Università degli Studi di Padova

Corso di laurea in Medicina e Chirurgia

Dipartimento di Scienze Cardio-Toraco-Vascolari e Sanità Pubblica
UOC di Pneumologia
Direttore: Prof.ssa Marina Saetta

TESI DI LAUREA

Post Coronavirus Disease 2019 (COVID-19) pulmonary fibrosis: a 12-month follow-up study

Relatore: Prof. Paolo Spagnolo

Correlatrice: Dott.ssa Elisabetta Balestro

Laureando: Matteo Bovo

Anno Accademico 2021-2022

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1. ABSTRACT

BACKGROUND Since the beginning of the pandemic over 540 million people have been infected by SARS-CoV-2 including over 6 million of deaths, reported by WHO. Despite these large numbers, the long-term pulmonary consequences of COVID-19 remain unclear, indeed, studies with large follow-up periods are required.

Previous SARS pandemic and MERS epidemics demonstrated that symptoms and imaging abnormalities persist over time, hence, it has been suggested to monitor patients after acute SARS-CoV-2 pneumonia.

AIMS OF THE STUDY This study, as first endpoint, aims to estimate and characterize patients with pulmonary residuals as a late sequela of COVID-19 at 12 months from hospitalization for SARS-CoV-2, in terms of: radiological changes and functional impairment. As secondary endpoint the study focuses on the identification of predictors for the persistence of lung abnormalities.

METHODS In this observational cohort study, patients have been prospectively enrolled at the post-COVID clinic after hospital discharge, with visits comprehensive of previous history, physical examination, PFTs, chest HRCT, lung ultrasounds. The total number of eligible patients was of 421, all of them were previously admitted to the University Hospital of Padova from the 22nd of February 2020 until the 30th of April 2021. High Resolution CT was used to evaluate the persistence and the characteristics of radiological changes during follow-up visits. Based on the persistency of CT changes, the whole population was then categorized in two groups: 1) NOT-RECOVERY group when, at 12 months from hospital admission, CT still showed lung abnormalities; 2) RECOVERY group when resolution was gained along the follow-up. The CT images were scored through a semiquantitative scale, particularly analyzing the percentage of GGO, interstitial thickening (IT), consolidations (Co) and the presence or absence of bronchiectasis and curvilinear or linear band opacities for each of the five lung lobes. Then, for each patient, a medium lung involvement (in GGO, IT, Co) was obtained as mean among each lung lobe score. Both a univariate logistic regression analysis and a multivariate regression model were set, comparing the REC and the NOT-REC group, to detect the predictive factors to do not recover at follow-up, entailing radiologic sequelae.

RESULTS Among the 421 initially enrolled patients, 74 were *lost* because of the exclusion criteria. The whole population (n=347) was categorized into REC (those who radiologically recovered on chest HRCT along the 12-month follow-up, n=323 - 93.1%) and NOT-REC group (those who did not recover on HRCT at 12 months, n=24 - 6.9%).

The NOT-REC group resulted to be significantly older [respectively, 67.5y (53-71) vs 63y (62-76); *p*=0.019], more frequently current smokers (n=4, 16.7% vs n=12, 3.8% *p*=0.019); with worsen parameters concerning clinical course and disease severity at hospitalization, in particular: higher maximum FiO_2 required (75% vs 36%, *p*=0.01); longer hospital stay (17d vs 10d, *p*=0.001); lower P/F at admission (201 vs 295, *p*=0.015); and more frequently requiring high degree of care (n=12, 50% vs n=57, 17.6%, *p*=0.0006) and presenting dyspnoea (n=18 75% vs n=140 44.7%, *p*=0.0008). Comorbidities of the patients seemed to be equally distributed between the groups. Not even the PFTs did statistically differed between REC and NOT-REC, moreover, both groups presented dynamic lung volumes within normal range (FVCpred=99% and 95%; FEV1pred=97% and 92%). In the REC group the median time for recovering was of 133 days (73-204). On chest HRCT at 12 months in the NOT-REC group, the most frequent alterations were IT (n=21 88%, median extension of 4%); GGO (n=19 79%, median extension of 3.5%); linear and curvilinear band opacities (n=16 66%); bronchiectasis (n=7 29%). At multivariate, being *current-smoker* was an independent predictor for lung sequalae after 12 months from infection [OR 5.6 (95% CI: 1.41 - 22.12), *p* 0.01].

CONCLUSIONS The study demonstrates that after 12 months from hospital admission for COVID-19 pneumonia, a small percentage of our sample of hospitalized patients maintains abnormalities on HRCT (6.9%), which are exiguous in extension (<5%). The presence of these radiological sequelae does not impact the pulmonary function, which values are included within the normal range in both groups, since the first visit of follow-up. Finally, being current smoker at the time of being infected by SARS-CoV-2 is an independent predictive factor, at multivariate, to still present radiologic involvement at 12 months (OR 5.6), regardless of the pneumonia severity.

1. ABSTRACT (VERSIONE IN ITALIANO)

CONTESTO Dall'inizio della pandemia, i dati dell'OMS riportano oltre 540 milioni di infezioni da SARS-CoV-2, con oltre 6 milioni di morti. Nonostante l'entità di questi numeri, le conseguenze a lungo termine da COVID-19 restano incerte, rendendo necessari studi con ampi periodi di follow-up. Le precedenti pandemia di SARS ed epidemie di MERS avevano già dimostrato che i sintomi e le anomalie radiologiche persistevano nel tempo, quindi, è stato suggerito di monitorare i pazienti postumi da polmonite di SARS-CoV-2.

OBIETTIVI DELLO STUDIO Questo studio si pone come primo obiettivo stimare e tipizzare i pazienti con coinvolgimento polmonare come sequela tardiva da COVID-19 a un anno dall'ospedalizzazione per SARS-CoV-2, in termini di: anomalie radiologiche e disfunzione polmonare. Come secondo obiettivo, lo studio si focalizza nell'identificare i fattori predittivi di rischio per la persistenza di coinvolgimento polmonare al follow-up.

METODI In questo studio osservazionale di coorte, i pazienti sono stati prospetticamente arruolati negli ambulatori del post-COVID dopo la loro dimissione ospedaliera; con visite comprensive di anamnesi, esame obiettivo, PFR, HRCT del torace, ecografia toracica. Il numero totale di pazienti era 421, tutti precedentemente ospedalizzati nella Azienda Ospedaliera Universitaria di Padova dal 22 febbraio 2020 al 30 aprile 2021. La TC ad alta risoluzione è stata l'indagine diagnostica radiologica utilizzata durante le visite di follow-up. Basandosi sulla persistenza di anomalie alla TC, l'intera popolazione è stata suddivisa in due gruppi: 1) NOT-RECOVERY, quando a 12 mesi dall'ospedalizzazione erano ancora presenti alterazioni radiologiche alla TC; 2) RECOVERY, quando durante il periodo di follow-up i pazienti ottenevano la risoluzione del coinvolgimento polmonare. Le immagini TC sono state valutate attraverso una scala semiquantitativa, analizzando per ognuno dei cinque lobi polmonari la percentuale di vetro smerigliato (GGO), ispessimento interstiziale (IT), consolidazioni (Co), e la presenza/assenza di bronchiectasie, o di strie lineari o curvilinee. In seguito, per ogni paziente è stato ricavato un coinvolgimento medio del polmone (distinto per GGO, IT, Co) come media del punteggio assegnato al singolo lobo. Sono state utilizzate sia l'univariata che la multivariata, comparando i due gruppi, per valutare quali possibili fattori al

momento dell'infezione potessero essere predittivi di rischio per non riuscire a recuperare a livello radiologico al follow-up.

RISULTATI Tra i 421 pazienti arruolati inizialmente, 74 sono stati eliminati a causa dei criteri di esclusione. L'intera popolazione (n=347) è stata categorizzata in due gruppi, REC (coloro che hanno recuperato dal punto di vista radiologico alla HRCT del torace durante il periodo di follow-up, n=323 - 93.1%) e NOT-REC (coloro che non hanno recuperato alla HRCT a 12 mesi, n=24 - 6.9%).

Il gruppo NOT-REC è risultato significativamente più anziano [rispettivamente, 67.5y (53-71) vs 63y (62-76); *p*=0.019], più spesso *fumatore attivo* (n=4, 16.7% vs n=12, 3.8% *p*=0.019); con peggior decorso clinico e severità della malattia, in particolar modo: maggiore FiO_2 massima richiesta (75% vs 36%, *p*=0.01); ricoveri più lunghi (17d vs 10d, *p*=0.001); P/F all'ingresso inferiore (201 vs 295, *p*=0.015); con necessità più frequente di elevati livelli di assistenza (n=12, 50% vs n=57, 17.6%, *p*=0.0006) e più frequentemente con dispnea (n=18 75% vs n=140 44.7%, *p*=0.0008). Nessuna evidenza statistica è emersa comparando le comorbidità tra i due gruppi e nemmeno tra le spirometrie, i cui volumi dinamici sono risultati nella norma (FVCpred=99% e 95%; FEV1pred=97% e 92%). Nel gruppo REC, il tempo mediano per il recupero radiologico è stato di 133 days (73-204). Alle HRCT del gruppo NOT-REC a 12 mesi, le alterazioni più frequenti sono state IT (n=21 88%, estensione mediana di 4%); GGO (n=19 79%, estensione mediana di 3.5%); strie lineari e curvilinee (n=16 66%); bronchiectasie (n=7 29%). Alla multivariata, essere *fumatore attivo* è risultato un fattore indipendente di rischio di mantenere sequele polmonari a 12 mesi dall'infezione [OR 5.6 (95% CI: 1.41 - 22.12), *p* 0.01].

CONCLUSIONI Lo studio dimostra che a 12 mesi dal ricovero per polmonite da COVID-19, una piccola percentuale del nostro campione di pazienti ha mantenuto anomalie alla HRCT (6.9%), le quali sono esigue in estensione (<5%). La presenza di queste sequele radiologiche non hanno implicazioni sulla funzionalità respiratoria, i cui valori sono nei limiti di norma in entrambi i gruppi fin dalla prima visita di follow-up. Infine, essere *fumatori attivi* al momento dell'infezione da SARS-CoV-2 è un fattore predittivo indipendente di rischio, alla multivariata, per mantenere a 12 mesi anomalie radiologiche (OR 5.6), indipendentemente dalla gravità della polmonite.

2. ABBREVIATIONS

6MWT six minutes walking test
ABG arterial blood gas
AEC-I type I alveolar epithelial cell
AEC-II type II alveolar epithelial cell
AKI Acute Kidney Injury
ARDS Acute respiratory Distress Syndrome
BAL Bronchoalveolar lavage
BMI Body Mass Index
Co Consolidation
COPD chronic obstructive pulmonary disease
COVID-19 Coronavirus Infectious Disease – year 2019
CXR chest X-ray
DECT dual-energy CT
DLCO Diffusion Lung CO (carbon monoxyde) capacity
ECM extracellular matrix
ERS European Respiratory Society
FEV1 Forced expiratory volume in 1 second
FiO_2 fraction of inspired oxygen
FVC Forced Vital Capacity
GGO ground-glass opacification
HRCT high resolution computed tomography
HFNC high flow nasal cannula
ICU Intensive Care Unit
IFN-γ interferon-gamma
ILD interstitial lung disease
IMV invasive mechanical ventilation
IQR interquartile range
IT Interstitial Thickening
M-CSF monocyte colony-stimulating factor
mAb monoclonal Antibody
MERS-CoV – Middle East Respiratory Syndrome
mMRC modified Medical Research Council Dyspnea scale
MoAMs Monocyte-derived alveolar macrophages
NETs Neutrophil extracellular traps
NIV non-invasive ventilation
NPV negative predictive value
Nsps nonstructural proteins
OTI orotracheal intubation
ORF Open-Reading Frame
PASC (Post-Acute Sequelae of COVID-19)
$P/F = paO_2/FiO_2$
PICS post-intensive care syndrome
PDGF-A platelet-derived growth factor A
PFR Prove di funzionalità respiratoria
PFT pulmonary function testing
PPV positive predictive value
RBD Receptor-binding Domain
RT-PCR real time-polymerase chain reaction
SARS-CoV Severe Acute Respiratory Syndrome — Coronavirus
SPECT single-photo emission computed tomography
TGF-β
TLC Total lung capacity
TMPRSS2 Transmembrane protease serine 2
TraM tissue resident alveolar macrophages

3. INTRODUCTION

i. BACKGROUND

At the time of writing, June 2022, over 540 million confirmed cases of SARS-CoV-2 (Severe Acute Respiratory Syndrome - Coronavirus - 2) have been reported by the World Health Organization, including over 6 million deaths. Of which around 228 million distributed through Europe, 162 million throughout Americas, 64 million in Western Pacific, 58 million reported in South-East Asia, 22 million in Eastern Mediterranean and just 9 million of confirmed cases in the African continent (1).

ii. HISTORY OF PANDEMICS

In December 2019 in Wuhan, China, cases of unexplained pneumonia were diagnosed (2). Deep sequencing analysis from lower respiratory tract samples of the patients infected were rapidly executed, with results indicating a novel coronavirus, named SARS-CoV-2. The findings showed that it was a positive-stranded RNA virus belonging to the Coronaviridae family, new to humans (3). During the first weeks of the epidemic in Wuhan, because of an association noted between the early cases and the Wuhan Huanan Seafood Wholesale Market, a zoonotic spread of SARS-CoV-2 through this market was suggested. Nonetheless, it remains unclear the role of this market, as several early cases reported no link to the Huanan Market or any other market in Wuhan (4).

COVID-19 is the term used for SARS-CoV-2 related disease, it's characterized by both different degrees of severity, from an asymptomatic status to an acute respiratory distress; and by a wide spectrum of sign and symptoms, like fever, shortness of breath, cough, sore throat, myalgia or fatigue, ageusia or dysgeusia, anosmia, nausea, vomiting and diarrhoea (2,5).

In the subsequent months SARS-CoV-2 epidemic spread from Wuhan to Taiwan, South Korea, Japan, Thailand, Vietnam and Singapore. In February and March 2020 reached contemporaneously Middle East and Europe turning into a pandemic.

To do not mess up with terms, a few definitions by the *Oxford Advanced Learner's Dictionary* (6,7)*:*

- "An epidemic is a large number of cases of a particular disease or medical condition happening at the same time in a particular community;"
- "A pandemic is a disease that spreads over a whole country or the whole world regularly found in a particular place or among a particular group of people and difficult to get rid of."

Other definitions of pandemics taken by the Bulletin of the World Health Organization are "an epidemic occurring worldwide, or over a very wide area, crossing international boundaries and usually affecting a large number of people" or "an epidemic occurring worldwide" (8).

Hence, when COVID-19 was limited to Wuhan, it was an epidemic, but the geographical spread all over the world turned it into a pandemic, officially declared by WHO – World Health Organization on March 11, 2020.

Through history many pandemics have occurred; indeed, just across the last century the Spanish flu in 1918, SARS-CoV-1 in 2002, another influenza pandemic in 2009 and MERS-CoV in 2012 happened, among others (9).

At the beginning of 20th century, the *Spanish influenza pandemic* also called the *Great Influenza Pandemic of 1918* killed over 40 million people; caused by an A(H1N1) virus. A unique factor of this pandemic was that mortality was higher in younger people 20-40 years old.

Milder pandemics occurred after almost forty years, in 1957-1958, the *Asian Flu* due to A(H2N2) and later on in 1968 with the *Hong Kong Flu* to A(H3N2), causative of 1 to 4 million deaths each (10).

In 2002-2003, SARS-CoV-1 was the first virus of the millennium capable of spreading along the international air travels. It was quite similar to a cold or an influenza, and it could transmit through droplets. Thanks to strict measures of surveillance and control implemented by WHO and international and regional organizations in order to contain the disease, SARS-CoV-1 remained confined to China, never gaining the appellative of pandemic (11).

A decade later, in 2012, MERS-CoV - Middle East Respiratory Syndrome was identified for the first time in Saudi Arabia. Even though the 80% of the cases have been diagnosed in Saudi Arabia, strains of MERS have been isolated by dromedaries in Egypt, Qatar, and Oman. As zoonotic infection, it's renowned that humans are infected by MERS through direct or indirect contact with dromedary camels. Approximately 35% of the patients reported with MERS infection have died, therefore, due to this high fatality rate, the virus diffusion was limited by itself (12). There is a subtle difference between the fatality rate and mortality; the former calculates the number of death caused by a disease in patients affected by that specific disease, the latter considers general mortality due to a specific pathology in a population (9).

In epidemiology, the basic reproductive rate (R_0) is a very used parameter to measure the virus capacity to spread among people. It estimates the average number of secondary transmissions by one infected person: if R_0 is lower than 1 the pandemic can be considered under control, by contrast, if it is higher the pandemic is growing (13).

As reported by Petersen et al. in July 2020, R_0 was estimated to be 2.4 for SARS-CoV-1 in the early months, 0.9 for MERS-CoV and for the influenza pandemic 2.0 in 1918 and 1.7 in 2009 (9).

For SARS-CoV-2 initially was calculated 2.5, but later on during the successive waves of the pandemics a more accurate evaluation estimated a value around 5 or 6 (14). Liu et al. reviewed twelve studies and calculated that R_0 of COVID-19 ranged between 1.5 to 6.68, which on average resulted 3.28 (13,15).

The lowest R_0 was the MERS' one, indeed, its diffusion has been limited with less efforts; we deduce that the more lethal the virus is, the less is transmissible (9).

As already mentioned, the age distribution of patients with severe illness differs consistently between SARS-CoV-2 and influenza pandemics. The case fatality rate was much higher in older than 50 years old for the coronaviruses pandemics, reaching more than 16% in Italy as average of all regions for the over 80. Overall, a percentage >95% of SARS-CoV-2 deaths happen in patients over 45 years old and >80% in over 65 (14).

By contrast, the influenza pandemics of 1918 rate had inverted trends, affecting mostly younger people. The mean age at death was 27 for the 1918 pandemic, a real burden to bear added to the First World War losses (9).

Why could SARS-CoV-2 spread worldwide compared to its predecessors? It still remains unclear, what we know for sure is that since 2003 international flights from China has increased at least 10 times and high-speed trains have been implemented through all China regions, including Wuhan. Another crucial aspect that favoured SARS-CoV-2 transmission was that a COVID-19 patient is already infective a few days before symptoms manifestations (9). Furthermore, biochemically speaking, SARS-CoV-2 seems to have a higher affinity of the RBD to ACE2 compared to SARS-CoV-1 (16). Novel features of SARS-CoV-2 are its hypercoagulability, the excessive multi-organ immune system response and long-term sequelae (17).

iii. ABOUT SARS-COV-2

a. VIRUS CLASSIFICATION

SARS-CoV-2 is part of the *Betacoronavirus* genus in the *Coronaviridae* family.

The *Coronaviridae* family consists of four genera: α, β, γ and δ.

Alphacoronaviruses and *Betacoronaviruses* are responsible of respiratory and gastrointestinal diseases. In particular, SARS-CoV-2 is a member of the lineage B of the *Betacoronavirus,* also called *Sarbecovirus* (18).

The three *Betacoronavirus*es related to severe acute respiratory distress are MERS, SARS-CoV-1 and 2, while other *Betacoronaviruses* are associated to a normal cold symptomatology such as HcoV-OC43, HcoV-HKU1 (19).

b. VIRUS STRUCTURE

The *Betacoronavirus* are enveloped and ssRNA+, which means that they are encoded by a single positive-sense strand RNA genome of around 30,000 nucleotides including 14 ORFs (Open-Reading Frame). Four structural proteins are encoded by this large genome: S, E, M, N.

The nucleocapsid protein (N) is where the genome is allocated, surrounded by a lipid envelope, made by the other three proteins spike (S), envelope (E) and membrane (M).

While M and E proteins are crucial for the virus assembly, S protein has the receptor-binding domain (RBD). The spike protein is constituted by three segments: an intracellular tail, a transmembrane domain and a large ectodomain. S1 and S2 are the RBD, placed in the ectodomain of the spike protein.

For curiosity, the word *Corona* in Latin means *crown*, that is the typical aspect resembled by the *Coronaviridae* at the electrical microscope because of the peplomers, large protrusions on the virus surface due to the S proteins ectodomains.

On the 5'-end, ORF1a and ORF1b are located, encoding respectively two large polypeptides pp1a and pp1b, from which through post-transcriptional modifications derive 16 Nsps (nonstructural proteins), each one with a specific role.

The ACE2 receptor (Angiotensin Converting Enzyme 2) is SARS-CoV-2 target during viral infection, exposed by the host respiratory cell type 2.

During the infection, the S1 binds ACE2, while S2 fuses the host with the viral membranes, triggered by host cell proteases as furin, trypsin, cathepsin and TMPRSS2 (Transmembrane protease serine 2). Thereby, SARS-CoV-2 can release the viral genome into the infected cell. TMPRSS2 has a doble role, favoring the viral entry to the cell and working as spike protein priming. Once SARS-CoV-2 enters the cell, through endocytosis or independent endocytic pathway, is processed by two lysosomal proteases (cathepsin L and B) (2,14,20).

SARS-CoV-2 infects humans via the airways and may directly invade other organs such as kidneys (distal tubules), intestines, liver and pancreas.

c. VIRUS TRANSMISSION

The main via of transmission of the virus is human-to-human airborne spreading through droplets, especially small-particle in short-distances.

Also fomite transmission is possible, which means by contact with contaminated objects (14).

iv. PATHOPHYSIOLOGY OF THE DISEASE

SARS-CoV-2 pathophysiology involves several organs and systems.

First, there is *a direct viral toxicity* against multiple cells type in the respiratory tract, in particular the AEC-II, alveolar epithelial cells type II. Furthermore, it has been observed the expression of ACE2 and TMPRSS2 in nasal goblet secretory cells, colonocytes, esophageal cells, cholangiocytes, pancreatic β-cells and renal proximal tubules and podocytes.

A second pathophysiological mechanism is the *endothelial cell damage*, responsible for a thrombin overproduction, fibrinolysis inhibition and activation of the complement system. A proinflammatory state induce cytokine release that activates inflammatory cells. Severe COVID-19 is not uncommon to present at blood examinations a T cell deficit, probably due to a viral antagonism to IFN, and a general elevation of inflammatory marker (C-reactive protein, ferritin, fibrinogen, D-dimer, lactate dehydrogenase), as well as high IL-6 levels has been related to a worse prognosis. This context permits NETs formation (Neutrophil extracellular traps), that by themselves facilitate the activation of the intrinsic and extrinsic pathways of the coagulation. Thereby, COVID-19 is strictly related to a pro-thrombotic status (16).

Another pathophysiological mechanism of damage *involves the RAAS system*. As renowned, the Angiotensin converting enzyme II cleaves Ang I to obtain Ang 1-9, or cleaves Ang II into Ang 1-7, both ligand of the AT2 receptor and MAS receptor. AT2 receptor is the *doppelganger* of AT1, the main receptor of the RAAS system: while AT1 has a vasoconstrictor and pro-fibrogenic role, AT2 is vasodilator and anti-fibrogenic. SARS-CoV-2 is cause of a RAAS dysregulation that may explain the organ-specific manifestations of the disease (16).

Sometimes, because of a maladaptive resolution of lung injury, there are patients that evolve into a fibrotic scenario. A huge number of insults can lead to this

common end-stage where the main character is interpretated by the fibroblast (21).

At least a couple of recognized pathways involve the fibroblasts in the fibrotic process. When alveolar injury happens, these cells migrate to the lungs answering to inflammatory cytokines (e.g. IL-1β, IL-6 and TGF-β1), where they begin secreting extracellular matrix and they differentiate to myofibroblasts.

The second recognized pathway sees the mutual interaction between fibroblasts and MoAMs (Monocyte-derived Alveolar Macrophages): alveolar macrophages secrete PDGF-A (platelet-derived growth factor A) while fibroblasts produce M-CSF monocyte colony-stimulating factor. Both these factors can stimulate and recruit fibroblasts and macrophages, respectively. In a normal status this process is needed to repair the alveolar damage, but in a pathological condition it is chronically maintained, leading to an exaggerated fibrotic response (21).

The IMAGE 1 below represents the situation when SARS-CoV-2 infects AEC-II and TRaMs (tissue resident alveolar macrophages). Once CoV-reactive T cells recognize antigens phagocytized by TRaMs, they begin to secrete IFN-γ. In this way, further monocytes and CoV-reactive T-cells are recruited, establishing a loop circuit in which monocytes, in turn, become MoAMs that will present SARS-CoV-2 antigens on their MHC (21).

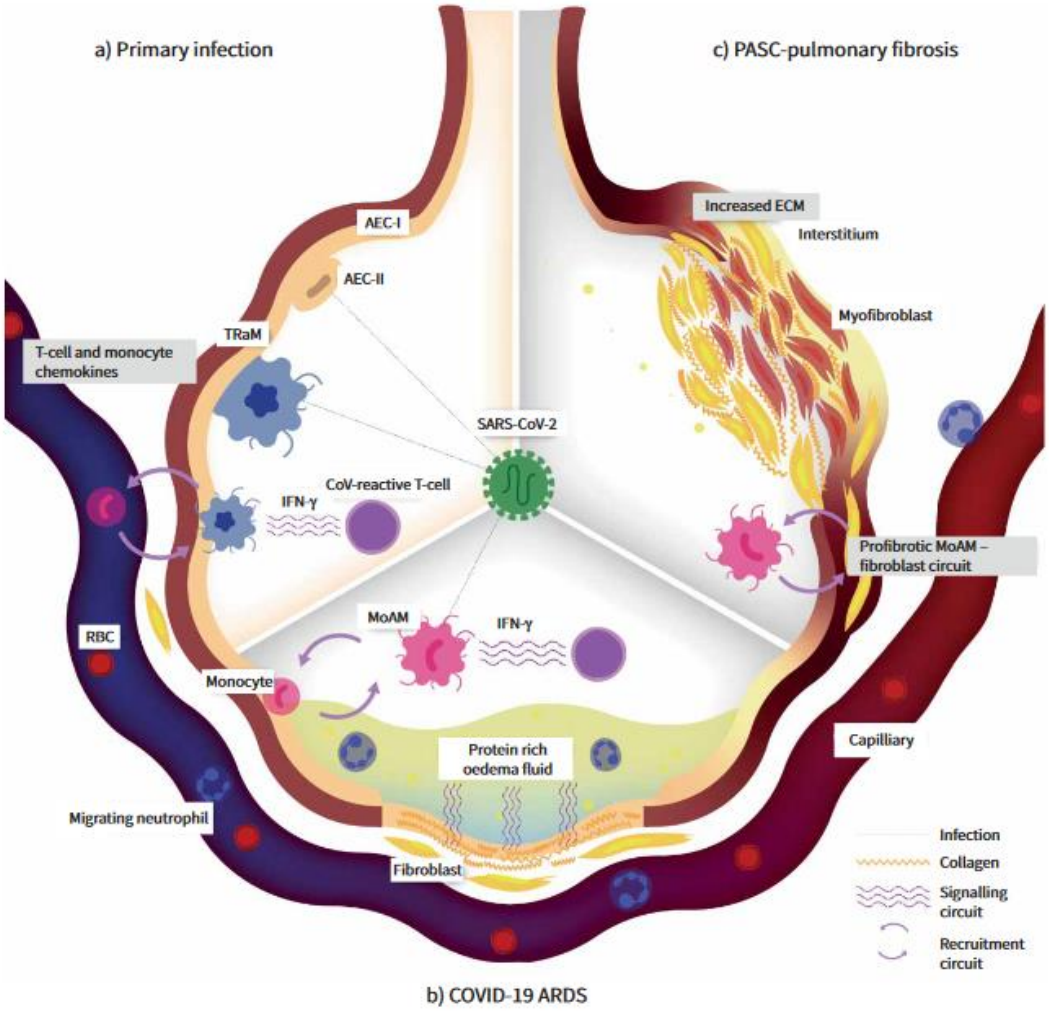


IMAGE 1: Representation of the interaction between SARS-CoV-2, AECs, TRaMs, CoV-reactive T-cells and fibroblasts.
Taken from *Mylvaganam RJ, Bailey JI, Sznajder JI, Sala MA. Recovering from a pandemic: pulmonary fibrosis after SARS-CoV-2 infection. Eur Respir Rev. 2021 Dec 31;30(162):210194.*

At this point remains to deepen when a prolonged inflammatory condition is maintained. Many different mechanisms can contribute to this state, but no evidence of causality has been demonstrated yet (21,22).

One reasonable cause are the *secondary infections*, detected or undetected. Since the profibrotic status may occur in patients that have cleared the virus, it means that SARS-CoV-2 has not a direct role in this process, but it may contribute to create a failure of the repairing process. It has been estimated a 16%-21% of secondary pulmonary infections due to SARS-CoV-2 (23,24).

A second correlation between the profibrotic status and SARS-CoV-2 infection is the duration of *mechanical ventilation* in an acute phase of the respiratory distress (21).

A third cause might be a *genetic predisposition*; in a study conducted by Grant et al. they compared BAL sample in a cohort of patients with new-onset (within 48h of intubation) respiratory failure due to COVID-19 with that of a group of patients affected by pneumonia secondary to pathogens other than SARS-CoV-2. They highlighted how COVID-19 has a longer severe clinical course, lasting 6-12 days between the onset of symptomatology and the acute distress, whereas 1-3 days for influenza A infection; furthermore, they measured higher levels of cytokines, and the transcriptome profile revealed a predisposition to IFN-γ gene response by immunity cells (25).

These three hypotheses may all contribute to a prolonged inflammatory state leading to fibrosis and maybe if all present at the same time they may have a synergic effect, but at the moment, the real cause of the fibrotic lesions remains unknown (21).

v. COVID-19 DISEASE

a. DIAGNOSIS

The gold standard for SARS-CoV-2 diagnosis is viral nucleic acid detection by RT-PCR investigating nasal or oropharyngeal sample (26). The 5^{th}-6^{th} day after symptoms onset, it is usually reached the peak of viral shedding in throat swab or sputum, when the detection rate hits 100% (26).

A quick diagnostic tool consists in the SARS-CoV-2 antigens by respiratory tract samples, usually positive in acute or early infections, but with inferior sensibility compared to the RT-PCR (26).

Serological testing is available as well, in which an enzyme-linked immunosorbent assay detects qualitatively anti-S protein IgG/IgM (26).

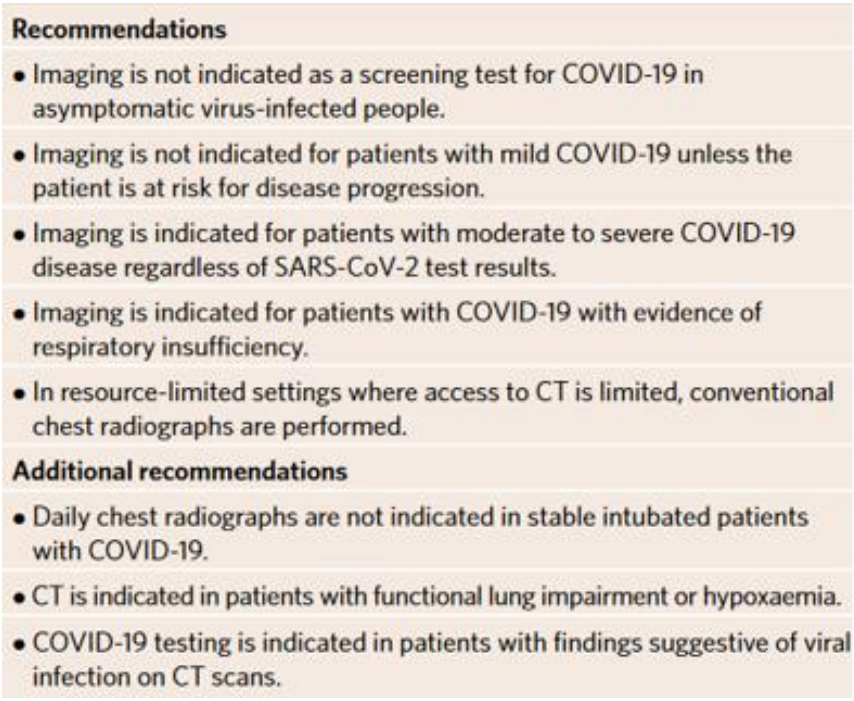
Other possible samples can be collected in saliva, faeces, BAL, pleural fluids, urine or blood (26).

At blood sample, it is common to find lymphopenia, thrombocytopenia, altered inflammatory markers like IL-6, TNFα, ferritin, C-reactive protein and increased

GOT, GPT, lactate dehydrogenase, D-dimer, prothrombin time, TnI, creatine phosphokinase (PT) and signs of acute liver injury (14).

Cases suspected for COVID-19, are used to undergo a radiographic evaluation, X-ray (CXR), CT and/or lung ultrasound. Usually CXR is sufficient, as gold standard investigation showing bilaterally consolidation and GGO, particularly in the lower sections of the lungs; but its NPV (negative predictive value) is not good enough to permit, if tested negative, an assured exclusion of lung involvement (14,26). Indeed, images obtained on CXR are not as specific as those collected with others techniques (e.g. chest CT) (26).

For cases with negative RT-PCR but with high clinical suspicion of COVID-19, there are evidence suggesting to perform a chest HRCT as confirmatory investigation, as shown at IMAGE 2 (26,27). HRCT has a very high sensitivity of 97%, quite low specificity of 25%, while the positive and negative predicted values are respectively 65% and 83% (27).



Recommendations
• Imaging is not indicated as a screening test for COVID-19 in asymptomatic virus-infected people.
• Imaging is not indicated for patients with mild COVID-19 unless the patient is at risk for disease progression.
• Imaging is indicated for patients with moderate to severe COVID-19 disease regardless of SARS-CoV-2 test results.
• Imaging is indicated for patients with COVID-19 with evidence of respiratory insufficiency.
• In resource-limited settings where access to CT is limited, conventional chest radiographs are performed.
Additional recommendations
• Daily chest radiographs are not indicated in stable intubated patients with COVID-19.
• CT is indicated in patients with functional lung impairment or hypoxaemia.
• COVID-19 testing is indicated in patients with findings suggestive of viral infection on CT scans.

IMAGE 2: Recommendations concerning COVID-19 imaging.
Taken from *Kevadiya BD, Machhi J, Herskovitz J, Oleynikov MD, Blomberg WR, Bajwa N, et al. Diagnostics for SARS-CoV-2 infections. Nat Mater. 2021 May;20(5):593–605.*

b. CLINICAL COURSE

- Acute manifestations of COVID-19

A meta-analysis by C. Fernández-de-las-Peñas et al. collected 33 studies for a total of 24 255 patients with previous SARS-CoV-2 infection; in the general sample the most frequent referred symptoms were fatigue in 63.4%, cough 60.2%, fever 55.3%, ageusia 46.0%, anosmia 45.7% and dyspnea 44.1% (28).

They analyzed patients subdivided in a group of previously hospitalized patient and a group of not hospitalized patients. At admission or at the onset of the disease, the most frequent symptoms in the hospitalized and the non-hospitalized groups were, respectively: cough (65.2% versus 56%), fever (59.45% versus 52.5%), fatigue (48.0% versus 71.89%), dyspnea (50.9% versus 38.9%), anosmia (34.3% versus 51.9%), ageusia (34.0% versus 51.8%) and myalgia (15.6% versus 59%). Hence, in a decrescendo order of frequency, symptoms resulted to be cough, fever, fatigue, dyspnea, anosmia and ageusia in hospitalized patients. While, in non-hospitalized patients, symptoms were fatigue, myalgia, cough, fever, anosmia and ageusia (28). It is curious to note that in the non-hospitalized group symptoms as chest pain, myalgias, sore throat, anosmia, diarrhea, nausea and vomiting, palpitations and vertigo were quite more frequent compared to the other group (28).

Extrapulmonary manifestations are frequent (14,16,28):

- venous thromboembolism: segmental pulmonary embolism, catheter-related thrombosis, deep vein thrombosis (particularly in severe COVID-19);
- cardiac: acute coronary syndrome, chest pain, Takotsubo cardiomyopathy, myocardial injury or myocarditis, arrhythmias, new-onset atrial fibrillation, cardiogenic shock, ischemic stroke, acute cor pulmonale, intracardiac thrombus;
- neurologic: headaches, cognitive impairment, dizziness, encephalopathy (in critical patients), encephalitis, delirium, seizures, ataxia, myalgia, ageusia, anosmia, stroke, Guillain-Barré;
- renal: AKI, proteinuria, hematuria;
- endocrine: hyperglycemia, diabetic ketoacidosis;

- gastrointestinal: diarrhea, nausea, vomiting, anorexia, abdominal pain and it can alter the intestinal microbiome with prolonged viral fecal shedding usually 11 days on average more than respiratory samples;
- dermatological: skin rash, hair loss, petechiae, livedo reticularis, urticaria, vesicles.

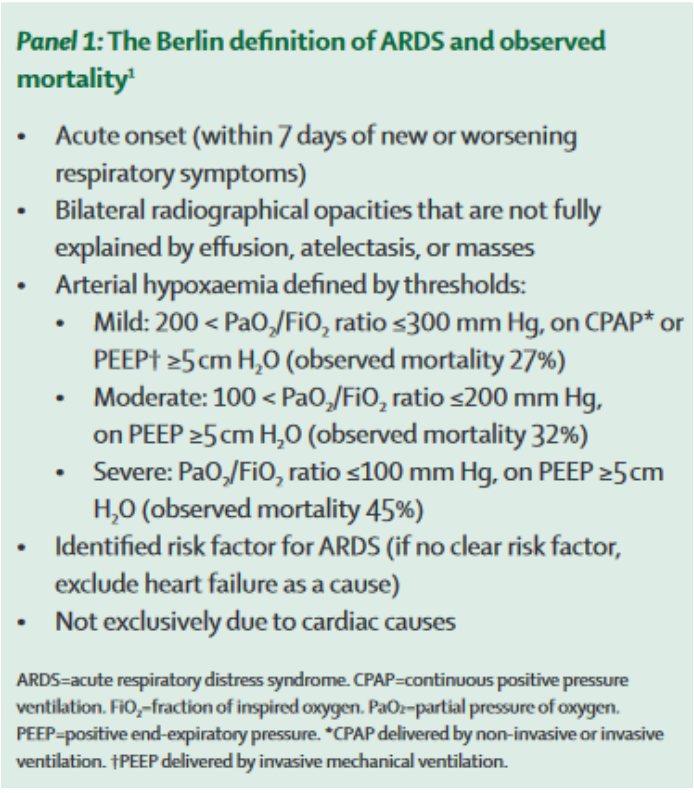
In a retrospective cross-sectional study conducted by G. Escalon and colleagues, investigating associations between intrathoracic complications in COVID-19 pneumonia and the outcomes, they analysed 976 patients who underwent chest imaging during the first month of the pandemic (29). They analysed chest X-rays looking for any of the followings: pneumothorax, chest tube, pneumomediastinum, pneumopericardium, subcutaneous emphysema, lobar collapse and pleural effusion; or chest CT in which, besides the previous abnormalities, they could search also pneumopericardium, associated to lobar collapse and pneumatocele airways findings (29). An overall prevalence of 1 out of 5 intrathoracic complications emerged (29). In particular, pleural effusion in 17% of patients, pneumothorax in 3% and pneumatoceles in 10%. From their results emerge also that intubated patients have higher risk of pneumothorax and pneumomediastinum, respectively 19% versus 6.5% and 10% versus 5% compared to never intubated patients (29). The authors support the theory that COVID-19 and ARDS increase the risk of barotrauma as cause of pneumothorax, pneumatocele and subcutaneous emphysema (29).
Finally, they conclude by saying that any of the intrathoracic complication they have looked for, if already manifested at presentation, they were predictor for a 11.2-fold higher risk for ICU admission ($p<0.0001$), 12.4-fold higher risk for intubation ($p<0.0001$), 0.49-fold lower probability of success in extubating ($p=0.02$) and a longer hospitalization (13 days versus 5 days $p<0.001$) (29). Fortunately, at least between patients with or without any intrathoracic complication no significative statistical variation in survival was found (29).

The most severe complication of COVID-19 is respiratory failure requiring mechanical ventilation (14).

In a retrospective cohort study by Wu et al., of 201 total patients infected by SARS-CoV-2 recruited, the 41.8% developed ARDS, of whom 52.4% died and 20% evolved into a severe ARDS (30). ARDS happening in COVID-19 disease is clinically comparable to ARDS in other etiologies. A quotable difference is that mechanically ventilated patients in a context of ARDS in COVID-19 have a 3-fold longer duration of the aforementioned ventilation compared to those patients with similar conditions caused by influenza virus (14 days versus 4 days) (21).

- What ARDS is

ARDS – Acute respiratory Distress Syndrome is a clinical situation with rapid onset, characterized by severe dyspnea evolving into respiratory failure and multi-organ failure (22,31). At X-rays it presents with bilateral radiopacity due to pulmonary oedema, but not related to a cardiogenic cause (22). Hypoxemia levels are used to classify ARDS in mild, moderate or severe disease, as shown at IMAGE 3 below (31).



***Panel 1:* The Berlin definition of ARDS and observed mortality[1]**

- Acute onset (within 7 days of new or worsening respiratory symptoms)
- Bilateral radiographical opacities that are not fully explained by effusion, atelectasis, or masses
- Arterial hypoxaemia defined by thresholds:
  - Mild: 200 < PaO_2/FiO_2 ratio ≤300 mm Hg, on CPAP* or PEEP† ≥5 cm H_2O (observed mortality 27%)
  - Moderate: 100 < PaO_2/FiO_2 ratio ≤200 mm Hg, on PEEP ≥5 cm H_2O (observed mortality 32%)
  - Severe: PaO_2/FiO_2 ratio ≤100 mm Hg, on PEEP ≥5 cm H_2O (observed mortality 45%)
- Identified risk factor for ARDS (if no clear risk factor, exclude heart failure as a cause)
- Not exclusively due to cardiac causes

ARDS=acute respiratory distress syndrome. CPAP=continuous positive pressure ventilation. FiO_2=fraction of inspired oxygen. PaO_2=partial pressure of oxygen. PEEP=positive end-expiratory pressure. *CPAP delivered by non-invasive or invasive ventilation. †PEEP delivered by invasive mechanical ventilation.

IMAGE 3: The Berlin definition of ARDS.
Taken from *Meyer NJ, Gattinoni L, Calfee CS. Acute respiratory distress syndrome. 2021;398:17.*

Causes are various, usually pooled by a direct or indirect damage involving alveolar permeability, such as infectious (sepsis) or toxic (31).

In 2016, before COVID-19 pandemic, a large observational study investigating ARDS incidence in ICUs spread over fifty different countries, comprehensive of 29 144 patients admitted to participating ICUs, found a 10.4% of fulfilling-criteria patients for ARDS (32). Among these patients with acute respiratory distress, a third had a mild form, almost a half a moderate presentation and less than a quarter a severe form (32).

When SARS-CoV-2 happened, clinicians' interest for ARDS suddenly increased because of the huge number of patients affected by COVID-19 and at the same time complicated by respiratory failure (22). According to data of a retrospective study conducted in Poland, it seems that among all their 116 539 COVID-19 discharged patients in 2020, the prevalence of ARDS was of 3.6%, (higher in males 4.4%, and lower in females 2.9%), and given the extreme diffusion of SARS-CoV-2, ARDS has become an overwhelming condition (22,33).

Furthermore, it has been noted that COVID-19-related ARDS have higher frequency of thrombosis, coagulopathy and endothelial dysfunction (22). Indeed, since the first period of the pandemic, surprising incongruences like the severity of hypoxemia occurring in a compliant lung have been highlighted, possibly explained by a loss in lung perfusion and in hypoxic vasoconstriction regulation (34).

ARDS consists of three phases: (31)

- Exudative: with typical alveolar edema and neutrophilic lung infiltration which determinate alveolar damage and hyaline membranes; it lasts less than seven days (31). This is due to permeability of the endothelial cells (pneumocytes type I and II) that are injured by the infection and leads to accumulation of fluids and generation of pro-inflammatory cytokines (31). Dyspnea and tachypnea in this phase occur, and imaging shows radiopacity of at least three quarters of the lungs, representative of pulmonary edema but not distinguishable from cardiogenic pulmonary edema (31).
- Proliferative: characterized by interstitial inflammation and early fibrotic changes, happening from the first to the third week (31). In this phase

patients are used to improve and get off mechanical ventilation, even though they still complain symptoms and hypoxemia (31). It is typical the change from a mostly neutrophilic infiltrate to a lymphocyte enriched one (31).

- Fibrotic: with interstitial fibrosis and emphysema-like bullae formation, usually after the third week (31). In this phase an oxygen support and/or long-term mechanical ventilation may be required. Further risks are development of pneumothorax, higher dead space and pulmonary hypertension (31).

c. TREATMENT

Clinical course should be divided in two different phases, a first one due to viral replication and second one driven more by the systemic inflammation and cytokine dysregulation than the cytopathic effect of SARS-CoV-2 (35). Based on these statements, antiviral drugs will be more effective in the early phase, in order to reduce the viral load which is strictly related to the severity of the disease; while anti-inflammatory treatment is more beneficial later on in the clinical course (14,35).

At time, antivirals and immunomodulators are the most investigated treatment approaches but only general principles can be stated with reliance (14).

- Glucocorticoids:

The ERS guidelines, as shown at IMAGE 4, strongly recommend with moderate level of evidence to use corticosteroids for patients with COVID-19 requiring oxygen, non-invasive or invasive mechanical ventilation. On the other side, they recommend not to use them for those COVID-19 patients requiring hospitalisation but not requiring supplementary oxygen or ventilatory support (36).

In a controlled, unblinded trial, Horby and colleagues recruited 6425 patients hospitalized for COVID-19, of whom a part receiving oral or intravenous dexamethasone (6 mg once daily for 10 days) and the other with usual treatment.

As first endpoint, mortality within the 28th day after randomization, resulted that for patients in the corticosteroid group also receiving oxygen through not invasive or invasive mechanical ventilation, mortality was statistically lower (for those with invasive mechanical ventilation 29.3% versus 41.4%, while 23.3% versus 26.2% for those with non-invasive oxygen support, both versus those receiving glucocorticoids but not receiving respiratory support) (37).

Nevertheless, dexamethasone treatment compared to usual care alone does not ameliorate the primary endpoint in those patients who, at randomization, were not receiving respiratory support (37).

These results find a strong scientific rationale in using anti-inflammatory treatments for the most severe disease cases because of the inflammation and the dysregulated cytokine release in COVID-19 (36).

Dexamethasone is the glucocorticoid with stronger evidence of benefit compared to prednisone or methylprednisolone (14).

Those patients requiring high dosage of glucocorticoids should be monitored for the possible side effects, like hyperglycaemia and co-infections (14).

- Anticoagulation:

The ERS guidelines recommend anticoagulating hospitalised patients with COVID-19 with strong class of recommendation and very low quality of evidence (36).

The incidence of venous thromboembolism is surprisingly variable, from 0 to 85% depending on the considered study (36).

- anti-IL-6 receptor antagonist mAb:

The ERS guidelines suggest with conditional class of recommendation and low evidence to use this kind of drug in COVID-19 hospitalised patients requiring oxygen or ventilatory support and to do not give it to those patients who do not require supplementary oxygen (36).

Before receiving anti-IL-6 receptor antagonist mAb, patients should have already received or should be receiving glucocorticoids, unless contraindications (36).

These are based on a meta-analysis conducted by Chalmers et al. for the ERS (36) amounting around 6300 patients including both groups treated with and without anti-IL-6. The drug used as mAb anti-IL6 was, for most cases, tocilizumab.
Overall, a significant effect on mortality of this treatment was not found, even though, it was found in the two largest studies RECOVERY (4116 patients enrolled) and REMAP-CAP (803 total patients) (36). The RECOVERY results show a significant reduction in 28-day mortality compared to the usual care group alone (31% versus 35%, rate ratio 0.85 p=0.0028) and reduced risk of progression of non-invasive oxygen support at baseline to an invasive mechanical ventilation or death (33% versus 42%, risk ratio 0.84, p<0.0001) (38). The REMAP-CAP recruited ICU COVID-19 patients receiving organ support; it shows how patients treated with tocilizumab or sarilumab have a higher number of organ support-free days compared to control, a 1,64 (CI 95%) more effective approach and an improved 90-day survival of 1,61 (CI 95%) (39).

- Non-invasive ventilatory support:

Recommendations by the ERS guidelines indicate HFNC or non-invasive CPAP delivered through either a helmet or a face-mask for COVID-19 patients with hypoxemic acute respiratory failure without an immediate indication for invasive mechanical ventilation (36).

- Chloroquine and Hydroxychloroquine:

With strong class of recommendations, the ERS guidelines indicate not to offer hydroxychloroquine to both hospitalised and outpatients with COVID-19 (36).
Initial studies had shown how these two drugs could be effective against some kinds of viruses in vitro, including SARS-CoV-2 (36,40). Further trials, finally, demonstrated that no beneficial effects could be obtained by this treatment in vivo (36).

- Azithromycin:

In absence of bacterial infection, azithromycin is not indicated for both hospitalized patients and outpatients (36).

- Remdesivir:

The ERS guidelines suggests not to treat with Remdesivir hospitalized patients for COVID-19 who require mechanical ventilation, but there is no recommendation regarding the use of this drug in hospitalized COVID-19 patients not requiring invasive mechanical ventilation (36).

One only trial (ACTT1) referred an amelioration in time to recovery and length of hospitalization among patients who received Remdesivir versus placebo (1062 total patients, 15 days versus 10 days for recovery, rate ratio 1.29, $p<0.001$ and reduced hospitalization from 17 days to 12 days) (36,41). Nevertheless, this is not in accordance with other larger clinical trials, such as SOLIDARITY that found no beneficials in mortality. Furthermore, this last statement is confirmed by a meta-analysis of trials comprehended by SOLIDARITY itself and including ACTT1 (36).

- Neither lopinavir-ritonavir, nor colchicine, nor interferon-beta are recommended by the ERS guideline for hospitalized COVID-19 patients management (36).

Therapy	Recommendations	Strength of recommendation	Quality of Evidence
Corticosteroids	1) The panel recommends offering treatment with corticosteroids for patients with COVID-19 requiring oxygen, noninvasive ventilation or invasive mechanical ventilation	Strong	Moderate
	2) The panel recommends NOT to offer treatment with corticosteroids for patients with COVID-19 requiring hospitalisation but not requiring supplementary oxygen or ventilatory support	Strong	Moderate
IL-6 receptor antagonist monoclonal antibody	3) The panel suggests offering IL-6 receptor antagonist monoclonal antibody therapy to hospitalised patients with COVID-19 requiring oxygen or ventilatory support	Conditional	Low
	4) The panel suggests NOT to offer IL-6 receptor antagonist monoclonal antibody to patients not requiring supplementary oxygen	Conditional	Low
Hydroxychloroquine	5) The panel recommends NOT to offer hydroxychloroquine to patients with COVID-19, including hospitalised patients and outpatients	Strong	Moderate
Azithromycin	6) The panel suggests NOT to offer azithromycin to hospitalised patients with COVID-19 in the absence of bacterial infection	Conditional	Very low
Azithromycin and hydroxychloroquine	7) The panel suggests NOT to offer hydroxychloroquine and azithromycin in combination to patients with COVID-19	Conditional	Moderate
Colchicine	8) The panel suggests NOT to offer colchicine for hospitalised patients with COVID-19	Conditional	Very Low
Lopinavir–ritonavir	9) The panel recommends NOT to offer lopinavir–ritonavir to hospitalised patients with COVID-19	Strong	Low
Remdesivir	10) No recommendation is made regarding the use of remdesivir in patients hospitalised with COVID-19 and not requiring invasive mechanical ventilation	None	Moderate
	11) The panel suggests not to offer remdesivir to patients hospitalised with COVID-19 infection who require invasive mechanical ventilation	Conditional	Moderate
Interferon-β	12) The panel suggests NOT to offer interferon-β to hospitalised patients with COVID-19	Conditional	Very low
Anticoagulation	13) The panel recommends offering a form of anticoagulation to hospitalised patients with COVID-19	Strong	Very low
Noninvasive ventilatory support	14) We suggest HFNC or noninvasive CPAP delivered through either a helmet or a facemask for patients with COVID-19 and hypoxaemic acute respiratory failure without an immediate indication for invasive mechanical ventilation	Conditional	Very low

In the document, high-flow nasal cannula oxygen therapy (HFNC) is integrated in the term "noninvasive ventilatory support". IL: interleukin; COVID-19: coronavirus disease 2019; CPAP: continuous positive airway pressure.

IMAGE 4: Recommendations and evidence for COVID-19 therapeutic strategies by ERS guidelines, Feb 2021.
Taken from *Chalmers JD, Crichton ML, Goeminne PC, Cao B, Humbert M, Shteinberg M, et al. Management of hospitalised adults with coronavirus disease 2019 (COVID-19): a European Respiratory Society living guideline. Eur Respir J. 2021 Apr;57(4):2100048.*

Since bacterial overinfection is not common in COVID-19 management, usually empiric antibiotics are needed only when diagnosis is not certain (14).

The most indicated anti-pyretic drug is acetaminophen, while NSAIDs category is suspected to have a worse clinical outcome in COVID-19 (14).

A particular attention has to be given for immunosuppressive agents because in these cases, patients have a higher risk to develop severe COVID-19 or to die (14).

vi. LONG-COVID

First, it is important to define terminology that nowadays have entered in the colloquial vocabulary about COVID-19 consequences. The ERS claims that 'Acute-COVID-19' are those symptoms occurring within the fourth week of SARS-CoV-2 infection. While 'Post-COVID-19 syndrome" or 'Long-COVID' refer to all those signs and symptoms that patients may develop during or after SARS-CoV-2 infection and continue for more than 12 weeks (42).

Others as 'long-term COVID syndrome or phases' or 'persistent post-COVID phases' are all synonymous (42). Patients with long COVID may complain different clinical entities like PICS (post-intensive care syndrome, the equivalent of Acute post-COVID), post-viral fatigue syndrome (when fatigue is the leading manifestation) or permanent organ damage (42).

PASC (Post-Acute Sequelae of COVID-19) has been coined to describe a persistent state of symptoms or complications affecting patients for a period >4 weeks (43,44). It can be subdivided into short-term PASC (1 month), intermediate-term (2-5 months) and long-term (≥ 6 months). On average, 54-55% of patients complained at least one PASC at 1 month, as well as at 2-5 months and at 6 months (44). In accordance with these results, a multicentre observational study conducted in Spain, analysed 797 patients for a period of 6 months. A 63.9% presented at least one sequela (45).

Data stratified for the income of the country shows how 56% of patients present at least one PASC in low-income countries, a 2% higher compared to the high-income countries (44).

A low- or middle-income country is defined as a median gross income equal or inferior to $12,535 (by contrast, high-income country > $12,536) (44).

Considering the extreme distribution of SARS-CoV-2 all over the world and the high incidence of these sequalae, some authors suspect that in low- and middle-income countries their health-care system will not be capable of managing the burden caused by this pandemic and its consequences (44).

COVID-19 is demonstrated to have several manifestations and sequelae such as pulmonary, hematologic, cardiovascular, neurologic, psychiatric, gastrointestinal, endocrine, renal, dermatological. Mechanisms of instauration of PASC can be explained by a direct effect of the virus (for instance: relapse, hyperinflammatory immune response, autoimmunity, neurotropism) or an indirect effect acting on mental health (post-traumatic stress, social isolation and economic factors) of not less importance (43,44).

We will mainly focus on the respiratory implications of the disease; anyway, just to quote some of the long term COVID-19 sequelae related to other organs: (43,44)

- venous thromboembolism;
- cardiac: chest pain and palpitation, dyspnea, myocardial fibrosis, arrythmias, autonomic dysfunction;
- neurologic: headaches, memory deficits, cognitive impairment, difficulty concentrating;
- mental health: depression, non-restorative sleep, anxiety disorders;
- mobility impairment and systemic symptoms: mobility decline, decreased exercise tolerance, functional impairment, fatigue, muscle weakness, general and joint pain, weight loss, flu-like symptoms, fever;
- renal: resolution of AKI;
- endocrine: worse control of glycemia, subacute thyroiditis and bone demineralization;
- gastrointestinal: diarrhea, nausea, vomiting, anorexia, abdominal pain, decreased appetite
- dermatological: skin rash, hair loss.

Since SARS-CoV-2 pandemic began in 2019 we still miss a lot to know about its consequences, in particular the long-term ones (21).

- RESPIRATORY MANIFESTATIONS:

A review by Groff et al. reported that a median of 29.7% patients complained dyspnea at visits, assessed with the modified Medical Research Council Dyspnea Scale (mMRC); cough in 13.1%, increased oxygen requirement in 65%, GGO

(Ground-glass opacification) in 23.1%, restrictive pattern on spirometry 10% and lung fibrosis 7% but chest imaging abnormalities overall in 62.2%; unless it remains unclear if the studies included in the review are considering short- intermediate- or long- term PASC (44).

The mMRC is a scale used to measure the degree of disability implied in patients by breathlessness on day-to-day activities. As defined by Rajala et al. for the ERS, it ranges from 0 up to 4; where 0 correspond to the absence of breathlessness, except on strenuous exercise; 1 to shortness of breath when hurrying on a flat ground or walking up a slight hill; 2 when patients walk slower than coetaneous on a flat ground because of breathlessness or has to stop catch a breath when walking at their own pace on the level; 3 if patients stop walking after around 100 meters or after few minutes on the level to take a breath; 4 patients are breathlessness when dressing or undressing and cannot leave the house. It is a self-rating scale (46).

A prospective cohort study conducted by E. M. Jutant et al. shows, at 4 months from discharge for COVID-19, how 16.3% of patients had a new-onset dyspnea and 4.8% a new-onset cough (in a total of 478 patients recruited) (47). Comparing the group with the new-onset dyspnea with patients without the onset of dyspnea they curiously noted that the first group was characterized by younger people with no differences concerning diabetes and hypertension prevalence, but with a more severe clinical presentation of COVID-19, longer hospital stays or ICU hospitalization and higher incidence of pulmonary embolism at presentation (47).
In patients with new-onset dyspnea, 23.1% manifested fibrotic lesions on HRCT (High Resolution Computed Tomography), and more often have cough and decreased value of the FVC and TLC (with a reduction of, respectively, 6.5% p=0.02 and 5.1% p=0.04, but in both cases not inferior to 80% of the predicted value), while no variations in DLCO were reported (47).

It has been suggested to monitor patients after acute SARS-CoV-2 infection for pulmonary fibrosis because previous both SARS pandemic and MERS epidemics demonstrated that symptoms and imaging abnormalities persisted over time (21). Pulmonary fibrosis as end-stage of the lung injury reparative process is considered

the most severe and long-term impact manifestation of COVID-19 in pulmonary function (21). A study published by Das et al. with the purpose of investigating if, after infection by MERS-CoV, could some patients may develop lung fibrosis (48). They collected radiological images at 32 and 320 days with a median follow-up of 43 days and resulted that the 36% with fibrosis were significantly older, with a higher number of ICU hospitalization in days and with a higher peak of lactate dehydrogenase levels (48).

The most relevant alterations observed on chest HRCT at 4 months by Jutant's group were 53.3% reticulations, 42.1% GGO, 19.3% fibrotic lesions (47). In the majority of patients (around 70%) the parenchyma was involved by lesions in <10% and in 97% of cases fibrotic lesions had an extension <25% (47). Furthermore, analyzing patients with fibrotic lesions features, these resulted to be older (+4.7 years, 61.2 years old median in the fibrotic group p=0.03) and they had manifested a worse COVID-19 course, with a longer hospitalization (16 days longer, 27 median days in the fibrotic group p<0.001), a longer intubation (10 days longer, 28 median days in the fibrotic group p=0.03), a more frequent access to ICU unit (a percentage differences between the two groups of +40.5%, 87.9% median in the fibrotic group p<0.001) and a higher incidence of acute pulmonary embolism (+27.8%, 39.4% in the fibrotic group p<0.001). Concerning the PFTs, patients with fibrotic lesions had a decrease of the VC (-11% of the predicted, 80.6% in the fibrotic group p=0.007), TLC (-10% of the pred, 74.1% in the fibrotic group p<0.001) and DLCO (-16.4% of the pred, 73.3% in the fibrotic group p<0.001) compared to non-fibrotic patients (47).

Nevertheless, no differences were observed about sex ratio, Body Mass Index (BMI), smoking status or comorbidities (47). Indeed, also in clinic presentation no differences were reported, as such, new-onset dyspnea and cough had a similar incidence in patients without fibrotic lesions (47). Even if new-onset dyspnea at 4 months from acute COVID-19 was common, the association with fibrotic lesions and reduced DLCO were rare (47).

The mMRC score and the 6MWT did not differed in the fibrotic lesion group and the non-fibrotic (47). The 6MWT is used in chronic respiratory disease to evaluate

physical capacities under effort, to consider the prognosis and to evaluate treatment response (49).

This demonstrate how post-COVID-19 symptoms cannot be simply related to parenchymal radiological findings (47). Even cardiological consequences as cause of the new-onset dyspnea were excluded having no differences on echocardiography between the groups with and without dyspnea (47). The authors retain that an hypothetical explanation of these symptoms should be investigated through a multifactorial approach including pulmonary embolism sequelae, lung implications and impaired muscular responsiveness (47).

In general, at four months, fibrotic lesions were not common, and it was not clear if they were irreversible or in evolution. There were authors who aimed that fibrosis needs to be clearly defined and they speculate about a possible origin of fibrotic lesions in areas of consolidation in the context of an organizing pneumonia, that may regress with time or with steroid treatment (50). In this regard, Jutant et al. highlighted in their study that, at that time, only a little percentage of patients had received a corticosteroid treatment (47).

A prospective, observational cohort study conducted in Lombardy, probably the most seriously affected region of Italy by the pandemic, investigated in 312 patients, stratified in maximum ventilatory support, the DLCO impairment as primary outcome, and the others PFT values, the mMRC scale, CXR alterations, and the 6MWT as secondary outcomes (51). At 6 months after hospital dismissal their results show a 58% DLCO impairment, usually mild and 44% radiological manifestation. Curiously, the two groups with higher DLCO impairment were those with the lowest and the highest levels of care required, respectively, “oxygen only” and the “invasive mechanical ventilation” group and not those with “CPAP”. The authors justify this circumstance with the less specific treatment offered to the oxygen-only group during the hospitalization, in particular regarding the administration of corticosteroids and prophylactic heparin (51). With a logistic regression model, they found that those patients who had required IMV support had a higher risk of radiological abnormalities, but these patients had not a higher risk for DLCO impairment which did not statistically differ to that found in the

oxygen-only group. Furthermore, the study shows how prophylactic heparin was protective against the DLCO impairment (51).

Dealing with post-COVID it is of undoubtedly importance to distinguish the follow-up period or the evolution during the acute disease (i.e. previous hospitalization or not) (28).

In the aforementioned meta-analysis by C. Fernández-de-las-Peñas (28) 1 month after the onset of symptoms, hospitalized patients and non-hospitalized, respectively, referred: cough (26.6% versus 13.9%), skin rashes (14% versus 2.5%), ageusia (11.4% versus 18.3%), anosmia (11.1% versus 19.9%), confusion (9.3% versus 7.0%), dyspnea (9.2% versus 15.7%), fatigue (7.7% versus 11.8%) and headache (1.1% versus 10.9%).

At 2 months after the onset of symptoms, respectively, symptoms referred by the aforementioned groups were: fatigue (53.9% versus 63.2%), dyspnea (24.4% versus 39.9%), joint pain (22.9% versus 10.4%), chest pain (21.0% versus 28.5%), cough (13.8% versus 40.7%), anosmia (11.5% versus 37.7%), sore throat (4.2% versus 67.0%) and headache (11.3% versus 48.2%) (28).

At 3 or more months after the onset of symptoms, symptoms referred by hospitalized patients and non-hospitalized were fatigue (38.5% versus 29.8%), dyspnea (33.3% versus 19.1%), cough (10.4% versus 6.7%), myalgia (9.7% versus 12.6%), joint pain (9.4% versus 11.2%), palpitations (9.1% versus 11.1%), anosmia (8.1% versus 15.5%), chest pain (7.7% versus 14.9%), and ageusia (7.6% versus 13.2%) (28).

The authors highlighted how one month after the acute phase of the disease, symptoms frequency reduced drastically, whereas at 2 months increased again for some of them (i.e. dyspnea prevalence in general sample at onset, 1 month, 2 months and at 3 months: 44.1% -> 13.2% -> 27.2% -> 26.3%; fatigue: 63.4% -> 11.7% -> 56.2% -> 35.3%; chest pain 16.5% -> 6.6% -> 23.6% -> 9.4%). (28) A weakness of this meta-analysis, recognized also by the authors, is that a real long-term sequela should be considered after 3 months from the onset of COVID-19 while within the month from acuteness it could still be referred to as residual symptomatology of the disease (28).

Cocconcelli and co-authors set a prospective 6-month follow-up study in their post-COVID clinic, collecting a total of 320 patients after hospitalization and aiming to explore clinical and radiological predictors of pulmonary fibrosis due to SARS-CoV-2 infection (52). At 6 months only 20% of patients still presented a radiological involvement. This group, mostly elder and man, was characterized by a worse gas impairment at admission, a worse clinical course, and radiologically by reticulations and consolidations at admission (52). In particular, higher levels of consolidation and reticulation on HRCT at admission, even when adjusted for gender, pack-years of smoking and P/F ratio in a multivariate analysis, remained significantly associated to the risk of maintaining alterations on HRCT (52).

This information is in accordance with the results published by Huang at al. in their bidirectional cohort study at 6 months from discharging, for a total of 1733 patients, in which they demonstrate how the radiological lung involvement at admission during the acute phase of SARS-CoV-2 independently predicts with the incidence of fibrotic-like changes at follow-up (52,53).

Another 6-month prospective study conducted by Han, found a 35% of patients with fibrotic-like lesions on CT in a total of 114 previously hospitalized patients; and independent predictors were: age (>50 years OR 8.5 p=0.01; duration of hospitalization >17 days OR 5.5 p=0.004; ARDS OR 13 p <0.001; CT score>=18 over 25 OR 4.2 p=0.02) (42,54).

In a longitudinal 12 months follow-up study conducted in China by X. Wu et al. they decided to exclude all patients with comorbidities (such as hypertension, diabetes, neoplastic or cardiovascular disease, asthma, COPD) and those who did require mechanical ventilation for a total number of 83 patients (55).

Anomalies in PFTs were noted between patients with versus without HRCT residual lesions at each follow-up visit (3, 6 and 12 months). Median DLCO was slightly reduced at 3 and 6 months, but reverted >80% at 12 months. Median FVC did not show reduction over the entire follow-up period (55).

At 3-month visit they found 81% of dyspnoeic patients, scored mMRC ≥1 and 6% with an mMRC ≥2. Just 5% of patients at 12 months still complained dyspnoea (55).

About persistence of radiological lesions at 12 months their results showed that almost a quarter (24%) of patients had a non-complete resolution. A general percentage of patients presenting any alteration at 3, 6, 9 and 12 months after dismissal, respectively were 78% -> 48% -> 27% -> 24% (55).
The evolution of GGO over the follow-up year (respectively at 3, 6, 9, 12 months) was 78% -> 46% -> 24% -> 23%; about interlobular septal thickening 34% -> 13% -> 5% -> 5%; reticular opacity 33% -> 16% -> 4% -> 4%; subpleural curvilinear opacity 11% -> 5% -> 1% -> 1% (55).
Among their sample of patients who did not require mechanical ventilation, none of them showed signs of established fibrotic lesions at 12 months from discharge; those characterized by any of the radiological changes had a statistically significant correlation to a longer hospitalization, to a higher HRCT pneumonia score during hospitalization, they had needed HFNC and/or NIV and they performed a worse PFT (55).

Zhan Y. group in a 12-month follow-up, among the 121 recruited patients, those with a non-severe clinic had no chest-CT remnants and less symptoms, whereas patients who had had a major clinical involvement showed up with a case-to-case different clinical presentation, since the absence of symptoms to multi-organ complication that drastically reduced the quality of life. They demonstrated, in the multivariable analysis, that the disease severity in the acute phase was associated with pulmonary diffusion abnormality and percentage change of CT score (56).

- MANAGEMENT OF LONG-COVID

There are still discordances about how to manage SARS-CoV-2 infection sequalae and one problematic, as already mentioned, is the confusing variety of terms and definitions used for the post-COVID condition. The ERS suggests that patients with prolonged symptomatology between 6-12 weeks after acute infection should undergo a COVID-19 clinic evaluation, especially for mild and moderate cases and, after a first assessment consider if further follow-up visits are needed (42).

For severe cases who had needed ICU, instead, the first follow-up visit should be planned 4 to 6 weeks after discharge (42).

About timing of the first follow-up visits, the British Thoracic Society Guidance on clinic-radiological follow-up of COVID-19 patients suggest to classify all those patients which accomplish at least one of the following in one single group (who require a first clinical review at 4 or 6 weeks after discharge): being managed on ICU or HDU, being discharged with a new-oxygen prescription, having a protracted dependency on high FiO_2, CPAP and bi-level non-invasive ventilation severely affected by COVID-19 pneumonia, or any clinical situation that the discharging team have concerning about (57). All those patients with a mild or moderate course of COVID-19 pneumonia who did not require ICU or HDU care and were typically managed on the ward or in the community can await 12 weeks for a first evaluation after acuteness and a CXR in order to confirm the complete resolution or minor insignificant changes (e.g. atelectasis).

The ERS was wondering whether exist features during the acute phase that may predict the long-term consequences of COVID-19 or not, as for instance, pulmonary fibrosis (PF) (42). It has been noted that the persistence of symptoms was not statistically correlated with the severity of acuteness, whereas among all the studies included to draft the ERS guidelines, the strongest predictor has resulted to be the age (42). Other predictive factors for the persistence of symptoms were the female gender and the number of symptoms during the first week of infection (42). This may be explained because elderly people lungs are more prone to fibrotic response or because they may be affected by an interstitial lung disease and a trigger like an infection may have larger implications than usual (42,58). In this process a key role is played by the extracellular matrix dysregulation, as sustained by Meiners et al. (58).

The persistence of symptoms implies that these patients may need for help for daily living activities as feeding, dressing, bathing, driving, housekeeping and so on since they see reduced their own autonomy. For these patients, a rehabilitating approach is effective (42).

Even if the initial status of the patients did not correlate with the persistence of symptoms, as already confirmed for other causes of ARDS, ARDS itself and severity during acuteness have been demonstrated to be predictors of PF, which manifests through decreased DLCO in a context of a restrictive syndrome, persistent GGO and fatigue or anxiety up to the 8th month after the onset of symptoms (42). In these studies COVID-19 severity was evaluated by mechanical ventilation utilization and lasting rather than other modalities of ventilation, imaging opacity score on discharge or time of hospitalization, but also for blood analyses as higher LDH at admission and IL-6, low T lymphocytes (42). The strongest risk factors for DLCO reduction were the high flow oxygen therapy and the mechanical ventilation, anyway, it is not clear the long-term evolution of this alteration (42).

It remains uncertain whether SARS-CoV-2 infection thromboembolism can be causative of a chronic thromboembolic pulmonary hypertension or pulmonary arterial hypertension or not (42). A follow-up evaluation should consider echocardiogram and contrast enhanced CT for those patients with persisting dyspnoea at 3 or 6 months after acuteness without evidence of abnormalities in HRCT and normal values on PFT except for the DLCO (42). Nevertheless, to exclude a pulmonary embolism contrast enhanced CT is not enough and SPECT or DECT should be proposed (42).

The ideal PFTs should regard TLC, expiratory flow rates and DLCO. There is a certain consensus among the researchers about the high incidence of DLCO alteration, in some cases one possible explanation might be the low haemoglobin levels of the patient with which should be corrected the DLCO value (42).
Mylvaganam et al. propose to screen all the asymptomatic or mild disease during acute infection patients and with no PASC only with PFTs at 3 months from discharge. For mild disease cases and no PASC it may also be considered a CXR, while for patients with mild disease and PASC a HRCT should be performed. Independently on PASC presentation, if patients had a moderate or severe clinical course the screening exams to consider are PFT and HRCT. (21)

Another unclear matter is about the nature of the fibrotic lesions visible on HRCT, with the risk of naming fibrosis something that actually it is not (42). Hence some fibrotic-like lesions could evolve over time or being irreversible instead. Particularly, when organizing pneumonia presents bronchial distortion there is more risk to misdiagnose it with fibrosis (42). In early-phases there is a higher probability of resolution of the imaging, thereby experts hesitate in reporting a scan as fibrotic (42,59). Furthermore, as already discussed, it is not clear if these fibrotic manifestations on HRCT have an infectious origin (due to ARDS) or if they may be caused by trauma caused by mechanical ventilation. In this regard, ERS guidelines claimed that further studies are necessary to better understand the origins of fibrosis in COVID-19 patients (42). In addition, they consider of utility, in the follow-up process, a chest CT investigation for previously hospitalized patients, or for the most severe cases or for those with a new-onset or persistent respiratory symptomatology (42).

Concerning this issue, Wells catalogues a list of reasons for which he is sceptical in calling the fibrotic-like lesions visible on HRCT as 'post-COVID ILD' (42,59). One has already been mentioned talking about the organizing pneumonia and bronchial distortion, a second one is that chest HRCT interpretation frequently does not find a confirmation in histological tissue. Furthermore, it is evident the regression of these lesions over time, but, this oppose to the concept of irreversibility implied by the term fibrosis (59).

Dealing with long-COVID management, to prevent from a high risk status of viral shedding, some authors consider PCR test at least once weekly in immunodeficient patients (42).

Increasingly, a rise of interest has been seen in the psychological entourage of COVID-19 patients while recovering in the post-pandemic time (42). An early approach in order to reduce the long-term effects of depression, anxiety, insomnia or psychosomatic manifestations has been demonstrated to be more effective (42).

The ERS guidelines affirmed that tele-health appointments were comparable to traditional in person visits, even though, studies about the cost-effectiveness are still missing (42).

4. AIMS OF THE STUDY

Several studies have already reported pulmonary consequences after either 3 or 6 months from SARS-CoV-2 pneumonia but very few data have been published concerning a longer observational period (21,44,47,51,52,54,60). Indeed, a wider knowledge of COVID-19 lung sequelae, after one year from infection, is required.

This study aims to estimate and characterize patients with pulmonary residuals as a late sequela of COVID-19, in terms of: radiological changes and functional impairment.

As secondary endpoint the study focuses on the identification of predictors for the persistence of lung abnormalities.

5. MATERIALS AND METHODS

i. STUDY POPULATION AND STUDY DESIGN

In this observational cohort study, patients have been prospectively enrolled at the post-COVID clinic after hospital discharge. The total number of eligible patients was of 421, all of them were previously admitted to the University Hospital of Padova from the 22nd of February 2020 until the 30th of April 2021.

For studying purposes, we completed the recruiting process in April 2021 which allowed to collect the data until April 2022, for a global period of one-year follow-up.

During hospitalization, positivity to SARS-CoV-2 was confirmed by a nasal or oropharyngeal swab RT-PCR.

High Resolution Computed Tomography (HRCT) was used to evaluate the persistence and characteristics of radiological changes during follow-up visits.

Based on the persistency of CT changes, the whole population was than categorized in two groups: 1) NOT-RECOVERY group, when at 12 months from hospital admission, CT still showed lung abnormalities; 2) RECOVERY group, when resolution was gained along the follow-up.

In this latter group, the mean and median recovery time at CT was then reported.

The predictive factors investigated were gender, age, smoking history (current, non-smoker, former and pack-years), BMI, presence of cardiovascular disease, respiratory disease, autoimmune disease, metabolic disease, oncologic disease, the maximum required FiO_2 during hospitalization, duration of the hospitalization, if high or low degree of care, P/F at admission, several therapeutic drugs and the PFT performed at first follow-up visit.

ii. INCLUSION AND EXCLUSION CRITERIA:

- Inclusion criteria:

1. Previous hospitalization for SARS-CoV-2 pneumonia at University Hospital of Padova from late February 2020 to April 2021
2. Patients aged ≥18
3. Patients with at least one HRCT at the last follow-up visit to demonstrate lung recovery

Patients have been prospectively enrolled for the study when they presented at follow-up visits after hospital discharge.

- Exclusion criteria:

1. Having only one or more CXR as unique radiological investigation
2. Missing at follow-up visits
3. Absence of HRCT imaging at 12 months

The exclusion criterion “missing at follow-up visits” gather all those patients who did not continue the follow-up visits whenever indicated by physicians, but also those patients who presented radiological abnormalities at a previous follow-up visit on chest HRCT (e.g. radiological alteration at 5 or 7 or 9 months from hospital admission) and for any reason they decided to stop follow-up.

One other reason of exclusion was for those patients who did have mild alterations at HRCT with normal PFTs, but had difficulties in performing further CT scan or to come back to follow-up visits due to comorbidities or for social reasons. The IMAGE 5 below describes the flow-chart of the study.

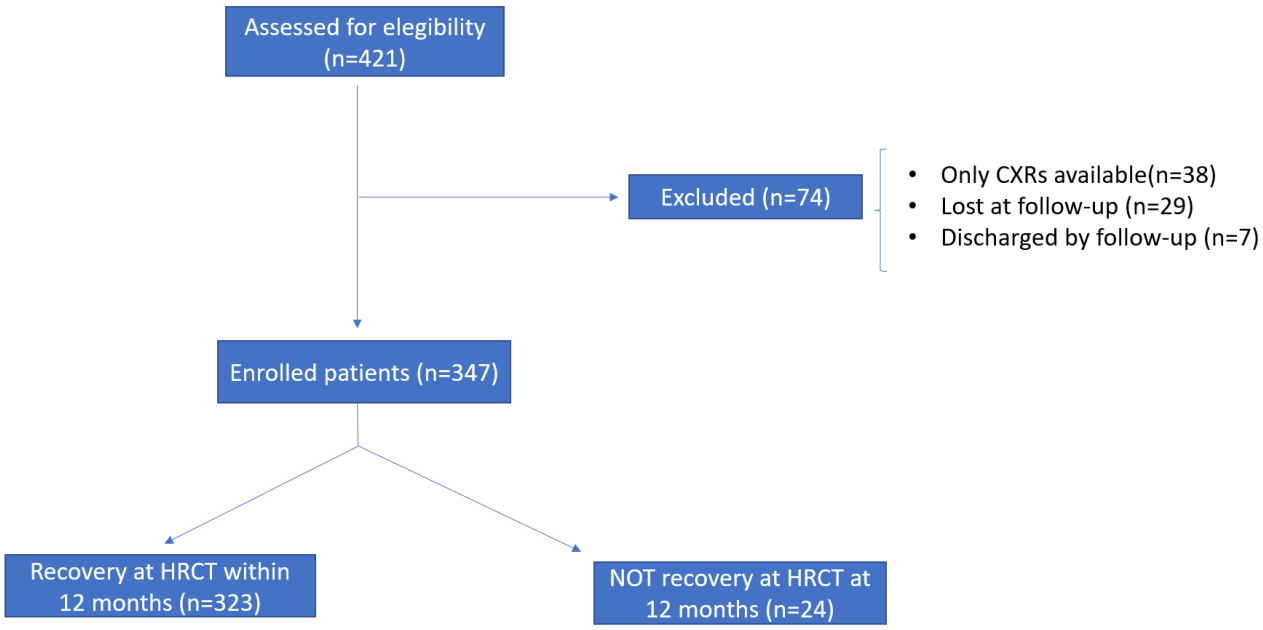


IMAGE 5: flow chart of the study. CXR= chest-X-ray, HRCT= High Resolution Computed Tomography

iii. FOLLOW-UP VISITS:

After hospital discharge, patients were evaluated for the first visit at the post-COVID clinic within the first month.

The visits consisted of comprehensive previous history, physical examination, with a particular focus on lung ultrasound, Pulmonary Function Test (PFT), 6 minutes walking test (6MWT) and the evaluation of a chest HRCT. Then, all the required dates of admission, hospital stay, discharge, follow-up visits and others have been collected using hospital medical records.

In details, for each patient were collected the following data: demographics, gender, age, BMI, smoking history (current, former, non-smoker); comorbidities (cardiovascular, respiratory, autoimmune, dyslipidemic, others); the following dates: positiveness to SARS-CoV-2, hospital admission and discharge, follow-up visits, chest imaging; severity of the pulmonary disease (room air, cannula/mask/reservoir, HFNC, NIV, OTI); arterial blood gas examination (ABG) and FiO_2 at admission, P/F, mMRC; any symptom among the followings: fever, asthenia, dyspnea, muscular alteration, anosmia/ageusia, gastrointestinal, cough, headache; positivity to chest auscultation (crackles, normal, other); therapy along the

hospitalization and use of corticosteroids at discharge; PFT values; presence of B-lines at lung ultrasound; blood sample values.

The first follow-up visit was indicated within the first month, then, the followings were programmed every 3-6 months up to one year after COVID-19 pneumonia. PFTs were performed in accordance with the guidelines published by the ATS and the ERS (49,61). The PFT values considered were those performed at the first follow-up visit.

The first chest CT was required within the first month after hospital discharge, then, according to the clinicians, when pulmonary impairment was suspected, based on physical, functional, and previous radiological imaging, a chest CT was prescribed every 3 or 6 months.

iv. RADIOLOGICAL EVALUATION

The total number of patients visited at discharge from hospital was 421, of which 74 were deleted because of the exclusion criteria, reaching the study population which included 347 patients. Then, we categorized the whole population, using HRCT, into recovery group (REC) which consisted of 323 subjects and the not recovery group (NOT-REC) which included 24 patients.

Chest imaging was analysed by a chest radiologist of the University Hospital of Padova. The CT images were scored through a semiquantitative scale, particularly analysing the percentage of GGO (alveolar score, AS), interstitial thickening (IT), consolidations (Co) and the presence or absence of bronchiectasis and curvilinear or linear band opacities for each of the five lung lobes. Then, for each patient, a medium lung involvement (in GGO, IT, Co) was obtained as mean among each lung lobe scored value. For dichotomic parameters, bronchiectasis and band opacities, the patient was considered affected by these abnormalities whenever at least one single lobe was involved.

Examples of chest HRCT and their radiological evaluation can be found at IMAGE 6, 7 and 8 below.

v. STATISTICAL ANALYSIS

All the continuous variables were described with median and interquartile range (IQR), whereas categorical variables were reported as absolute (n) and relative (%) values.

To compare demographic, hospitalization and follow-up data between the two groups (REC and NOT-REC), the Mann-Whitney U test was used for continuous variable (as independent samples), while the χ^2 test and the Fischer's exact test were used for nominal dichotomous variables, the latter for those cases with at least one value <5.

Both a univariate logistic regression analysis and a multivariate regression model were set to detect the predictive factors to do not recover at follow-up involving radiologic sequelae.

In this study the *p* values below 0.05 were considered statistically significant.

The statistics was performed with SPSS Software version 25.0 (US: IBM Corp., New York, NY, USA) and the statistical package GraphPad Prism 8.0 (GraphPad Software, Inc., La Jolla, CA, USA).

6. RESULTS

Among the 421 patients initially included in the study, at the time of data analysis 74 patients were excluded for different reasons: 38 subjects had never undergone an HRCT, 29 had decided to interrupt the visits and, finally, only 7 are those who still presented radiological alterations but were unavailable to reach the hospital.

The TABLE 1 summarizes the characteristics of the overall population (n=347), predominantly men (n=217 62.5%) with a median age of 63 years old (IQR 53-72) and a BMI of 27 (IQR 24-30). Current smokers were n=16 - 4.7%, non-smokers n=216 - 62.8% and former smokers n=114 - 33.1%.

The most frequent comorbidities were the cardiovascular ones (n=174 - 50.1%), followed by the metabolic diseases (n=158 - 45.5%). Almost the entire population manifested fever during hospitalization (n=331 98.8%), cough was present in 54% as second most frequent symptom and dyspnoea in almost a half, asthenia and anosmia/ageusia in around a third of the overall population. Less frequent

symptoms reported during hospitalization were muscular pain or myalgia, gastrointestinal symptoms and headache.

Regarding treatment, the most administered therapies were heparins (n=281 - 81.2%) and corticosteroids (n=238 - 68.8%), then also azithromycin was frequently used (n=209 - 60.4%) and ceftriaxone (n=142 - 41.0%). After dismissal, corticosteroids were assumed by n=195 - 56.3%.

At functional test, the overall population showed normal lung volumes, FVCabs and pred of 3.3 litres 92% (2.8-4.0; 81%-104%) and FEV1abs and pred of 2.8 litres, 95% (2.3-3.3; 84%-137%).

TABLE 1 - Baseline and characteristics of patients who assisted at follow-up in the post-COVID clinic according to HRCT recovery at 12 months

	Available data	Overall population (n=347)	REC (n=323)	NOT-REC (n=24)	*p*
Demographic data					
Male – *n (%)*	347	217 (62.5)	200 (61.9)	17 (70.8)	0.51
Age at admission – *years*	347	63 (53-72)	63 (53-71)	67.5 (62-76)	**0.019**
BMI - (kg/m^2)	247	27 (24-30)	27 (25-30)	26 (24-30)	0.23
Smoking history					
pack years	334	0 (0-5)	0 (0-5)	3.1 (0-21)	0.06
Current – *n (%)*	344	16 (4.7)	12 (3.8)	4 (16.7)	**0.019**
Non-smoker – *n (%)*	344	216 (62.8)	205 (64)	11 (45.8)	0.083
Former – *n (%)*	344	114 (33.1)	105 (32.8)	9 (37.5)	0.66
Comorbidities:					
Cardiovascular diseases - n (%)	347	174 (50.1)	161 (49.8)	13 (54.2)	0.83
Respiratory diseases - n (%)	347	50 (14.4)	44 (13.6)	6 (25)	0.13
Autoimmune diseases - n (%)	347	52 (14.9)	45 (13.9)	7 (29.1)	0.068
Metabolic diseases - n (%)	347	158 (45.5)	147 (45.5)	11 (45.8)	0.99
Oncologic diseases - n (%)	347	57 (16.4)	52 (16.1)	5 (20.8)	0.57
Hospitalization characteristics					
FiO_2 max during hospitalization - %	337	36 (27-70)	36 (24-66)	75 (32-100)	**0.01**
Hospitalization - days	347	10 (6-17)	10 (6-16)	17 (10-41)	**0.001**
High degree of care – n (%)	347	69 (19.9)	57 (17.6)	12 (50.0)	**0.0006**
P/F at admission	205	295 (218-342)	295 (223-343)	201 (101-314)	**0.015**
Symptoms during hospitalization					
Fever	335	331 (98.8)	289 (92.3)	22 (100)	0.39
Asthenia	335	119 (35.5)	112 (35.8)	7 (31.8)	0.70
Dyspnea	335	158 (47.2)	140 (44.7)	18 (75.0)	**0.0008**
Anosmia/Ageusia	335	104 (31.0)	96 (30.7)	8 (33.3)	0.58
Muscular alterations	335	61 (18.2)	55 (17.6)	6 (25.0)	0.25
Headache	335	36 (10.7)	35 (11.2)	1 (4.2)	0.33
Gastrointestinal	335	74 (22.1)	69 (22.0)	5 (20.8)	0.94
Cough	335	182 (54.3)	168 (53.7)	14 (58.3)	0.36
Treatment during hospitalization					
Hydroxychloroquine/chloroquine	346	132 (38.2)	121 (37.6)	11 (45.8)	0.51
Azithromycin	346	209 (60.4)	196 (60.9)	13 (54.2)	0.52
Ceftriaxone	346	142 (41.0)	132 (41.0)	10 (41.7)	0.99
Other antibiotics	346	116 (33.5)	101 (31.4)	15 (62.5)	**0.003**
Lopinavir/ritonavir	346	65 (18.8)	58 (18.0)	7 (29.2)	0.18
Remdesivir	346	107 (30.9)	98 (30.4)	9 (37.5)	0.50
Other antivirals	346	6 (1.7)	5 (1.6)	1 (4.2)	0.35
Tocilizumab	346	18 (5.2)	15 (4.7)	3 (12.5)	0.12
Corticosteroids	346	238 (68.8)	217 (67.4)	21 (87.5)	**0.04**
Heparins	346	281 (81.2)	262 (81.4)	19 (79.2)	0.79
IgG plasma	346	67 (19.4)	62 (19.3)	5 (20.8)	0.79
Corticosteroids during follow-up					
Corticosteroids	346	195 (56.3)	179 (55.4)	16 (69.6)	0.20
PFT at first follow-up					
FVCabs – litres	288	3.3 (2.8-4.0)	3.3 (2.8-4.0)	3.2 (2.4-4.0)	0.38
FVCpred - %	287	92 (81-104)	92 (81-104)	97 (70-103)	0.79
FEV1abs -litres	343	2.8 (2.3-3.3)	2.8 (2.3-3.3)	2.6 (2.0-3.5)	0.37
FEV1pred - %	343	95 (84-137)	95 (84-137)	99 (81-121)	0.74

Then, the whole population of the study was categorized into REC (those who radiologically recovered on chest HRCT along the 12-month follow-up, n=323 - 93.1%) and NOT-REC group (those who did not recover on HRCT at 12 months, n=24 - 6.9%).

Concerning demographics (TABLE 1), the two groups were comparable for gender and BMI, whereas the NOT-REC resulted to be significantly older than the REC group [respectively, 67.5 y (IQR 62-76) vs 63 y (IQR 53-71), *p* 0.019].

Current smokers were significantly more frequent in the NOT-REC group (n=4, 16.7% vs n=12, 3.8%; *p* 0.019), whereas non-smoker, former smoker and the number of pack-years did not differ between the two groups.

Comorbidities seemed to be equally distributed between the groups, showing differences neither in cardiovascular nor respiratory nor autoimmune nor metabolic nor oncologic diseases.

Among all the symptoms reported at hospitalization, only dyspnoea showed a significant difference between the groups, indeed, 18 patients (75%) of the NOT-REC group complained it compared to the 140 patients (44.7%) of the REC group, *p*=0.0008.

Regarding all characteristics concerning hospital stay and disease severity, NOT-REC group displayed statistically significant and worsen parameters compared with the REC group, indicating that the former were affected by a more severe pneumonia, in particular: the median of the maximum FiO_2 reached during the hospitalization was higher [75% (IQR 32-100%) vs 36% (IQR 24-66%), *p* 0.01], the median duration of hospitalization was longer [17d IQR (10-41) vs 10d (IQR 6-16d), *p* 0.001], the median of the PaO2/FiO_2 ratio at admission was lower [201 (IQR 101-314) vs 295 (IQR 223-343), *p* 0.015] and the high degree of care resulted more frequent (n=12, 50% vs n=57, 17.6%; *p* 0.0006).

Among all treatment, two differed between the NOT-REC and the REC groups, in particular: *other antibiotics* (n=15, 62.5% vs n=101, 31.4% p=0.003) and *corticosteroids* (n=21, 87.5% vs n=217, 67.4% p=0.04) were more frequently used, during the hospital stay, in the NOT-REC group. For all the other drugs, including the administration of corticosteroids after discharge, we did not find any statistical difference between the two groups.

Pulmonary functional parameters were similar between REC and NOT-REC, both in FVC abs or pred and FEV abs or pred, moreover, both groups presented dynamic lung volumes within normal range (respectively, FVCpred=99% and 95%; FEV1pred=97% and 92%).

In the REC group the mean time for recovering was of 143 ± 8.6 days and the median was of 133 days (IQR 73-204) from the first day of hospital admission to the date of the first negative HRCT (TABLE 2).

***TABLE 2* – Timing of recovery on HRCT in the REC group from hospital admission**	
days mean	143 (±8.6)
days median	133 (73-204)

- Radiological evaluation at 12 months after COVID-19 pneumonia

Radiological features were characterized and scored in the NOT-REC group (n=24) at 12 months and results are summarized in TABLE 3. The most frequent alteration is the interstitial thickening, in 21 patients (88%), with a median extension of 4%; followed by the GGO in 19 patients (79%) with a median of 3.5%, and the consolidations in only 2 patients (8%) with an extension <1%. The linear and curvilinear band opacities were reported in 16 patients (66%) and the bronchiectasis in 7 (29%).

TABLE 3 - Lung abnormalities on 12-month chest HRCT in the NOT-REC group (n=24)

Ground-glass opacities	
Ground-glass opacities, n (%)	19 (79%)
Extent of Ground-glass opacities	3.5%
Interstitial thickening	
Interstitial thickening, n (%)	21 (88%)
Extent of Interstitial thickening	4%
Consolidations	
Consolidations, n (%)	2 (8%)
Extent of Consolidations	<1%
Bronchiectasis	
Bronchiectasis, n (%)	7 (29%)
Curvilinear and linear band opacities	
Curvilinear and linear band opacities, n (%)	16 (66%)

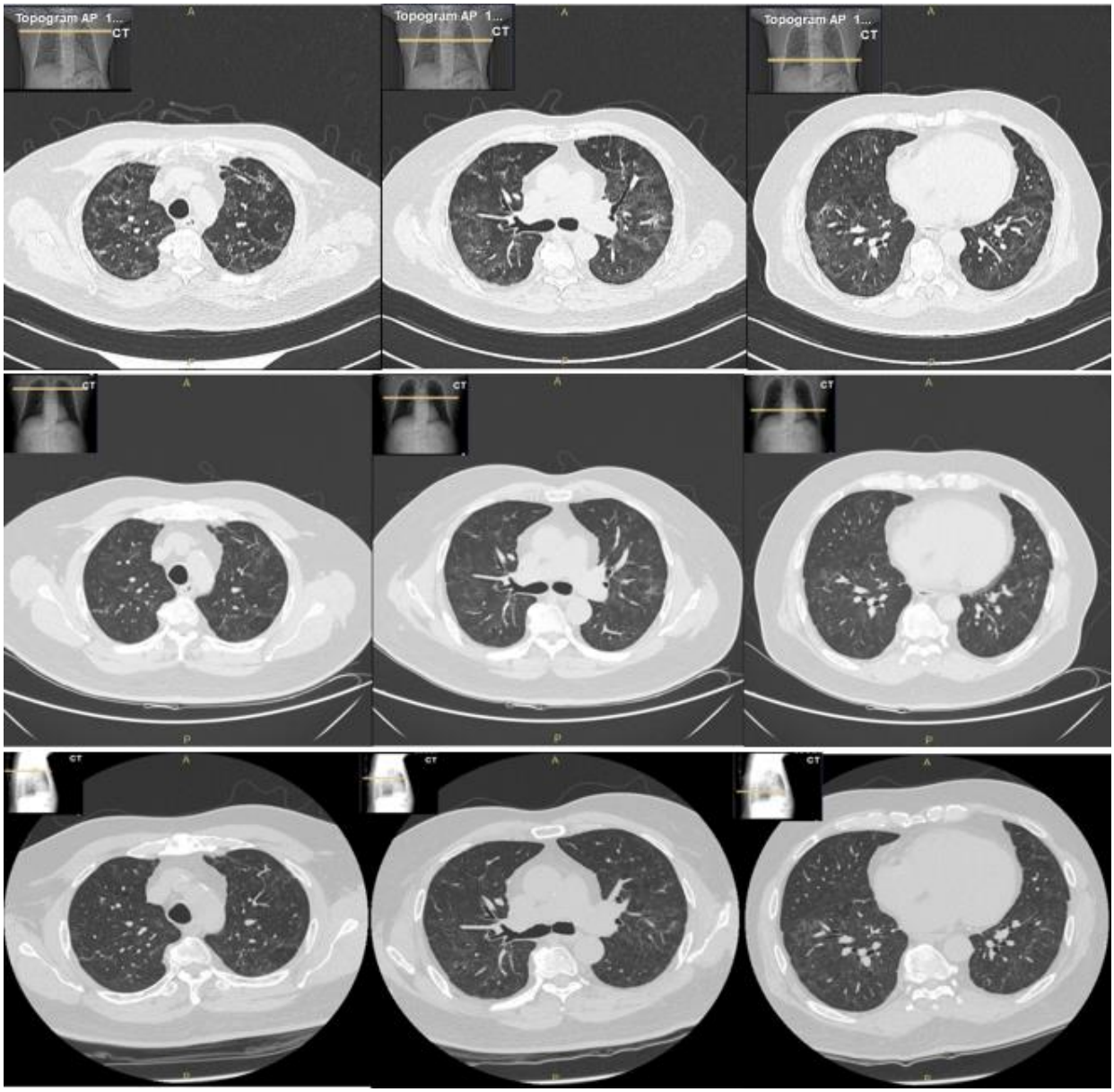


IMAGE 6: images from chest HRCTs of a NOT-REC patient, male 62 years old.
1st row, at 2 months from hospital admission with wide areas of GGO;
2nd row, at 6 months from hospital admission, reduced but still persisting GGO lung involvement;
3rd row, at 12 months from hospital admission, further ameliorations, (evaluated trough the semi-quantitative grading as follows: 22% of mean GGO extension of the five lobes; 4% of mean IT; 0% Co; 0 bronchiectasis; 1 of linear and curvilinear band opacities).

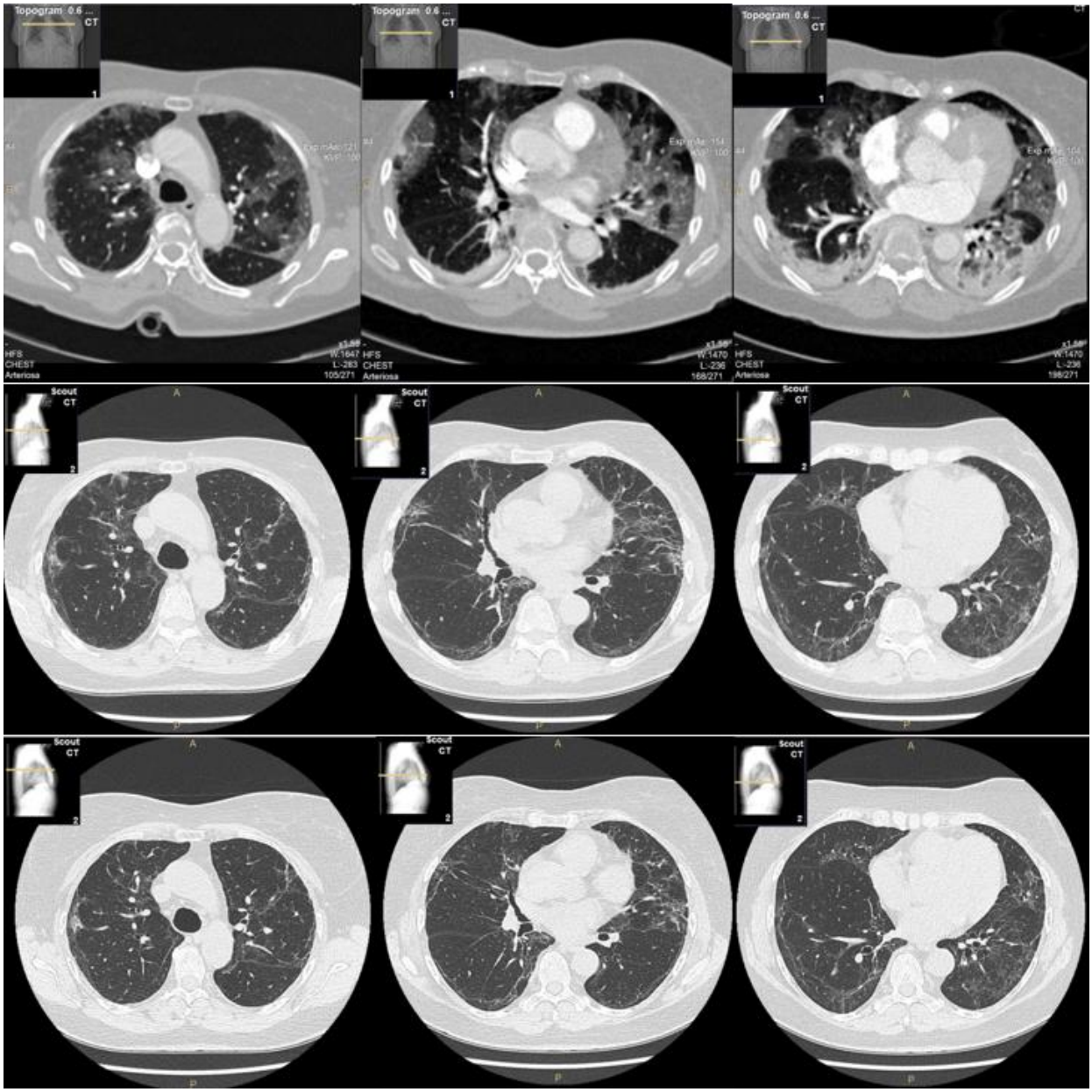


IMAGE 7: images from chest HRCTs of a NOT-REC patient, female 71 years old.
1st row, at hospitalization: acute COVID-19 pneumonia with consolidations and GGO spread in all lung lobes;
2nd row, at 5 months from hospital admission: interstitial thickening, particularly in the lower 2/3 lung fields with dystelectatic band opacities associated;
3rd row, at 12 months from hospital admission: almost unvaried compared to previous, (evaluated trough the semi-quantitative grading as follows: 7% of mean GGO extension of the five lobes; 10% of mean IT; 0% Co; 1 bronchiectasis; 1 of linear and curvilinear band opacities).

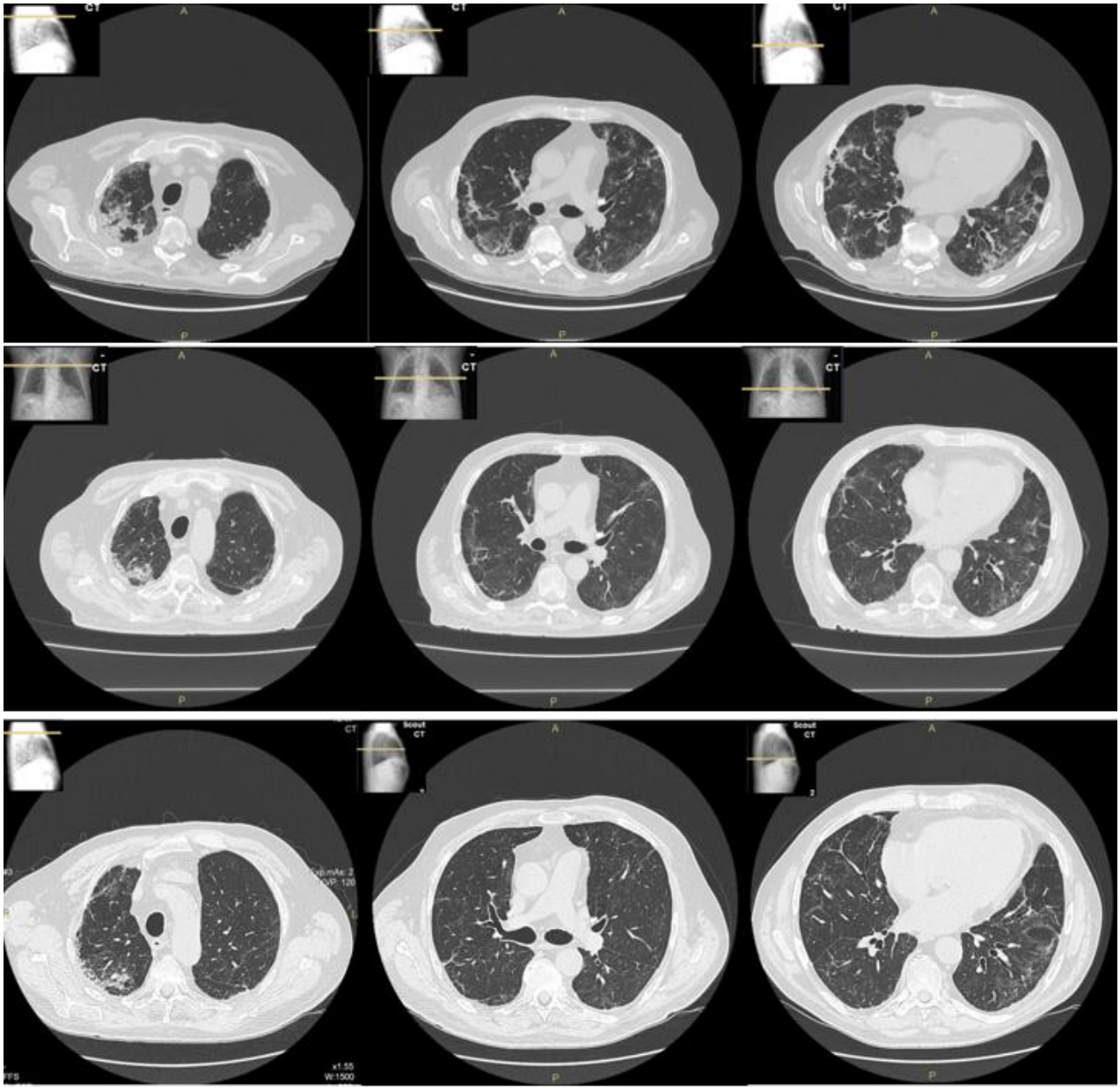


IMAGE 8: images from chest HRCTs of a NOT-REC patient, male 84 years old.
1st row, at 1 month from hospital admission: diffuse GGO, also with consolidations in particular in the apical subpleural region and right basal;
2nd row, at 6 months from hospital admission: ameliorated, GGO persist in the lung bases and the medium lobe; still showing interstitial thickening with bronchiectasis;
3rd row, at 12 months from hospital admission: sclerosis of the apical subclavian regions, atelectatic thickening in right upper lobe, diffused bronchiectasis and dystelectasis (evaluated trough the semi-quantitative grading as follows: 0% of mean GGO extension of the five lobes; 21% of mean IT; 0% Co; 1 bronchiectasis; 1 of linear and curvilinear band opacities).

- Predictors of post COVID-19 pulmonary sequelae

By univariate analysis we found that a*ge ≥63 years [*OR 2.8 (95% CI 1.09 - 7.33, *p* 0.03)] and *current-smoker*, [OR 5.2 (95% CI 1.53 - 17.53), *p* 0.008] were dependent predictors for having persistent radiological changes at 12 months of follow-up after COVID-19 pneumonia. Among characteristics of hospital stay, *days ≥10 days* [OR 2.9 (CI 1.14 – 7.61), *p* 0.03]; *high degree of care* [OR 4.7 (CI 1.99 – 10.92), *p* 0.0001]; *FiO_2 max ≥36%* [OR 2.6 (CI 1.01 – 6.78), *p* 0.047] and *other antibiotics [*OR 3.6 (CI 1.54 – 8.61), *p* 0.003], were also dependent risk factors which predict post COVID-19 pulmonary changes (TABLE 4).

At multivariate, adjusted for previous risk factor, we observed that only smoking history, particularly current smoking habit resulted a significant independent predictor for lung sequelae after 12 months from infection [OR of 5.6 (CI 1.41 – 22.12), *p* 0.01].

TABLE 4 – Univariate and Multivariate models applied between the two group REC and NOT-REC

	Univariate		Multivariate	
	CI (95%)	***p***	**CI (95%)**	***p***
Demographic data				
Age ≥ 63	2.8 (1.09 - 7.33)	**0.03**	2.6 (0.96 – 7.18)	0.06
BMI ≥ 27	0.4 (0.15 - 1.07)	0.07		
Smoking history				
Current	5.2 (1.53 - 17.53)	**0.008**	5.6 (1.41 – 22.12)	**0.01**
Non-smoker	2.0 (0.86 – 4.58)	0.11		
Comorbidities				
Cardiovascular diseases	1.2 (0.52 – 2.73)	0.68		
Respiratory diseases	2.1 (0.79 – 5.61)	0.13		
Autoimmune diseases	2.5 (0.99- 6.48)	0.051		
Metabolic diseases	1.0 (0.44 – 2.33)	0.98		
Oncologic diseases	1.4 (0.49 – 3.84)	0.55		
Hospitalization characteristics				
Hospitalization days ≥ 10	2.9 (1.14 – 7.61)	**0.03**	1.0 (0.25 – 3.29)	0.89
High degree of care	4.7 (1.99 – 10.92)	**0.0001**	2.6 (0.83 – 8.35)	0.10
FiO_2 max ≥ 36%	2.6 (1.01 – 6.78)	**0.047**	1.1 (0.31 – 3.86)	0.89
P/F ≥ 295	0.4 (0.14 – 1.25)	0.12		
Treatment during hospitalization				
Hydroxychloroquine/chloroquine	1.4 (0.61 – 3.24)	0.42		
Azithromycin	0.7 (0.33 – 1.75)	0.52		
Ceftriaxone	1.1 (0.44 - 2.38)	0.95		
Other antibiotics	3.6 (1.54 – 8.61)	**0.003**	2.4 (0.84 – 3.28)	0.10
Lopinavir/ritonavir	1.9 (0.74 – 4.73)	0.18		
Remdesivir	1.4 (0.58 – 3.24)	0.47		
Other antivirals	2.7 (0.31- 24.59)	0.36		
Tocilizumab	2.9 (0.78 – 10.90)	0.11		
Corticosteroids	3.4 (0.98 – 11.61)	0.052		
Heparins	0.88 (0.32 – 2.46)	0.81		
Corticosteroids during follow-up				
Corticosteroids	1.8 (0.74 – 4.59)	0.19		

7. DISCUSSION

As experienced by other Coronaviruses pandemic and epidemics, symptoms and radiological alterations were used to persist over time (21,48).

As previously investigated by our group (Cocconcelli et al.) we already demonstrated that pulmonary sequelae of COVID-19 at 6 months interested a small part of the population hospitalized for SARS-CoV-2 pneumonia (52).

This study wants to further investigate the SARS-CoV-2 sequelae in a larger cohort of patients and with a 12-month follow-up period from hospital admission for COVID-19 pneumonia. The main focus is based on radiologic abnormalities on chest HRCT, which is the criterion trough which patients have been classified into the two groups, if recovered along the visits or not.

In this study we found that only 24 patients (6.4%) of the overall population still presented radiologic changes at chest CT. This is not much in accordance with the study conducted in China by X. Wu et al., whose results at 12 months show that 24% of patients had a non-complete resolution at chest HRCT (55). Anyway, our data is more in line with the 19% of patients with fibrotic involvement at 4 months by Jutant and co-authors (47), whose patients, at least a further percentage, would have probably recovered at 12 months; as well as the 20% of patients with chest HRCT involvement at 6 months by Cocconcelli et al. (52).

In the overall population of the study, the median BMI shows how there is a tendency towards being overweight in this sample of hospitalized patients for COVID-19. In accordance with the study conducted by Jutant and co-authors, the pre-COVID-19 comorbidities are comparable in distribution to our overall population, in particular for the cardiovascular, respiratory and metabolic diseases (47). Our population who does not recover within the follow-up is older than that who does recover (67.5 vs 63 years, *p* 0.019), as found also by Jutant and Han, but in opposition to the study conducted by Zhan whose data, at 12 months, adjusted for the multivariate analysis indicates a negative correlation between age and the percentage of CT changed score (47,54,56). The Age ≥63 years old was confirmed as statistically relevant at the univariate analysis with an increased risk of developing abnormalities on HRCT at 12 months of OR 2.8 (1.09 - 7.33) *p* 0.03, but

not at multivariate, even if close to the threshold of significancy OR 2.6 (0.96 – 7.18) *p* 0.06, as shown in TABLE 4.

In a Fischer's exact test between the two groups at issue seems that being current smokers can be a risk factor (n=4, 16.7% vs n=12, 3.8%, *p* =0.019). This cannot find a support in literature since some studies report no statistic differences among the groups, and other authors do not distinguish the smoking-history in current, former or not-smoker. Nevertheless, at multivariate analysis, the study by Zhan found a *p* value for being current-smoker as risk factor for an increase of percentage change of CT score quite close to significancy [OR 13.05 (-1.53 to 27.62), *p* 0.08] (47,52,56). In both the univariate and multivariate analysis this variable was confirmed as relevant, respectively with an increased risk of OR 5.2 (1.53 - 17.53) *p* 0.008 and OR 5.6 (1.41 – 22.12) *p* 0.01. Therefore, being active smoker is an independent risk factor, regardless of the severity of the pneumonia and any other variable, to maintain radiological sequelae in hospitalized patients after a period of one year of follow-up, implying the 560% of risk of non-active smokers.

It is evident that the NOT-REC patients had a worse clinical course compared to patients who recovered on HRCT along the follow-up period. Indeed, median maximum FiO_2 required was 2-fold higher in NOT-REC patients (75% vs 36% *p*=0.01) and was recognized as a risk factor for lung sequelae at the univariate model. Moreover, subjects in the NOT-REC group presented also the worse clinical condition at admission with lower values of P/F ratio, higher degree of cares (NIV and/or IOT), (as shown in TABLE 1). Even if a strong correlation resulted between the severity of the acute phase and the sequelae, none of the indicators received further confirmations as independent predictors at the multivariate analysis.

This is in line with previous studies, indeed, it has been reported that patients who presented a more severe acute COVID-19 pneumonia, as indicated by ventilatory support, gas exchange index, and duration of hospital stay, are those who still present radiologic involvement at follow-up (at 4, 6 or 12 months) (47,51,52,54–56). As confirmed in literature, also dyspnoea was more frequent in the NOT-REC group, this is a further signal of worse clinical presentation during acuteness (47).

In this regard we may speculate that the category other antibiotics resulted statistically significant because those patients who did not recover at 12 months

were those critical patients who needed a wider approach to manage acute COVID-19 pneumonia, with an OR at univariate of 3.6.

The NOT-REC group received more frequently corticosteroids during hospitalization, this could be apparently difficult to explain, but it may characterize critical ill patients during the acute phase. Therefore, it could be of interest to know if they would have not taken corticosteroids during hospitalization how their clinical and radiological history would have progressed. Anyway, we highlight that the *p* value is quite close to the threshold of significancy. We consider that the use of corticosteroids after hospitalization in this case is not a confounder since the difference between the percentage of administration of the two groups was not significant.

Furthermore, in our cohort of patients, COVID-19 pneumonia not even at the first visit did imply a worsening of the respiratory function neither in the overall population nor in both groups compared [NOT-REC group: FEV1pred 99% (IQR 81-121%), FVC 97% (IQR 70-103%)]. This finding is not in line with Jutant's study at 4 months of follow-up, where they found significant differences in the group with fibrotic lesions in terms of VC(%) 80±20, TLC(%) 74± 13, DLCO(%) 73± 18 compared to those without fibrotic lesions and DLCO <70% in 41%. Even the Steinbeis group in Germany demonstrated that at 12 months the degree of pulmonary function impairment apparently still correlates with severity during the acute phase, but it improves over time (62).

Whenever recovery on HRCT was reached both the mean and the median time required overcame 4 months.

Analysing the characteristics of radiological features presented by the NOT-REC patients on HRCT at 12 months, we observed that the most frequent changes were the fibrotic interstitial thickening (88%), the GGO (79%) and the curvilinear and linear band opacities (66%). This finding is in accordance with Han and co-authors' study, that found the thickening of the pleura more frequently in the "fibrotic-like changes on CT" group and a worse CT score in general in the same group of patients (54). In this regard, Cocconcelli and co-authors found differences at 6 months of follow-up in the extension of alveolar score, consolidations and interstitial score

among the groups compared (52); and also Huang et al. at 6 months found a more severe involvement in those patients who required a higher degree of care, with the GGO as most frequent radiological entity, followed by the irregular lines (53).
Overall, in our study the lung involvement at 12 months was minimal since the median involvement reached the 4% for IT and 3.5% for GGO (with the maximum lung involvement of 21% and 22% in one patient, regarding IT and GGO respectively). Of course, it remains to be elucidated if the fibrotic lesions are strictly caused by the infection or if the contribution of mechanical ventilation lung injury should be considered (21,42,59).

We decided to exclude those patients that during hospitalization only underwent CXRs (38 patients) and did not perform a CT investigation. This because of the different sensibility and specificity, between CXR and CT, to detect lung abnormalities. In order to evaluate patients through the same tool, we considered more appropriate that those who had neither a CT from hospitalization nor a CT at follow-up visits had to be excluded, rather than compare patients with different radiological modalities. These patients were usually those who manifested a milder form of COVID-19, indeed, CT scan was not considered of utility for those hospitalized patients who were in good clinical status and did not present important consolidation at CXRs.
Conversely, for those patients who had a longer hospitalization, chest CT was necessary to better understand the pneumonia evolution, moreover, when pulmonary embolism was suspected a chest Angio CT was prescribed as well.
Regarding the group of discharged patients, the decision of interrupting the follow-up was taken by clinicians whenever they considered that, despite the persistent abnormalities on HRCT, the patient could not benefit from further visits and a specialistic evaluation was not necessary anymore. The clinical reports of these seven patients reported that the HRCTs of two of them were unvaried compared to the previous one, whereas in four patients the radiological sings were improving, always compared to the previous HRCT. All of them were asymptomatic or pauci-symptomatic and only one patient still presented lineage B bilaterally at lung ultrasounds.

Due to the nature itself of the observational study, this investigation is not thought to demonstrate a relation of causality between a possible predictive factor and an event, as in this case would be for the virus and the fibrosis; but the study can provide the strength of either a more or less probable correlation between the risk factors and the development of fibrosis.

- Points of strength and limits of the study

To the best of our knowledge, few data have been published with such a large cohort of consecutive patients and with such a long follow-up.

The study correlates the radiological features of patients with huge amount of clinical data. This allows to better understand the phenotype of those patients who have an increased risk to suffer from SARS-CoV-2 sequelae.

Another strength point of the study is the characterization of the radiological sequelae with a quantitative and also a qualitative score. There is no universal way to quantify the impact of pneumonia on CT evaluation. In addition to our previous published data (52), as already done by Liu et al. (63), we decided to include qualitative features to better classify the pulmonary involvement.

One first limit is the incomplete PFTs, that often were missing the DLCO. This is a useful parameter, particularly for the interstitial diseases of the lung that permits to understand the quality of gas exchanges. Many studies have reported a reduction in DLCO at 3 or 6 months after SARS-CoV-2 infection (47,51,55). However at 12 months Wu et al. results show how it reverted to normality, so the long-term trend is not still clear and need further clarifications (55).

Those patients who missed at follow-up could be considered another limitation of the study, since we are not aware if they would have presented persistency or resolution of pulmonary changes.

Another possible bias affecting the study might be the different therapeutic management among the various pandemic waves. For instance, in the first wave, drugs as chloroquine or hydroxychloroquine, colchicine, lopinavir-ritonavir and interferon-beta were commonly used. Since the second wave of the pandemic, the

use of glucocorticoids, anticoagulants, Remdesivir and anti IL-6 mAb have been implemented.

Furthermore, this is not a multicentre study, but it is based in data collected in one single hospital.

Finally, a last possible confounder could be the vaccine campaign, but for the timing of this study it did not affect the outcomes since patients enrolled were hospitalized, if anything, in April 2021. In Italy the vaccine campaign officially began with the administrative order on 12 March 2021, except for health givers who received it a few months earlier (64).

8. CONCLUSIONS

The study demonstrates that after 12 months from hospital admission for COVID-19 pneumonia, a small percentage of our sample of hospitalized patients maintains abnormalities on HRCT (6.9%), which are exiguous in extension (median <5%). The presence of these radiological sequelae does not impact the pulmonary function, which values are included within the normal range in both groups, since the first visit of follow-up.

Finally, being current smoker at the time of being infected by SARS-CoV-2 is an independent predictive factor, at multivariate regression model, to still present radiologic changes at 12 months, regardless of the pneumonia severity.

9. ETHICS STATEMENT

This study did not undergo the evaluation of an ethic committee.

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