Rassegna di Patologia dell'Apparato Respiratorio **2020**;35:10-18
doi: 10.36166/2531-4920-A004

Commentario

Pathogenesis of coronavirus disease 2019 (COVID-19)

Patogenesi della malattia da coronavirus 2019 (COVID-19)

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In December 2019 a cluster of fatal interstitial pneumonias caused by a previously unknown coronavirus was identified in Wuhan, China. Now a pandemic is a great alarm because of the mortality rate – significantly higher in males, elderly and fragile subjects – and because of the overwhelming need of intensive care unit (ICU) beds to treat patients. This virus, whose genome was soon after identified, is now named SARS-CoV-2 because it has 75,9% genomic sequence identity to the SARS-CoV that caused an epidemic in 2002-2003, and the disease, a zoonosis, is called COVID-2019. The causative virus has replication modalities that are similar to the SARS-CoV virus and it uses angiotensin converting enzyme 2 (ACE2) (a protein that has a pivotal role in hindering the appearance of acute lung injury) and TMPRSS2 (a cytoplasmatic protease) as receptors to enter into human cells. These receptors are expressed in different human tissues but – for the "COVID-19" pathogenetic point of view – the most important cells are type I and type II pneumocytes, endothelial cells, enteric cells, cells of the nasal mucosa and probably the olfactory nervous cells. The importance of viral load and genetic predispositions in the pathogenesis of COVID-19 has not been explored yet but it is intuitive that they should be important.

Pathology of COVID-19 is still not yet well known. In lungs alveolar edema mainly made by granular proteinaceous material, diffuse alveolar damage, acute fibrinous and organizing pneumonia (AFOP), organizing pneumonia, infiltration of macrophages and lymphocytes, micro-thrombi and thrombi in the pulmonary arteries have been reported. Thrombosis in the vessels outside the lungs, liver steatosis and inflammation, myocardial inflammatory infiltrates made mainly by lymphocytes, enlargement of lymph nodes and spleen are also part of the pathologic spectrum (liver steatosis, lymph nodes enlargement and splenomegaly , thrombosis are easily documented by CT scan). Quite recently a case of hemorrhagic, necrotizing encephalitis has also been reported.

Laboratory profile is characterized by an increase of "acute phase" inflammatory proteins such as Reactive C Protein (RCP), of LDH, D-dimer, ferritin, interleukin 6 and in a significant number of cases by lymphopenia.

Emerging evidence suggests that some patients may respond to SARS-

Ricevuto il 5-4-2020
Accettato il 22-4-2020

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Conflitto di interessi
Gli autori dichiarano di non avere nessun conflitto di interesse con l'argomento trattato nell'articolo.

Come citare questo articolo: Poletti V, Ravaglia C, Tardella M, et al. Pathogenesis of coronavirus disease 2019 (COVID-19). Rassegna di Patologia dell'Apparato Respiratorio 2020;35:10-18. https://doi.org/10.36166/2531-4920-A004

CoV-2 with an exuberant "cytokine storm" reaction with a clinical and laboratory scenario similar to that observed during bacterial sepsis, macrophage activation syndrome (MAS) or chimeric antigen receptor T (CAR-T) related cytokine release.
Therefore the pathogenesis of the disease may be probably divided in two phases: the first one in which replication of viruses (with viremia) and cytopathic damage related to this proliferation are prominent (this phase appears at the beginning and innate immunity is part of the game and in fragile patients may lead to death) and a second phase (mainly related to the appearance of adaptive immunity) in which a "cytokine storm" – and organs damage related to it – predominate.
In the second phase cytokines such as interleukin 1 (IL-1), Interleukin-6 and, endothelial damage with a procoagulant balance seem to have a pivotal role. The problem of fibrotic sequelae is appearing.
Treatment strategies should be tailored according to the pathogenesis above summarized and probably in the near future also anti-fibrotic drugs could have a role.
All these concepts have been reported in the following comic strips. References are reported in the strips.

Nel dicembre 2019 è stato identificato a Whuan, in Cina, un cluster di polmoniti interstiziali fatali causate da un coronavirus fino ad allora sconosciuto. In questo momento una pandemia dovuta a questo virus rappresenta un grande problema sanitario ed economico. Questo virus è stato recentemente denominato SARS-CoV-2 poiché ha un genoma identico per il 75% circa con quello del SARS-CoV, coronavirus che causò un'epidemia nel 2002-2003 e la malattia dovuta al SARS-CoV-2, una zoonosi, è stata chiamata COVID-2019. Il virus causale ha modalità di replicazione simili al virus SARS-CoV e utilizza l'enzima di conversione dell'angiotensina 2 (ACE2) (la proteina che ha un ruolo fondamentale nell'ostacolare la comparsa di lesioni polmonari acute) e TMPRSS2 (una proteasi citoplasmatica della cellula ospite) come recettori per entrare nelle cellule umane. Questi recettori sono espressi in diversi tessuti umani ma – dal punto di vista patogenetico "COVID-19" – le cellule più importanti sono gli pneumociti di tipo I e di tipo II, le cellule endoteliali, le cellule epiteliali enteriche, le cellule della mucosa nasale e probabilmente le cellule olfattive. Il ruolo patogenetico della carica virale e della predisposizione genetica non è stato ancora ben definito anche se appare intuitivo che entrambi possano avere un peso significativo.
La patologia COVID-19 non è ancora ben nota. Si ha edema alveolare polmonare, costituito principalmente da materiale proteinaceo granuloso, aspetti di danno alveolare diffuso, polmonite fibrinosa acuta con organizzazione (AFOP), polmonite organizzativa, infiltrazione di macrofagi e linfociti, micro-trombi e trombi nelle arterie polmonari. Fanno inoltre parte dello spettro patologico trombosi dei vasi al di fuori dei polmoni, steatosi e infiammazione epatica, infiltrati infiammatori del miocardio determinati principalmente da linfociti, ingrossamento dei linfonodi e della milza (steatosi epatica, ingrossamento dei linfonodi e splenomegalia sono facilmente documentabili mediante TC). Molto recentemente è stato riportato anche un caso di encefalite emorragica e necrotizzante.
Il profilo laboratoristico è caratterizzato da un aumento delle proteine infiammatorie nella "fase acuta" come proteina C-reattiva (RCP), lattato deidrogenasi (LDH), D-dimero, ferritina, interleuchina 6 e, in un numero significativo di casi, da linfopenia.
Recenti evidenze suggeriscono che alcuni pazienti possano rispondere al SARS-CoV-2 con una esuberante "tempesta citochinica" con uno scenario clinico e di laboratorio simile a quello osservato durante la sepsi batterica, la sindrome da attivazione macrofagica o da rilascio di citochine in corso di terapia con CAR-T.
Pertanto la patogenesi della malattia può essere probabilmente divisa in due fasi: la prima in cui sono predominanti la replicazione dei virus (con viremia) e il danno citopatico correlato a questa proliferazione (questa fase appare all'inizio e l'immunità innata è parte fondamentale del gioco) e una seconda fase (principalmente correlata alla comparsa di immunità adattativa) in cui prevale una "tempesta citochinica" – e danni agli organi ad essa correlati.
Nella seconda fase sembrano avere un ruolo fondamentale le citochine come l'interleuchina 1 (IL-1), l'interleuchina 6 (IL-6) e il danno endoteliale con significativo aumento del D-dimero e trombofilia. Il problema delle sequele fibrotiche sta iniziando ad emergere.
Le strategie terapeutiche devono essere personalizzate in base alla patogenesi sopra riassunta e probabilmente anche i farmaci antifibrotici potranno avere un ruolo.
Tutti questi concetti sono stati riportati nel seguente fumetto. I riferimenti bibliografici sono riportati nelle strisce.

PATHOGENESIS OF COVID-19

- Virology (classification; replication; invasion of human cells; viral transmission; role of cellular receptors)
- Pathology of COVID-19: clues for the understanding of COVID-19
- Laboratory markers (suggesting activation of a «cytokine storm»)
- «Two steps» clinical (and pathogenetic) profile
- Radiologic features sustaining the «two steps» profile
- IL-6 (and other cytokines)
- Animal model and IL-6
- D-dimer and thrombophilia
- The problem of «fibrotic sequelae»
- Treatment strategies based on pathogenetic considerations

CORONAVIRUSES ARE CLASSIFIED AS A FAMILY WITHIN THE *NIDOVIRALES* ORDER

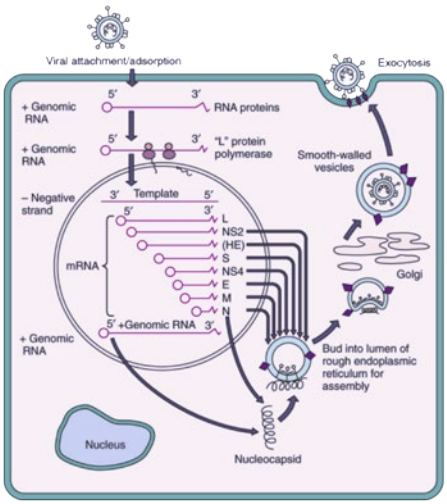
THE CORONAVIRUS SUBFAMILY IS FURTHER CLASSIFIED INTO FOUR GENERA: ALPHA, BETA, GAMMA AND DELTA CORONAVIRUSES

THE HUMAN CORONAVIRUSES ARE IN TWO OF THESE GENERA:
ALPHA CORONAVIRUSES (HCoV-229 and HCoV-NL63)
BETA CORONAVIRUSES (HCoV-HKU1, HCoV-OC43, MERS, SARS-CoV, SARS-CoV-2)

SARS-CoV-2 shares 79.5% sequence identity to SARS-CoV and 96.2% overall genome sequence identity to RaTG13, which is a short RdRp region from a bat coronavirus.

Population genetic analyses of 103 genomes of SARS-CoV-2 indicates that there are two major types of viruses (designated L and S), currently circulating between humans. The type L is predominant (70%).

He F, et al. *J Med Virol* 2020. *In press*
Tang K. *Nat Sci Rev* 2020. *In press*



(modified from Murray P, Rosenthal K, Pfaller M. *Medical Microbiology 19° Ed*, 2020)

VIRAL REPLICATION

Translation of genome occurs in two phases:
(1) the early phase produces an RNA polymerase (L), and
(2) the late phase, from a negative-sense RNA template, yields structural and nonstructural proteins.

Virus assembles at the rough endoplasmic reticulum.

Non-structural proteins
3-chymotrypsin-like protease
Papain-like protease
Helicase
RNA-dependent RNA polymerase

Structural proteins
Envelope protein
Membrane protein
Spike protein
Nucleocapside protein

TRANSMISSION

Person-to-person spread of SARS-CoV-2 is thought to occur mainly via respiratory droplets, resembling the spread of influenza. With droplet transmission, virus released in the respiratory secretions when a person with infection coughs, sneezes, or talks can infect another person if it makes direct contact with the mucous membranes.

Infection can also occur if a person touches an infected surface and then touches his or her eyes, nose, or mouth.

Droplets typically do not travel more than six feet (about two meters) and do not linger in the air; however, in one letter to the editor, SARS-CoV-2 remained viable in aerosols under experimental conditions for at least three hours.

Viral RNA levels appear to be higher soon after symptom onset compared with later in the illness.

Transmission of SARS-CoV-2 from asymptomatic individuals. *«Substantial undocumented infection facilitates the rapid dissemination of SARS-CoV 19 (Science 2020)»*

Unlike SARS and MERS, patients diagnosed as COVID-19 have presented with high viral loads even those who have no fever or mild symptoms.»*Holshue ML, DeBolt C, Lindquist S, et al. First Case of 2019 Novel Coronavirus in the United States. N Engl J Med. 2020»*

R0= 2.5/>4
R-naught is the virus's basic reproductive number
He X, et al. Temporal dynamics in viral shedding and transmission of COVID-19. *Nature Med* 2020. *In press.*

Viral load in feces in children
The oro-fecal modality of transmission could be important mainly in children

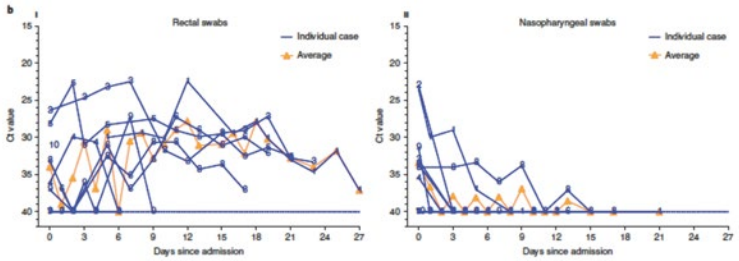


Fig. 1. Chronology of major epidemiological events and molecular testing results of n = 10 independent pediatric patients confirmed with SARS-CoV-2 infection. Chronological changes in Ct values of *Orflab* and *N* genes using real-time RT-PCR after hospital admission. The Ct values of *Orflab* and *N* genes on real-time RT-PCR detected in rectal swabs obtained from *n* = 10 independent patients (i) and Ct values in nasopharyngeal swabs from *n* = 10 independent cases (ii). The Ct value is supposed to be inversely related to viral RNA copy numbers and a value of 40 means the virus is molecularly undetectable.

(modified from Xu Y, Li X, Zhu B, et al. *Nat Med* 2020;26:502-5.)
Zheng S, et al. *BMJ* 2020. *In press.*

ACE2, A TRANSMEMBRANE PROTEIN IS THE RECPETOR FOR SARS-CoV AND SARS-CoV-2

The biophysical and structural analysis indicated that S protein of SARS-CoV-2 binds ACE2 with approximately10- to 20- fold higher affinity than S protein of SARS-CoV[24]. The high affinity of S protein for human ACE2 may facilitate the spread of SARS-CoV-2 in human populations. Meanwhile, SARS-CoV-2 does not use other coronavirus receptors, such as aminopeptidase N and dipeptidyl peptidase 4 (DPP4) to enter cells[3].

He F, et al. *J Med Virol* 2020. *In press*
Sungnak W, Huang N, Bécavin, C, et al. SARS-CoV-2 entry factors are highly expressed in nasal epithelial cells together with innate immune genes. *Nat Med* 2020. *In press.*

(Yan R, Zhang Y, Li Y, et al. *Science* 2020;367:1444-8.)

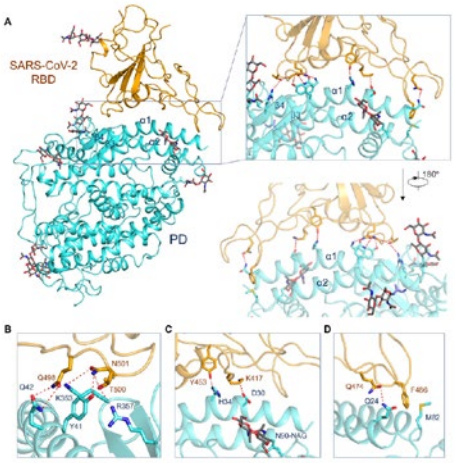
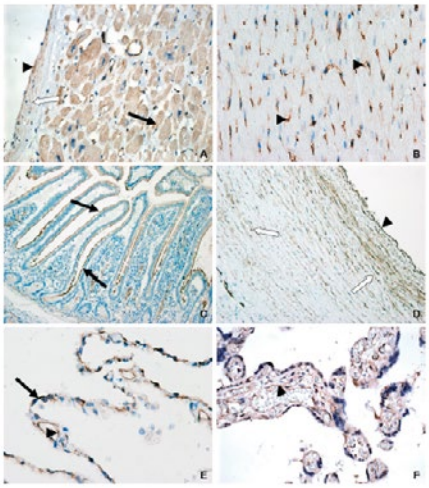


Fig. 4. Interactions between SARS-CoV-2-RBD and ACE2. (A) The protease domain (PD) of ACE2 mainly engages the α1 helix in the recognition of the RBD. The α2 helix and the linker between β3 and β4 also contribute to the interaction. Only one RBD-ACE2 is shown. (B to D) Detailed analysis of the interface between SARS-CoV-2-RBD and ACE2. Polar interactions are indicated by red, dashed lines.

Hikmet F, et al.
The protein expression of ACE2 in human tissues. *In press*

ACE2 enzyme is expressed in numerous humans tissues (also in nasal mucosa)

PROBABLY THE EXPRESSION OF THESE RECEPTORS IN HUMAN TISSUES VARIES BETWEEN PERSONS AND THESE COULD EXPLAIN THE CLINICAL VARIABILITY OF COVID-19



(Hamming I, Cooper ME, Haagmans BL, et al. *J Pathol* 2007;212:1-11.)

Figure 3. Immunohistochemical staining pattern of ACE2 in several organs. In the healthy human heart (A), ACE2 is expressed in cardiomyocytes (closed arrow), vascular endothelium (arrowhead), and smooth muscle cells (open arrow). In rat heart (B), ACE2 is predominantly expressed in vascular endothelium (arrowhead). (C) ACE2 expression in human small intestine (jejunum); abundant staining can be found in the brush border of enterocytes (arrow). In human aorta (D), ACE2 is expressed in the endothelium (arrowhead) and vascular smooth muscle cells (open arrow). In healthy human lung (E), ACE2 is present in alveolar epithelial cells (arrow) and in capillary endothelial cells (arrowhead). In human placenta (F), positive staining for ACE2 is found in the placental villi [syncytiotrophoblast, cytotrophoblast, vascular endothelium (arrowhead), and smooth muscle cells]

Loss of ACE2 expression precipitates severe acute lung failure

Injection of recombinant human ACE2 attenuates acute lung failure in ACE2 KO as well as wild-type mice

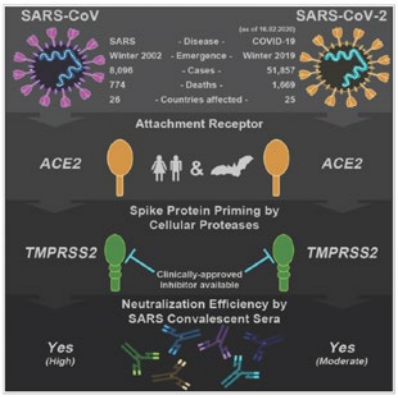
Increased ACE2 may play a role in reducing the initial leakage over the alveolar capillary interface

ACE2 exhibits protective effects against LPS-induced acute lung injury

Exp Mol Pathol 2020

Graphical Abstract

A Human protease
(TMPRSS2) is important
for viral invasion of human cells



(modified from Hoffmann M, Kleiner-Weber H, Schroeder S, et al. *Cell* 2020 Mar 4. doi: 10.1016/j.cell.2020.02.052. [Epub ahead of print]

PATHOLOGY OF COVID-19

- LUNG (DAD, alveolar proteinaceous, granular edema; diffuse alveolar damage with cytopathic changes in type II pneumocytes; acute fibrinous organizing pneumonia; organizing pneumonia, microthrombi, platelets accumulation, infiltration of macrophages and-at least in biopsies-lymphocytes)

- OTHER ORGANS
  - Spleen (enlarged in CT scan)
  - Kidney
  - Heart (myocarditis, pericardial effusion)
  - Liver (steatosis,inflammation)
  - Lymph nodes (enlarged in the mediastinal/hilar stations)
  - Vessels (thrombi-microthrombi; vasculitis?)
  - Central Nervous System (CNS): acute hemorrhagic necrotizing encephalopathy
  - Skin (varicella-like exantem)

MICRO (and macro) VASCULAR DAMAGE SEEMS TO BE PRESENT IN DIFFERENT SITES
Varga Z, et al. *Lancet* 2020, Apr 17.
Rockx B, et al. Comparative pathogenesis of COVID-19, MERS, and SARS in a nonhuman primate model. *Science* 2020. *In press.*
Marzano AV, et al. *J Am Acc Dermatol* 2020. *In press.*

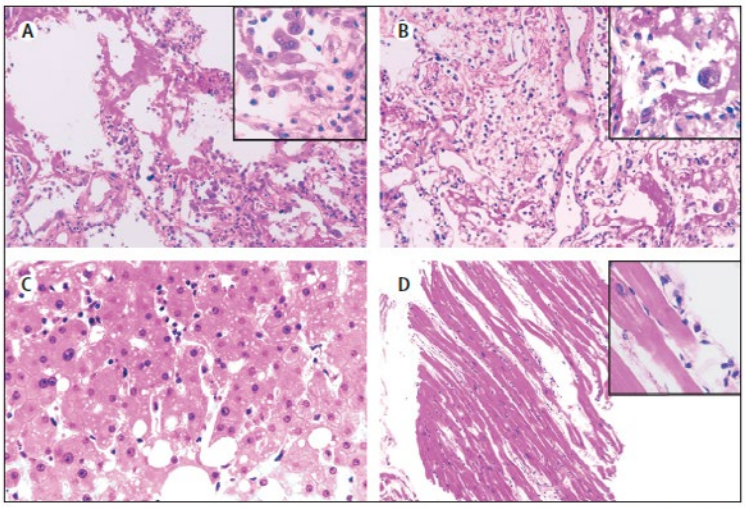
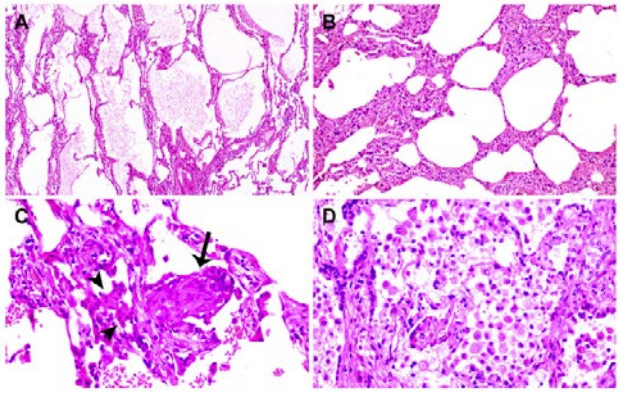


Figure 2: **Pathological manifestations of right (A) and left (B) lung tissue, liver tissue (C), and heart tissue (D) in a patient with severe pneumonia caused by SARS-CoV-2**
SARS-CoV-2=severe acute respiratory syndrome coronavirus 2.

LRM, in press

Biopsy samples were taken from lung, liver, and heart tissue of the patient. Histological examination showed bilateral diffuse alveolar damage with cellular fibromyxoid exudates (figure 2A, B). The right lung showed evident desquamation of pneumocytes and hyaline membrane formation, indicating acute respiratory distress syndrome (ARDS; figure 2A). The left lung tissue displayed pulmonary oedema with hyaline membrane formation, suggestive of early-phase ARDS (figure 2B). Interstitial mononuclear inflammatory infiltrates, dominated by lymphocytes, were seen in both lungs. Multinucleated syncytial cells with atypical enlarged pneumocytes

(Xu Z, Shi L, Wang Y, et al. *Lancet Respir Med* 2020;8:420-2.)



(Xu Z, Shi L, Wang Y, et al. *Lancet Respir Med* 2020;8:420-2.)

Figure 3. Histologic changes of coronavirus disease 2019 pneumonia in case 2. (*A*) Evident proteinaceous and fibrin exudate; (*B*) diffuse expansion of alveolar walls and septa owing to fibroblastic proliferations and type II pneumocyte hyperplasia, consistent with early diffuse alveolar damage pattern; (*C*) plugs of proliferating fibroblasts or "fibroblast balls" in the interstitium (arrow); (*D*) abundant macrophages infiltrating airspaces and type II pneumocyte hyperplasia.

LAB PROFILE (and PROGNOSTIC MARKERS)

- Platelet count
- Lymphocytes
- LDH
- Troponin (high sensitive cardiac)
- CPK
- D-dimer
- Serum ferritin
- Serum IL-6

Zhou T, et al. *Lancet* 2020

diac injury [6]. Significantly high blood levels of cytokines and chemokines were noted in patients with COVID-19 infection that included IL1-β, IL1RA, IL7, IL8, IL9, IL10, basic FGF2, GCSF, GMCSF, IFNγ, IP10, MCP1, MIP1α, MIP1β, PDGFB, TNFα, and VEGFA. Some of the severe cases that were admitted to the intensive care unit showed high levels of pro-inflammatory cytokines including IL2, IL7, IL10, GCSF, IP10, MCP1, MIP1α, and TNFα that are reasoned to promote disease severity [6]. Other factors: IL-37, CCL2,…pro-fibrotic proteins,……..

Activation of NLRP-3 inflammasome

EMERGING EVIDENCE SUGGESTS THAT SOME PATIENTS MAY RESPOND TO SARS-COV-2 WITH AN EXUBERANT «CYTOKINE STORM» REACTION
(bacterial sepsis like scenario; hemophagocytic syndrome; T-CAR related cytokine release syndrome, «TAFRO» like syndrome)

Swanson KV, et al. *Nature Rev Immunol* 2019
Hirano T, Murakami M. COVID-19: a new virus, but a familiar receptor and cytokine release syndrome. *Immunity* 2020. *In press.*
Giamarellos-Bourboulis EJ, Netea MG, Rovina N, et al. Complex immune dysregulation in COVID-19 patients with severe respiratory failure. *Cell Host Microbe* 2020;27:1-9.

COVID-19: CLINICAL PROFILE

*Fourteen percent of confirmed cases have been "severe," involving serious pneumonia and shortness of breath
*Another 5 percent of patients confirmed to have the disease developed respiratory failure, septic shock, and/or multi-organ failure – what the agency calls "critical cases" potentially resulting in death.
*Roughly 2.3 percent of confirmed cases did result in death
*A «two step» clinical profile observed in at least a subset of pts

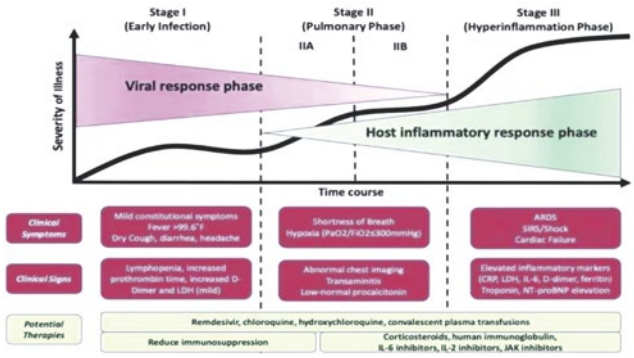
STAGES OF ILLNESS

***REPLICATIVE STAGE.** VIRAL REPLICATION OCCURS OVER A PERIOD OF SEVERAL DAYS.
AN INNATE RESPONSE OCCURS. RELATIVELY MILD SYMPTOMS MAY BE PRESENT DUE TO DIRECT VIRAL CYTOPATHIC EFFECTS BUT THIS RESPONSE MAY FAIL TO CONTAIN THE VIRUS.

***ADAPTIVE IMMUNITY STAGE**. AN ADAPTIVE IMMUNE RESPONSE EVENTUALLY KICKS INTO GEAR.
THIS LEADS TO FALLING TITERS OF VIRUS AND MAY LEAD TO RECOVERY. HOWEVER (IN A LARGE MINORITY OF CASES) IT MAY ALSO INCREASE LEVELS OF INFLAMMATORY CYTOKINES AND DETERMINATE TISSUE DAMAGE, CAUSING CLINICAL DETERIORATION.

Moore JB, June CH. Cytokine release syndrome in severe COVID-19. *Science* 2020. *In press.*

THESE TWO STEPS/STAGES OVERLAP AFTER 6-7 DAYS OF SYMPTOMS



(modified from Siddiqi HK, Mehra MR. *J Heart Lung Transplant* 2020 Mar 20. doi: 10.1016/j.healun.2020.03.012. [Epub ahead of print]

A Two-Step clinical (and biological) profile is also suggested by CT Scan findings in asymptomaic and symptomatic patients

112 Pts included in the study

- ❖ 82 (73%) Asymptomatic — 44 (54%) had lung opacities on CT
- ❖ 30 (27%) Symptomatic — 24 (80%) had lung opacities on CT

- ✓ Asymptomatic Pts showed more GGO over consolidation (80%)
- ✓ Symptomatic cases more frequently showed consolidation over GGO (38%)

The CT severity score was higher in symptomatic cases than asymptomatic pts, particularly in the lower lobes

Inui S, et al. Chest CT findings in cases from the Cruise Ship «Diamond Princess» with coronavirus disease 2019 (COVID-19). *Radiology* 2020

A TWO STEP CLINICAL PROFILE IS ALSO SUGGESTED BY CT SCAN CHANGES OVER TIME

*TWO TYPES OF HRCT PATTERN?

- **ACUTE LUNG INJURY/DAD** (ground glass attenuation, «crazy paving» pattern
In the early phase in which viral replication predominates

- **ORGANIZING PNEUMONIA/PERILOBULAR PATTERN** (alveolar consolidation, archiform lines)
when the «cytokine storm» phase starts

***Pulmonary thrombosis may be documented in patients with high D-dimer levels**

Saleshi S, et al. *AJR Am J Roentgenol* 2020. *In press.*

INTERLEUKIN 6 (and other cytokines including IL-1) HAVE A PATHOGENETIC ROLE AND MAY BE A TARGET FOR SPECIFIC DRUGS

IL-6 is produced by many cell types. Analogous to IL-1, IL-6 may act at the systemic level like a hormone and takes part in the «acute phase» inflammation and in the maintanance of the blood-brain barrier. IL-6 may also be secreted by macrophages in response to «patterns» to lead to the toxic shock syndrome.

Perricone C, et al. *J Autoimmun* 2020. *In press.*

AN EXPERIMENTAL MODEL OF VIRAL INFECTION DOCUMENTING THE PATHOGENETIC ROLE OF IL-6

Published in final edited form as:
J Immunol. 2019 February 01; 202(3): 871–882. doi:10.4049/jimmunol.1800927.

Critical adverse impact of interleukin-6 in acute pneumovirus infection*

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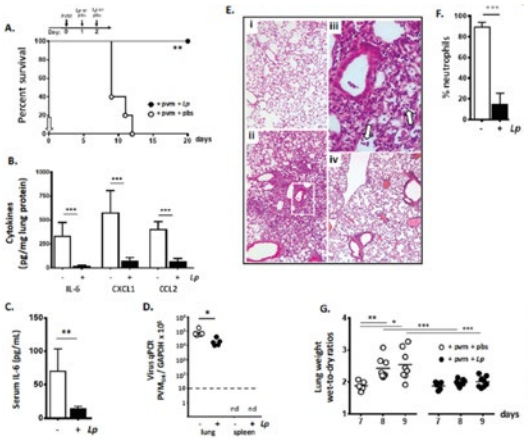

Fig. 1. *Lactobacillus plantarum* at the respiratory tract results in profound suppression of local inflammation.

A. Survival of wild-type mice inoculated intranasally with pneumonia virus of mice (PVM) at day 0 followed by intranasal inoculation with *L. plantarum* (*Lp*; 10^5 cells in 50 μL) or diluent (pbs) alone on days 1 and 2; arrow indicates the start of experiment on day 0. $^{**}p<0.01$ log-rank. n = 5 – 8 mice per group. **B.** Proinflammatory cytokines IL-6, CXCL1 and CCL2 detected in lung tissue homogenates of PVM-infected mice treated with *L. plantarum* or diluent control in A. (day 8); $^{***}p<0.001$ one-way ANOVA, n = 5 mice per group. **C.** IL-6 detected in systemic circulation of PVM-infected mice treated with *L. plantarum* or diluent alone as in A.; day 8, $^{**}p<0.01$, Mann-Whitney U-test. **D.** Detection of virus in lung and spleen tissue by qPCR in PVM-infected mice treated with *L. plantarum* or diluent alone as in A., $^{*}p<0.05$, Mann-Whitney U-test, nd, virus not detected, n = 5 mice per group; dashed line denotes lower limit of detection. **E.** Microscopic histology: (**i.**) original magnification 10x, lung tissue from uninfected control; (**ii.**), original magnification 10x, PVM infection followed by diluent as in A., day 8; (**iii.**) area within box in (ii.) is enlarged, original magnification 40x, arrows denote neutrophils in alveoli; (**iv.**), original magnification 10x, PVM infection followed by *L. plantarum* as in A. **F.** Neutrophil recruitment to the airways in PVM-infected mice treated with *L. plantarum* or diluent alone as in A.; day 8, $^{***}p<0.001$ Student's t-test, n = 5 mice per group. **G.** Wet-to-dry ratios determined for

In conclusion, we have examined the role of IL-6 in promoting pathology in acute respiratory virus infection with PVM, a natural rodent pathogen that generates local inflammation in lung tissue. We showed here that IL-6 has a critical negative impact in PVM infection. Among our results, we found that PVM was substantially less lethal in *IL-6*-deficient than in wild-type mice and was associated with reduced neutrophil recruitment to the lungs. Likewise, administration of immunobiotic *L. plantarum* to the respiratory mucosa

D-dimer, coagulopathy and COVID-19

- Marked elevated D-dimer correlates with a worse prognosis and might be associated with thrombophilia, disseminated intravascular coagulation
- Autoptic reports point out the presence of thrombi in systemic and pulmonary vessels
- Anticoagulant therapy mainly with LMW heparins seems to be associated with decreased mortality in severe COVID-19

Tang N, et al. *J Thromb Haemost* 2020. *In press*
Magro C, et al. *Trans Res* 2020. *In press*

THE PROBLEM OF LUNG FIBROTIC SEQUELAE IS APPEARING

PATIENTS MAINLY DISCHARGED BY ICU DEVELOP A LUNG FIBROTIC PATTERN

- «fibrosing organizing pneumonia» CT pattern

- «fibrosing NSIP» CT pattern

Pro-fibrotic mechanisms appear during the subacute phase and probably are also incited by mechanical ventilation

Hypothetical cytokines or pathways involved: VEGF, PDGF, EDGF, TF, Wnt pathway, etc

TREATMENT STRATEGIES BASED ON PATHOGENETIC CONSIDERATIONS

- Vaccination
- Immunoglobulins from recovered Pts

- Steroids
- Azythromycin
- LMW Heparins
- Tocilizumab, Sarilumab
- Kanakinumab (?)
- Colchicine
- JAK Inhibitors (Baricitinib, Tofacitinib, Upadacitinib, Ruxolitinib, ...)
- BTK-inhibitors (Ibrutinib, ...)
- Eculizumab (?)
- Nintedanib/Pirfenidone?

- Inhibitors of viral replication (including chloroquine/ hydroxycholoroquine)
- Protease inhibitors (Camostat Mesylate, Nafamostat Mesylate)
- Monoclonal Abs against Spike proteins

- Mantain O_2 level
- C-PAP; NIV; Mechanical Ventilation

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