Single-nucleus lung transcriptomics and inflammatory responses in lethal COVID-19 reveal potential drugs in advanced-stage clinical trials

Andrés López-Cortés (aalc84@gmail.com)
Facultad de Ciencias de la Salud Eugenio Espejo, Universidad UTE, Quito, Ecuador
https://orcid.org/0000-0003-1503-1929

Santiago Guerrero
Latin American Network for the Implementation and Validation of Clinical Pharmacogenomics Guidelines (RELIVAF-CYTED), Madrid, Spain https://orcid.org/0000-0003-3473-7214

Esteban Ortiz-Prado
One Health Research Group, Faculty of Medicine, Universidad de Las Américas, Quito, Ecuador
https://orcid.org/0000-0002-1895-7498

Verónica Yumiceba
Institut für Humangenetik Lübeck, Universität zu Lübeck, Lübeck, Germany https://orcid.org/0000-0001-6998-7913

Antonella Vera-Gupi
Integrated Research and Treatment Center, Center for Sepsis Control and Care (CSCC), Jena University Hospital, Jena, Germany

Ángela León Cáceres
Heidelberg Institute of Global Health, Faculty of Medicine, University of Heidelberg, Germany
https://orcid.org/0000-0002-4517-9409

Katherine Simbaña-Rivera
One Health Research Group, Faculty of Medicine, Universidad de Las Américas, Quito, Ecuador
https://orcid.org/0000-0002-8130-5361

Ana María Gómez-Jaramillo
Faculty of Medicine, Pontifical Catholic University of Ecuador, Quito, Ecuador
https://orcid.org/0000-0003-2180-7316

Patricia Guevara-Ramírez
Latin American Network for the Implementation and Validation of Clinical Pharmacogenomics Guidelines (RELIVAF-CYTED), Madrid, Spain https://orcid.org/0000-0002-4829-3653

Jennyfer M. García-Cárdenas
Facultad de Ciencias de la Salud Eugenio Espejo, Universidad UTE, Quito, Ecuador
https://orcid.org/0000-0001-9035-7668

Alejandro Cabrera-Andrade
Grupo de Bio-Quimioinformática, Universidad de Las Américas, Quito, Ecuador  https://orcid.org/0000-0001-9702-6618

**Gabriela Echeverría-Garcés**  
Latin American Network for the Implementation and Validation of Clinical Pharmacogenomics Guidelines (RELIVAF-CYTED), Madrid, Spain  https://orcid.org/0000-0002-1941-7864

**Lourdes Puig San Andrés**  
BIOscience Research Group, Quito, Ecuador  https://orcid.org/0000-0002-1388-3151

**Doménica Cevallos-Robalino**  
BIOscience Research Group, Quito, Ecuador  https://orcid.org/0000-0002-7820-4359

**Jhommara Bautista**  
BIOscience Research Group, Quito, Ecuador  https://orcid.org/0000-0002-9717-8096

**Isaac Armendáriz-Castillo**  
Facultade de Ciencias, Universidade da Coruña, A Coruña, Spain  https://orcid.org/0000-0003-2636-2707

**Andy Pérez-Villa**  
Latin American Network for the Implementation and Validation of Clinical Pharmacogenomics Guidelines (RELIVAF-CYTED), Madrid, Spain  https://orcid.org/0000-0001-6758-4299

**Andrea Abad-Sojos**  
BIOscience Research Group, Quito, Ecuador  https://orcid.org/0000-0003-2919-7487

**María José Ramos-Medina**  
BIOscience Research Group, Quito, Ecuador

**Ariana León-Sosa**  
BIOscience Research Group, Quito, Ecuador

**Estefanía Abarca**  
BIOscience Research Group, Quito, Ecuador

**Álvaro A. Pérez-Meza**  
Biotechnology Engineering Career, Faculty of Life Sciences, Universidad Regional Amazónica Ikiam, Tena, Ecuador

**Karol Nieto-Jaramillo**  
BIOscience Research Group, Quito, Ecuador

**Andrea V. Jácome**  
Faculty of Medicine, Universidad de Las Américas, Quito, Ecuador

**Andrea Morillo**  
BIOscience Research Group, Quito, Ecuador

**Fernanda Arias-Erazo**  
BIOscience Research Group, Quito, Ecuador

**Luis Fuenmayor-González**  
BIOscience Research Group, Quito, Ecuador  https://orcid.org/0000-0001-6141-7692

**Nikolaos C Kyriakidis**
Research Article

Keywords: lethal COVID-19, inflammatory proteins, drugs, clinical trials

DOI: https://doi.org/10.21203/rs.3.rs-808746/v1

License: ☑️ ⭐️ This work is licensed under a Creative Commons Attribution 4.0 International License. Read Full License
Abstract

There is pressing urgency to identify drugs that allow treating COVID-19 patients effectively. Respiratory failure is the leading cause of death in patients with severe COVID-19, and the host inflammatory response at the lungs remains poorly understood. Therefore, we retrieved data from postmortem lungs from COVID-19 patients and performed in-depth in silico analyses of single-nucleus RNA sequencing data, inflammatory protein interactome network, functional enrichment, and shortest pathways to cancer hallmark phenotypes to reveal potential therapeutic targets and drugs in advanced-stage COVID-19 clinical trials. Herein, we analyzed transcriptomics data of 719 inflammatory response genes across 19 cell types (116,313 nuclei) from lung autopsies. The functional enrichment analysis of the 233 significantly expressed genes showed that the most relevant biological annotations were: inflammatory response, innate immune response, cytokine production, interferon production, macrophage activation, thymic stromal lymphopoietin, blood coagulation, IL-1 and megakaryocytes in obesity, NLRP3 inflammasome complex, and the TLR, JAK-STAT, NF-κB, TNF, oncostatin M, AGE-RAGE signaling pathways. Subsequently, we identified 34 essential inflammatory proteins with both high-confidence protein interactions and shortest pathways to inflammation, cell death, glycolysis, and angiogenesis. Lastly, we propose five small molecules involved in advanced-stage COVID-19 clinical trials: baricitinib, pacritinib, and ruxolitinib are tyrosine-protein kinase JAK2 inhibitors, losmapimod is a MAP kinase p38 alpha inhibitor, and eritoran is a TLR4/MD-2 antagonist. After being thoroughly analyzed in COVID-19 clinical trials, these drugs can be considered for treating severe COVID-19 patients.

Introduction

The first zoonotic transmission of the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) occurred in China in late December 2019, and it is the etiological agent of the coronavirus disease 2019 (COVID-19). Since the World Health Organization (WHO) declared the outbreak of COVID-19 as a pandemic on March 11, 2020, the SARS-CoV-2 infection has led to more than 200 million cases and more than 4 million deaths globally.

SARS-CoV-2 is the seventh CoV known to infect humans, along with HKU1, OC43, NL63, 229E, SARS-CoV, and Middle East respiratory syndrome (MERS). The novel coronavirus is a single-stranded positive-sense RNA virus of about 30 kb in length, which encompasses a 5' terminal cap, 14 open reading frames (ORFs) encoding for 29 proteins, and a 3' poly A tail. ORF1a and ORF1ab encode 16 non-structural proteins (nsps) involved in antiviral response (nsp1), viral replication (the nsp3-nsp4-nsp6 complex), the protease 3Cpro (nsp5), the RNA polymerase (the nsp7-nsp8 complex), the single strand RNA binding protein (nsp9), the methyltransferase activity (nsp10 and nsp16), the RNA-dependent RNA polymerase (nsp12), the helicase/triphosphatase (nsp13), the 3'-5' exonuclease (nsp14), the uridine-specific endoribonuclease (nsp15), and the RNA-cap (nsp16). The remaining genes encode structural proteins: the spike (S) glycoprotein, the nucleocapsid (N) protein, the membrane (M) glycoprotein, the envelope (E) protein, and several accessory proteins.
At the molecular level, amino-acid changes that result in reduced fitness are generally removed by negative selection, whereas changes that increase virus fitness are maintained by positive selection. The most significant mutation observed in SARS-CoV-2 is probably the D614G substitution in the S1 subunit of the S protein. This mutation confers a 20% increase in infectivity and is associated with a higher ACE2-binding affinity. Additionally, SARS-CoV-2 fitness is also enhanced in presence of the E484K mutation, which increases resistance to antibodies.

This transmissibility advantage was further increased to approximately 50% by the emergence of the B.1.1.7 variant. Subsequently more variants have emerged, some of them capable of escaping monoclonal antibodies, partially eluding the polyclonal immune responses induced by previous infection or even allowing re-infections. It should be noted that recent improvements in immune escape are linked to mutations that alter the N-terminal domain (NTD) rather than the receptor-binding domain (RBD) of the S protein, where early and functionally important alterations prevailed. However, improved transmissibility, rather than immunoevasion or increased lethality, are considered as the main route for the virus to become fitter and more viable.

The variants that are being carefully monitored include: A) Variants of concern (VOCs): characterized by increased transmissibility, cause a more severe manifestation of the disease or significant reduction in neutralizing antibodies generated during a previous infection or after vaccination, reduced effectiveness of treatments, or diagnostic detection failures. This group includes the B.1.1.7 (Alpha), B.1.351 (Beta), B.1.617.2 (Delta), and P1 (Gamma) variants. B) Variants of interest (VOIs): present reduced neutralization by antibodies induced by previous infection or vaccines, reduced efficacy of treatments, potential diagnostic escape and expected increase in transmissibility or severity of COVID-19. Among them, we find the B.1.427 / 429 (Epsilon), B.1.525 (Eta), B.1.526 (Iota), B.1.617.1 (Kappa), C.37 (Lambda), B.1.617.3, P.2 (Zeta), and B.1.621 variants. C) Variants under monitoring: could have properties similar to those of VOCs, however precise information is still lacking. D) High consequence variants: those that have significantly reduced the efficacy of vaccines in relation to previously circulating variants, and additionally cause failures in their diagnosis; however, there are not SARS-CoV-2 variants that rise to the level of high consequence yet. It is expected that more variants will emerge over time that will need to be closely monitored, since they are a potential threat to public health. Nevertheless, this will not happen indefinitely because the virus will reach its maximum transmission point, therefore, new variants will not acquire more advantages in terms of infectivity. Thereafter, virus infectivity will stabilize and experience occasional and minimal variations.

SARS-CoV-2, enriched by the previously mentioned genomic variants, has the ability to infect human body cells—especially in the lung microenvironment—through the angiotensin-converting enzyme 2 (ACE2) protein receptor. Lung homeostasis necessitates a fine balance between tolerance mechanisms against non-pathogenic agents, pro-inflammatory immune system activation to fight off respiratory tract infections and anti-inflammatory and pro-fibrotic processes that will minimize the immune-mediated tissular lesion and promote tissue remodeling and repair. These complex mechanisms are mediated by a
variety of tissue-resident and recruited cell types. The pulmonary alveolar epithelium is mainly composed of alveolar type I cells (AT1), which are essential for the gas-exchange function of the lungs, and alveolar type II cells (AT2), which are best known for their functions in synthesizing and secreting pulmonary surfactant factors. Airway epithelial cells are central players in mucociliary clearance of the lungs. They produce a variety of antimicrobial substances, cytokines, and growth factors that mediate leukocyte recruitment, modulation of innate and adaptive immunity, and tissue repair. They also constitute the first cells that contact invading pathogens and are responsible for early pathogen recognition and induction of the antiviral state through pro-inflammatory cytokines and type I interferon secretion. Pulmonary endothelial cells are localized in the interface between the pulmonary tissue and the bloodstream. Their strategic location is also reflected in their pleiotropic functions that range from gas interchange to regulating vascular tone and facilitating immune cell recruitment and diapedesis upon receiving pro-inflammatory stimuli. Mast cells are innate immune cells, involved in immune defense and surveillance. They are filled with secretory granules, which upon activation, release bioactive mediators to fight pathogens or induce allergic reactions. Macrophages are key sentinel cells residing in peripheral tissues that detect pathogen invasion or tissue damage and initiate acute inflammatory processes triggering recruitment and activation of innate and – in second step- adaptive immune responses. Macrophages are also key inducers of the respiratory burst, professional antigen-presenting cells and tissue repair and remodeling mediators. Dendritic cells form a heterogeneous population of the immune system that have a wide array of immune functions. Conventional dendritic cells bridge the innate and adaptive immune responses as they are constantly sampling antigens from the airways and/or the infected lung tissue, migrate to T-cell areas of secondary lymphoid organs and present it to T lymphocytes thereby activating them. Monocytes, subsets of leukocytes mostly originated from myeloid progenitors in the bone marrow, are able to differentiate into macrophages or dendritic cells in peripheral tissues. They seed tissues with enough macrophages to replace their loss through infection and tissue damage and can adopt specific macrophage or dendritic cell phenotypes depending on the cytokine milieu they encounter upon arrival to the inflamed tissue. Natural killer (NK) cells are lymphocytes of the innate immune system, that play a main role in anti-viral and anti-tumor responses. They can identify and kill infected or stressed cells by releasing perforin and granzymes or by death receptor signaling (FasL/Fas interactions and the subsequent induction of apoptosis). NK cells can also release IFNγ upon activation, thereby contributing to naive T helper cell activation and differentiation and classical activation of macrophages. The CD4+ T helper cell population orchestrates the innate and adaptive immune responses in acute and chronic viral infections by secreting a panel of immunomodulatory cytokines. These cells also play a key role for the establishment of long-term cellular and humoral antigen-specific immunity, which is the basis of long-term protection induced by a plethora of viral infections and vaccines. The cytotoxic CD8+ T cells play a pivotal role in controlling infections caused by intracellular pathogens. These cells can be considered the adaptive immunity counterparts of NK cells, but unlike their innate immunity counterparts CD8+ T cells are activated by specific pathogen or tumor-derived antigen presented on class I major histocompatibility complex molecules (MHC I). The three major mechanisms of action of these cells are quite similar to NK cell functions: a) direct killing of...
infected or tumor cells by release of perforin and granzymes, b) indirect destruction of cells via death receptor signaling (Fas/FasL interactions), and c) secretion of cytokines that can direct and potentiate immune responses of nearby cells. Treg cells are potent immunosuppressive cells that play a vital role in maintaining immune homeostasis and in the prevention of autoimmune responses by suppressing the activation of conventional T-cells. B cells have a key role in the humoral adaptive immune response and –once activated- are responsible for the production of antigen-specific immunoglobulins. Plasma blasts are terminally differentiated populations of effector B cells cells, which produce antibodies, providing immunity during initial exposure to a pathogen and mediating the protective effects of vaccination. Fibroblasts are key cells in the wound repair process and tissue scarring. They participate in the immune response by producing cytokines and chemokines that initiate the recruitment and retention of bone marrow-derived immune effector cells. Smooth muscle cells provide the main support for the vessel wall structure and regulate vascular tone to maintain intravascular pressure and tissue perfusion. Lastly, neuronal cells release neurotransmitters and neuropeptides that allow fast communication with immune cells, maintaining homeostasis and fighting infections. Neuroimmune interactions are also implicated in several chronic inflammatory conditions.

Previous studies have reported profound SARS-CoV-2-induced transcriptional and immunological changes in animal models, as well as in bronchoalveolar lavage fluid (BALV), nasopharyngeal, and human blood samples. However, the respiratory failure is the leading cause of death in patients with severe COVID-19 disease and the host inflammatory response at the lung tissue level remains poorly understood. To shed light on this physiological response, we retrieved data from COVID-19 autopsies and performed in-depth in silico analyses of single-nucleus RNA sequencing (snRNA-seq) data, inflammatory protein-protein interactome (iPPI) network, miRNome enrichment, gene ontology (GO), and the shortest paths to cancer hallmark phenotypes to reveal potential therapeutic targets and drugs in advanced stage COVID-19 clinical trials.

**Methods**

**Protein sets.** We have retrieved a total of 719 inflammatory response proteins from the David Bioinformatics Resource (https://david.ncifcrf.gov/) using the gene ontology (GO) term: 0006954 inflammatory response. We have also retrieved the 332 human proteins physically interacting with 26 of the 29 SARS-CoV-2 proteins proposed by Gordon et al.

**Single-nucleus RNA sequencing data.** Melms et al have previously published the molecular single-cell lung atlas of lethal COVID-19 through the snRNA-seq technology needed to profile hard-to-dissociate tissues. Motivated by this study, we performed an in-depth in silico analysis comparing the transcriptional data of 719 genes involved in the inflammatory response between 9608 alveolar type I cells, 11341 alveolar type II cells, 7332 airway epithelial cells, 1845 B cells, 7586 CD4 + T cells, 3561 CD8 + T cells, 2814 cycling NK / T cells, 1083 dendritic cells, 5386 endothelial cells, 21472 fibroblast cells, 25960 macrophages, 1438 mast cells, 3464 monocytes, 2141 NK cells, 2017 neuronal cells, 5391 plasma...
cells, 1437 smooth muscle cells, 649 Treg cells, and 1788 other epithelial cells. The snRNA-seq database was taken from the ‘COVID-19 Studies’ section of the Single Cell Portal (https://singlecell.broadinstitute.org/single_cell/covid19), and the transcriptomics data of 116,313 nuclei was taken from ‘Columbia University / NYP COVID-19 Lung Atlas’ study (https://singlecell.broadinstitute.org/single_cell/study/SCP1219/columbia-university-nyp-covid-19-lung-atlas?cluster=UMAP&spatialGroups=--&annotation=cell_type_intermediate--group--study&subsample=all#study-summary) 51.

The criteria of the analysis of the lung transcriptomics data was the following: ‘uniform manifold approximation and projection (UMAP)’ as load cluster, ‘cell type intermediate’ as selected annotation, and ‘all cells’ as subsampling threshold. Additionally, we adjusted the mRNA expression taking into account the Z-scores, that is, overexpressed mRNAs with Z-scores ≥ 2 and underexpressed mRNAs with Z-scores ≤ -2. Regarding visualization of transcriptomics data, we designed dot plots to visualize the percentage of cells expressing a certain gene, box plots to compare the mean Z-score across cell types, and scatter plots of 2D UMAPs to visualize significantly expressed multiple genes per subpopulation cell, and biological annotations across cell types.

**Functional enrichment analysis.** The functional enrichment analysis gives curated signatures of gene sets generated from omics-scale experiments 11,53,54. We performed the enrichment analysis to validate the correlation between significantly expressed genes and biological annotations related to lethal COVID-19. The enrichment was calculated using g:Profiler version e101_eg48_p14_baf17f0 (https://biit.cs.ut.ee/gprofiler/gost) to obtain significant annotations (Benjamini-Hochberg FDR q-value < 0.001) related to gene ontology: biological processes, the Kyoto Encyclopedia of Genes and Genomes (KEGG) signaling pathways, Reactome signaling pathways, and Wikipathways 53–57. Lastly, the expression of genes involved in significant annotations was visualized in scatter plots, and the significant terms related to lethal COVID-19 pathology were manually curated.

**miRNome enrichment analysis.** The Gene Set Enrichment Analysis (GSEA) (https://www.gsea-msigdb.org/gsea/index.jsp) is a powerful analytical method for interpreting gene expression data that share common biological functions or regulations 58. Therefore, we performed a miRNome enrichment analysis using the ‘microRNA targets’ option to compute overlaps between miRNAs and significantly expressed mRNAs in > 50% of lung cells from lethal COVID-19 patients. Lastly, we proposed the most significant miRNAs with a false discovery rate (FDR) q-value < 0.01.

**Inflammatory protein-protein interactome network.** The iPPI network with zero node addition and a highest confidence cutoff of 0.9 was created between the human proteins physically associated with SARS-CoV-2 and human proteins involved in the pulmonary inflammatory response. This network was generated using the human proteome from the Cytoscape StringAPP 59,60, which imports protein interactions from the STRING database 60. The degree centrality represents the number of edges the node has in a network 11,61,62, and it was calculated using the CytoNCA app 63. The network elements were organized through the organic layout producing a clear representation of complex networks, and the iPPI
network was visualized through the Cytoscape software v.3.7.1. Finally, we ranked the inflammatory response proteins with the highest protein-protein interactions to the human-SARS-CoV-2 proteins.

**Shortest paths from inflammatory response proteins to cancer hallmark phenotypes.** CancerGenNet ([https://signor.uniroma2.it/CancerGeneNet/](https://signor.uniroma2.it/CancerGeneNet/)) is a resource that links frequently altered proteins to cancer hallmark phenotypes. This bioinformatic tool, curated by SIGNOR, is based on experimental information that allows to infer likely paths of causal interactions linking proteins to oncogenic phenotypes. The shortest distance scores or paths from proteins to cancer phenotypes were programmatically implemented using the shortest path function of *igraph* R package. Hence, we calculated the shortest distance scores of positive regulation from the inflammatory response proteins with the highest confidence interactions to the human-SARS-CoV-2 proteins to the inflammation, cell death, angiogenesis, and glycolysis hallmark phenotypes.

**Drugs involved in current COVID-19 clinical trials.** The Open Targets Platform version 21.06 ([https://www.targetvalidation.org](https://www.targetvalidation.org)) is comprehensive and robust data integration for access to and visualization of potential drug targets associated with several diseases including COVID-19. This platform has developed the COVID-19 Target Prioritization Tool ([https://covid19.opentargets.org/](https://covid19.opentargets.org/)) that integrates molecular data from the ChEMBL database to provide an evidence-based framework to support decision-making on potential drug targets for COVID-19. Lastly, this platform shows all drugs in clinical trials associated with target proteins, detailing its modality, mechanism of action, phase, status, type of drug, target class, and clinical trial number.

**Results**

**Single-nucleus RNA sequencing data.** Single-nucleus biology is a powerful approach of omics medicine, needed to profile hard-to-dissociate tissues, that provides unprecedented resolution to the cellular underpinnings of biological processes in order to find druggable targets for complex diseases. Here, we identified 233 inflammatory response genes with significant expression in 116,313 nuclei belonging to 19 different lung cell types. Genes with the highest mean Z-score (3.26) and the most significant p-value (0.001) were identified in neural cells, followed by B cells (3.24; 0.001), mast cells (3.14; 0.002), fibroblast cells (3.0; 0.003), alveolar type II cells (2.96; 0.003), cycling NK / T cells (2.94; 0.003), endothelial cells (2.89; 0.004), macrophages (2.88; 0.004), airway epithelial cells (2.76; 0.006), alveolar type I cells (2.74; 0.006), NK cells (2.73; 0.006), dendritic cells (2.70; 0.007), smooth cells (2.68; 0.007), Treg cells (2.67; 0.008), plasma cells (2.62; 0.009), monocytes (2.47; 0.014), other epithelial cells (2.41; 0.016), CD4 + T cells (2.39; 0.017), and CD8 + T cells (2.24; 0.025) (Fig. 1).

Figure 2 shows scatter plots of significant mean log normalized gene expression and dot plots of genes with the highest percentage of cells expressing per lung cell type. *MECOM* has the highest percentage of cells expressing in alveolar type I cells, *LRRK2* in alveolar type II cells, *ELF3* in airway epithelial cells, *PXK* in B cells, *CAMK4* in CD4 + T cells, *AOAH* in CD8 + T cells, *HMGB1* in cycling NK / T cells, *CIITA* in dendritic cells, *RBPJ* in macrophages, *KIT* in mast cells, *SLC11A1* in monocytes cells, *APP* in neuronal cells, *AOAH*
in NK cells, \textit{CALCRL} in endothelial cells, \textit{RORA} in fibroblasts, \textit{ASH1L} in plasma cells, \textit{FN1} in smooth muscle cells, and \textit{SGMS1} in Treg cells. Lastly, the 26 inflammatory response genes significantly expressed in more that 50% of lung cells were \textit{ABR}, \textit{ACER3}, \textit{AOAH}, \textit{APP}, \textit{ASH1L}, \textit{ATM}, \textit{CALCRL}, \textit{CAMK1D}, \textit{CAMK4}, \textit{CD163}, \textit{CIITA}, \textit{EGFR}, \textit{FN1}, \textit{HDAC9}, \textit{IL18R1}, \textit{IL1R1}, \textit{KIT}, \textit{LRRK2}, \textit{LYN}, \textit{MECOM}, \textit{PRKCA}, \textit{PRKCZ}, \textit{RBPJ}, \textit{RORA}, \textit{SLC11A1}, and \textit{SLIT2} (Supplementary Table 1).

**Functional enrichment analysis.** The functional enrichment analysis was performed using g:Profiler to obtain significant GO: biological processes, KEGG signaling pathways, Reactome signaling pathways, and Wikipathways related to lethal COVID-19 (Benjamini-Hochberg FDR q < 0.001)\textsuperscript{11,53,54}. Figure 3 shows scatter plots of significantly expressed genes (n = 233) in lung cells of lethal COVID-19 autopsies. After a manual curation of biological annotations, the most significant GO terms were inflammatory response (5.9 x 10\textsuperscript{−241}), cytokine production (9.5 x 10\textsuperscript{−62}), innate immune response (1.0 x 10\textsuperscript{−30}), macrophage activation (1.1 x 10\textsuperscript{−29}), toll-like receptor signaling pathway (3.8 x 10\textsuperscript{−15}), type I and II interferon production (1.8 x 10\textsuperscript{−13}), the Janus Kinase (JAK) / Signal Transducers and Activators of Transcription (STAT) signaling pathway (9.0 x 10\textsuperscript{−8}), NF-κB signaling pathway (2.0 x 10\textsuperscript{−6}), thymic stromal lymphopoietin (TSLP) (4.5 x 10\textsuperscript{−6}), TNF signaling pathway (4.9 x 10\textsuperscript{−6}), blood coagulation (5.6 x 10\textsuperscript{−6}), oncostatin M signaling pathway (5.9 x 10\textsuperscript{−6}), AGE-RAGE signaling pathway (5.9 x 10\textsuperscript{−6}), IL-1 and megakaryocytes in obesity (6.8 x 10\textsuperscript{−6}), and NLRP3 inflammasome complex (2.5 x 10\textsuperscript{−4}) (Supplementary Table 2).

**miRNome enrichment analysis.** After identifying the significantly expressed genes in lung cells of lethal COVID-19 autopsies, we performed the GSEA analysis to compute overlaps between miRNAs and mRNAs\textsuperscript{58}. Figure 4 shows a circos plot of the 18 most significant miRNAs (FDR q-value < 0.01) overlapped with the 19 significantly expressed genes in > 50% of lung cells. The most significant miRNAs were MIR6867_5P (q = 0.002), MIR2662 (q = 0.002), MIR32_3P (q = 0.002), MIR548A0_5P-MIR548AX (q = 0.003), MIR570_3P (q = 0.004), MIR338_5P (q = 0.005), MIR144_3P (q = 0.005), MIR4711_3P (q = 0.005), MIR628_5P (q = 0.005), MIR548AJ_3P-MIR548X_3P (q = 0.005), MIR1290 (q = 0.006), MIR4496 (q = 0.007), MIR12136 (q = 0.007), MIR9718 (q = 0.007), MIR875_3P (q = 0.008), MIR4698 (q = 0.008), MIR3941 (q = 0.009), and MIR4789_3P (q = 0.009).

**Inflammatory protein-protein interactome network.** We generated the iPPI network encompassing 265 nodes and 2052 edges (Fig. 5). Of them, 159 pulmonary inflammatory response proteins had a mean of degree centrality of 8 and 108 human-SARS-CoV-2 proteins had a mean of degree centrality of 7.2. The top ten inflammatory response proteins with the highest degree centrality were APP (38), NFKB1 (36), STAT3 (34), C3 (31), ITGAM (29), FN1 (26), PTAFR (24), JAK2 (22), EGFR (20), and LYN (20). The top ten human-SARS-CoV-2 proteins with the highest degree centrality were GNB1 (29), GNG5 (25), RHOA (23), ITGB1 (22), STOM (20), RAB14 (20), PRKAR2B (17), RAB8A (17), PRKACA (17), and ANO6 (16). Additionally, 111 pulmonary inflammatory response proteins had the highest confidence interactions (cutoff = 0.9) with human-SARS-CoV-2 proteins, being the top ten: C3 (11 interactions), FN1 (11), NFKB1
Shortest pathways from inflammatory response proteins to cancer hallmark phenotypes. We analyzed the 111 pulmonary inflammatory response proteins with the highest confidence interactions (cutoff = 0.9) to human-SARS-CoV-2 proteins in order to find the shortest pathways toward inflammation, cell death, angiogenesis, and glycolysis according to Iannuccelli et al. Figure 6A shows box plots encompassing proteins with the shortest distance scores to cancer hallmark phenotypes related to COVID-19 pathology. Cell death was the phenotype with the shortest mean of distance score (2.82), followed by inflammation (3.06), glycolysis (3.12), and angiogenesis (3.79). Figure 6B shows a Venn diagram integrating inflammatory proteins with shortest pathways to biological phenotypes related to COVID-19. We found 34 essential inflammatory response proteins with shortest pathways simultaneously to inflammation, glycolysis, cell death, and angiogenesis (Supplementary Table 4). Figure 6C details the ranking of inflammatory response proteins with positive response and shortest distance to cell death, inflammation, glycolysis, and angiogenesis. The top ten essential proteins with shortest pathways of positive regulation to cell death were ATM (1.20), NFKBIA (1.42), TNFRSF1B (1.64), APP (1.73), MAPK14 (1.73), PRKCZ (1.93), TLR4 (1.97), JAK2 (2.26), TGFB1 (2.35), and MECOM (2.36). The top ten essential proteins with shortest pathways of positive regulation to inflammation were PTGS2 (0.53), PRKCZ (1.39), NFKB1A (1.71), MAPK14 (1.92), TNFRSF1B (2.40), TLR4 (2.44), ATM (2.45), MECOM (2.57), PIK3CG (2.63), and EGFR (2.64). The top ten essential proteins with shortest pathways of positive regulation to glycolysis were ATM (1.67), CD28 (1.84), EGFR (1.98), TNFAIP3 (2.00), HGF (2.03), CYLD (2.09), PRKCZ (2.21), EPHA2 (2.25), JAK2 (2.27), and PIK3CG (2.34). The top ten essential proteins with shortest pathways of positive regulation to angiogenesis were TGFB1 (0.86), STAT3 (1.97), MAPK14 (1.98), EGFR (2.48), JAK2 (2.58), ATM (2.91), NFKBIA (2.96), PRKCZ (3.13), TLR4 (3.35), and PTAFR (3.46) (Supplementary Table 5). Lastly, Fig. 7 details all shortest pathways and distance scores of positive regulation from the 34 essential proteins to the inflammation phenotype.

Drugs involved in current COVID-19 clinical trials. Figure 8 details the current status of COVID-19 clinical trials regarding to essential inflammatory proteins, according to the Open Targets Platform. There are 5 drugs (small molecules) that are being analyzed in 8 clinical trials in advanced stages (phases III and IV) for 3 essential inflammatory proteins. Baricitinib is a tyrosine-protein kinase JAK1/2 inhibitor that acts on the JAK proteins and it is being studied in 4 clinical trials in phase III (NCT04640168, NCT04693026, NCT04421027, and NCT04401579). Similarly, pacritinib and ruxolitinib are tyrosine-protein kinase JAK1/2 inhibitors. They are currently been evaluated in a phase III clinical trial NCT04362137, respectively. Losmapimod is a MAP kinase p38 alpha inhibitor that acts on the MAPK14 protein and it is being studied in a phase III clinical trial (NCT04511819). Lastly, eritoran is a toll-like receptor 4/MD-2 antagonist that acts on the TLR4 protein and it is being studied in one clinical trial in phase IV (NCT02735707).

Discussion
Since the identification of patient zero in China, a wide spectrum of clinical features have been discovered in severe COVID-19. For instance, dyspnea, acute respiratory distress syndrome (ARDS), respiratory failure, lung edema, severe hypoxemia, cardiac arrhythmias, lymphopenia, hyperferritinemia, rhabdomyolysis, intravascular coagulopathy, and pulmonary thromboembolism. Nowadays, it is known that SARS-CoV-2 not only causes respiratory tract infection, but also skin, kidneys, blood, and central neural system pathologies. Therefore, it is imperative to continuously review the clinical manifestations and physiopathological mechanisms of the SARS-CoV-2 infection, especially with the appearance of new genomic variants.

Single-cell biology provides unprecedented resolution to the cellular underpinnings of biological processes in order to find therapeutically actionable targets for complex diseases. Melms et al have previously published the molecular single-cell lung atlas of lethal COVID-19. Motivated by this study, we performed an in-depth in silico analysis comparing the transcriptional data of 719 inflammatory response genes across 19 lung cell types belonging to COVID-19 autopsies. The functional enrichment analysis of the 233 significantly expressed inflammatory genes showed that the most significant biological annotations were inflammatory response (5.9 x 10^{-241}), cytokine production (9.5 x 10^{-62}), innate immune response (1.0 x 10^{-30}), macrophage activation (1.1 x 10^{-29}), TLR signaling pathway (3.8 x 10^{-15}), type I and II interferon production (1.8 x 10^{-13}), JAK-STAT signaling pathway (9.0 x 10^{-8}), NF-κB signaling pathway (2.0 x 10^{-6}), thymic stromal lymphopoietin (4.5 x 10^{-6}), TNF signaling pathway (4.9 x 10^{-6}), blood coagulation (5.6 x 10^{-6}), oncostatin M signaling pathway (5.9 x 10^{-6}), AGE-RAGE signaling pathway (5.9 x 10^{-6}), IL-1 and megakaryocytes in obesity (6.8 x 10^{-6}), and the NLRP3 inflammasome complex (2.5 x 10^{-4}).

The innate immune response is the first line of defense against new invading pathogens. Pattern recognition receptors (PRRs) are capable of recognizing molecules with conserved motifs commonly shared by pathogen groups. Recognition by these receptors triggers innate immune responses and induces multiple IFN and pro-inflammatory cytokine secretion in COVID-19 patients. Upon ligand recognition PRRs initiate a signaling pathway that activate key transcription factors, such as NF-κB, AP-1, and interferon regulatory factors (IRF3 and IRF7) that induce pro-inflammatory cytokines and type I interferon. Type I IFNs are responsible for inducing the JAK-STAT signaling pathway to activate IFN-stimulated genes and develop the “anti-viral state” in the infected organism.

Interferon is a cytoplasmic glycoprotein with antiviral activity. This cytokine is another contributing factor in the humoral immunological response against respiratory viruses. Through a bronchoalveolar lavage in severely ill patients, evidence of local induction of interferon and the stimulation of interferon genes was found. In contrast, minimal levels of interferon were found in peripheral blood of severely ill patients. This increased cytokine production with limited interferon levels might be due to an antagonist mechanism of the nsp1 protein against interferon signaling. Regarding the genetics underlying severe COVID-19, Zhang et al concluded that genetics may determine the clinical course of
SARS-CoV-2 infection identifying mutations in genes involved in the regulation of type I and III IFN immunity \(^\text{86}\), and Bastard et al identified high titers of neutralizing autoantibodies against type I IFN-\(\alpha\)2 and IFN-\(\omega\) in 10% of patients with severe COVID-19 \(^\text{87}\).

Macrophages are cells that perform crucial functions in the immune system, from the phagocytosis of the viruses and bacteria to maintaining homeostasis \(^\text{88}\). Precisely, macrophages produce high amounts of pro-inflammatory cytokines in patients with ARDS, who present an activated state known as cytokine storm or macrophage activation syndrome \(^\text{89}\). The overexpression of cytokines (i.e., TNF-\(\alpha\), IL-2, IL-10, IL-1, and IL-6) leads to a hyperinflammatory response, which has been reported as a remarkable feature of SARS-CoV-2 infection \(^\text{90–92}\). IL-6 plays a main role in the severity of COVID-19, while TNF-\(\alpha\) and IL-1\(\beta\) trigger the NF-\(\kappa\)B signaling pathway \(^\text{93,94}\). The excessive production of cytokines leads to development of pathological symptoms, such as lung damage, cell death, severe pneumonia, ARDS, lung fibrosis, and multiple organ failure \(^\text{93,95}\). Hence, this cytokine storm plays a crucial role in the progression of SARS-CoV-2 infection and is considered as one of the main causes of lethal COVID-19 \(^\text{92,93}\).

TNF is considered as one of the most important pro-inflammatory cytokines, affecting different parts of the immune system and regulating various pathological and physiological processes \(^\text{96}\). Therefore, the TNF\(\alpha\)-NF-\(\kappa\)B axis is considered as a potential therapeutic target in COVID-19 \(^\text{97}\). Initially, NF-\(\kappa\)B is present within the cytoplasm, after activation of IkB through phosphorylation of IkB kinase, NF-\(\kappa\)B is activated and translocated to the nucleus where it regulates the transcription of various target genes \(^\text{98,99}\). To date, SARS-CoV-2-mediated NF-\(\kappa\)B activation has been observed in several cells such as macrophages of liver, kidney, lung, central nervous system, cardiovascular system, and gastrointestinal system. This causes a chronic production of IL-1, IL-2, IL-6, IL-12, TNF-\(\alpha\), LT-\(\alpha\), LT-\(\beta\), GM-CSF, and several chemokines, leading to the aforementioned pathological symptoms \(^\text{100}\). Catanzaro \(\text{et al}\) have recently published a report analyzing the role of the TNF\(\alpha\)-NF-\(\kappa\)B pathway in COVID-19. In their report, it was suggested that inhibiting this axis may prevent pulmonary complications in COVID-19 patients \(^\text{97}\). This was also observed in SARS-CoV infection. NF-\(\kappa\)B expression was elevated in the lungs of recombinant SARS-CoV-1-infected mice, while NF-\(\kappa\)B inhibitors reduced SARS-CoV-related expanding survival of these mice \(^\text{101}\).

The cytokine signaling depends on the JAK and STAT which are phosphorylated and activated upon cytokines binding to their receptors. The STAT homodimers translocate into the nucleus, where they upregulate the transcription of several genes that participate not only in cytokine production but also in apoptosis, immune regulation, and cell cycle differentiation \(^\text{102}\). In the context of SARS-CoV-2 infection, inhibition of the JAK-STAT pathway seems as promising approach to prevent cytokines storm in fatal cases or in patients with comorbidities that express high levels of inflammatory markers such as of IL-6, TNF\(\alpha\), IL-17a, GM-CSF, and G-CSF \(^\text{103}\). In fact, the GenOMICC GWAS study suggests that individuals with a variant on chromosome 19: 10,466,123 that affects expression of tyrosine kinase 2 (TYK2), member of the JAK family, could be associated with a host-driven inflammatory response that leads to severe lung injury \(^\text{104}\). Thus, several clinical trials have shown that baricitinib, a JAK inhibitors possesses a good
safety and efficacy profiles in reducing cytokine levels of severe COVID-19 patients without side effects. Nevertheless, the JAK/STAT pathway is also necessary to mediate the immune response to clear viral infections and prolonged inhibition of the pathway could lead to immunosuppression and prolonged infections. For instance, SARS-CoV-2 is able to hijack the JAK/STAT pathway in order to increase its proliferation by evading the immune response. Li et al showed that SARS-CoV-2 infected cell had a decreased expression of JAK1, JAK2, TYK2, and STAT2 proteins. This is explained by action of viral nsp1, ORF6, and ORF8 that prevent the phosphorylation of STAT1 and STAT3 to inhibit IFN production. Therefore, the timeline for administration of JAK/STAT inhibitors should be carefully analyzed since reducing the hyperinflammation could affect viral clearance. Due to the narrow therapeutic window of JAK/STAT inhibitors, dosage should aim to restore the immune response homeostasis.

The incidence of thrombotic events in COVID-19 patients responsible for strokes and heart attacks raises the concern about the abnormal coagulation patterns and poor prognosis in the actual pandemic. Tang et al reported that 71.4% of non-surviving COVID-19 patients met the criteria for disseminated intravascular coagulation and presented high levels of coagulation-related biomarkers such as D-dimer and fibrin degradation products. The mechanisms of the coagulopathy are not clear; however, some reports indicate that dysregulated immune responses are involved in such processes. Exacerbation of inflammatory cytokines promoting proliferation of megakaryocytes, lymphocyte cell-death, hypoxia, endothelial damage contributing to ischemia and organ dysfunction, and the association between autoantibodies and neutrophil extracellular traps seem to be involved in the abnormal thrombotic events in COVID-19 patients.

Oncostatin M is a cytokine involved in homeostasis and chronic inflammation that has pleiotropic functions such as cell differentiation and proliferation, and it is present in hematopoietic, immunological, and inflammatory networks. One of the most important functions of oncostatin M is the stimulation of the chemokines CCL1, CCL7 and CCL8 in primary human dermal fibroblasts at a faster kinetics than IL-1β or TNF-α. In 2020, it was proposed as a new mortality biomarker in patients with acute respiratory failure supported by venous-venous extracorporeal membrane oxygenation. In the case of COVID-19, an increase of OSM plasma levels and other inflammatory mediators was detected; this finding was correlated with the severity of disease and the increase of bacterial products in plasma. Finally, OSM is curiously elevated in obese patients and upon recognition by its specific receptor (OSMRβ) induces obesity and insulin resistance conditions.

Obesity is one of the main risk factors associated with lethal COVID-19, and levels of pro-inflammatory cytokines increase under this pathology. Low NAD+ levels in obese individuals decrease the activity of SIRT1, a molecule that modulates cytokine production. However, the excess of amino acid availability hyperactivates the mTOR signaling pathway increasing viral replication and inflammatory response. Additionally, because adipose tissue has a considerable level of ACE2 expression, viral
shedding increases, as well as the production of pro-inflammatory factors. This inflammatory process contributes to thrombotic problems, a probable cause of multiorgan failure, which has been evidenced by the presence of elevated levels of megakaryocytes in COVID-19 autopsies.

Thymic stromal lymphopoietin is an epithelial cytokine normally produced by airway epithelial cells. It has been associated with T-helper type 2 (Th2) responses in allergic diseases, highlighting its role in inflammatory disease pathogenesis. It has been discovered that TSLP can be triggered by respiratory viral infections, bacteria, allergens and injuries. TSLP acts upon cells with TSLP receptor such as hematopoietic progenitor cells, eosinophils, basophils, mast cells, airway smooth muscle cells, group 2 innate lymphoid cells, lymphocytes, dendritic cells and monocytes/macrophages. When several immune mediators were measured in patient's plasma suffering from influenza A (H1N1) and COVID-19, TSLP levels were significantly upregulated in COVID-19 patients. This fact suggests a possible contribution of TSLP in COVID-19 pathogenesis and perhaps aids differential diagnosis. Besides, since TSLP concentration was reported to be higher in severely affected than in mild and moderated COVID-19 cases, it may be potentially used as a biomarker for disease severity.

Optimal NLRP3 inflammasome activation is crucial for host immune defense against several pathogenic infections. SARS-CoV-2 activates inflammasomes, which are large multiprotein assemblies that are broadly responsive to pathogen-associated cellular insults, leading to secretion of cytokines and an inflammatory form of cell death. However, excessive activation can lead to systemic inflammation and tissue damage which are detrimental to the host. Patients with severe COVID-19 have been found to have higher serum concentrations of pro-inflammatory cytokines and chemokines such as granulocyte-colony stimulating factor (GCSF), monocyte chemoattractant protein 1 (MCP1), TNF, IL-6, and IL-1β compared with healthy individuals. A unified mechanism for NLRP3 inflammasome activation has not been proposed yet; however, some researchers have found that SARS-CoV-2 ORF-8b interacts with the LRR domain of NLRP3 inflammasome activating IL-1β secretion in THP-1 macrophages. Findings suggest that SARS-CoV-2 infection leads to NLRP3 inflammasome activation, caspase-1 cleavage, and the release of IL-1β stimulating pyroptosis in peripheral blood mononuclear cells from severe COVID-19 patients.

In a biological system approach, SARS-CoV-2 employs a suite of virulent proteins that interacts with host targets to extensively rewire the flow of information and cause COVID-19. The human proteins physically associated with SARS-CoV-2 are the first line of host proteins, which also interact with proteins involved in a wide spectrum of signaling pathways and biological processes within lung cells. In this study, we identified 111 pulmonary inflammatory response proteins with the highest confidence interactions to human-SARS-CoV-2 proteins, being the top ten: C3, FN1, NFKB1, RPS19, CTSC, HSPD1, APP, ITGAM, SNAP23, and MAPK14.

Subsequently, we analyzed these 111 inflammatory response proteins to identify those with the shortest pathways to four cancer hallmark phenotypes. Inflammation is a hallmark of cancer observed in patients...
with SARS-CoV-2 infection\textsuperscript{135}. The chronic inflammatory process causes cell death\textsuperscript{136,137}, angiogenesis\textsuperscript{138}, and during the peak of inflammation, immune cells preferentially use glycolysis as a source of energy\textsuperscript{139}. These facts provide a biological rationale to analyze and prioritize the inflammatory response proteins with the shortest distance scores to these biological phenotypes. Consequently, we identified 34 essential inflammatory response proteins highly associated with cell death, glycolysis, and angiogenesis. These proteins were: PTGS2, PRKCZ, NFKBIA, MAPK14, TNFRSF1B, TLR4, ATM, MECOM, PIK3CG, EGFR, JAK2, LYN, CYLD, PRKCQ, STAT3, TGFB1, RBPJ, TNFAIP3, NOTCH1, IGF1, CD28, CCL5, PTAFR, FPR1, EDNRA, EDNRB, CYSLTR1, CNR2, HGF, EPHA2, FN1, CSF1, PTGFR, and APP.

The SARS-CoV-2 infection of lung epithelial cells activates caspase-8 to trigger the three major cell death pathways, including apoptosis, pyroptosis, and necroptosis. Cell death and inflammatory responses are intimately linked during SARS-CoV-2 infection\textsuperscript{140}. Lastly, analysis of postmortem lung sections of lethal COVID-19 patients has revealed that inflammatory responses from lung epithelial cells may induce infiltration of inflammatory cells that trigger strong immune pathogenesis\textsuperscript{137}.

Recent studies showed that SARS-CoV-2 rewire human monocytes in a high glucose culture medium. This induces viral replication and cytokine production, and might be the reason why people suffering from diabetes, obesity and other related metabolic diseases are more susceptible to developing severe COVID-19\textsuperscript{139}. For instance, people with type 2 diabetes show an increased glucose metabolism due to hyperglycemia, which may boost SARS-CoV-2 pathogenesis\textsuperscript{139}. Codo \textit{et al.} proved that glycolytic flux is essential for SARS-CoV-2 impact\textsuperscript{141}. Through several assays, they inhibited glycolysis by blocking 2-deoxy-D-glucose (2-DG) and glycolytic enzymes 6-phospho-fructo-2-kinase/fructose-2,6-biphosphatase-3 (PFKFB3) and lactate dehydrogenase A (LDH-A), as a consequence, they observed that both viral replication and cytokine response stopped\textsuperscript{141}. The metabolic transcription factor HIF-1\textalpha activity and related genes are strongly stimulated in SARS-CoV-2 infected blood monocytes isolated from severe COVID-19 patients\textsuperscript{141}. HIF-1\textalpha is also a major glycolysis regulator, when inhibited, viral replication and cytokine expression were also blocked. Overall, these experiments showed that high glucose concentration and glycolysis are essential for SARS-CoV-2 replication, inflammatory response, and upregulation of ACE2\textsuperscript{141}.

Angiogenesis occurs in response to the activation of acute inflammation or chronic systemic hypoxia pathways that increase the expression of proteins and factors (HIF-1\textalpha, VEGF, NO) associated with its development\textsuperscript{142}. During the SARS-CoV-2 infection, local endothelial damage, known as endotheliitis, is associated with acute inflammation of the outermost endovascular layers, triggering a cascade of reactions that result in endothelial inflammation, platelet aggregation, and impaired laminar flow\textsuperscript{138,143}. In the context of COVID-19 disease, the reported vasoconstriction and subsequent hypoxia, stimulate the formation of new blood vessels by promoting branching of pre-existing blood vessels (intussusception) and \textit{de novo} angiogenesis that contributes to the already established systemic hypoxia\textsuperscript{144}. This process together with the systemic hypoxia observed in severe COVID-19 patients cause a structural and
functional reorganization of the pullmonary tissue, which ultimate function is to allow an adequate gas exchange between the tissue and the cells \(^{142}\).

Regarding drugs against COVID-19 disease, in this study we propose five small molecules (ruxolitinib, baricitinib, pacritinib, losmapimod, and eritoran) that after being thoroughly analyzed in COVID-19 clinical trials, these drugs can be considered for treating severe COVID-19 patients.

A systematic review and meta-analysis published by Walz et al concluded that Janus kinase-inhibitor treatment is significantly associated with positive clinical outcomes in terms of mortality, intensive care unit admission, and discharge \(^{145}\). Ruxolitinib is a tyrosine-protein kinase JAK1/2 inhibitor \(^{146}\) that is currently used for myelofibrosis and polycythemia vera, both hematologic malignancies. The use of ruxolitinib in these diseases is based on its ability of being a kinase inhibitor, which mediates the signaling of a number of cytokines and growth factors that are important in hematopoiesis and immune function. Based on this principle, it is reasonable then, from a clinical point of view, to use this drug to specifically manage cytokine storm in COVID-19 \(^{147}\). According to Yan et al, ruxolitinib normalized interferon signature genes and all complement gene transcripts induced by SARS-CoV-2 in lung epithelial cell lines. They proposed that combination therapy with JAK inhibitors and drugs that normalize NF-κB-signaling could potentially have clinical application for severe COVID-19 \(^{146}\). Baricitinib is a tyrosine-protein kinase JAK1/2 inhibitor \(^{148}\) mainly used for rheumatoid arthritis, and among its pharmacological properties it has an antiviral effect on the entry of a virus \(^{149}\). At the moment, baricitinib is approved by the WHO, the Food and Drug Administration (FDA) of the United States, and the National Institutes of Health (NIH) for emergent use in severe pneumonia due to COVID-19 \(^{150}\). The use of baricitinib is indicated in COVID-19 critically ill patients with high oxygen needs despite the use of dexamethasone (the only approved corticosteroid), however it should not be used when IL-6 inhibitors such as tocilizumab have been started, given that its combined use has not yet been tested as well as its safety. The known efficacy of Baricitinib is from emerging data from an unpublished article where the 27.8% of participants receiving baricitinib vs 30.5% receiving placebo progressed (primary endpoint, odds ratio 0.85, 95% CI 0.67–1.08; \(p = 0.18\)), and the all-cause mortality was 8.1% for baricitinib and 13.1% for placebo, corresponding to a 38.2% reduction in mortality (hazard ratio [HR] 0.57, 95% CI 0.41–0.78; nominal \(p = 0.002\)) \(^{151}\). Pacritinib is also a protein-kinase inhibitor mainly focused on JAK2 and FLT3 protein targets. This small molecule has been developed for the treatment of myelofibrosis \(^{152}\). On the other hand, losmapimod is a MAP kinase p38 alpha inhibitor that has been investigated for the prevention of chronic obstructive pulmonary disease and cardiovascular disease \(^{153}\). The therapeutic hypothesis for the use of losmapimod in COVID-19 is that increased mortality is caused by p38 MAPK-mediated exaggerated acute inflammatory response resulting in SARS-CoV-2 infection. Lastly, eritoran is a Toll-like receptor 4 / MD-2 antagonist that downregulates the intracellular generation of pro-inflammatory cytokines IL-6 and TNF-alpha in human monocytes, and has been developed for the treatment of severe sepsis. Shirey et al examined how antagonizing TLR4 signaling has been effective experimentally in ameliorating acute lung injury and lethal infection in challenge models triggered by acute lung injury-inducing viruses \(^{154}\).
Considering the enormous pressure that health systems are facing due to the COVID-19 pandemic and the continuous need to present and implement comprehensive health strategies that can address the global situation; mainly after the emergence of different variants, it is imperative to recognize the urgent need to diminish the gaps between research and the implementation of public health measures. In fact, it may be unprecedented in the history of science to know how many research articles related to COVID-19 have been submitted and published. However, according to Park et al., the research community has emphasized on “the new norm of publishing: quantity over quality” and this is also related to the well known problems that clinical trials faced even before the pandemic. This is of particular interest to our research given that we acknowledge that clinical trials are essential in evidence-based medicine, and consequently, in the decision making process of public health policies and strategies. Relevantly, the need to smartly invest not only in randomized clinical trials but also in large-scale clinical trials with master protocols and conducted by coordinated and collaborative structures, as also supported by Park et al. These clinical trials networks are essential to coordinate actions between clinical researchers and health practitioners, also promoting knowledge sharing, leadership, and cost-time reductions. In addition, it is critical to decentralize, improve and increase clinical trials in low and middle-income countries, as current evidence shows large inequalities and concentrations of funds and information in high-income countries. This holds true especially for Latin America, one of the most affected regions in the world by the pandemic.

The role of health research is fundamental in the response to COVID-19, considering the importance of data sharing and assuring efficiency, equity, and effectiveness in the diverse processes. Contradictorily, a large number of clinical trials might never be completed and others are done with doubtful methodologies. Thus, analyzing potential drugs targets for COVID-19, especially the ones which can serve for severe cases, need an urgent and efficient development of well designed and managed clinical trials, which can provide potential interventions that help people to live longer, diminish long-term effects, manage pain and/or possible disabilities; not to mention the possible positive effects on the reduction of hospitalization costs, both at the individual level and in terms of possible savings for the national health system. As another study also mentioned, the potential and benefits of repositioning clinical trials are directed to use the already available information of safe and affordable generic drugs and propose “potential, prompt, cost-effective, and safe solutions for the public and global health problems, with a human-centered approach”. This is also conveyed by the Pan American Health Organization (PAHO), which adds to the benefits, the idea of having already pharmaceutical formed supply chains.

Finally, as other authors have contributed, the current global research situation must be guided towards a collaborative and synergetic approach instead of being conceived as a competitive and isolated process. The COVID-19 pandemic assures the need to eliminate structural barriers that increase health inequalities, and in this perspective, benefits, knowledge, and of course potential treatments must be available for all, in order to achieve universal health coverage and equity.

**Declarations**
Data Availability Statement

The datasets generated for this study are included in this published article (and its Supplementary Information files).

Author Contributions

AL-C conceived the subject and the conceptualization of the study. NCK gave conceptual advice and valuable scientific input. AL-C, PG-R, AA-S, AL-S, EA, AAP-M, KN-J, AM, and FA-E did data curation and supplementary data. AL-C did funding acquisition. All authors wrote and edited the manuscript. Lastly, all authors reviewed and approved the manuscript.

Conflict of Interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Funding

No information

Acknowledgments
This work was supported by the Latin American Society of Pharmacogenomics and Personalized Medicine (SOLFAGEM).

References

1. Tay, M. Z., Poh, C. M., Rénia, L., MacAry, P. A. & Ng, L. F. P. The trinity of COVID-19: immunity, inflammation and intervention. *Nat. Rev. Immunol.* (2020) doi:10.1038/s41577-020-0311-8.

2. Sanders, J. M., Monogue, M. L., Jodlowski, T. Z. & Cutrell, J. B. Pharmacologic Treatments for Coronavirus Disease 2019 (COVID-19): A Review. *JAMA - Journal of the American Medical Association* (2020) doi:10.1001/jama.2020.6019.

3. WHO. COVID-19 Weekly Epidemiological Update 35. *World Heal. Organ.* 1–3 (2020).

4. Ortiz-Prado, E. et al. Clinical, molecular, and epidemiological characterization of the SARS-CoV-2 virus and the Coronavirus Disease 2019 (COVID-19), a comprehensive literature review. *Diagnostic Microbiology and Infectious Disease* (2020) doi:10.1016/j.diagmicrobio.2020.115094.

5. Ziegler, C. G. K. et al. SARS-CoV-2 Receptor ACE2 Is an Interferon-Stimulated Gene in Human Airway Epithelial Cells and Is Detected in Specific Cell Subsets across Tissues. *Cell* (2020) doi: 10.1016/j.cell.2020.04.035.

6. Zhou, P. et al. A pneumonia outbreak associated with a new coronavirus of probable bat origin. *Nature* (2020) doi:10.1038/s41586-020-2012-7.

7. Wu, A. et al. Genome Composition and Divergence of the Novel Coronavirus (2019-nCoV) Originating in China. *Cell Host Microbe* 27, 325–328 (2020).

8. Zhang, L. et al. Crystal structure of SARS-CoV-2 main protease provides a basis for design of improved α-ketoamide inhibitors. *Science* (2020) doi:10.1126/science.abb3405.

9. Gao, Y. et al. Structure of the RNA-dependent RNA polymerase from COVID-19 virus. *Science (80-.)* eabb7498 (2020) doi:10.1126/science.abb7498.

10. Gordon, D. E. et al. A SARS-CoV-2 protein interaction map reveals targets for drug repurposing. *Nature* (2020) doi:10.1038/s41586-020-2286-9.

11. López-Cortés, A. et al. In silico Analyses of Immune System Protein Interactome Network, Single-Cell RNA Sequencing of Human Tissues, and Artificial Neural Networks Reveal Potential Therapeutic Targets for Drug Repurposing Against COVID-19. *Front. Pharmacol.* 12, 1–24 (2021).

12. Lo Presti, A., Rezza, G. & Stefanelli, P. Selective pressure on SARS-CoV-2 protein coding genes and glycosylation site prediction. *Heliyon* 6, (2020).

13. Zhou, B. et al. SARS-CoV-2 spike D614G change enhances replication and transmission. *Nature* 592, (2021).

14. McCormick, B. K. D., Jacobs, J. L. & Mellors, J. W. The emerging plasticity of SARS-CoV-2. *Science (80-.)* 371, 1306–1308 (2021).
15. States, U. *et al.* Article Emergence and rapid transmission of SARS-CoV-2 II Article Emergence and rapid transmission of SARS-CoV-2 B. 1. 1. 7 in the United States. *Cell* **184**, 2587–2594.e7 (2021).

16. Burioni, R. & Topol, E.. Has SARS-CoV-2 reached peak fitness? *Nat Med* (2021) doi:https://doi.org/10.1038/s41591-021-01421-7.

17. Boehm, E. *et al.* Novel SARS-CoV-2 variants: the pandemics within the pandemic. *Clin. Microbiol. Infect.* (2021) doi:10.1016/j.cmi.2021.05.022.

18. CDC. SARS-CoV-2 Variant Classifications and Definitions. *Variant Surveillance* (2021).

19. ECDC. SARS-CoV-2 variants of concern as of 8 July 2021. *Situation updates on COVID-19* (2021).

20. Harvey, W. T. *et al.* SARS-CoV-2 variants, spike mutations and immune escape. *Nat. Rev. Microbiol.* **19**, (2021).

21. Burton, D. & Topol, E. Toward superhuman SARS-CoV-2 immunity? *Nat Med* **27**, 5–6 (2021).

22. Wang, Y. *et al.* Pulmonary alveolar type I cell population consists of two distinct subtypes that differ in cell fate. *Proc. Natl. Acad. Sci. U. S. A.* **115**, (2018).

23. Hiemstra, P. S., McCray, P. B. & Bals, R. The innate immune function of airway epithelial cells in inflammatory lung disease. *Eur. Respir. J.* **45**, (2015).

24. Weitnauer, M., Mijošek, V. & Dalpke, A. H. Control of local immunity by airway epithelial cells. *Mucosal Immunology* vol. 9 (2016).

25. Yoo, J. K., Kim, T. S., Hufford, M. M. & Braciale, T. J. Viral infection of the lung: Host response and sequelae. *Journal of Allergy and Clinical Immunology* vol. 132 (2013).

26. Niethamer, T. K. *et al.* Defining the role of pulmonary endothelial cell heterogeneity in the response to acute lung injury. *Elife* **9**, (2020).

27. Espinosa, E. & Valitutti, S. New roles and controls of mast cells. *Current Opinion in Immunology* vol. 50 (2018).

28. Biswas, S. K. & Mantovani, A. *Macrophages: Biology and role in the pathology of diseases.* *Macrophages: Biology and Role in the Pathology of Diseases* (2014). doi:10.1007/978-1-4939-1311-4.

29. Schraml, B. U. & Reis e Sousa, C. Defining dendritic cells. *Current Opinion in Immunology* vol. 32 (2015).

30. Murray, P. J. Immune regulation by monocytes. *Seminars in Immunology* vol. 35 (2018).

31. Bi, J. & Tian, Z. NK cell exhaustion. *Frontiers in Immunology* vol. 8 (2017).

32. van Eeden, C., Khan, L., Osman, M. S. & Tervaert, J. W. C. Natural killer cell dysfunction and its role in covid-19. *International Journal of Molecular Sciences* vol. 21 (2020).

33. Maucourant, C. *et al.* Natural killer cell immunotypes related to COVID-19 disease severity. *Sci. Immunol.* **5**, (2020).

34. Culley, F. J. Natural killer cells in infection and inflammation of the lung. *Immunology* vol. 128 (2009).
35. Peng, X. et al. Sharing CD4 + T Cell Loss: When COVID-19 and HIV Collide on Immune System. *Frontiers in Immunology* vol. 11 (2020).

36. Zhang, N. & Bevan, M. J. CD8 + T Cells: Foot Soldiers of the Immune System. *Immunity* vol. 35 (2011).

37. Savage, P. A., Klawon, D. E. J. & Miller, C. H. Regulatory T Cell Development. *Annu. Rev. Immunol.* 38, (2020).

38. Gladstone, D. E., Kim, B. S., Mooney, K., Karaba, A. H. & D’Alessio, F. R. Regulatory T Cells for Treating Patients With COVID-19 and Acute Respiratory Distress Syndrome: Two Case Reports. *Ann. Intern. Med.* 173, (2020).

39. Shuwa, H. A. et al. Alterations in T and B cell function persist in convalescent COVID-19 patients. *Med* 2, (2021).

40. Nutt, S. L., Hodgkin, P. D., Tarlinton, D. M. & Corcoran, L. M. The generation of antibody-secreting plasma cells. *Nat. Rev. Immunol.* 15, (2015).

41. Smith, R. S., Smith, T. J., Blieden, T. M. & Phipps, R. P. Fibroblasts as sentinel cells. Synthesis of chemokines and regulation of inflammation. *The American journal of pathology* vol. 151 (1997).

42. Wang, G., Jacquet, L., Karamariti, E. & Xu, Q. Origin and differentiation of vascular smooth muscle cells. *J. Physiol.* 593, (2015).

43. Blake, K. J., Jiang, X. R. & Chiu, I. M. Neuronal Regulation of Immunity in the Skin and Lungs. *Trends in Neurosciences* vol. 42 (2019).

44. Blanco-Melo, D. et al. Imbalanced host response to SARS-CoV-2 drives development of COVID-19. *Cell* (2020) doi:10.1016/j.cell.2020.04.026.

45. Liao, M. et al. The landscape of lung bronchoalveolar immune cells in COVID-19 revealed by single-cell RNA sequencing. *medRxiv* (2020) doi:10.1101/2020.02.23.20026690.

46. Butler, D. et al. Shotgun transcriptome, spatial omics, and isothermal profiling of SARS-CoV-2 infection reveals unique host responses, viral diversification, and drug interactions. *Nat. Commun.* 12, 1–17 (2021).

47. Wilk, A. J. et al. A single-cell atlas of the peripheral immune response in patients with severe COVID-19. *Nat. Med.* 26, 1070–1076 (2020).

48. Dong, Y. et al. Epidemiology of COVID-19 among children in China. *Pediatrics* 145, (2020).

49. Zhou, F. et al. Clinical course and risk factors for mortality of adult inpatients with COVID-19 in Wuhan, China: a retrospective cohort study. *Lancet* 395, 1054–1062 (2020).

50. Huang, D. W., Sherman, B. T. & Lempicki, R. A. Systematic and integrative analysis of large gene lists using DAVID bioinformatics resources. *Nat. Protoc.* 4, 44–57 (2009).

51. Melms, J. C. et al. A molecular single-cell lung atlas of lethal COVID-19. *Nature* (2021) doi:10.1038/s41586-021-03569-1.

52. Slyper, M. et al. A single-cell and single-nucleus RNA-Seq toolbox for fresh and frozen human tumors. *Nat. Med.* 26, (2020).
53. Raudvere, U. et al. g:Profiler: a web server for functional enrichment analysis and conversions of gene lists (2019 update). *Nucleic Acids Res.* (2019) doi:10.1093/nar/gkz369.

54. Reimand, J. et al. g:Profiler-a web server for functional interpretation of gene lists (2016 update). *Nucleic Acids Res.* (2016) doi:10.1093/nar/gkw199.

55. Slenter, D. N. et al. WikiPathways: A multifaceted pathway database bridging metabolomics to other omics research. *Nucleic Acids Res.* (2016) doi:10.1093/nar/gkx1064.

56. Jassal, B. et al. The reactome pathway knowledgebase. *Nucleic Acids Res.* (2020) doi:10.1093/nar/gkz1031.

57. Ogata, H. et al. KEGG: Kyoto encyclopedia of genes and genomes. *Nucleic Acids Research* vol. 27 29–34 (1999).

58. Subramanian, A. et al. Gene set enrichment analysis: A knowledge-based approach for interpreting genome-wide expression profiles. *Proc. Natl. Acad. Sci. U. S. A.* 102, 15545–15550 (2005).

59. Doncheva, N. T., Morris, J. H., Gorodkin, J. & Jensen, L. J. Cytoscape StringApp: Network Analysis and Visualization of Proteomics Data. *J. Proteome Res.* (2019) doi:10.1021/acs.jproteome.8b00702.

60. Szklarczyk, D. et al. STRING v10: protein-protein interaction networks, integrated over the tree of life. *Nucleic Acids Res.* 43, D447-52 (2015).

61. López-Cortés, A. et al. Gene prioritization, communality analysis, networking and metabolic integrated pathway to better understand breast cancer pathogenesis. *Sci. Rep.* 8, 16679 (2018).

62. López-Cortés, A. et al. OncoOmics approaches to reveal essential genes in breast cancer: a panoramic view from pathogenesis to precision medicine. *Sci. Rep.* 10, 5285 (2020).

63. Tang, Y., Li, M., Wang, J., Pan, Y. & Wu, F. X. CytoNCA: A cytoscape plugin for centrality analysis and evaluation of protein interaction networks. *BioSystems* (2015) doi:10.1016/j.biosystems.2014.11.005.

64. Shannon, P. et al. Cytoscape: a software environment for integrated models of biomolecular interaction networks. *Genome Res.* 13, 2498–504 (2003).

65. Iannuccelli, M. et al. CancerGeneNet: Linking driver genes to cancer hallmarks. *Nucleic Acids Res.* (2020) doi:10.1093/nar/gkz871.

66. Perfetto, L. et al. SIGNOR: A database of causal relationships between biological entities. *Nucleic Acids Res.* (2016) doi:10.1093/nar/gkv1048.

67. Carvalho-Silva, D. et al. Open Targets Platform: New developments and updates two years on. *Nucleic Acids Res.* (2019) doi:10.1093/nar/gky1133.

68. Gaulton, A. et al. The ChEMBL database in 2017. *Nucleic Acids Res.* (2017) doi:10.1093/nar/gkw1074.

69. Gawel, D. R. et al. A validated single-cell-based strategy to identify diagnostic and therapeutic targets in complex diseases. *Genome Med.* (2019) doi:10.1186/s13073-019-0657-3.

70. Stephenson, E. et al. Single-cell multi-omics analysis of the immune response in COVID-19. *Nat. Med.* 27, (2021).
71. Montenegro, F. et al. Acute respiratory distress syndrome (ARDS) caused by the novel coronavirus disease (COVID-19): a practical comprehensive literature review. Expert Rev. Respir. Med. (2020) doi:10.1080/17476348.2020.1820329.

72. Terpos, E. et al. Hematological findings and complications of COVID-19. American journal of hematology (2020) doi:10.1002/ajh.25829.

73. Zhang, Y. et al. Coagulopathy and Antiphospholipid Antibodies in Patients with Covid-19. N. Engl. J. Med. 382, e38 (2020).

74. Fogarty, H. et al. COVID-19 Coagulopathy in Caucasian patients. Br. J. Haematol. (2020) doi:10.1111/bjh.16749.

75. Rotzinger, D. C., Beigelman-Aubry, C., von Garnier, C. & Qanadli, S. D. Pulmonary embolism in patients with COVID-19: Time to change the paradigm of computed tomography. Thrombosis Research (2020) doi:10.1016/j.thromres.2020.04.011.

76. Delorey, T. M. et al. COVID-19 tissue atlases reveal SARS-CoV-2 pathology and cellular targets. Nature 595, (2021).

77. Alberts, B. et al. Innate Immunity. in Molecular Biology of the Cell vol. 4 (Garland Science, 2002).

78. Amarante-Mendes, G. P. et al. Pattern recognition receptors and the host cell death molecular machinery. Frontiers in Immunology vol. 9 (2018).

79. Boechat, J. L., Chora, I., Morais, A. & Delgado, L. The immune response to SARS-CoV-2 and COVID-19. Pulmonology 1–15 (2021) doi:https://doi.org/10.1016/j.pulmoe.2021.03.008.

80. Channappanavar, R. et al. IFN-I response timing relative to virus replication determines MERS coronavirus infection outcomes. J. Clin. Invest. (2019) doi:10.1172/JCI126363.

81. Perlman, S. & Dandekar, A. A. Immunopathogenesis of coronavirus infections: Implications for SARS. Nat. Rev. Immunol. 5, 917–927 (2005).

82. Lopez, L., Sang, P. C., Tian, Y. & Sang, Y. Dysregulated interferon response underlying severe covid-19. Viruses vol. 12 (2020).

83. Billiau, A. Interferon: The pathways of discovery. I. Molecular and cellular aspects. Cytokine and Growth Factor Reviews vol. 17 381–409 (2006).

84. Acharya, D., Liu, G. Q. & Gack, M. U. Dysregulation of type I interferon responses in COVID-19. Nature Reviews Immunology vol. 20 397–398 (2020).

85. Samudrala, P. K. et al. Virology, pathogenesis, diagnosis and in-line treatment of COVID-19. Eur. J. Pharmacol. 883, (2020).

86. Zhang, Q. et al. Inborn errors of type I IFN immunity in patients with life-threatening COVID-19. Science (80-.). 370, (2020).

87. Bastard, P. et al. Autoantibodies against type I IFNs in patients with life-threatening COVID-19. Science (80-.). 370, (2020).

88. Gracia-Hernandez, M., Sotomayor, E. M. & Villagra, A. Targeting Macrophages as a Therapeutic Option in Coronavirus Disease 2019. Front. Pharmacol. 11, (2020).
89. Otsuka, R. & Seino, K. I. Macrophage activation syndrome and COVID-19. *Inflamm. Regen.* **40**, (2020).
90. Zhang, X., Zhang, Y., Qiao, W., Zhang, J. & Qi, Z. Baricitinib, a drug with potential effect to prevent SARS-COV-2 from entering target cells and control cytokine storm induced by COVID-19. *Int. Immunopharmacol.* **86**, 106749 (2020).
91. Kahn, R. *et al.* Mismatch between circulating cytokines and spontaneous cytokine production by leukocytes in hyperinflammatory COVID-19. *J. Leukoc. Biol.* **109**, 115–120 (2021).
92. Rabaan, A. A. *et al.* Role of inflammatory cytokines in covid-19 patients: A review on molecular mechanisms, immune functions, immunopathology and immunomodulatory drugs to counter cytokine storm. *Vaccines* **9**, (2021).
93. Rowaiye, A. B. *et al.* Attenuating the effects of novel COVID-19 (SARS-CoV-2) infection-induced cytokine storm and the implications. *J. Inflamm. Res.* **14**, 104673 (2021).
94. Tjan, L. H. *et al.* Early Differences in Cytokine Production by Severity of Coronavirus Disease 2019. *J. Infect. Dis.* **223**, 1145–1149 (2021).
95. Mustafa, M. I., Abdelmoneim, A. H., Mahmoud, E. M. & Makhawi, A. M. Cytokine Storm in COVID-19 Patients. Its Impact on Organs and Potential Treatment by QTY Code-Designed Detergent-Free Chemokine Receptors. *Mediators Inflamm.* 2020, (2020).
96. Choudhary, S., Sharma, K. & Silakari, O. The interplay between inflammatory pathways and COVID-19: A critical review on pathogenesis and therapeutic options. *Microb. Pathog.* **150**, 104673 (2021).
97. Catanzaro, M. *et al.* Immune response in COVID-19: addressing a pharmacological challenge by targeting pathways triggered by SARS-CoV-2. *Signal Transduct. Target. Ther.* **5**, (2020).
98. Zhou, Q., Mrowietz, U. & Rostami-Yazdi, M. Oxidative stress in the pathogenesis of psoriasis. *Free Radical Biology and Medicine* vol. 47 891–905 (2009).
99. *Recent Pat. Inflamm. Allergy Drug Discov.* **3**, 40–48 (2009).
100. Hariharan, A., Hakeem, A. R., Radhakrishnan, S., Reddy, M. S. & Rela, M. The Role and Therapeutic Potential of NF-kappa-B Pathway in Severe COVID-19 Patients. *Inflammopharmacology* vol. 29 (2021).
101. DeDiego, M. L. *et al.* Inhibition of NF- B-Mediated Inflammation in Severe Acute Respiratory Syndrome Coronavirus-Infected Mice Increases Survival. *J. Virol.* **88**, (2014).
102. Luo, W. *et al.* Targeting JAK-STAT Signaling to Control Cytokine Release Syndrome in COVID-19. *Trends Pharmacol. Sci.* **41**, 531–543 (2020).
103. Rojas, P. & Sarmiento, M. JAK/STAT Pathway Inhibition May Be a Promising Therapy for COVID-19-Related Hyperinflammation in Hematologic Patients. *Acta Haematol.* **144**, 1 (2021).
104. Pairo-Castineira, E. *et al.* Genetic mechanisms of critical illness in COVID-19. *Nat. 2020 5917848 591*, 92–98 (2020).
105. F, C. *et al.* Baricitinib therapy in COVID-19: A pilot study on safety and clinical impact. *J. Infect.* **81**, 318–356 (2020).
106. Satarker, S. et al. JAK-STAT Pathway Inhibition and their Implications in COVID-19 Therapy. *Postgrad. Med.* **133**, 1 (2020).

107. MG, W., M, O., MB, F. & RS, B. Severe acute respiratory syndrome coronavirus evades antiviral signaling: role of nsp1 and rational design of an attenuated strain. *J. Virol.* **81**, 11620–11633 (2007).

108. JY, L. et al. The ORF6, ORF8 and nucleocapsid proteins of SARS-CoV-2 inhibit type I interferon signaling pathway. *Virus Res.* **286**, (2020).

109. Tang, N., Li, D., Wang, X. & Sun, Z. Abnormal coagulation parameters are associated with poor prognosis in patients with novel coronavirus pneumonia. *J. Thromb. Haemost.* (2020) doi:10.1111/jth.14768.

110. Vinayagam, S. & Sattu, K. SARS-CoV-2 and coagulation disorders in different organs. *Life Sciences* (2020) doi:10.1016/j.lfs.2020.118431.

111. Biswas, S. et al. Blood clots in COVID-19 patients: Simplifying the curious mystery. *Med. Hypotheses* **146**, (2021).

112. Blasco, A. et al. Assessment of Neutrophil Extracellular Traps in Coronary Thrombus of a Case Series of Patients with COVID-19 and Myocardial Infarction. *JAMA Cardiol.* **6**, (2021).

113. Richards, C. D. The Enigmatic Cytokine Oncostatin M and Roles in Disease. *ISRN Inflamm.* 2013, (2013).

114. Hintzen, C., Haan, C., Tuckermann, J. P., Heinrich, P. C. & Hermanns, H. M. Oncostatin M-Induced and Constitutive Activation of the JAK2/STAT5/CIS Pathway Suppresses CCL1, but Not CCL7 and CCL8, Chemokine Expression. *J. Immunol.* **181**, (2008).

115. Setiadi, H. et al. Oncostatin M as a Biomarker to Predict the Outcome of V-V ECMO Supported Patients with Acute Pulmonary Failure. *J. Hear. Lung Transplant.* **39**, (2020).

116. Arunachalam, P. S. et al. Systems biological assessment of immunity to mild versus severe COVID-19 infection in humans. *Science* (80-.). **369**, 1210–1220 (2020).

117. Sanchez-Infantes, D. & Stephens, J. M. Adipocyte Oncostatin Receptor Regulates Adipose Tissue Homeostasis and Inflammation. *Frontiers in Immunology* vol. 11 (2021).

118. Michalakis, K. & Ilias, I. SARS-CoV-2 infection and obesity: Common inflammatory and metabolic aspects. *Diabetes Metab. Syndr. Clin. Res. Rev.* **14**, (2020).

119. Miller, R., Wentzel, A. R. & Richards, G. A. COVID-19: NAD + deficiency may predispose the aged, obese and type2 diabetics to mortality through its effect on SIRT1 activity. *Med. Hypotheses* **144**, (2020).

120. Philips, A. M. & Khan, N. Amino acid sensing pathway: A major check point in the pathogenesis of obesity and COVID-19. *Obes. Rev.* **22**, (2021).

121. Belančić, A., Kreso, A. & Rački, V. Potential pathophysiological mechanisms leading to increased COVID-19 susceptibility and severity in obesity. *Obesity Medicine* vol. 19 (2020).

122. Campbell, R. A., Boilard, E. & Rondina, M. T. Is there a role for the ACE2 receptor in SARS-CoV-2 interactions with platelets? *J. Thromb. Haemost.* **19**, (2021).
123. Rapkiewicz, A. V. et al. Megakaryocytes and platelet-fibrin thrombi characterize multi-organ thrombosis at autopsy in COVID-19: A case series. EClinicalMedicine 24, (2020).

124. Kato, A., Favoreto, S., Avila, P. C. & Schleimer, R. P. TLR3- and Th2 Cytokine-Dependent Production of Thymic Stromal Lymphopoietin in Human Airway Epithelial Cells. J. Immunol. 179, (2007).

125. Choreño-Parra, J. A. et al. Clinical and Immunological Factors That Distinguish COVID-19 From Pandemic Influenza A(H1N1). Front. Immunol. 12, (2021).

126. Caterino, M. et al. Dysregulation of lipid metabolism and pathological inflammation in patients with COVID-19. Sci. Rep. 11, (2021).

127. Kelley, N., Jeltema, D., Duan, Y. & He, Y. The NLRP3 Inflammasome: An Overview of Mechanisms of Activation and Regulation. Int. J. Mol. Sci. 20, (2019).

128. Vora, S. M., Lieberman, J. & Wu, H. Inflammasome activation at the crux. Nat. Rev. Immunol. doi:10.1038/s41577-021-00588-x.

129. Lee, S., Channappanavar, R. & Kanneganti, T.-D. Coronaviruses: Innate Immunity, Inflammasome Activation, Inflammatory Cell Death, and Cytokines. Trends Immunol. 41, 1083–1099 (2020).

130. Shi, C.-S., Nabar, N. R., Huang, N.-N. & Kehrl, J. H. SARS-Coronavirus Open Reading Frame-8b triggers intracellular stress pathways and activates NLRP3 inflammasomes. Cell Death Discov. 2019 51 5, 1–12 (2019).

131. Rodrigues, T. S. et al. Inflammasome activation in COVID-19 patients. medRxiv 2020.08.05.20168872 (2020) doi:10.1101/2020.08.05.20168872.

132. Vidal, M., Cusick, M. E. & Barabási, A. L. Interactome networks and human disease. Cell (2011) doi:10.1016/j.cell.2011.02.016.

133. Pan, A., Lahiri, C., Rajendiran, A. & Shanmugham, B. Computational analysis of protein interaction networks for infectious diseases. Brief. Bioinform. (2016) doi:10.1093/bib/bbv059.

134. Kumar, N., Mishra, B., Mehmood, A., Mohammad Athar & M Shahid Mukhtar. Integrative Network Biology Framework Elucidates Molecular Mechanisms of SARS-CoV-2 Pathogenesis. iScience (2020) doi:10.1016/j.isci.2020.101526.

135. Saini, K. S. et al. Repurposing anticancer drugs for COVID-19-induced inflammation, immune dysfunction, and coagulopathy. British Journal of Cancer (2020) doi:10.1038/s41416-020-0948-x.

136. Lee, S. J., Channappanavar, R. & Kanneganti, T. D. Coronaviruses: Innate Immunity, Inflammasome Activation, Inflammatory Cell Death, and Cytokines. Trends Immunol. 41, 1083–1099 (2020).

137. Li, S. et al. SARS-CoV-2 triggers inflammatory responses and cell death through caspase-8 activation. Signal Transduct. Target. Ther. 5, (2020).

138. Ackermann, M., Mentzer, S. J., Kolb, M. & Jonigk, D. Inflammation and intussusceptive angiogenesis in COVID-19: Everything in and out of flow. Eur. Respir. J. 56, (2020).

139. Ardestani, A. & Azizi, Z. Targeting glucose metabolism for treatment of COVID-19. Signal Transduct. Target. Ther. 6, 1–2 (2021).
140. Amaral, M. P. & Bortoluci, K. R. Caspase-8 and FADD: Where Cell Death and Inflammation Collide. *Immunity* **52**, (2020).

141. Codo, A. C. *et al.* Elevated Glucose Levels Favor SARS-CoV-2 Infection and Monocyte Response through a HIF-1α/Glycolysis-Dependent Axis. *Cell Metab.* **32**, (2020).

142. Ortiz-Prado, E., Dunn, J. F., Vasconez, J., Castillo, D. & Visor, G. Partial pressure of oxygen in the human body: a general review. *Am. J. Blood Res.* (2019).

143. Price, L. C., McCabe, C., Garfield, B. & Wort, S. J. Thrombosis and COVID-19 pneumonia: The clot thickens! *European Respiratory Journal* vol. 56 (2020).

144. Huertas, A. *et al.* Endothelial cell dysfunction: A major player in SARS-CoV-2 infection (COVID-19)? *European Respiratory Journal* vol. 56 (2020).

145. Walz, L. *et al.* JAK-inhibitor and type I interferon ability to produce favorable clinical outcomes in COVID-19 patients: a systematic review and meta-analysis. *BMC Infect. Dis.* **21**, (2021).

146. Yan, B. *et al.* SARS-CoV-2 drives JAK1/2-dependent local complement hyperactivation. *Sci. Immunol.* **6**, 1–20 (2021).

147. Nct. Phase 3 Randomized, Double-blind, Placebo-controlled Multi-center Study to Assess the Efficacy and Safety of Ruxolitinib in Patients With COVID-19 Associated Cytokine Storm (RUXCOVID). https://clinicaltrials.gov/show/NCT04362137 (2020).

148. Fridman, J. S. *et al.* Selective Inhibition of JAK1 and JAK2 Is Efficacious in Rodent Models of Arthritis: Preclinical Characterization of INCB028050. *J. Immunol.* **184**, (2010).

149. Cantini, F. *et al.* Immune Therapy, or Antiviral Therapy, or Both for COVID-19: A Systematic Review. *Drugs* vol. 80 (2020).

150. COVID-19 Treatment Guidelines Panel. Coronavirus Disease 2019 (COVID-19) Treatment Guidelines. Disponible en: https://covid19treatmentguidelines.nih.gov/. *Natl. Inst. Heal.* 2019, (2020).

151. Marconi, V. C. *et al.* Baricitinib plus Standard of Care for Hospitalized Adults with COVID-19. *medRxiv* (2021).

152. William, A. D. *et al.* Discovery of the macrocycle 11-(2-pyrrolidin-1-yl-ethoxy)-14,19-dioxo-5,7, 26-triaza-tetracyclo[19.3.1.1(2,6).1(8,12)]heptacosa-1(25),2(26),3,5,8,10,12(27),16,21,23-decaene (SB1518), a potent Janus Kinase 2/Fms-like tyrosine kinase-3 (JAK2/FLT3) inhibitor for the treatment of myelofibrosis and lymphoma. *J. Med. Chem.* **54**, (2011).

153. Willette, R. N. *et al.* Differential effects of p38 mitogen-activated protein kinase and cyclooxygenase 2 inhibitors in a model of cardiovascular disease. *J. Pharmacol. Exp. Ther.* **330**, (2009).

154. Shirey, K. A., Blanco, J. C. G. & Vogel, S. N. Targeting TLR4 Signaling to Blunt Viral-Mediated Acute Lung Injury. *Frontiers in Immunology* vol. 12 (2021).

155. Park, J. J. H. *et al.* How COVID-19 has fundamentally changed clinical research in global health. *The Lancet Global Health* vol. 9 (2021).

156. Global coalition to accelerate COVID-19 clinical research in resource-limited settings. *The Lancet* vol. 395 (2020).
157. WHO. *WHO Coronavirus (COVID-19) Dashboard*. *World Health Organisation* (2021).

158. Scoggins, J. F. & Ramsey, S. D. A National cancer clinical trials system for the 21st century: Reinvigorating the NCI cooperative group program. *Journal of the National Cancer Institute* vol. 102 (2010).

159. Altay, O. et al. Current Status of COVID-19 Therapies and Drug Repositioning Applications. *iScience* 23, 101303 (2020).

**Figures**
Figure 1

Significantly expressed genes across lung cell types. Box plots show the number of significantly expressed genes, their Z-scores, and p-values per each lung cell type. Neural cells were the cell type with the highest mean Z-score, followed by B cells, mast cells, fibroblast cells, alveolar type II cells, cycling natural killer / T cells, endothelial cells, macrophages, airway epithelial cells, alveolar type I cells, natural

CD8+ T cells
CD4+ T cells
Other epithelial cells
Monocytes
Plasma cells
Treg cells
Smooth cells
Dendritic cells
Natural killer cells
Alveolar type I cells
Airway epithelial cells
Macrophages
Endothelial cells
Cycling natural killer / T cells
Alveolar type II cells
Fibroblast cells
Mast cells
B cells
Neuronal cells

Figure 1

Significantly expressed genes across lung cell types. Box plots show the number of significantly expressed genes, their Z-scores, and p-values per each lung cell type. Neural cells were the cell type with the highest mean Z-score, followed by B cells, mast cells, fibroblast cells, alveolar type II cells, cycling natural killer / T cells, endothelial cells, macrophages, airway epithelial cells, alveolar type I cells, natural
killer cells, dendritic cells, smooth cells, Treg cells, plasma cells, monocytes, other epithelial cells, CD4+ T cells, and CD8+ T cells.

Figure 2

Transcriptomics data of 116,313 lung nuclei from lethal COVID-19 patients. UMAPs show the mean log normalized expression of significantly expressed genes per lung cell type. Dot plots show the ranking of genes with the highest percentage of cells expressing. UMAP: uniform manifold approximation and
projection for dimension reduction; NK: natural killer; OG: overexpressed genes; and UG: underexpressed genes.

Figure 3

Functional enrichment analysis. UMAPs show the most significant genes per lung cell type involved in gene ontology biological processes and signaling pathways. The most significant biological term was inflammatory response, followed by cytokine production, innate immune response, macrophage
activation, Toll-like receptor signaling pathway, interferon production, JAK-STAT signaling pathway, NF-κB signaling pathway, thymic stromal lymphopoietin, TNF signaling pathway, blood coagulation, oncostatin M signaling pathway, AGE-RAGE signaling pathway, IL-1 and megakaryocytes in obesity, and NLRP3 inflammasome complex. UMAP: uniform manifold approximation and projection for dimension reduction.

Figure 4

miRNome enrichment analysis. Circos plot show a GSEA to compute overlaps between the 18 most significant miRNAs (FDR q-value < 0.01) and the 19 significantly expressed genes in > 50% of lung cells.
The most significant miRNA was MIR6867_5P, followed by MIR2662, MIR32_3P, MIR548A0_5P_MIR548AX, MIR570_3P, MIR338_5P, MIR144_3P, MIR4711_3P, MIR628_5P, MIR548AJ_3P_MIR548X_3P, MIR1290, MIR4496, MIR12136, MIR9718, MIR875_3P, MIR4698, MIR3941, and MIR4789_3P. GSEA: gene set enrichment analysis.
Inflammatory protein-protein interactome network. iPPI network made up of 265 nodes and 2052 edges. Of them, 159 pulmonary inflammatory response proteins had a mean of degree centrality of 8 and 108 human-SARS-CoV-2 proteins had a mean of degree centrality of 7.2. The top ten inflammatory response proteins with the highest degree centrality were APP, NFKB1, STAT3, C3, ITGAM, FN1, PTAFR, JAK2, EGFR, and LYN. The top ten human-SARS-CoV-2 proteins with the highest degree centrality were GNB1, GNG5, RHOA, ITGB1, STOM, RAB14, PRKAR2B, RAB8A, PRKACA, and ANO6. Additionally, 111 pulmonary inflammatory response proteins had the highest confidence interactions (cutoff = 0.9) with human-SARS-CoV-2 proteins, being the top ten: C3, FN1, NFKB1, RPS19, CTSC, HSPD1, APP, ITGAM, SNAP23, and MAPK14.
Figure 6

Shortest paths to cancer hallmark phenotypes. A) Box plots encompassing inflammatory response proteins with the shortest mean of distance score per phenotype. Cell death was the phenotype with the shortest paths, followed by inflammation, glycolysis, and angiogenesis. B) Venn diagram of inflammatory response proteins with shortest paths to hallmarks of cancer related to COVID-19. C)
Ranking of the most essential proteins with shortest paths to cell death, inflammation, glycolysis, and angiogenesis.

Figure 7

Essential proteins with the shortest distance score to the inflammation phenotype. The essential proteins with positive regulation to inflammation were PTGS2, PRKCZ, NFKBIA, MAPK14, TNFRSF1B, TLR4, ATM,
MECOM, PIK3CG, EGFR, JAK2, LYN, CYLD, PRKCQ, STAT3, TGFB1, RBPJ, TNFAIP3, NOTCH1, IGF1, CD28, CCL5, PTAFR, FPR1, EDNRA, EDNRB, CYSLTR1, CNR2, HGF, EPHA2, FN1, CSF1, PTGFR, and APP.

**Figure 8**

Drugs involved in advanced-stage COVID-19 clinical trials. Drug name, class type, druggable target, structure, pharmacological indication, and clinical trial number related to small molecules involved in advanced-stage COVID-19 clinical trials.

**Supplementary Files**

This is a list of supplementary files associated with this preprint. Click to download.

- [SupplementaryDataset.xlsx](#)