



HAL
open science

Adjunct immunotherapies for the management of severely ill COVID-19 patients

Srinivasa Reddy Bonam, Srini V Kaveri, Anavaj Sakuntabhai, Laurent Gilardin, Jagadeesh Bayry

► **To cite this version:**

Srinivasa Reddy Bonam, Srini V Kaveri, Anavaj Sakuntabhai, Laurent Gilardin, Jagadeesh Bayry. Adjunct immunotherapies for the management of severely ill COVID-19 patients. *Cell Reports Medicine*, 2020, 1 (2), pp.100016. 10.1016/j.xcrm.2020.100016 . inserm-02555342v2

HAL Id: inserm-02555342

<https://inserm.hal.science/inserm-02555342v2>

Submitted on 1 May 2020 (v2), last revised 26 Jun 2020 (v3)

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.



Distributed under a Creative Commons Attribution - NonCommercial - NoDerivatives 4.0 International License

Journal Pre-proof

Adjunct immunotherapies for the management of severely ill COVID-19 patients

Srinivasa Reddy Bonam, Srinivasa V. Kaveri, Anavaj Sakuntabhai, Laurent Gilardin, Jagadeesh Bayry



PII: S2666-3791(20)30021-5

DOI: <https://doi.org/10.1016/j.xcrm.2020.100016>

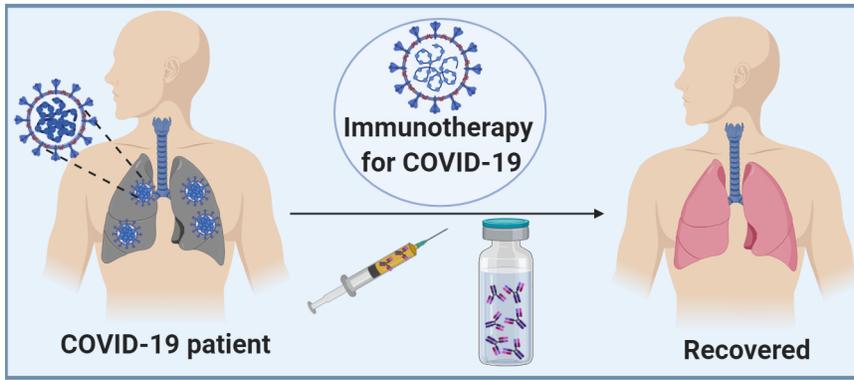
Reference: XCRM 100016

To appear in: *Cell Reports Medicine*

Please cite this article as: Bonam, S.R., Kaveri, S.V, Sakuntabhai, A., Gilardin, L., Bayry, J., Adjunct immunotherapies for the management of severely ill COVID-19 patients, *Cell Reports Medicine* (2020), doi: <https://doi.org/10.1016/j.xcrm.2020.100016>.

This is a PDF file of an article that has undergone enhancements after acceptance, such as the addition of a cover page and metadata, and formatting for readability, but it is not yet the definitive version of record. This version will undergo additional copyediting, typesetting and review before it is published in its final form, but we are providing this version to give early visibility of the article. Please note that, during the production process, errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

© 2020 The Author(s).



Journal Pre-proof

Adjunct immunotherapies for the management of severely ill COVID-19 patients

Srinivasa Reddy Bonam¹, Srini V Kaveri¹, Anavaj Sakuntabhai², Laurent Gilardin³, Jagadeesh Bayry^{1,*}

¹Institut National de la Santé et de la Recherche Médicale ; Centre de Recherche des Cordeliers, Equipe - Immunopathologie et Immunointervention Thérapeutique, Sorbonne Université, Université de Paris, Paris F-75006, France

²Unité de Génétique Fonctionnelle des Maladies Infectieuses, Institut Pasteur; CNRS UMR2000 Génomique Évolutive, Modélisation et Santé, Institut Pasteur, Paris F-75015, France

³Hôpital Bégin, Saint Mandé F-94160, France

*Correspondence to (lead author): jagadeesh.bayry@crc.jussieu.fr

Abstract

Coronavirus disease 2019 (COVID-19) is caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). It has infected millions with more than 181,000 fatal cases as of 22nd April 2020. Currently, there are no specific COVID-19 therapies. Most patients depend on mechanical ventilation. Current COVID-19 data clearly highlight that cytokine storm and activated immune cell migration to the lungs characterize the early immune response to COVID-19 that causes severe lung damage and development of acute respiratory distress syndrome. In view of uncertainty associated with immunosuppressive treatments such as corticosteroids and their possible secondary effects, including risks of secondary infections, we suggest immunotherapies as an adjunct therapy in severe COVID-19 cases. Such immunotherapies based on inflammatory cytokine neutralization, immunomodulation and passive viral neutralization, not only reduce inflammation, inflammation-associated lung damage, or viral load, but could also prevent intensive care unit hospitalization and dependency on mechanical ventilation both of which are limited resources.

Keywords: SARS-CoV-2; COVID-19; Cytokine storm syndrome; Immunotherapy; Cytokines; Monoclonal antibody; Inflammation; Passive immunotherapy; IVIG; Convalescent plasma; Hyperimmune globulin

Introduction

Coronavirus disease 2019 (COVID-19) is caused by "Severe Acute Respiratory Syndrome Coronavirus 2" (SARS-CoV-2). It has affected over 2.6 million people with more than 181,000 fatal cases as of 22nd April 2020 (<https://coronavirus.jhu.edu/map.html>). While the majority of cases result in mild symptoms such as fever and cough, about 10-15% progress to severe pneumonia and respiratory failure requiring intensive care unit hospitalization and mechanical ventilation. The death rate in intensive care unit (ICU) admitted patients could reach up to 26%. Age and other health problems such as chronic pulmonary or cardiac disease conditions are the risk factors that determine the mortality rate.¹⁻⁴ A systematic review and meta-analysis confirmed that old age and/or associated comorbidity are the common risk factors that predict death in all three major respiratory diseases caused by coronaviruses including COVID-19, Severe Acute Respiratory Syndrome (SARS) and Middle East Respiratory Syndrome (MERS).⁵ Obesity is another risk factor for the development of severe SARS-CoV-2.⁶ Though anti-viral molecules and hydroxychloroquine are under investigation to avert severe disease,⁷⁻¹¹ currently, there are no specific therapies for COVID-19, particularly when severe acute respiratory distress syndrome (ARDS) occurs. Most COVID-19 patients with ARDS required intensive care unit hospitalization and prolonged supportive mechanical ventilator management.^{1,2,4} ARDS can lead to other complications, including secondary bacterial infection and lung fibrosis.

Inflammatory response to COVID-19

Recent clinical data suggest that severe COVID-19 is due to an overreactive immune response leading to a cytokine storm and development of acute respiratory distress syndrome.^{12,13} Plasma obtained from COVID-19 patients, in particular moribund patients, demonstrated increased

concentrations of various inflammation-related cytokines and chemokines implicated in the recruitment of immune cells including interleukin (IL)-1 β , tumor necrosis factor (TNF)- α , IL-2, IL-6, IL-7, IL-8, IL-9, IL-17, granulocyte-colony stimulating factor (G-CSF), granulocyte-macrophage colony-stimulating factor (GM-CSF), interferon (IFN)- γ , C-X-C motif chemokine 10 (CXCL10), chemokine ligand 2 (CCL2), CCL3 and CCL4.^{1,14,15} In addition, COVID-19 patients showed relatively increased neutrophil counts in the blood.¹⁵ On the other hand, monocytes, basophils and T cells, especially CD4⁺ and CD8⁺ T cells, were decreased in the peripheral blood, possibly due to their migration to the lungs.¹³⁻¹⁷ However, these T cells exhibited enhanced activated markers (CD69, CD38 and CD44).¹⁷

Single-cell RNA sequencing of bronchoalveolar lavage (BAL) fluid immune cells from a small number of mild to severely ill COVID-19 patients suggested that balance in the lung macrophage populations is highly dysregulated in severely ill COVID-19 patients. These data indicate that monocyte-derived macrophages (FCN1⁺) rather than FABP4⁺ alveolar macrophages contribute to lung inflammation and lung damage in severe cases.¹⁸ This study thus confirms another report based on lung postmortem biopsy of a patient that revealed the presence of bilateral diffuse alveolar damage and recruitment of monocytes.¹³ The study by Liao et al. also observed an increased proportion of clonally expanded CD8⁺ T cells in the BAL fluid of mild cases as compared to severe cases.¹⁸ Another investigation that compared BAL fluid immune cells in various respiratory pathologies highlighted a more prominent surge of neutrophils in COVID-19 patients as compared to pneumonia caused by other pathogens.¹⁹ By metatranscriptome sequencing of BAL fluid and global functional analyses of differentially expressed genes, this report also identified strong upregulation of numerous type I IFN-inducible genes.¹⁹

However, caution needs to be exercised while interpreting these data due to potential influence of therapies such as IFN α -2b, ant-viral and/or steroids on the landscape of immune cells and immune signatures of BAL fluid or lungs. Nevertheless, these reports confirm the proposition that an influx of immune cells to the lungs follows SARS-CoV-2 infection (**Figure 1**).

Severely ill COVID-19 patients also displayed reduced peripheral blood regulatory T cells (Tregs), the immune suppressor cells critical for reducing inflammation and inflammation-associated tissue damage.¹⁵ Another report suggested that activated GM-CSF⁺IFN γ ⁺ pathogenic Th1 cells that secrete GM-CSF promote inflammatory CD14⁺CD16⁺ monocyte responses with enhanced IL-6.¹⁷ GM-CSF⁺IFN γ ⁺ Th1 cells and inflammatory monocytes were positively correlated with the severe pulmonary syndrome characteristic of COVID-19 patients.¹⁷ The simultaneous increase in IL-1 receptor antagonist (IL-1RA) and IL-10 also suggest that anti-inflammatory responses, though induced, are not sufficient to reduce inflammation and eventually lead to severe lung damage.

Biomarkers that could predict ARDS in COVID-19 patients

Various reports have shown that higher inflammation-related biomarkers such as plasma C-reactive protein (CRP), ferritin and IL-6 were significantly associated with higher risks of developing ARDS.²⁰⁻²⁴ Of note, IL-6 levels were associated with course of the disease and death from COVID-19.^{20,23,24}

A recent systematic review and meta-analysis of 30 studies conducted in China (26 studies, of which 13 are from Wuhan), Australia, USA and Korea that included 53,000 patients with COVID-19, has confirmed that raised CRP (41.12-67.62 mg/L in severe vs 12.00-21.48 in mild cases) and ferritin (654.26-2087.63 ng/mL vs 43.01-1005.97) are found in seriously ill COVID-19 patients.⁵ The massive increase in plasma ferritin levels is indicative of haemophagocytic lymphohistiocytosis activation syndrome in these patients. These studies thus provide a rationale for targeting inflammatory mediators for the management of severely ill COVID-19 patients.

The use of corticosteroids in COVID-19 patients

Currently, there is no clear evidence for the use of steroids in SARS-CoV-2 infections and their use is highly debated, particularly with respect to the window of treatment, dose and management of patients in cases of bacterial co-infection. A retrospective cohort analysis of 201 patients from Wuhan suggested that methylprednisolone might benefit patients who develop ARDS (n=88) by reducing the death rate.²⁰ A retrospective analysis of hospitalized patients with severe COVID-19 pneumonia (n=46) indicated that an early low-dose steroid therapy (1-2 mg/kg/day) in 26 patients for a short duration (5-7 days) reduced the oxygen requirement period and improved disease course.²⁵

However, COVID-19 patients treated with methylprednisolone were sicker and had a higher pneumonia severity index than those patients who did not receive methylprednisolone.²⁰ Recently, an additional retrospective matched case-control study involving 31 paired patients in several ICUs in China showed that treatment with steroids for 8 days (4-12 days) in addition to antiviral drugs was associated with a 39% 28-day death rate, compared to 16% in a propensity

score matched control group.²⁶ Clinical evidence is also lacking for the use of steroids in other coronavirus diseases (SARS-CoV, MERS-CoV),^{27,28} and steroids might even impair viral clearance mechanisms of patients and predispose them to secondary infections. Double-blinded randomized clinical trials involving a large number of patients should be conducted to evaluate the use of steroids in severely ill COVID-19 patients with reduction in intensive care unit requirement as the primary endpoint.

Therefore, as per WHO interim guidance of 13th March 2020,²⁹ and some local guidelines (covidprotocols.org),³⁰ steroids are not recommended at present as a treatment for COVID-19. If steroids are used for a particular pathological condition, low dose and short duration of treatment is recommended. Similarly, as per a survey from the French National Agency for Medicines and Health Products Safety (ANSM), non-steroidal anti-inflammatory drugs (NSAIDs) like ibuprofen are also not recommended due to infectious complications and kidney, cardiac and gastrointestinal adverse effects.³¹

In view of an uncertain prognosis and complications due to generalized immunosuppression from corticosteroid and NSAID treatment, immunotherapies based on immunomodulatory approaches appear promising in the treatment of COVID-19 to regulate inflammation and anti-viral immune responses. We believe that the immunotherapies discussed next could result in a quicker therapeutic response with minimal adverse effects.

Immunotherapies as an adjunct to COVID-19 patient management

The current clinical data on COVID-19 patients clearly highlight that cytokine storm and an influx of activated immune cells to the lungs characterize the early immune response to COVID-19 that causes severe lung damage. Therefore, we suggest host-directed immunotherapies as an adjunct therapy in severe cases to not only reduce inflammation and inflammation-associated lung damage, but also to prevent ICU hospitalization and dependency on mechanical ventilation that are limited resources in the context of SARS-CoV-2 pandemic. In fact, several immunotherapeutic approaches that target either inflammatory mediators, passively neutralize SARS-CoV-2, or prevent viral entry are under evaluation at various centers (**Table 1 & Figure 2**), thus giving perfect examples of such drug repurposing. In addition, lessons learnt from the management of other severe respiratory diseases as in the case of SARS, MERS and severe influenza provide a roadmap for tackling ARDS and cytokine storm in severely ill COVID-19 patients.^{27,32,33}

Immunotherapies that target specific inflammatory molecules and pathways

An increase in IL-6 leading to lung tissue damage has been observed in a majority of the COVID-19 patients.^{13,14,16,17} Of note, similar to severely ill COVID-19 cases, elevated serum levels of IL-6, TNF- α and IFN- γ have been consistently observed in cytokine release syndrome (CRS) that is common in the patients receiving T cell-engaging immunotherapies (bispecific antibody constructs or chimeric antigen receptor (CAR) T cell therapies).³⁴ Targeting either IL-6 (siltuximab, a chimeric monoclonal antibody) or IL-6 receptor (tocilizumab, a recombinant humanized monoclonal antibody) led to rapid resolution of CRS symptoms in those patients.³⁴ Severe CRS can also lead to hypotension, pulmonary edema and cardiac dysfunction that are

commonly observed in severely ill COVID-19 patients. These data provide justification for targeting IL-6 in severely ill COVID-19 patients.

Both anti-IL-6 receptor antibodies (tocilizumab, and a human monoclonal antibody sarilumab (Kevzara®)), and anti-IL-6 antibody siltuximab (Sylvant®) are under evaluation in COVID-19 patients at various centers (**Table 1**). An immunosuppressed patient with severe COVID-19-related lung disease was successfully treated with tocilizumab and therapy led to a partial decrease in the pulmonary infiltrates.³⁵ Unpublished data based on off-label use of tocilizumab along with lopinavir and methylprednisolone in 21 severely ill COVID-19 patients suggested that the addition of tocilizumab not only quickly reduced their clinical symptoms (fever, oxygen requirement) and inflammation, but also normalized peripheral blood T cell counts in the majority of patients.³⁶

Additional targets include TNF- α (infliximab, a chimeric monoclonal antibody to TNF- α ; etanercept, a recombinant fusion TNF-R that binds soluble TNF- α) and IFN- γ (emapalumab, a fully human IgG1 monoclonal antibody against IFN- γ) (**Table 1**). Of note, clinical trials in idiopathic pneumonia syndrome have reported favorable response to etanercept therapy.^{37,38}

GM-CSF represents another target for severe cases of COVID-19. GM-CSF stimulates the influx of granulocytes and monocytes from bone marrow, and hence further enhances inflammatory responses. A randomized Phase 1b/2, double-blind, placebo-controlled clinical trial is currently recruiting patients to investigate the therapeutic efficacy of a humanized anti-GM-CSF IgG1 monoclonal antibody TJ003234 in severely ill COVID-19 patients (NCT04341116). In addition,

another human monoclonal antibody that targets GM-CSF (gimsilumab; Roivant Sciences Ltd., Altasciences Co. Inc.) is under consideration for treating ARDS in COVID-19 patients.^{39,40}

Likewise, baricitinib, a selective Janus kinase (JAK1/JAK2) inhibitor, inhibits gp130 family cytokines (such as IL-6, IL-12, IL-23 and IFN- γ).⁴¹ In addition, baricitinib inhibits the AP2-associated protein kinase 1 and cyclin G-associated kinases that are regulators of viral endocytosis process and hence could block viral infection of cells.⁴¹ Similarly, anakinra (Kineret®), a recombinant human IL-1RA and FDA-approved Janus kinase 2 inhibitor, fedratinib (Inrebic®) also possess the capacity to suppress inflammation (**Table 1**).⁴² Though, JAK inhibitors can impair type I IFN-mediated anti-viral responses, recent data from metatranscriptome sequencing of BAL fluid from eight COVID-19 patients highlight that SARS-CoV-2 induces strong type I IFN responses with an overexpression of a large number of type I IFN-inducible genes implicated in inflammation.¹⁹ Therefore, use of JAK inhibitors might help in alleviating inflammation, but dose and window of treatment should be tailored to ensure that anti-viral response of the patient is not totally curtailed.

Type I IFN ($\beta 1\alpha$ and $\beta 2\alpha$) is under consideration for severe COVID-19 (**Table 1**). Type I IFN signals through JAK-STAT (signal transducers and activators of transcription) pathway and upregulates IFN-stimulated genes that kill viruses in the infected cells. Our knowledge on the role of type I IFN in the pathogenesis of SARS-CoV-2 infection is not complete. Current evidence, however, reveals an overexpression of a large number of type I IFN-inducible genes with immunopathogenic potentials in COVID-19 patients.¹⁹ Also, results on the use of IFN- α in

SARS patients are inconclusive.²⁷ Therefore, care should be taken regarding the dosage of type I IFN and timing of treatment to avoid deleterious effects.

Intravenous immunoglobulin therapy

Additional immunotherapies that could be considered for COVID-19 include immunomodulators that have broad anti-inflammatory effects such as pooled normal IgG or intravenous immunoglobulin (IVIG) therapy. Obtained from the pooled plasma of several thousand healthy donors, IVIG is one of the most widely used immunotherapies for a large number of autoimmune and inflammatory diseases.^{43,44} A recent open-label trial in three patients reported benefits of IVIG therapy (0.4 g/kg for five days) in severe SARS-CoV-2-induced pneumonia⁴⁵ thus providing another option for the management of COVID-19 patients. Another multicenter retrospective cohort study indicated that IVIG therapy could benefit critical COVID-19 patients, but dose (0.1-0.5 g/kg) and duration of therapy (5-10 days) were too variable.⁴⁶ However, grouping of patients based on the dose of IVIG (>15g/day or ≤15g/day) and timing of infusion (>7 or ≤7 days since hospitalization) implied that beneficial effects are associated with high dose and early IVIG treatment.⁴⁶ A retrospective study on 58 severe or critically ill cases of COVID-19 also found that adjunct therapy with IVIG within 48 hours of hospitalization reduced hospital stay and ventilator use, and improved 28-day mortality.⁴⁷

Other than IL-6 and CRP, inflammatory parameters following IVIG immunotherapy were not evaluated in detail in these patients.⁴⁶ Based on the vast literature on the mechanisms of IVIG, we suggest that IVIG suppresses the activation and secretion of inflammatory cytokines from the innate immune cells, blocks the inflammatory Th1 and Th17 responses, exerts scavenging effects

on complement cascade and possibly enhances Tregs.^{48,49} In addition, IVIG contains antibodies to diverse pathogens and their antigens,^{50,51} and hence secondary infections could also be prevented in IVIG-treated patients.

Though these studies provide a pointer for initiating a randomized clinical trial with a large number of patients (NCT04261426) (**Table 1**), certain key aspects need to be considered. First, the cost associated with IVIG immunotherapy could be considered prohibitive, and second, the necessity to reserve IVIG for patients whose survival essentially depends on this immunotherapy, particularly primary immunodeficient patients, must be weighed. Currently, there is a worldwide shortage of IVIG and the COVID-19 pandemic will affect the collection of plasma from donors for IVIG production. Therefore, during this emergency period, providing IVIG to those patients who depend on it should take priority over initiating an IVIG trial in COVID-19 patients.

Passive immunotherapy

Studies on COVID-19 patients' sera confirmed the presence of seroconverted antibodies, such as IgG, IgM, and IgA with varying kinetics and sensitivities.^{52,53} Findings in a monkey SARS-CoV-2 infection model confirmed that primary infection could protect the host from reinfection perhaps aided by humoral immune responses.⁵⁴ Therefore, passive immunization of SARS-CoV-2 by using convalescent plasma, intravenous hyperimmune globulin (containing high concentrations of neutralizing antibodies obtained from the pooled plasma of large number of recovered patients) or neutralizing monoclonal antibodies represent another potential therapeutic option.

A meta-analysis reported a significantly reduced mortality rate in SARS-CoV-infected patients following infusion of convalescent plasma. These patients recorded a rapid decline in their virus load.⁵⁵ Although not conclusive, a report with MERS-CoV also suggested that the use of convalescent plasma could be an option for COVID-19.⁵⁶ Similarly, a meta-analysis of patients with Spanish influenza pneumonia concluded that influenza-convalescent human blood products reduced the death risk.⁵⁷ Hyperimmune globulin treatment in severe H1N1 influenza within five days of symptoms also reduced viral load and mortality in patients.⁵⁸

Development of neutralizing monoclonal antibodies in cases of SARS-CoV and MERS-CoV focused mainly on spike (S) protein as a target.⁵⁹ Several monoclonal antibodies have demonstrated efficacy in experimental models.^{60,61} Either antibody libraries consisting of entire V_H and V_L genes of B cells or antigen-specific antibodies isolated from the memory B cells of convalescent COVID-19 patients could be used to obtain potent neutralizing monoclonal antibodies to SARS-CoV-2. Various laboratories are actively exploring this option.⁶² As S proteins of SARS-CoV and SARS-CoV-2 are phylogenetically closely related, SARS-CoV neutralizing monoclonal antibodies with strong cross-neutralizing activity on SARS-CoV-2 represent an additional possibility.^{63,64}

It is important to validate the neutralizing activity of aforementioned plasma or antibody preparations before using with COVID-19 patients; particularly in the case of convalescent plasma as neutralizing activity might significantly differ in mildly vs. severely ill patients. Because, neutralizing monoclonal antibodies or hyperimmune globulins are pre-validated for

their neutralizing activity, they do not pose such a dilemma. It is essential to note that the median seroconversion time for IgM and IgG was day 12 and day 14 respectively following onset of COVID-19.^{65,66} At day-15 following onset of disease, the presence of antibodies was 94.3% in case of IgM and 79.8% for IgG.⁶⁵ Therefore, convalescent plasma and hyperimmune globulins should be collected at least three-weeks post SARS-CoV-2 infection to increase the chances of obtaining high titer neutralizing antibodies.

A preliminary report from China suggested that convalescent plasma therapy leads to improvement in 91 of 245 COVID-19 patients treated.⁶⁷ An uncontrolled trial on transfusion of 400 mL of convalescent plasma (with viral neutralizing titers more than 1:40) in five patients critically ill with COVID-19 led to resolution of pulmonary lesions, and reduction in the severity of the disease and viral loads.⁶⁸ Similarly, treatment of ten severely ill COVID-19 patients with 200 mL of convalescent plasma containing viral neutralizing antibody titers more than 1:640 (A dilution of plasma that neutralized 100 TCID₅₀ (50% tissue-culture-infective dose) of SARS-CoV-2) led to reduced CRP levels, undetectable viremia and improved clinical symptoms.⁶⁹ Several randomized Phase 2 and 3 clinical trials are already planned with convalescent plasma or hyper immunoglobulin in many centers (**Table 1**) and the plasma fractionation industry has offered support for such efforts.^{70,71}

Meplazumab, a humanized anti-CD147 IgG2 monoclonal antibody is under evaluation for COVID-19 pneumonia.⁷² This antibody therapy was aimed at preventing CD147-mediated SARS-CoV-2 viral entry and T cell chemotaxis. Preliminary data in a small number of patients

indicated that meplazumab improved clinical course of the disease, and normalized the peripheral lymphocyte count and CRP level.⁷²

Treg targeted therapies

Tregs are classically known to suppress anti-microbial protective immunity by affecting the activation of both innate and adaptive immune cells. However, Tregs are also important for preventing inflammation-associated tissue damage in acute infections.^{73,74} As mentioned earlier, the Treg number is affected in COVID-19 patients, which explains in part severe inflammation and lung damage observed in seriously ill COVID-19 patients. Treg therapy by adoptive transfer of ex vivo expanded autologous Tregs has been evaluated in autoimmune diseases. However, considering the cost and time required for the expansion of clinical-grade Tregs, adoptive Treg therapy is not feasible for COVID-19. Alternatively, therapies that enhance Treg number in vivo or use Treg-derived molecules would benefit severely ill COVID-19 patients.

IL-2-based therapies to expand Tregs: IL-2 complexed to a monoclonal antibody that recognizes the CD122/ γ c (dimeric IL-2 receptor) epitope of IL-2 was constructed in order to selectively activate Tregs that constitutively express trimeric IL-2R (dimeric IL-2 and CD25). This monoclonal antibody-IL-2 complex displayed promising results in experimental models of several autoimmune and inflammatory diseases.⁷⁵ Recently, a human anti-IL-2 antibody named F5111.2 has been designed that potentiates Tregs by a structure-based mechanism wherein this antibody stabilizes IL-2 in a conformation that results in the preferential STAT5 phosphorylation of Tregs.⁷⁶ Though not yet tested in humans, such approaches could boost Treg number in COVID-19 patients and reduce inflammation. On the other hand, low dose-IL-2 therapy has been explored in the clinic for several autoimmune diseases to expand Tregs.⁷⁷ But due to the fact that

COVID-19 patients are reported to have high IL-2 in the blood, this option might not be useful. Our opinion is that neutralization of cytokines or immunomodulatory therapies are preferred over Treg expansion strategies in view of the fact that due to cytokine storm, Tregs might not be fully functional unless inflammatory cytokines are nullified.

Treg-derived therapeutic molecules: Cytotoxic T-lymphocyte-associated protein 4 (CTLA-4) plays a critical role in Treg-mediated suppression by interacting with CD80/86 on antigen presenting cells and inducing trans-endocytosis of those co-stimulatory molecules. A recombinant CTLA-4 fusion protein consisting of the Fc portion of IgG1 (Abatacept, Belatacept) has been approved for the therapy of rheumatoid arthritis.⁷⁸ This therapeutic molecule could also be tested for reducing inflammation in COVID-19 patients. The clinical spectra of SARS-CoV-2 infection in check-point inhibitor-treated cancer patients and abatacept/belatacept-treated autoimmune patients should provide a valuable indicator for the use of abatacept/belatacept.

Natural killer (NK) cell-based immunotherapy

Currently, cellular therapies have not been given much consideration for COVID-19, possibly due to uncertainties of such approaches, time and economic reasons. Nevertheless, because of their anti-viral effector functions, several NK cell-based approaches are under consideration.

Little is known about the efficacy of NK cell therapy in infectious diseases. Conventional treatment in combination with NK cells (twice a week, $0.1-2 \times 10^7$ NK cells/kg body weight) is under clinical evaluation (NCT04280224) (**Table 1**). However, the source of NK cells for the therapy is not precise in this trial. Recently, FDA has given permission to Celularity Inc, to

investigate an universal NK cell therapy CYNK-001 in COVID-19 patients.⁷⁹ CYNK-001 contains human placental CD34⁺ stem cell-derived NK cells and are culture-expanded.

An innovative NK cell-based therapy with multipronged actions called NKG2D-ACE2 CAR-NK cell therapy is currently recruiting patients (NCT04324996) (**Table 1**). NKG2D-ACE2 CAR-NK cells secrete IL-15 (to ensure long-term survival of NK cells) and GM-CSF neutralizing scFv (to prevent CRS). In addition to clearance of virus-infected cells through ACE2, these NK cells can competitively inhibit SARS-CoV-2 infection of susceptible cells including alveolar epithelial cells. Although the strategy looks rational, toxicity due to clearance of virus-infected lung cells, should be closely monitored.

In addition to NK cell therapy, other cell-based therapies like SARS-CoV-2-specific T cell therapies are also on the horizon.

Other therapeutic options for COVID-19

Many anti-viral drugs such as protease inhibitors (camostat mesilate, lopinavir/ritonavir, and darunavir/cobicistat), nucleoside analogues (remdesivir, ribavirin), neuraminidase inhibitors (oseltamivir), RNA polymerase inhibitors (favipiravir), viral fusion inhibitor (umifenovir), and viral endonuclease inhibitor (baloxivir marboxil) are under evaluation as a treatment for SARS-CoV-2.⁸⁻¹⁰ An early compassionate-use of remdesivir for 10 days in a small cohort of hospitalized COVID-19 patients, reported clinical improvement in 36 of 53 treated patients (68%).¹⁰ Lopinavir/ritonavir, and remdesivir were also found to be effective for MERS-CoV and SARS-CoV.⁸ Ribavirin however, did not show benefits for MERS-CoV and SARS-CoV and was

associated with severe side effects like hemolytic anemia and liver dysfunction.^{27,32} The window of treatment is critical with anti-viral drugs, and they need to be used early in a disease. Late initiation of therapy with anti-viral drugs, particularly after 10-14 days post appearance of clinical symptoms, might lead to a poor outcome.⁷

Though the study design and results were controversial, data from a small number of patients suggested utility of hydroxychloroquine for COVID-19 treatment.¹¹ The follow-up study by the same group in a cohort of 1061 COVID-19 patients demonstrated that a hydroxychloroquine and azithromycin combination when given during the early phase of the disease, prevents aggravation of the disease and viral persistence.⁸⁰ Further details of this study are awaited. A randomized trial also observed improved pneumonia in hydroxychloroquine-treated patients.⁸¹ Chloroquine and hydroxychloroquine possess a unique ability to increase the pH of lysosomes and hence prevent SARS-CoV-2 viral fusion and replication. In vitro studies on chloroquine suggested anti-SARS-CoV-2 effects of these molecules.^{82,83} Interestingly, previous studies have demonstrated anti-inflammatory activity of chloroquine for the treatment of autoimmune diseases. But other clinical trials did not support the use of hydroxychloroquine, particularly in hospitalized COVID-19 patients with hypoxic pneumonia or severe COVID-19 cases.^{84,85} Therefore, it appears that any efficacy hydroxychloroquine might have requires that it be given during the early phase of COVID-19 before progression to more severe disease. However, due to side effects, particularly cardiac, it is recommended to use chloroquine and hydroxychloroquine under strict medical surveillance. More evidence from randomized clinical trials is required

Conclusion and perspectives

The immediate surge in COVID-19 positive cases prompted clinicians all around the world to explore a range of therapeutic interventions for severely ill COVID-19 patients. Due to the lack of specific therapies and time required to develop potent vaccines for SARS-CoV-2, current investigations are focused on available anti-viral drugs.

As inflammation is the hallmark of SARS-CoV-2 infections, immunotherapies that target various players of inflammation are currently being investigated. We propose that a combination of anti-viral drugs and anti-inflammatory immunotherapies during early phase of the disease would function in a synergistic manner and provide maximum benefits. Anti-viral drugs would reduce the viral load and hence lessen the stimuli for inflammatory responses, while immunotherapies would correct the dysregulated inflammatory response and prevent lung damage. Care must be taken regarding an appropriate window of treatment and selecting the dose of anti-inflammatory immunotherapies as excessive suppression of inflammation might reduce the capacity of the patient to mount anti-viral responses. Data from randomized trials are urgently needed to confirm the utility of the aforementioned immunotherapies in the management of critically ill COVID-19 patients.

Note:

The majority of the articles referred to in this manuscript are unpublished and have not yet undergone the peer-review process. They were obtained from the various preprint servers.

Acknowledgements

We thank three anonymous reviewers for their suggestions. Supported by the Institut National de la Santé et de la Recherche Médicale (INSERM), Sorbonne Université, Université de Paris, Institut Pasteur, and Agence Nationale de la Recherche : ANR-19-CE17-0021(BASIN).

Author Contributions

Article Conception, J.B.

Literature Survey and Writing, S.R.B., L.G., and J.B.

Editing and Reviewing, S.V.K., and A.S.

Final Approval, All the authors.

Declaration of Interests

The authors declare no competing interests

References

1. Huang, C., Wang, Y., Li, X., Ren, L., Zhao, J., Hu, Y., Zhang, L., Fan, G., Xu, J., Gu, X., et al. (2020). Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. *Lancet* 395, 497-506.
2. Guan, W.-J., Ni, Z.-Y., Hu, Y., Liang, W.-h., Ou, C.-q., He, J.-x., Liu, L., Shan, H., Lei, C.-l., Hui, D.S.C., et al. (2020). Clinical Characteristics of Coronavirus Disease 2019 in China. *New Engl. J. Med.* Published online February 28, 2020. <https://doi.org/10.1056/NEJMoa2002032>
3. Zhou, P., Yang, X.L., Wang, X.G., Hu, B., Zhang, L., Zhang, W., Si, H.R., Zhu, Y., Li, B., Huang, C.L., et al. (2020). A pneumonia outbreak associated with a new coronavirus of probable bat origin. *Nature* 579, 270-273.
4. Grasselli, G., Zangrillo, A., Zanella, A., Antonelli, M., Cabrini, L., Castelli, A., Cereda, D., Coluccello, A., Foti, G., Fumagalli, R., et al. (2020). Baseline Characteristics and Outcomes of 1591 Patients Infected With SARS-CoV-2 Admitted to ICUs of the Lombardy Region, Italy. *JAMA*. Published online April 6, 2020. <https://doi.org/10.1001/jama.2020.5394>
5. Zhao, X., Zhang, B., Li, P., Ma, C., Gu, J., Hou, P., Guo, Z., Wu, H., and Bai, Y. (2020). Incidence, clinical characteristics and prognostic factor of patients with COVID-19: a systematic review and meta-analysis. *medRxiv*. Published online March 20, 2020. <https://doi.org/10.1101/2020.03.17.20037572>
6. Simonnet, A., Chetboun, M., Poissy, J., Raverdy, V., Noulette, J., Duhamel, A., Labreuche, J., Mathieu, D., Pattou, F., Jourdain, M., et al. (2020). High prevalence of obesity in severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) requiring invasive mechanical ventilation. *Obesity*. Published online April 9, 2020. <https://doi.org/10.1002/oby.22831>
7. Cao, B., Wang, Y., Wen, D., Liu, W., Wang, J., Fan, G., Ruan, L., Song, B., Cai, Y., Wei, M., et al. (2020). A Trial of Lopinavir-Ritonavir in Adults Hospitalized with Severe Covid-19. *N. Engl. J. Med.* Published online March 18, 2020. <https://doi.org/10.1056/NEJMoa2001282>
8. Singh, A.K., Singh, A., Shaikh, A., Singh, R., and Misra, A. (2020). Chloroquine and hydroxychloroquine in the treatment of COVID-19 with or without diabetes: A systematic search and a narrative review with a special reference to India and other developing countries. *Diabetes, Metab. Syndrome Clin. Res. Rev.* 14, 241-246
9. Belhadi, D., Peiffer-Smadja, N., Lescure, F.-X., Yazdanpanah, Y., Mentré, F., and Laouénan, C. (2020). A brief review of antiviral drugs evaluated in registered clinical trials for COVID-19. *medRxiv*. Published online March 27, 2020. <https://doi.org/10.1101/2020.03.18.20038190>
10. Grein, J., Ohmagari, N., Shin, D., Diaz, G., Asperges, E., Castagna, A., Feldt, T., Green, G., Green, M.L., Lescure, F.-X., et al. (2020). Compassionate Use of Remdesivir for Patients with Severe Covid-19. *New Engl. J. Med.* Published online April 10, 2020. <https://doi.org/10.1056/NEJMoa2007016>
11. Gautret, P., Lagier, J.C., Parola, P., Hoang, V.T., Meddeb, L., Mailhe, M., Doudier, B., Courjon, J., Giordanengo, V., Vieira, V.E., et al. (2020). Hydroxychloroquine and azithromycin as a treatment of COVID-19: results of an open-label non-randomized clinical trial. *Int. J. Antimicrob. Agents* 105949. Published online March, 20 2020. <https://doi.org/10.1016/j.ijantimicag.2020.105949>
12. Zhou, Y., Fu, B., Zheng, X., Wang, D., Zhao, C., Sun, R., Tian, Z., Xu, X., and Wei, H. (2020). Pathogenic T cells and inflammatory monocytes incite inflammatory storm in severe COVID-19 patients. *Natl. Sci. Rev.* Published online March 13, 2020. <https://doi.org/10.1093/nsr/nwaa041>
13. Xu, Z., Shi, L., Wang, Y., Zhang, J., Huang, L., Zhang, C., Liu, S., Zhao, P., Liu, H., Zhu, L., et al. (2020). Pathological findings of COVID-19 associated with acute respiratory distress syndrome. *Lancet Respir. Med.* Published online February 18, 2020. [https://doi.org/10.1016/S2213-2600\(20\)30076-X](https://doi.org/10.1016/S2213-2600(20)30076-X)
14. Wang, W., He, J., and Wu, S. (2020). The definition and risks of Cytokine Release Syndrome-Like in 11 COVID-19-Infected Pneumonia critically ill patients: Disease Characteristics and

- Retrospective Analysis. medrxiv. Published online February 27, 2020. <https://doi.org/10.1101/2020.02.26.20026989>
15. Qin, C., Zhou, L., Hu, Z., Zhang, S., Yang, S., Tao, Y., Xie, C., Ma, K., Shang, K., Wang, W., et al. (2020). Dysregulation of immune response in patients with COVID-19 in Wuhan, China. *Clin. Infect. Dis.* Published online March 12, 2020. <https://doi.org/10.1093/cid/ciaa248>
 16. Diao, B., Wang, C., Tan, Y., Chen, X., Liu, Y., Ning, L., Chen, L., Li, M., Liu, Y., and Wang, G. (2020). Reduction and functional exhaustion of T cells in patients with coronavirus disease 2019 (COVID-19). medrxiv. Published online February 20, 2020. <https://doi.org/10.1101/2020.02.18.20024364>
 17. Zhou, Y., Fu, B., Zheng, X., Wang, D., Zhao, C., qi, Y., Sun, R., Tian, Z., Xu, X., and Wei, H. (2020). Aberrant pathogenic GM-CSF+ T cells and inflammatory CD14+ CD16+ monocytes in severe pulmonary syndrome patients of a new coronavirus. *bioRxiv*. Published online February 20, 2020. <https://doi.org/10.1101/2020.02.12.945576>
 18. Liao, M., Liu, Y., Yuan, J., Wen, Y., Xu, G., Zhao, J., Chen, L., Li, J., Wang, X., Wang, F., et al. (2020). The landscape of lung bronchoalveolar immune cells in COVID-19 revealed by single-cell RNA sequencing. medRxiv. Published online February 26, 2020. <https://doi.org/10.1101/2020.02.23.20026690>
 19. Zhou, Z., Ren, L., Zhang, L., Zhong, J., Xiao, Y., Jia, Z., Guo, L., Yang, J., Wang, C., Jiang, S., et al. (2020). Overly Exuberant Innate Immune Response to SARS-CoV-2 Infection. SSRN. Published online March 24, 2020. https://papers.ssrn.com/sol3/papers.cfm?abstract_id=3551623
 20. Wu, C., Chen, X., Cai, Y., Xia, J.a., Zhou, X., Xu, S., Huang, H., Zhang, L., Zhou, X., Du, C., et al. (2020). Risk Factors Associated With Acute Respiratory Distress Syndrome and Death in Patients With Coronavirus Disease 2019 Pneumonia in Wuhan, China. *JAMA Intern. Med.* Published online March 13, 2020. <https://doi.org/10.1001/jamainternmed.2020.0994>
 21. Bo Zhou, J.S., Yadan Wang, Xiancang Ma (2020). Utility of Ferritin, Procalcitonin, and C-reactive Protein in Severe Patients with 2019 Novel Coronavirus Disease. Research Square Published online March 19, 2020. <https://doi.org/10.21203/rs.3.rs-18079/v1>
 22. Luo, X., Zhou, W., Yan, X., Guo, T., Wang, B., Xia, H., Ye, L., Xiong, J., Jiang, Z., Liu, Y., et al. (2020). Prognostic value of C-reactive protein in patients with COVID-19. medRxiv. Published online March 23, 2020. <https://doi.org/10.1101/2020.03.21.20040360>
 23. Liu, T., Zhang, J., Yang, Y., Ma, H., Li, Z., Zhang, J., Cheng, J., Zhang, X., Zhao, Y., Xia, Z., et al. (2020). The potential role of IL-6 in monitoring severe case of coronavirus disease 2019. medRxiv. Published online March 10, 2020. <https://doi.org/10.1101/2020.03.01.20029769>
 24. Zhou, F., Yu, T., Du, R., Fan, G., Liu, Y., Liu, Z., Xiang, J., Wang, Y., Song, B., Gu, X., et al. (2020). Clinical course and risk factors for mortality of adult inpatients with COVID-19 in Wuhan, China: a retrospective cohort study. *Lancet* 395, 1054-1062
 25. Wang, Y., Jiang, W., He, Q., Wang, C., Wang, B., Zhou, P., Dong, N., and Tong, Q. (2020). Early, low-dose and short-term application of corticosteroid treatment in patients with severe COVID-19 pneumonia: single-center experience from Wuhan, China. medRxiv. Published online March 12, 2020. <https://doi.org/10.1101/2020.03.06.20032342>
 26. Lu, X., Chen, T., Wang, Y., Wang, J., Zhang, B., Li, Y., and Yan, F. (2020). Adjuvant corticosteroid therapy for critically ill patients with COVID-19. medRxiv. Published online April 11, 2020. <https://doi.org/10.1101/2020.04.07.20056390>
 27. Stockman, L.J., Bellamy, R., and Garner, P. (2006). SARS: systematic review of treatment effects. *PLoS Med.* 3, e343
 28. Arabi, Y.M., Mandourah, Y., Al-Hameed, F., Sindi, A.A., Almekhlafi, G.A., Hussein, M.A., Jose, J., Pinto, R., Al-Omari, A., Kharaba, A., et al. (2018). Corticosteroid Therapy for Critically Ill Patients with Middle East Respiratory Syndrome. *Am. J. Respir. Crit. Care Med.* 197, 757-767.
 29. Clinical management of severe acute respiratory infection when COVID-19 is suspected. Department of Communications, WHO team. Published online March 13, 2020.

- [https://www.who.int/publications-detail/clinical-management-of-severe-acute-respiratory-infection-when-novel-coronavirus-\(ncov\)-infection-is-suspected](https://www.who.int/publications-detail/clinical-management-of-severe-acute-respiratory-infection-when-novel-coronavirus-(ncov)-infection-is-suspected)
30. Katherine H. Walker, J.C.P. (2020) Brigham and Women's Hospital COVID-19 Clinical Guidelines. Brigham and Women's Hospital. Published online March 15, 2020. <https://covidprotocols.org/protocols/04-therapeutics-and-clinical-trials>
 31. Nonsteroidal anti-inflammatory drugs (NSAIDs) and serious infectious complications - Information Point. Agence Nationale de Sécurité du Médicament et des Produits de Santé. Published online March 18, 2019. <https://www.ansm.sante.fr/S-informer/Points-d-information-Points-d-information/Anti-inflammatoires-non-steroidiens-AINS-et-complications-infectieuses-graves-Point-d-Information>
 32. Momattin, H., Mohammed, K., Zumla, A., Memish, Z.A., and Al-Tawfiq, J.A. (2013). Therapeutic Options for Middle East Respiratory Syndrome Coronavirus (MERS-CoV) – possible lessons from a systematic review of SARS-CoV therapy. *Int. J. Infect. Dis.* *17*, e792-e798
 33. Liu, Q., Zhou, Y.-H., and Yang, Z.-Q. (2016). The cytokine storm of severe influenza and development of immunomodulatory therapy. *Cell. Mol. Immunol.* *13*, 3-10
 34. Shimabukuro-Vornhagen, A., Godel, P., Subklewe, M., Stemmler, H.J., Schlosser, H.A., Schlaak, M., Kochanek, M., Boll, B., and von Bergwelt-Baildon, M.S. (2018). Cytokine release syndrome. *J Immunother Cancer* *6*, 56
 35. Michot, J.-M., Albiges, L., Chaput, N., Saada, V., Pommeret, F., Griscelli, F., Balleyguier, C., Besse, B., Marabelle, A., Netzer, F., et al. (2020). Tocilizumab, an anti-IL6 receptor antibody, to treat Covid-19-related respiratory failure: a case report. *Ann. Oncol.* Published online April 2, 2020. <https://doi.org/10.1016/j.annonc.2020.03.300>
 36. Xu, X., Han, M., Li, T., Sun, W., Wang, D., Fu, B., Zhou, Y., Zheng, X., Yang, Y., and Li, X. (2020). Effective treatment of severe COVID-19 patients with Tocilizumab. *ChinaXiv*. Published online March 20, 2020. <https://doi.org/10.12074/202003.00026>
 37. Yanik, G.A., Ho, V.T., Levine, J.E., White, E.S., Braun, T., Antin, J.H., Whitfield, J., Custer, J., Jones, D., Ferrara, J.L., et al. (2008). The impact of soluble tumor necrosis factor receptor etanercept on the treatment of idiopathic pneumonia syndrome after allogeneic hematopoietic stem cell transplantation. *Blood* *112*, 3073-3081
 38. Yanik, G.A., Grupp, S.A., Pulsipher, M.A., Levine, J.E., Schultz, K.R., Wall, D.A., Langholz, B., Dvorak, C.C., Alangaden, K., Goyal, R.K., et al. (2015). TNF-receptor inhibitor therapy for the treatment of children with idiopathic pneumonia syndrome. A joint Pediatric Blood and Marrow Transplant Consortium and Children's Oncology Group Study (ASCT0521). *Biol. Blood Marrow Transplant.* *21*, 67-73
 39. Cabana, J.-A. (2020) Altasciences completes phase I study on gimsilumab for ARDS in COVID-19. Altasciences. Published online March 27, 2020. <https://www.altasciences.com/press-release/altasciences-completes-phase-i-study-gimsilumab-ards-covid-19>
 40. Davis, P. (2020) Roivant Announces Development of Anti-GM-CSF Monoclonal Antibody to Prevent and Treat Acute Respiratory Distress Syndrome (ARDS) in Patients with COVID-19. Roivant Sciences Published online March 18, 2020. <https://roivant.com/roivant-announces-development-of-anti-gm-csf-monoclonal-antibody-to-prevent-and-treat-acute-respiratory-distress-syndrome-ards-in-patients-with-covid-19/>
 41. Richardson, P., Griffin, I., Tucker, C., Smith, D., Oechsle, O., Phelan, A., and Stebbing, J. (2020). Baricitinib as potential treatment for 2019-nCoV acute respiratory disease. *Lancet* *395*, e30-e31
 42. Wu, D. and Yang, X.O. (2020). TH17 responses in cytokine storm of COVID-19: An emerging target of JAK2 inhibitor Fedratinib. *J. Microbiol. Immunol. Infect.* Published online March 11, 2020. <https://doi.org/10.1016/j.jmii.2020.03.005>
 43. Bayry, J., Negi, V.S., and Kaveri, S.V. (2011). Intravenous immunoglobulin therapy in rheumatic diseases. *Nat. Rev. Rheumatol.* *7*, 349-359

44. Perez, E.E., Orange, J.S., Bonilla, F., Chinen, J., Chinn, I.K., Dorsey, M., El-Gamal, Y., Harville, T.O., Hossny, E., and Mazer, B. (2017). Update on the use of immunoglobulin in human disease: a review of evidence. *J. Allergy Clin. Immunol.* *139*, S1-S46
45. Cao, W., Liu, X., Bai, T., Fan, H., Hong, K., Song, H., Han, Y., Lin, L., Ruan, L., and Li, T. (2020). High-dose intravenous immunoglobulin as a therapeutic option for deteriorating patients with Coronavirus Disease 2019. *Open Forum Infect. Dis.* Published online March 21, 2020. <https://doi.org/10.1093/ofid/ofaa102>
46. Shao, Z., Feng, Y., Zhong, L., Xie, Q., Lei, M., Liu, Z., Wang, C., Ji, J., Li, W., Liu, H., et al. (2020). Clinical Efficacy of Intravenous Immunoglobulin Therapy in Critical Patients with COVID-19: A multicenter retrospective cohort study. *medRxiv.* Published online April 15, 2020. <https://doi.org/10.1101/2020.04.11.20061739>
47. Xie, Y., Cao, S., Li, Q., Chen, E., Dong, H., Zhang, W., Yang, L., Fu, S., and Wang, R. (2020). Effect of regular intravenous immunoglobulin therapy on prognosis of severe pneumonia in patients with COVID-19. *J. Infect.* Published online April 10, 2020. <https://doi.org/10.1016/j.jinf.2020.03.044>
48. Schwab, I. and Nimmerjahn, F. (2013). Intravenous immunoglobulin therapy: how does IgG modulate the immune system? *Nat. Rev. Immunol.* *13*, 176-189
49. Galeotti, C., Kaveri, S.V., and Bayry, J. (2017). IVIG-mediated effector functions in autoimmune and inflammatory diseases. *Int. Immunol.* *29*, 491-498
50. Bayry, J., Lacroix-Desmazes, S., Kazatchkine, M.D., and Kaveri, S.V. (2004). Intravenous immunoglobulin for infectious diseases: back to the pre-antibiotic and passive prophylaxis era? *Trends Pharmacol. Sci.* *25*, 306-310
51. Shopsin, B., Kaveri, S.v., and Bayry, J. (2016). Tackling difficult Staphylococcus aureus infections: antibodies show the way. *Cell Host Microbe* *20*, 555-557
52. Long, Q.-X., Deng, H.-J., Chen, J., Hu, J., Liu, B.-z., Liao, P., Lin, Y., Yu, L.-h., Mo, Z., Xu, Y.-y., et al. (2020). Antibody responses to SARS-CoV-2 in COVID-19 patients: the perspective application of serological tests in clinical practice. *medRxiv.* Published online March 20, 2020. <https://doi.org/10.1101/2020.03.18.20038018>
53. Okba, N.M.A., Muller, M.A., Li, W., Wang, C., GeurtsvanKessel, C.H., Corman, V.M., Lamers, M.M., Sikkema, R.S., de Bruin, E., Chandler, F.D., et al. (2020). SARS-CoV-2 specific antibody responses in COVID-19 patients. *medRxiv.* Published online March 20, 2020. <https://doi.org/10.1101/2020.03.18.20038059>
54. Bao, L., Deng, W., Gao, H., Xiao, C., Liu, J., Xue, J., Lv, Q., Liu, J., Yu, P., Xu, Y., et al. (2020). Reinfection could not occur in SARS-CoV-2 infected rhesus macaques. *bioRxiv.* Published online March 14, 2020. <https://doi.org/10.1101/2020.03.13.990226>
55. Mair-Jenkins, J., Saavedra-Campos, M., Baillie, J.K., Cleary, P., Khaw, F.-M., Lim, W.S., Makki, S., Rooney, K.D., Group, C.P.S., Nguyen-Van-Tam, J.S., et al. (2014). The Effectiveness of Convalescent Plasma and Hyperimmune Immunoglobulin for the Treatment of Severe Acute Respiratory Infections of Viral Etiology: A Systematic Review and Exploratory Meta-analysis. *J. Infect. Dis.* *211*, 80-90
56. Ko, J.-H., Seok, H., Cho, S.Y., Ha, Y.E., Baek, J.Y., Kim, S.H., Kim, Y.-J., Park, J.K., Chung, C.R., and Kang, E.-S. (2018). Short communication Challenges of convalescent plasma infusion therapy in Middle East respiratory coronavirus infection: a single centre experience. *Antivir. Ther.* *23*, 617-622
57. Luke, T.C., Kilbane, E.M., Jackson, J.L., and Hoffman, S.L. (2006). Meta-Analysis: Convalescent Blood Products for Spanish Influenza Pneumonia: A Future H5N1 Treatment? *Ann. Intern. Med.* *145*, 599-609
58. Hung, I.F.N., To, K.K.W., Lee, C.-K., Lee, K.-L., Yan, W.-W., Chan, K., Chan, W.-M., Ngai, C.-W., Law, K.-I., Chow, F.-L., et al. (2013). Hyperimmune IV Immunoglobulin Treatment: A Multicenter Double-Blind Randomized Controlled Trial for Patients With Severe 2009 Influenza A(H1N1) Infection. *Chest* *144*, 464-473

59. Jiang, S., Hillyer, C., and Du, L. (2020). Neutralizing Antibodies against SARS-CoV-2 and Other Human Coronaviruses. *Trends Immunol.* Published online April 2, 2020. <https://doi.org/10.1016/j.it.2020.03.007>
60. Sui, J., Li, W., Murakami, A., Tamin, A., Matthews, L.J., Wong, S.K., Moore, M.J., Tallarico, A.S.C., Olurinde, M., Choe, H., et al. (2004). Potent neutralization of severe acute respiratory syndrome (SARS) coronavirus by a human mAb to S1 protein that blocks receptor association. *Proc. Natl. Acad. Sci. U. S. A.* *101*, 2536-2541
61. ter Meulen, J., van den Brink, E.N., Poon, L.L., Marissen, W.E., Leung, C.S., Cox, F., Cheung, C.Y., Bakker, A.Q., Bogaards, J.A., van Deventer, E., et al. (2006). Human monoclonal antibody combination against SARS coronavirus: synergy and coverage of escape mutants. *PLoS Med.* *3*, e237.
62. Ye, L., Chen, X., Li, R., Pan, Z., Qian, C., Yang, Y., You, R., Zhao, J., Gao, L., Li, Z., et al. (2020). Human monoclonal antibodies block the binding of SARS-CoV-2 spike protein to angiotensin converting enzyme 2 receptor. *medRxiv.* Published online April 11, 2020. <https://doi.org/10.1101/2020.04.06.20055475>.
63. Yuan, M., Wu, N.C., Zhu, X., Lee, C.-C.D., So, R.T.Y., Lv, H., Mok, C.K.P., and Wilson, I.A. (2020). A highly conserved cryptic epitope in the receptor-binding domains of SARS-CoV-2 and SARS-CoV. *Science* eabb7269. Published online April 03, 2020. <https://doi.org/10.1126/science.abb7269>
64. Wang, C., Li, W., Drabek, D., Okba, N.M.A., van Haperen, R., Osterhaus, A.D.M.E., van Kuppeveld, F.J.M., Haagmans, B.L., Grosveld, F., and Bosch, B.-J. (2020). A human monoclonal antibody blocking SARS-CoV-2 infection. *bioRxiv.* Published online March 12, 2020. <https://doi.org/10.1101/2020.03.11.987958>
65. Zhao, J., Yuan, Q., Wang, H., Liu, W., Liao, X., Su, Y., Wang, X., Yuan, J., Li, T., Li, J., et al. (2020). Antibody responses to SARS-CoV-2 in patients of novel coronavirus disease 2019. *Clin. Infect. Dis.* Published online March 28, 2020. <https://doi.org/10.1093/cid/ciaa344>
66. Guo, L., Ren, L., Yang, S., Xiao, M., Chang, D., Yang, F., Dela Cruz, C.S., Wang, Y., Wu, C., Xiao, Y., et al. (2020). Profiling Early Humoral Response to Diagnose Novel Coronavirus Disease (COVID-19). *Clin. Infect. Dis.* Published online March 21, 2020. <https://doi.org/10.1093/cid/ciaa310>
67. Huaxia (2020) China puts 245 COVID-19 patients on convalescent plasma therapy. Published online February 28, 2020. http://www.xinhuanet.com/english/2020-02/28/c_138828177.htm
68. Shen, C., Wang, Z., Zhao, F., Yang, Y., Li, J., Yuan, J., Wang, F., Li, D., Yang, M., and Xing, L. (2020). Treatment of 5 Critically Ill Patients With COVID-19 With Convalescent Plasma. *JAMA.* Published online March 27, 2020. <https://doi.org/10.1001/jama.2020.4783>
69. Duan, K., Liu, B., Li, C., Zhang, H., Yu, T., Qu, J., Zhou, M., Chen, L., Meng, S., Hu, Y., et al. (2020). Effectiveness of convalescent plasma therapy in severe COVID-19 patients. *Proc. Natl. Acad. Sci. USA* Published online April 6, 2020. <https://doi.org/10.1073/pnas.2004168117>
70. Takeda Initiates Development of a Plasma-Derived Therapy for COVID-19. Published online March 4, 2020. <https://www.takeda.com/newsroom/newsreleases/2020/takeda-initiates-development-of-a-plasma-derived-therapy-for-covid-19>
71. Grifols Announces Formal Collaboration with US Government to Produce the First Treatment Specifically Targeting COVID-19. Published online March 25, 2020. <https://www.grifols.com/en/view-news/-/news/grifols-announces-formal-collaboration-with-us-government-to-produce-the-first-treatment-specifically-targeting-covid-19>
72. Bian, H., Zheng, Z.-H., Wei, D., Zhang, Z., Kang, W.-Z., Hao, C.-Q., Dong, K., Kang, W., Xia, J.-L., Miao, J.-L., et al. (2020). Meplazumab treats COVID-19 pneumonia: an open-labelled, concurrent controlled add-on clinical trial. *medRxiv.* Published online March 24, 2020. <https://doi.org/10.1101/2020.03.21.20040691>
73. Wing, J.B., Tanaka, A., and Sakaguchi, S. (2019). Human FOXP3(+) Regulatory T Cell Heterogeneity and Function in Autoimmunity and Cancer. *Immunity* *50*, 302-316

74. Stephen-Victor, E., Bosschem, I., Haesebrouck, F., and Bayry, J. (2017). The Yin and Yang of regulatory T cells in infectious diseases and avenues to target them. *Cell. Microbiol.* *19*, e12746
75. Webster, K.E., Walters, S., Kohler, R.E., Mrkvan, T., Boyman, O., Surh, C.D., Grey, S.T., and Sprent, J. (2009). In vivo expansion of T reg cells with IL-2-mAb complexes: induction of resistance to EAE and long-term acceptance of islet allografts without immunosuppression. *J. Exp. Med.* *206*, 751-760
76. Trotta, E., Bessette, P.H., Silveria, S.L., Ely, L.K., Jude, K.M., Le, D.T., Holst, C.R., Coyle, A., Potempa, M., and Lanier, L.L. (2018). A human anti-IL-2 antibody that potentiates regulatory T cells by a structure-based mechanism. *Nat. Med.* *24*, 1005-1014
77. Abbas, A.K., Trotta, E., D, R.S., Marson, A., and Bluestone, J.A. (2018). Revisiting IL-2: Biology and therapeutic prospects. *Sci. Immunol.* *3*, eaat1482
78. Rosenblum, M.D., Gratz, I.K., Paw, J.S., and Abbas, A.K. (2012). Treating human autoimmunity: current practice and future prospects. *Sci. Transl. Med.* *4*, 125sr121
79. Slater, H. (2020). FDA Accepts IND for NK Cell Therapy CYNK-001 to Treat Patients with COVID-19. CancerNetwork home of the journal ONCOLOGY. Published online April 3, 2020. <https://www.cancernetwork.com/immuno-oncology/fda-accepts-ind-nk-cell-therapy-cynk-001-treat-patients-covid-19>
80. Million, M., Lagier, J-C., Gautret, P., Colson, P., Fournier, P-E., Amrane, S., Hocquart, M., Mailhe, M., Esteves-Vieira, V., Doudier, B., et al. (2020). Early treatment of 1061 COVID-19 patients with hydroxychloroquine and azithromycin, Marseille, France. Published online April 9, 2020. <https://www.mediterranee-infection.com/early-treatment-of-1061-covid-19-patients-with-hydroxychloroquine-and-azithromycin-marseille-france/>
81. Chen, Z., Hu, J., Zhang, Z., Jiang, S., Han, S., Yan, D., Zhuang, R., Hu, B., and Zhang, Z. (2020). Efficacy of hydroxychloroquine in patients with COVID-19: results of a randomized clinical trial. medRxiv. Published online April 10, 2020. <https://doi.org/10.1101/2020.03.22.20040758>
82. Wang, M., Cao, R., Zhang, L., Yang, X., Liu, J., Xu, M., Shi, Z., Hu, Z., Zhong, W., and Xiao, G. (2020). Remdesivir and chloroquine effectively inhibit the recently emerged novel coronavirus (2019-nCoV) in vitro. *Cell Res.* *30*, 269-271
83. Liu, J., Cao, R., Xu, M., Wang, X., Zhang, H., Hu, H., Li, Y., Hu, Z., Zhong, W., and Wang, M. (2020). Hydroxychloroquine, a less toxic derivative of chloroquine, is effective in inhibiting SARS-CoV-2 infection in vitro. *Cell Discov.* *6*, 16. <https://doi.org/10.1038/s41421-020-0156-0>
84. Mahevas, M., Tran, V.-T., Roumier, M., Chabrol, A., Paule, R., Guillaud, C., Gallien, S., Lepeule, R., Szwebel, T.-A., Lescure, X., et al. (2020). No evidence of clinical efficacy of hydroxychloroquine in patients hospitalized for COVID-19 infection with oxygen requirement: results of a study using routinely collected data to emulate a target trial. medRxiv. Published online April 14, 2020. <https://doi.org/10.1101/2020.04.10.20060699>.
85. Molina, J.M., Delaugerre, C., Le Goff, J., Mela-Lima, B., Ponscarne, D., Goldwirt, L., and de Castro, N. (2020). No evidence of rapid antiviral clearance or clinical benefit with the combination of hydroxychloroquine and azithromycin in patients with severe COVID-19 infection. *Med. Mal. Infect.* Published online March 30, 2020. <https://doi.org/10.1016/j.medmal.2020.03.006>

Table 1| Immunotherapies under evaluation for COVID-19*

Immunomodulator/company	Mechanism	Stage of development	Identifier
Anakinra/Swedish Orphan Biovitrum	IL-1 receptor antagonist	Phase 3	NCT04324021
Anakinra/Hellenic Institute for the Study of Sepsis		Phase 2	NCT04339712
Anakinra/Assistance Publique - Hôpitaux de Paris		Phase 2	NCT04341584
Anakinra/University Hospital, Ghent		Phase 3	NCT04330638
Baricitinib/Lisa Barrett	TNF inhibitor	Phase 2	NCT04321993
Baricitinib/Hospital of Prato		Phase 3	NCT04320277
TJ003234 (Anti-GM-CSF Monoclonal Antibody)/ I-Mab Biopharma Co. Ltd	GM-CSF neutralization	Phase 1b/2	NCT04341116
Bevacizumab/Qilu Hospital of Shandong University	Anti-vascular endothelial growth factor	Phase 3	NCT04275414, NCT04305106
CD24Fc/OncoImmune, Inc.	Regulation of innate response to inflammatory stimuli and tissue injury by binding to danger-associated molecular patterns and Siglec G/10	Phase 3	NCT04317040
Eculizumab (Soliris)/Hudson Medical	Inhibition of complement activation by binding to terminal complement protein C5	NA	NCT04288713
Emapalumab/Swedish Orphan Biovitrum	Blockade of IFN- γ mediated signaling pathways	Phase 2	NCT04324021
Fingolimod (FTY720)/First Affiliated Hospital of Fujian Medical University	Sphingosine-1-phosphate receptor regulator	Phase 2	NCT04280588
Interferon β 1 α /Institut National de la Santé et de la Recherche Médicale, France	Inhibition of viral replication by inducing type I IFN-stimulated genes	Phase 3	NCT04315948
Recombinant human interferon α 1 β /Shanghai Jiao Tong University School of Medicine		Phase 3	NCT04320238
Recombinant human interferon α 2 β /Tongji Hospital		Early Phase 1	NCT04293887
Meplazumab/Tang-Du Hospital	Prevention of CD147-mediated SARS-CoV-2 viral entry and T cell chemotaxis	Phase 2	NCT04275245
Siltuximab/University Hospital, Ghent	IL-6 neutralization	Phase 3	NCT04330638
Sarilumab/Regeneron Pharmaceuticals		Phase 2	NCT04315298
Sarilumab/Assistance Publique - Hôpitaux de Paris		Phase 2/3	NCT04324073
Sarilumab/Sanofi		Phase 2/3	NCT04327388
Sarilumab/Nova Scotia Health Authority		Phase 2	NCT04321993

Tocilizumab/National Cancer Institute, Naples	IL-6 receptor antagonist	Phase 2	NCT04317092
Tocilizumab/Hoffmann-La Roche		Phase 3	NCT04320615
Tocilizumab/University of L'Aquila		NA	NCT04332913
Tocilizumab/University Hospital, Ghent		Phase 3	NCT04330638
RoActemra or Kevzara/Marius Henriksen		Phase 2	NCT04322773
Intravenous immunoglobulin (IVIG)/Peking Union Medical College Hospital	Suppression of innate and adaptive inflammatory responses and concomitant enhancement of anti-inflammatory processes.	Phase 2/3	NCT04261426
Immunoglobulin from cured patients/Wuhan Union Hospital, China	Virus neutralization	NA	NCT04264858
Anti-SARS-CoV-2 inactivated convalescent plasma/Shanghai Public Health Clinical Center		NA	NCT04292340
Human coronavirus immune plasma /Sidney Kimmel Comprehensive Cancer Center at Johns Hopkins		Phase 2	NCT04323800
Convalescent plasma/Mayo Clinic		Phase 2	NCT04325672
Hyperimmune plasma/Foundation IRCCS San Matteo Hospital		NA	NCT04321421
Convalescent plasma/Baylor Research Institute		Phase 1	NCT04333251
Convalescent plasma/Mazandaran University of Medical Sciences, Iran		NA	NCT04327349
Convalescent plasma/Universidad del Rosario, Colombia		Phase 2/3	NCT04332835
Convalescent plasma/Universidad del Rosario, Colombi		Phase 2	NCT04332380
NK cell treatment/Xinxiang medical university		Anti-viral action	Phase 1
NKG2D-ACE2 CAR-NK Cells/Chongqing Public Health Medical Center	GM-CSF neutralization, CRS prevention, inhibition of viral infection of alveolar epithelial cells and clearance of virus-infected cells	Phase 1/2	NCT04324996

NA, Not Applicable.

*The drugs tabulated here are included in clinical trials (<https://clinicaltrials.gov/>) either alone or in combination with other drugs/supplements

Figure Legends

Figure 1. Cytokine storm in COVID-19 infection. Lungs are the primary organs affected by SARS-CoV-2. A dysregulated cytokine response (i.e., cytokine storm) due to an influx of activated immune cells following SARS-CoV-2 infection results in pulmonary edema leading to damaged alveoli and formation of scarred interstitium culminating in a reduced gas exchange process. The figure was created with the support of BioRender.com under the paid subscription.

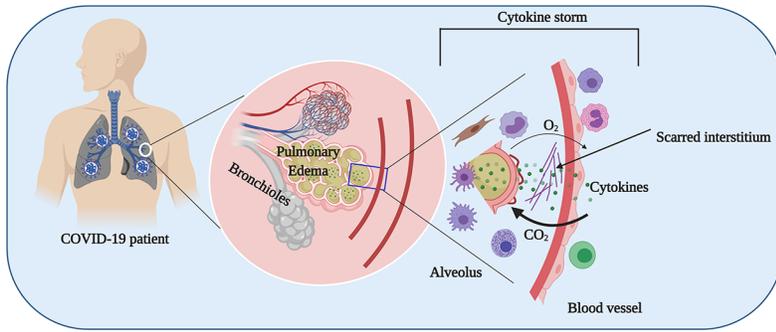
Figure 2. Immunotherapeutic strategies for severely ill COVID-19 patients. Diversified immunotherapeutic approaches against severe COVID-19 cases are illustrated. The figure was created with the support of BioRender.com under the paid subscription.

In Brief

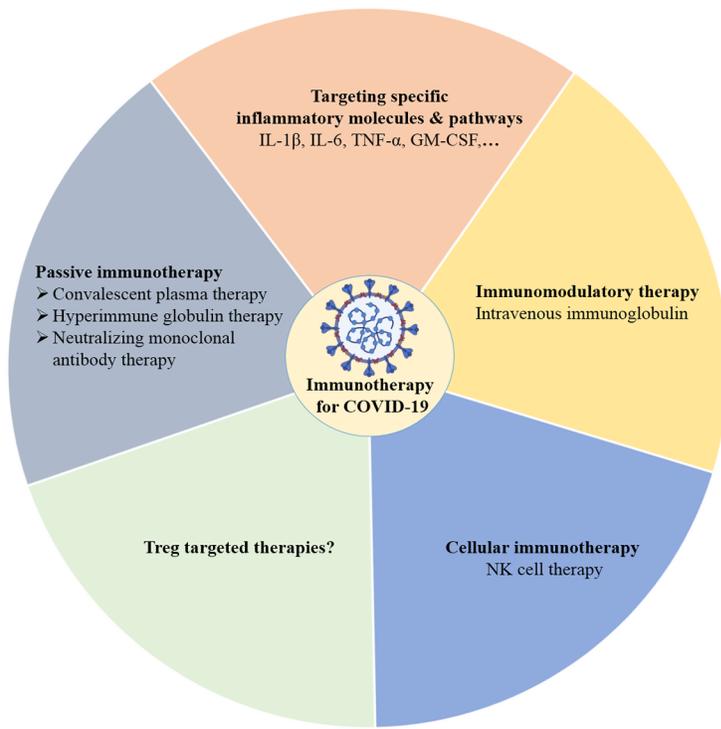
An overreactive immune response, cytokine storm and development of acute respiratory distress syndrome characterize severe cases of COVID-19. Bonam et al. present a Perspective on diverse immunotherapeutic approaches based on the neutralization of inflammatory cytokines, immunomodulation and passive viral neutralization for treating severely ill COVID-19 patients.

Highlights

- Inflammation is the hallmark of SARS-CoV-2 infection
- Cytokine storm characterizes severely ill COVID-19 cases
- Immunotherapies represent promising options for the management of severe COVID-19
- Immunotherapies either neutralize cytokines, SARS-CoV-2 or exert immunomodulation



Journal Pre-proof



Journal Pre-proof