inflammatory cytokines that cause the "cytokine storm", a great therapeutical benefit can be achieved for COVID-19 disease.

In December 2019, a severe lung disease with respiratory syndrome induced by SARS-CoV-2 (coronavirus-2) appeared in Wuhan, China (1). A few months later, after so many people were infected, it was learned that it was a widespread pandemic that until today has not yet been eradicated.

COVID-19 pandemic, caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), is potentially life-threatening and has affected almost all countries globally, but it is hoped that through vaccination, the situation will improve considerably (2). The serious pandemic that is taking place due to the SARS-CoV-2 infection that causes COVID-19 mainly affects the upper and lower respiratory tracts, causing pneumonia and death. COVID-19 has caused unexpected mortality among the elderly, adults and now also children, causing significant psychological stress and economic repercussions.

At the onset of SARS-CoV-2 infection, the virus is attacked by innate immune cells, including mast cells (MCs) and macrophages (3). SARS-CoV-2 infection induces an innate and adaptive immune response to protect the host. However, innate immune cells such as macrophages and mast cells (MCs), which are supposed to fight the virus, release an overproduction of pro-inflammatory cytokines with serious problems locally and systemically. Therefore, innate immunity is important for the clearance of the SARS-CoV-2 virus, but cells that generate it, such as macrophages and MCs, can contribute to the aggravation of

Keywords: *mast cell, inflammation, immunity, cytokine, cytokine storm, lung, SARS-CoV-2, COVID-19*

Corresponding author:
Professor Pio Conti,
Immunology Division,
Postgraduate Medical School,
University of Chieti-Pescara,
66013 Chieti, (CH), Italy
e-mail: pioconti@unich.it

0393-974X (2022)

COVID-19. The SARS-CoV-2 virus uses the ACE2 receptor, an angiotensin II peptidase, by activating pattern recognition receptor (PRR) signaling and DAMP-mediated signaling pathways.

The intervention of macrophages in the inflammatory processes induced by many chemical and biological compounds, including SARS-CoV-2, mainly causes the secretion of powerful pro-inflammatory cytokines of the IL-1 family in macrophages and MCs. IL-1, IL-6, TNF and IL-33 released by those cells produce an autocrine action on the inflammatory network. The overproduction of inflammatory cytokines can cause the so-called "cytokine storm", an excessive immune response that can end in fatal respiratory distress due to organ failure. Along with other immune cells, MCs are involved in the defence against viruses, including SARS-CoV-2 (4). In severe COVID-19, an accumulation of MCs in the lung tissue has been seen to cause edema, clotting and thrombosis.

The cytokine storm is considered the culmination of severe inflammation, such as that caused by SARS-CoV-2, which occurs with high concentrations of cytokines both systemically and in the tissues, together with high fever and many disrupted biological parameters. Removing the cause of this severe inflammatory phenomenon is certainly helpful in treating the disease and can be lifesaving. The main cytokines that intervene in the cytokine storm are IL-1, IL-6, TNF and IL-33, all produced after viral infection by macrophages and MCs. The immune and inflammatory response is manifested more severely when there is a high viral load, triggering the "cytokine storm". Furthermore, it should be noted that the host's unregulated innate and adaptive immune responses against SARS-CoV-2 depend on virulence, viral replication, and the number of cytokines produced by immune cells, including MCs.

Mast cells (MCs)

MCs arise from hematopoietic bone marrow cells that migrate and mature in vascularized tissues. They reside near blood vessels, epithelial, nerves, in the tissue of the airways, in the tissue of the gastrointestinal tract, and other strategic places in the body, where they can encounter and attack the external agents. MCs also reside in the central nervous system (CNS) and possess a series of receptors to recognise microbes that can induce neuroinflammation. Two populations of MCs are currently known in mice.

The first are CTMCs, of the connective tissue, which reside in the skin, peritoneum and gastrointestinal tract. The second are MMCs, belonging to the gastric mucosa, which contain low levels of histamine and heparin. MMCs are more active and increase in infectious processes. MCs contain numerous granules, a reservoir of chemical mediators, that can be released into the cellular environment seconds after an allergic stimulus. In adaptive immunity, Th2 cells and other immune cells produce IL-4, IL-5 and IL-13, which induce MC activation and IgE production, protecting the body from microbial invasion. Cytokines IL-17 and IL-22 activate phagocytes by triggering an innate immune response to microorganisms producing cytokines, including IL-1, TNF, and IL-33, that activate the MCs, inducing inflammatory processes without degranulation.

Therefore, MCs are involved in allergy and anaphylactic reactions when the IgE binds to its FceRI receptor, quickly causing aggregation and degranulation. They can also be activated by antigens of microorganisms, including viral ones, inducing the *de novo* synthesis (late release) of cytokines such as IL-1, IL-6, IL-33, and TNF. The immediate allergic activation system of MCs and the delayed one with the production of cytokines seem to be distinct and remain unclear. Shedding light on this topic could help in the therapy of the inflammatory process (5). Microorganisms such as SARS-CoV-2 activate TLR2 and TR4 of MCs, which selectively release pro-inflammatory cytokines, in turn activating PAR-2 on endothelial cells and causing increased vascular permeability, leukocyte recruitment and inflammation. MCs also have chemokine receptors that promote virus binding and are a source of cytokines and chemokines that can be induced by various inflammatory stimuli (6); therefore, it is pertinent to think that MCs intervene in SARS-CoV-2 infection where the innate and adaptive immune system is activated.

TNF

Inflammatory cytokines play an important role in

the pathogenesis of infections where they are produced nonspecifically. IL-1 and TNF act on different receptors, but in inflammation, they have a similar effect. In fact, IL-1 induces TNF and vice versa. TNF is released by LPS-stimulated macrophages and can lyse some types of tumor cells. During infection with SARS-CoV-2, there can be a high production of TNF in the tissues and circulation, causing hypotension and shock that can lead to death in the patient. Within activated MCs, TNF can be produced and secreted through two different pathways; one with the induction of the mRNA and protein synthesis (late release), and the other associated with degranulation (immediate release), as TNF is the only cytokine pre-formed and stored in the secretory granules of MCs.

TNF also plays a role in fighting against microorganisms, including SARS-CoV-2. In fact, in inflammatory infections, there is an increase in macrophages, which produce high levels of TNF, stimulating the recruitment of leukocytes into the inflammatory sites. In COVID-19, TNF is one of the factors that participate in the phenomenon of the "cytokine storm", which can be lethal for the patient (7).

IL-1

The IL-1 family cytokine influences both immunological and inflammatory processes. IL-1 has a receptor similar to TLR, which binds microorganisms, including viruses. IL-1, which is formed through the activation of caspase-1, is a dominant cytokine in innate immunity and induces inflammation by binding to its IL-1R receptor (8). Viruses and other microorganisms, including SARS-CoV-2, bind to TLR, inducing inflammation. SARS-CoV-2 induces IL-1 secretion via TLR, causing an inflammatory response.

The inflammatory effect of IL-1 and that induced through the TLR are very similar. Several pro-inflammatory cytokines are involved in the initial phase of severe COVID-19, including IL-1, which participates in the "cytokine storm". COVID-19, which can be fatal, can occur after SARS-CoV-2 infection, with an overproduction of cytokines, including IL-1. This effect can be systemic, but above all, it can affect the lung with serious consequences. Hence, IL-1 is a key cytokine in the inflammatory response induced by SARS-CoV-2, and its production is mainly due to virus-activated macrophages and MCs. Therefore, activated macrophages secrete IL-1, which stimulates MCs to produce IL-6 and TNF, which participate in the inflammatory pathogenic processes.

In severe COVID-19, the serum levels of IL-1 are significantly increased, as well as the pathological effects. The use of IL-1 blockers can prevent multiple organ failure, septic shock, and acute respiratory deficiency syndrome.

IL-33

IL-33 is produced by various cells, including macrophages and MCs, and is a ligand of the ST2 receptor (9). The pro-IL-33 matures into IL-33 and binds to the transmembrane receptor ST2, recruiting MyD88, which activates the factor 6 associated with the TNF receptor (TRAF6), and subsequently the NFkB, resulting in the production of cytokines and chemokines, as can be seen from Fig. 1. IL-33 is an important cytokine in inflammatory diseases, including COVID-19. IL-33, also called "alarmin", is a key cytokine in acute and chronic inflammation in COVID-19, where MCs play a crucial role. Moreover, this cytokine plays an important role in the pathology of various hypersensitivity diseases where MCs are the protagonists (10).

MCs release IL-33 into the virus-damaged and

TIR domain

PRO-IL-33→IL-33→ST2→----------→MyD88→IRAK4, TRAF6→IKbeta, p50, p65→NFkB→CYTOKINES/CHEMOKINES

Fig. 1. *IL-33 binds to its receptor, ST2, and regulates the transcription genes of cytokines and chemokines.*

inflamed lung, dysregulating immune Treg cells and causing dysfunction of the entire immune system. In COVID-19, IL-33 not only mediates the inflammatory process but can also be involved in pulmonary fibrosis by acting on epithelial cells and fibroblasts.

Upon exposure to the SARS-CoV-2 virus or other pathogens, MCs become activated and produce IL-33, triggering the immune response. SARS-CoV-2 induces MCs to produce IL-33, which promotes the maturation, activation and survival of the cells, and secretes various cytokines, including IL-6. Therefore, inhibition of IL-33 can help treat inflammatory diseases, including that generated by SARS-CoV-2.

IL-6

IL-6, which is part of the IL-6 family, is made up of 10 members with has various functions both in health and in inflammatory pathological states (11). The cellular IL-6 receptor (IL-6R) is composed of IL-6Rα and the signal transducer gp130. Because IL-6 is expressed in many cells, it can interact with various cell types (12). IL-6 correlates with the clinical severity of inflammatory diseases where plasma levels of this cytokine increase, and the patient is more likely to die from septic shock. Unlike IL-1 and TNF, IL-6 does not cause activation of arachidonic acid metabolites, such as prostaglandins and leukotrienes, nor does it induce hypotension. IL-6 is a pleiotropic cytokine, which like other cytokines released by macrophages, mediates the immune response and collaborates with IL-1 and TNF in inducing the "cytokine storm" in COVID-19 disease.

The transcription of IL-6 genes is induced by various factors, including TLR ligands, IL-1 and TNF. The synergy of NF-κB and STAT3 can result in the hyperactivation of NF-κB, causing the release of pro-inflammatory cytokines. IL-1, an activator of NFkB, induces the production of IL-6 and virus infection, causing a systemic increase of IL-6 secreted by MCs and macrophages. In the immune system, IL-6 favours the formation of plasma cell antibodies and the activation of IL-6Ra or gp130, leading to the formation of JAK, together with gp130 (13).

Macrophages, MCs, and other cell types produce IL-6, which participates in the COVID-19 disease and increases the CRP (C reactive protein) levels, contributing to inflammation. Lowering the levels of IL-6 in COVID-19 can be of great therapeutic benefit.

REFERENCES

1. Ackermann M, Verleden SE, Kuehnel M, et al. Pulmonary Vascular Endothelialitis, Thrombosis, and Angiogenesis in Covid-19. N Engl J Med 2020; 383(2):120-128.
2. Abrams EM, Szefler SJ. COVID-19 and the impact of social determinants of health. Lancet Respir Med. 2020; 8(7):659-661.
3. Theoharides TC, Conti P. COVID-19 and Multisystem Inflammatory Syndrome, or is it Mast Cell Activation Syndrome? J Biol Regul Homeost Agents. 2020; 34(5):1633-1636.
4. Conti P, Ronconi G, Caraffa A, et al. Induction of pro-inflammatory cytokines (IL-1 and IL-6) and lung inflammation by Coronavirus-19 (COVI-19 or SARS-CoV-2): anti-inflammatory strategies. J Biol Regul Homeost Agents. 2020; 34(2):327-331.
5. Contini C, Enrica Gallenga C, Neri G, et al A new pharmacological approach based on remdesivir aerosolized administration on SARS-CoV-2 pulmonary inflammation: A possible and rational therapeutic application. Med Hypotheses. 2020; 144:109876.
6. Kempuraj D, Selvakumar GP, Ahmed ME, COVID-19, Mast Cells, Cytokine Storm, Psychological Stress, and Neuroinflammation. Neuroscientist. 2020; 26(5-6):402-414.
7. Ribeiro Dos Santos Miggiolaro AF, da Silva Motta Junior J, Busatta Vaz de Paula C, et al. Covid-19 cytokine storm in pulmonary tissue: Anatomopathological and immunohistochemical findings. Respir Med Case Rep. 2020; 31:101292.
8. Cavalli G, Colafrancesco S, Emmi G, et al. Interleukin 1alpha: a comprehensive review on the role of IL-1alpha in the pathogenesis and treatment of autoimmune and inflammatory diseases. Autoimmun Rev. 2021; 20(3):102763.
9. Taracanova A, Tsilioni I, Conti P, et al. Substance P and IL-33 administered together stimulate a marked secretion of IL-1beta from human mast cells, inhibited by methoxyluteolin. Proc Natl Acad Sci U S A. 2018; 115(40):E9381-E9390.

10. Theoharides TC, Petra AI, Taracanova A, et al. Targeting IL-33 in autoimmunity and inflammation. J Pharmacol Exp Ther. 2015; 354(1):24-31.
11. Conti P, Kempuraj D, Di Gioacchino M, et al. Interleukin-6 and mast cells. Allergy Asthma Proc. 2002;23(5):331-5.
12. Conti P, Bartle L, Barbacane RC, Synergistic activation of serum amyloid A (SAA) by IL-6 and IL-1 in combination on human Hep 3B hepatoma cell line. Role of PGE2 and IL-1 receptor antagonist. Immunol Invest. 1995; 24(3):523-35.
13. Coomes EA, Haghbayan H.Interleukin-6 in Covid-19: A systematic review and meta-analysis. Rev Med Virol. 2020; 30(6):1-9.