

Potential Pharmacological Options and New Avenues Using Inhaled Curcumin Nanoformulations for Treatment of Post-COVID-19 Fibrosis

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ABSTRACT

Coronavirus disease (COVID-19) is a highly infectious disease caused by SARS-CoV-2 that may lead to adverse respiratory problems and death. As of January 17th, 2021, it has transformed into a global health crisis with 93,194,922 reported cases and 2.2% case fatality rate. Severe form of COVID-19 may progress into acute respiratory distress syndrome (ARDS), which in turn leads to pulmonary fibrosis. This review aims to investigate the potential use of anti-inflammatory and antifibrotic drugs as well as inhaled curcumin nanoformulations for post-ARDS fibrosis. Anti-inflammatory and antifibrotic drugs can inhibit certain pathways in the development of post-ARDS fibrosis, but there are serious limitations in their use. Since transforming growth factor- β (TGF- β) is the most important pathway in fibrosis development and is up-regulated in SARS-CoV-2 infection, antifibrotic agents that target it may be an alternative treatment. This property can be found in curcumin. Complexing curcumin with nanoparticles has proven to be safe, cost-effective, and enhances its pharmacodynamic action. Unlike current drugs, curcumin nanoformulations can be generated as inhaled aerosol and given to COVID-19 ARDS patients receiving mechanical ventilation. Future research should clarify the recommended dosing and timing of curcumin aerosol delivery, its safety and efficacy, as well as its long-term effects in the lungs.

Keywords: Acute respiratory distress syndrome, COVID-19, Curcumin, Pulmonary fibrosis, Anti-inflammatory agents

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INTRODUCTION

Coronavirus disease (COVID-19) is currently the most important global health issue. It is caused by the highly pathogenic SARS-CoV-2 that may lead to adverse respiratory events and even death on some occasions. As of January 17th, 2021, there have been 93,194,922 reported cases of COVID-19 globally with a 2.2% case fatality rate.^[1] At least one comorbidity was reported in 76% of decedents, the most common being cardiovascular diseases and diabetes.^[2]

The first stage of COVID-19 is asymptomatic, while the second stage is marked by innate immune response in the upper respiratory tract. The third stage of the disease happens in around 20% of all cases, during which the virus infects the lower respiratory tract causing lung infiltration, acute respiratory distress syndrome (ARDS), and eventually fibrosis.^[3,4] This process is mediated by profibrotic pathways, one of which is transforming growth factor (TGF)- β that regulates fibroblast activation and extracellular matrix (ECM) organization.^[5]

As of today, there are no treatments that can completely reverse the occurrence of pulmonary fibrosis, but a few can slow its progression. Although corticosteroids and conventional antifibrotic drugs nintedanib and pirfenidone have a beneficial role in pulmonary fibrosis, their use is limited due to cost, adverse effects, and availability.^[6] Furthermore, their route of administration is only through oral route, which poses a problem for COVID-19 ARDS patients receiving mechanical ventilation in the intensive care unit (ICU).^[7-9] Meanwhile, active compounds from plants such as curcumin was proven to be useful in other respiratory illnesses, such as asthma or acute lung injury.^[10] In order to optimize its use in pulmonary diseases, however, a new drug delivery

method for curcumin is needed. Curcumin in combination with nanoparticles such as cyclodextrin and chitosan may be investigated as a potential targeted therapy. Hence, we propose a review on potential pharmacological options and new avenues using inhaled curcumin nanoformulations as a novel treatment for treating post-ARDS fibrosis in severe COVID-19.

MATERIALS AND METHODS

This review was assembled by searching, compiling, and analyzing various primary studies, systematic reviews, meta-analyses, and clinical trials that investigated current and future treatment options for post-ARDS-fibrosis. The sources were cited from Google Scholar, PubMed, ERS Journal, and Scopus published between 2007-2021. Several papers were related to the role of TGF- β in fibrosis, the current available treatments for pulmonary fibrosis, and inhaled curcumin nanoformulations.

The studies were selected by matching the keywords, identifying relevant updated information, and compiling them into a single comprehensive review. Inclusion criteria for the literature were studies discussing potential use of certain drugs and inhaled curcumin nanoformulations for treatment of post-COVID-19 pulmonary fibrosis. Exclusion criteria were inaccessible and non-English journals. Search terms that were used included ("ARDS" OR "COVID-19" OR "SARS-CoV-2" OR "coronavirus") AND ("pulmonary fibrosis") AND ("treatment" or "management") AND ("curcumin" OR "inhaled curcumin nanoformulation").

RESULTS AND DISCUSSION

Pathogenesis of Post-ARDS Fibrosis in Severe COVID-19

A. Development of ARDS and Pulmonary Fibrosis

Angiotensin converting enzyme 2 (ACE2) is the main receptor for SARS-CoV-2 and binding between ACE2 receptor and spike proteins will allow the virus to penetrate the host cell along the respiratory tract.^[11] ACE2 plays an important role in normal physiology as a negative regulator for the renin-angiotensin system (RAS). It is widely expressed in various organs, but its expression level in the respiratory tract is of primary interest in the case of COVID-19 pathophysiology. Recent study by Ortiz *et al.* utilizing single-cell RNA sequencing and immunohistochemistry discovered that ACE2 expression along the respiratory tract is highest in sinonasal cavity and alveolar type II cells, permitting easy entry for SARS-CoV-2.^[12]

Not long after virus entry into host cells, immune cells and respiratory epithelium will normally secrete interferons (IFN-I, IFN- α/β) which inhibit virus replication and lower virus titer. However, production of interferons is delayed in early stages of SARS-CoV-2 infection, resulting in uncontrolled viral spread down the respiratory tract. The virus applies an array of strategies to conceal itself from host immune response, such as modulating some RNA features and directly antagonizing IFN activity. On the other hand, proinflammatory cytokines (IL-1 β , IL-6, TNF, etc.) and chemokines (CCL2, CCL5, etc.) are elevated in number.^[13,14] This rapid release of proinflammatory cytokines may be triggered by downregulation of ACE2 by SARS-CoV-2. As ACE2 normally breaks down angiotensin-II which is the end product of RAS, reduced availability of this receptor leads to increased production of angiotensin II. Interaction between angiotensin II and its receptor AT1R will then upregulate gene expression of proinflammatory molecules as well as activate macrophages that contribute to “cytokine storm”.^[15]

Evidence from *in vitro* studies of human coronaviruses including SARS-CoV-2 point to the imbalance between innate antiviral response and proinflammatory response

as the main culprit of cytokine storm. It has been postulated that cytokine storm is the major contributor to development of ARDS and multiple organ failure following COVID-19. A result of excessive and dysregulated host immune response, cytokine storm happens when large numbers of cytokines are released into the blood too quickly. The excessive inflammatory mediators, along with uncontrollably high viral load, will recruit monocytes and neutrophils to the lung tissue.^[13,14] Eventually, there will be persistent disruption of alveolar-capillary barrier and fluid leakage into alveolar spaces – two hallmarks of ARDS.^[16] ARDS typically occurs in patients who develop severe COVID-19 infection, in which the virus already infiltrates the gas exchange units of lungs and mainly infects alveolar type II cells.^[3]

Following alveolar-capillary damage, there will be proliferation and migration of lung fibroblasts to the injury site (Figure 1).^[16] In order to promote tissue repair, they will be activated by fibroproliferative signals to produce extracellular matrix (ECM) which consists of type I collagen, type III collagen, fibronectin, fibrin, and other compounds. Normally, provisional ECM will be removed once the lung normal architecture is restored, and pulmonary edema fluid in alveolar spaces will also be cleared. However, if ARDS is not swiftly managed, persistent lung injury will drive uncontrolled fibroproliferation through upregulation of profibrotic pathways (platelet-derived growth factor (PDGF), fibroblast growth factor (FGF), interleukin-1 β , TGF- β and downregulation of antifibrotic pathways (hepatocyte growth factor, keratinocyte growth factor, prostaglandin E2).^[16,17] TGF- β is particularly important since it is known to be the most potent inducer of ECM production by fibroblasts (Figure 2).^[18] It also contributes to the balance between matrix metalloproteinase (MMP) and tissue inhibitors of MMP (TIMP).^[19] The pathologic events above lead to pulmonary fibrosis marked by excessive deposition of connective tissue, formation of scar tissue, and decline of lung function.^[16-19]

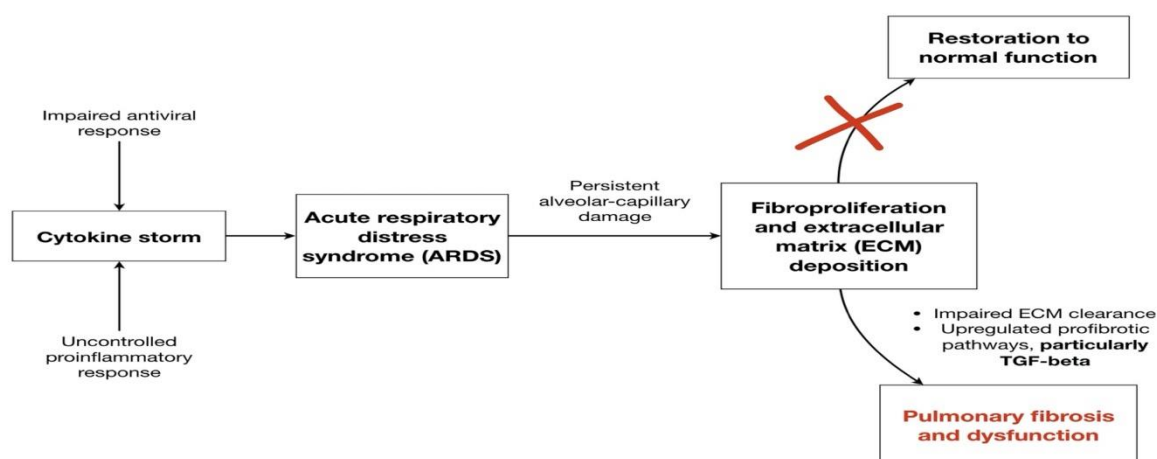


Figure 1: Key events in the progression of cytokine storm to acute respiratory distress syndrome (ARDS) to pulmonary fibrosis.

B. Relationship Between SARS-CoV-2 Infection and TGF- β Pathway

Interestingly, a recent research by Chen *et al.* discovered that SARS-CoV-2 infection appears to up-regulate the expression of TGF- β in the lungs.^[20] This leads to the hypothesis that the development of pulmonary fibrosis in most COVID-19 ARDS patients is caused by increased TGF- β expression. This upregulation is potentially caused by several mechanisms. One of them is when viral infection and its immune and inflammatory response stimulate massive activation of latent TGF- β in the lungs and blood. Consequently, more active TGF- β are pooled in the lung. Another mentions that the viral infection stimulates neutrophils infiltration to the lungs, where the neutrophils release and activate stored TGF- β then create a positive feedback loop to recruit more neutrophils. Last but not

least, the infection results in the apoptosis of pneumocytes, bronchial epithelial cells, and T lymphocytes, which triggers the infiltration of macrophage to the lungs and subsequent secretion of latent and active TGF- β . The latent TGF- β will then be activated by ROS, local proteases (elastase, furin, and plasmin), MMPs, and integrins (α V β 6).^[20-22]

Not only does TGF- β have potent pro-fibrogenic properties, but TGF- β also has potent anti-inflammatory and cell-cycle inhibitory properties. Hence, it is possible that SARS-CoV-2 utilizes TGF- β to dampen the immune response, thus leading to viral persistence.^[23,24] This is coherent with the findings that coronavirus infection increases the amount of regulatory T cells, which plays a role in delayed clearance of virus in the host.^[23,25]

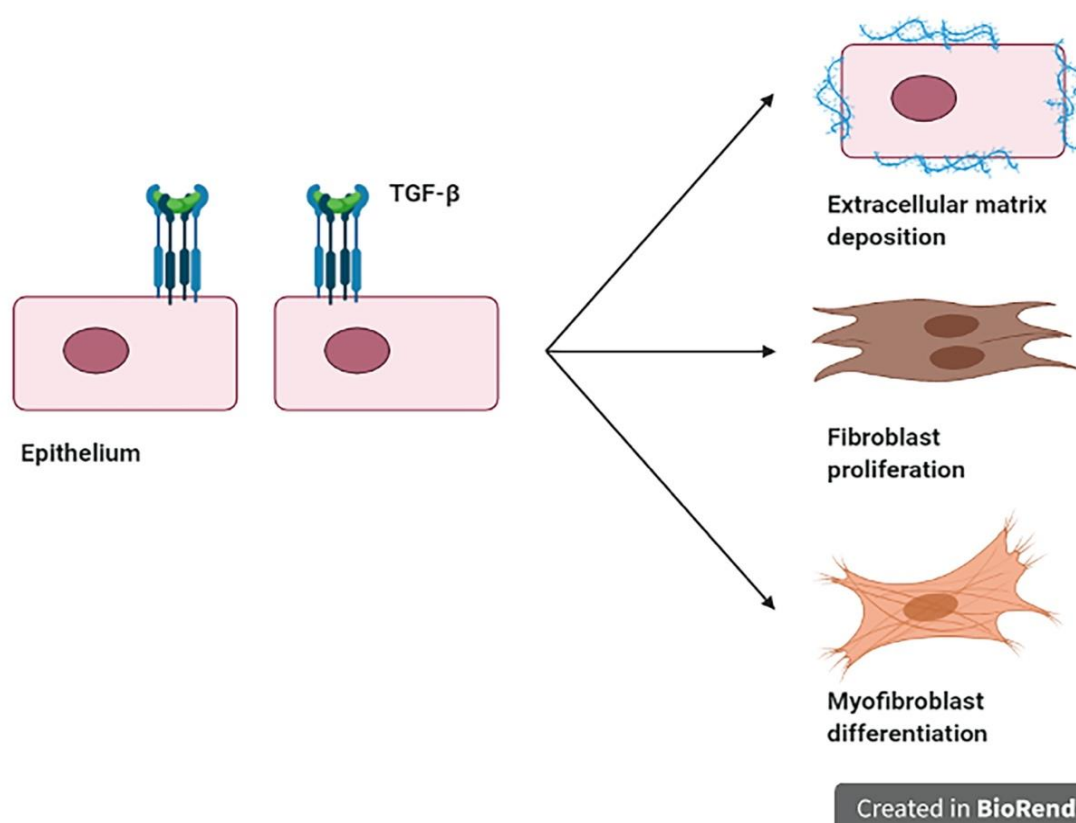


Figure 2: TGF- β in the development of pulmonary fibrosis. Modified from Biernacka A, Dobaczewski M, Frangogiannis NG. TGF- β signaling in fibrosis. *Growth Factors*. 2011;29(5):196–202. Created using BioRender.

Current Pharmacological Treatment Modalities for ARDS and Pulmonary Fibrosis

A. Corticosteroids

Since inflammation is key in both ARDS and pulmonary fibrosis, one of the current treatments proposed is corticosteroids - a drug that alleviates inflammation and fibrosis. Synthetic corticosteroids can exert their anti-inflammatory effect by inducing mitogen-activated protein kinase (MAPK) phosphatase (MKP)-1, thereby deactivating p38 MAPK that modulates the mRNA destabilizing function of tristetraprolin (TTP).^[26] Corticosteroids also inhibit phospholipase A2 and repress the genes responsible for transcription of cyclooxygenase-2. On the other hand, corticosteroid promotes the regulation of annexin A1 which leads to fewer migration of

neutrophils. This process is mediated by the reduction in levels of prostaglandin and leukotriene.^[27] These processes inhibit the transcription of cytokines and chemokines that contribute to the pathogenesis of ARDS. The development of ARDS can be influenced by several comorbidities such as pneumonia, sepsis, and aspiration of gastric contents. However, there have been limited clinical trials that examine the efficacy of corticosteroids in these high-risk patients. Previous studies found that administration of corticosteroids did not prevent ARDS in high-risk patients. In fact, ARDS developed more in corticosteroid-treated patients (64%) than normal patients (33%).^[28]

The use of corticosteroids to treat ARDS patients in severe COVID-19 is still controversial. A pilot randomized controlled trial by Meduri *et al.* in patients with ARDS onset within 72 hours found encouraging results - improvement in the lung injury score and reduced need for mechanical ventilation - when administered with methylprednisolone.^[29] Timely administration of glucocorticoids revealed a decrease in mortality rate in a retrospective study of 401 severe SARS patients.^[30] Villar *et al.* in a randomized controlled trial found that mechanical ventilation and overall mortality was reduced in patients with established moderate-to-severe ARDS that were given dexamethasone in the early stages.^[31] However, in persistent or late ARDS, fibrosis is more dominant than cellular inflammation. Thus, the therapeutic effect of corticosteroids at this stage would likely be negligible. A follow-up study by the National Institutes of Health (NIH) found that although corticosteroid-treated patients had more ventilator-free days, the length of stay in the ICU was not significantly different.^[29] Furthermore, patients randomized to corticosteroids were more prone to recurrent respiratory failure. It is thought that tapering of methylprednisolone increases risk of infection or neuromyopathy, resulting in recurrent inflammation and fibroproliferation in the lungs.^[29] In viral pneumonia that may lead to ARDS, administration of systemic corticosteroids in COVID-19 patients with ventilation has not been thoroughly investigated. Moreover, current research on COVID-19 proposes that corticosteroids can delay clearance of viral RNA and increase the risk of secondary infection.^[30,32]

Based on these studies, corticosteroids may be beneficial in the early stages of ARDS due to severe COVID-19 by alleviating the cytokine storm. Short-term use of glucocorticoid (3-5 days) like methylprednisolone can be given with a dose of 1-2 mg/kg/day.^[33] Nevertheless, it is unlikely that corticosteroids have any significant effect in late ARDS, where fibrosis is already dominant.^[34] The efficacy of low dose corticosteroid for management of post-COVID-19 pulmonary fibrosis has been investigated by a recent clinical trial. Subjects included COVID-19 patients that were discharged after 2 negative polymerase chain reaction (PCR) results with persistent radiological changes in follow-up chest computed tomography (CT) scan. The study patients received methylprednisolone 20 mg/day and followed up after 14 days with chest CT scan. However, the result of this clinical trial has not been published.^[33]

B. Nintedanib and Pirfenidone

A more promising treatment for post-COVID-19 pulmonary fibrosis would be nintedanib and pirfenidone. Nintedanib is an intracellular tyrosine kinase inhibitor that has been approved for the treatment of idiopathic pulmonary fibrosis (IPF) in the United States and Europe.^[35] Through oral administration, it broadly inhibits profibrotic mediators such as PDGF, FGF, VEGF, and TGF- β , thus subsequently inhibiting signaling pathway involved in fibrosis.^[36,37] This includes proliferation, migration, and differentiation of fibroblast and secretion of ECM by myofibroblast. Through *in vitro* and animal lung fibrosis models, it shows anti-inflammatory and antifibrotic properties.^[36]

Clinically, the efficacy of nintedanib in IPF was first observed in the phase II TOMORROW trial (NCT00514683).^[35,38] Among 4 various dose regimens (50 mg once daily, 50 mg twice daily, 100 mg twice daily, and 150 mg twice daily), oral administration of 150 mg twice

daily had the most promising result with a decreased annual decline rate in forced vital capacity (FVC) by 68%.^[38] Moreover, it decreased the incidence of acute exacerbations and improved the quality of life using the St. George's Respiratory Questionnaire score (SGRQ score). Administering 150 mg twice daily offered a good safety and tolerability profile, but prominent side effects included gastrointestinal symptoms (diarrhea, nausea, vomiting) that caused higher discontinuation, increased liver aminotransferase, and bleeding.^[35,38]

Based on the result of TOMORROW trial, the efficacy and safety of nintedanib 150 mg twice daily were observed in two replicate INPULSIS phase III trials.^[35,37,39,40] Consistent with the TOMORROW study, nintedanib decreased the annual decline rate in FVC with a mean difference of 109.9 ml/year (113.6 ml/year nintedanib vs 223.5 ml/year placebo). Although INPULSIS-1 failed to show any correlation with acute exacerbations, INPULSIS-2 showed a decrease in incidence of exacerbation. There is an acceptable safety and tolerability profile, with diarrhea being the most common adverse effect.^[37,39,40] The long-term benefits were consistently observed in INPULSIS-ON, an open-label extension involving 734 patients.^[41,42] Overall, a pooled meta-analysis using data from TOMORROW and INPULSIS trials confirmed the beneficial role of nintedanib by reducing annual rate of FVC decline by approximately 50% and improving the time to first acute exacerbation and SGRQ score.^[7] The decline in FVC was associated with lower mortality in IPF patients.^[40]

Despite having clear benefits in IPF, the efficacy of nintedanib in post-COVID-19 pulmonary fibrosis is not yet established. Currently, there is an ongoing phase III NINTECOR trial (NCT04541680) and phase IV ENDCOV-I trial (NCT04619680), investigating the benefits of nintedanib 150 mg twice daily on the changes in post-COVID-19 pulmonary fibrosis patient's FVC.^[43,44]

Other than nintedanib, another promising antifibrotic drug is pirfenidone (5-methyl-1-phenyl-2-[IH]-pyridone), a pyridone derivative approved for treatment of IPF.^[45,46] As an oral agent, it has a broad antifibrotic, anti-inflammatory, and antioxidative properties, which helps limit the damage from cytokine storm in ARDS.^[47] Its antifibrotic property result from inhibiting TGF- β -induced activation of fibroblast and differentiation to myofibroblast, and downregulating procollagen gene expression.^[48,49] Furthermore, pirfenidone is known to downregulate ACE2 receptors, strengthening the argument that this drug is potentially useful in the context of severe COVID-19.^[47]

The key clinical findings of the efficacy and safety of pirfenidone was observed in two concurrent phase III CAPACITY trials (CAPACITY-004 and CAPACITY-006) involving 435 and 344 patients, respectively. By week 72, administering pirfenidone 2403 mg/day significantly decreased the percentage of predicted FVC decline in CAPACITY-004 (8.0% pirfenidone vs 12.4% placebo), while it decreased in CAPACITY-006 (9.0% pirfenidone vs 9.6% placebo). Combining both data, it favored the use of pirfenidone for pulmonary fibrosis. Both studies also showed a good tolerability profile with no significant difference in the incidence of acute exacerbations.^[50]

Likewise, the efficacy of pirfenidone was observed in the ASCEND trial involving 555 patients. By 52-weeks, administering 2403 mg/day significantly decreased the annual rate of FVC decline with a mean difference of 193 ml/year (235.0 ml/year pirfenidone vs 428.0 ml/year placebo). Moreover, there was a 47.9% decrease in the proportion of patients who had a decline of 10% or more

in predicted FVC, or who died. There was an increase in survival beyond 533 weeks.^[51] The most common adverse effects reported were nausea, diarrhea, rashes, and/or photosensitivity. However, in some cases, elevated liver function test results were observed, thus it is contraindicated in patients with severe liver dysfunction.^[48,52]

With its antifibrotic, anti-inflammatory, and anti-apoptotic properties and ability to downregulate ACE receptors expression, pirfenidone is a likely therapeutic candidate for post-COVID-19 fibrosis. Currently, there is a phase II trial (NCT04607928) and a phase III trial (NCT04282902), exploring the role of pirfenidone.^[53,54]

Nevertheless, there are several limitations in the use of nintedanib and pirfenidone to manage pulmonary fibrosis associated with COVID-19. First, both drugs are currently only available in oral form - restricting their use in intubated or mechanically ventilated patients. This limits their use for severe COVID-19 patients who are in the

intensive care unit (ICU).^[9] Moreover, liver dysfunction is a common manifestation in SARS-CoV-2 infection as 22% of patients who are positive for COVID-19 are seen with elevated liver enzymes.^[55] Since both agents are hepatotoxic, risk and benefit ratio should be assessed before administration, especially in severe patients who consume antibiotics that are also hepatotoxic.^[9,37,56] The risk of acute pulmonary embolism is also elevated in COVID-19 patients, thus anticoagulant therapy may be used in severe cases or patients with coagulopathy. The use of nintedanib should be carefully assessed because it significantly increases the risk of bleeding when it is administered with an anticoagulant.^[37] Last but not least, the availability of pirfenidone and nintedanib is limited due to the price. The price for 1 pack of nintedanib in Belgium is €2,376.21, while that of pirfenidone is €2,472.85.^[57] Even worse, since both drugs require long-term administration, the overall cost presents huge financial burden for the general population. Therefore, a more affordable and safe option is highly needed.

Table 1: Summary of potential pharmacological options for post-ARDS fibrosis.

Drug	Mechanism of action	Advantages	Disadvantages	Clinical trials identifier for post COVID-19 fibrosis (from www.clinicaltrials.gov)
Corticosteroid ^[26-34]	<ul style="list-style-type: none"> Induces mitogen-activated protein kinase (MAPK) phosphatase (MKP)-1. Inhibits phospholipase A2 and represses gene transcription for cyclooxygenase 2. Upregulates annexin A1. 	<ul style="list-style-type: none"> Early administration alleviates cytokine storm 	<ul style="list-style-type: none"> Not effective in the later stage of acute respiratory distress syndrome (ARDS) because fibrosis is already dominant 	<ul style="list-style-type: none"> NCT04551781 (Completed, but results are not published yet)
Nintedanib ^[36-44]	<ul style="list-style-type: none"> Inhibits intracellular tyrosine kinase receptors and profibrotic mediators such as transforming growth factor-β (TGF-β) 	<ul style="list-style-type: none"> Slows down the annual decline of forced vital capacity (FVC) 	<ul style="list-style-type: none"> Only oral administration Hepatotoxic and increases risk of bleeding. Costly 	<ul style="list-style-type: none"> NCT04619680 (Recruiting) NCT04541680 (Recruiting)
Pirfenidone ^[45-54]	<ul style="list-style-type: none"> Disrupts transforming growth factor-β (TGF-β) signaling pathway which decreases fibroblast/myofibroblast activity. Decreases procollagen gene expression 	<ul style="list-style-type: none"> Slows down the annual decline of forced vital capacity (FVC) 	<ul style="list-style-type: none"> Only oral administration Hepatotoxic and increases risk of bleeding. Costly 	<ul style="list-style-type: none"> NCT04607928 (Recruiting) NCT04282902 (Recruiting)

Targeting TGF- β Pathway Using Curcumin for Post-ARDS-Fibrosis

A. Curcumin

Since the beginning of the disease outbreak in December 2019, various types of drugs have been investigated for potential use in COVID-19 patients including active compounds derived from plants. A meta-analysis of randomized controlled trials by Ang *et al.* provided evidence that combination therapy using herbal and Western medicine speeds up recovery and symptom relief for patients with COVID-19.^[58]

As herbal medicine contains certain active compounds that possess therapeutic potential, there is good rationale to utilize it to target a specific pathway involved in the disease development. Targeting TGF- β signaling pathway, which is a vital mediator in post-COVID-19 pulmonary fibrosis development, may actually be the answer. This mechanism of action is seen in curcumin, a natural polyphenolic compound with antifibrotic properties seen in a mouse model of viral-induced ARDS. Although curcumin does not modulate the expression of TGF- β 1, it works by significantly decreasing the expression of TGF- β receptor II, a receptor for TGF- β signaling pathway.^[59] Another research using a different pulmonary fibrosis model showed that curcumin reduces the expression of TGF- β protein or mRNA.^[60] Altogether, these mechanisms of action prevent TGF- β -mediated pulmonary fibrosis.^[61] Other than inhibiting TGF- β , curcumin also manages fibrosis through other pathways. Since collagen deposits are found in pulmonary fibrosis, curcumin can delay fibrosis progression by inhibiting the synthesis of collagen and procollagen I mRNA, thus decreasing collagen deposition. It also acts as an anti-inflammatory agent by decreasing the expression of cytokines (e.g., IL-1, IL-6), inflammatory mediators (NF- κ B, TNF- α , MMP-2, MMP-9, IFN- γ), infiltration of macrophages and lymphocytes, and subsequent fibrosis.^[60,62] Curcumin also gives strong inhibitory effect on bradykinin, which is involved in inflammatory events affecting the airway.^[63] Recent study by Boozari *et al.* further added that curcumin inhibits several extracellular toll-like receptors (TLR2, TLR4, TLR8) and intracellular toll-like receptor (TLR9).^[64]

Additional benefits of curcumin include antioxidant and antiviral properties. It also helps with pulmonary exudation and edema caused by inflammation.^[60,62] In fact, a number of *in vitro* and *in vivo* studies already studied the pleiotropic effect of curcumin on many diseases including prostate cancer, liver cancer, lung cancer, neurodegenerative disorders, and cardiovascular diseases.^[65] Overall, curcumin's pharmacodynamic profile

makes it suitable for the treatment of post-ARDS fibrosis, especially in the context of SARS-CoV-2 infection.

B. Benefits of Inhaled Curcumin Nanoformulations Over Current Treatments

In addition to its excellent pharmacodynamic profile, curcumin must be considered as an alternative treatment for post-ARDS fibrosis due to its wide availability particularly in Asian countries. Curcumin is mostly extracted from turmeric (*Curcuma longa*; *kunyit*), a flowering plant of the ginger family. The compound is recognized as generally safe with minimum toxicity profile, and its cheap cost further supports its use in herbal treatment.^[66] Despite its promising potential, therapeutic application of curcumin is still limited due to its poor oral bioavailability, hydrophobicity, and rapid metabolism in intestine and liver.^[67,68] It was discovered that curcumin only became detectable in the patient's serum in small amounts after its oral dosage was increased to 12 grams.^[68] Yang *et al.* suggested that curcumin's bioavailability is only 1% with a half-life of approximately 28.1 minutes.^[69] A recent review on curcumin's role as adjuvant therapy in COVID-19 case suggested that the optimal dose for curcumin administration must be high enough to exert its therapeutic actions but not too high that it becomes counterproductive.^[70] Preliminary evidence hinted that a curcumin dose as high as more than 7,500 mg/day may actually facilitate SARS-CoV-2 entry by upregulating ACE2 expression.^[71]

Therefore, researchers have begun experimenting with different formulations of curcumin *in vitro* and *in vivo* with hopes of enhancing its bioavailability. These formulations are known as curcumin nanoformulations, which refer to the process of complexing curcumin with small, nanoscale-sized molecules. Nanoparticle formulation as a drug delivery method is deemed revolutionary as it can stabilize an active compound in a physiological environment, increase its cellular uptake and bioavailability, and eventually make the drug more potent.^[67] For example, Arozal *et al.* recently developed a chitosan-sodium tripolyphosphate-based curcumin nanoformulation that resulted in higher curcumin plasma concentration and lower clearance.^[65] Previous studies have also studied the biological activity of another curcumin nanoformulation called cyclodextrin-curcumin complex (CDC) in lung tumor as well as other cancers, and all results were consistent in stating that CDC improved curcumin's cellular uptake, half-life, water solubility, and pharmacological properties (Figure 3A, B).^[67,68] The prospect of using nanoparticle-based drug appeals to researchers as its synthesis is relatively easy, straightforward, and cheap.^[67]

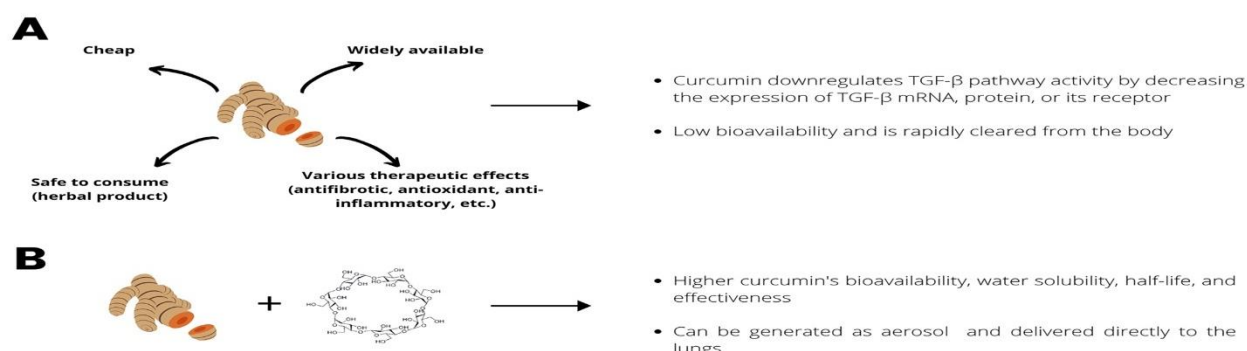


Figure 3: Curcumin's strengths, weaknesses, and mode of action in the treatment of pulmonary fibrosis **(A)**. Encapsulating curcumin in cyclodextrin (CDC), an example of curcumin nanoformulation, can optimize its use **(B)**.

Not only does it come with simplicity and low cost, but curcumin nanoformulations may also be a breakthrough antifibrotic treatment for severe COVID-19 ARDS where nintedanib and pirfenidone fail. Results from an autopsy study of 159 patients with ARDS concluded that oral antifibrotic therapy should ideally be administered within the first week of ARDS onset.^[72] Nevertheless, COVID-19 patients who exhibit signs of ARDS will most likely be put on mechanical ventilation immediately. These patients are obviously unable to take either nintedanib or pirfenidone, and intravenous access may already be occupied by other drugs such as analgesics and sedatives. As a result, inhaled drug administration becomes the main delivery route for mechanically ventilated patients; recent survey documented that 99% intensivists utilize aerosol therapy via either nebulizer or metered-dose inhaler.^[73] Curcumin nanoformulations can be generated as aerosol and later loaded into a nebulizer or inhaler.^[74,75] These devices can be attached to the ventilator circuit used in mechanical ventilation.^[75]

The first documented attempt to investigate the effectiveness of inhaled curcumin nanoformulations was done by Suresh *et al.*, who found a marked improvement in acute lung injury in mouse models through pulmonary administration of CDC.^[74] The researchers specifically designed the nanoformulation for inhalation purpose by mixing curcumin with hydroxypropyl- γ -cyclodextrin at high pH. *In vitro* experiment using Calu-3 human airway epithelial cell line proved that CDC had higher cellular permeability coefficient (P_{eff}) and better preservation of epithelial barrier than curcumin alone, and this was confirmed *in vivo* using fluorescent microscopy of the mice lungs after CDC administration. Furthermore, direct pulmonary administration of CDC reduced severity of acute lung injury by alleviating oxidative stress and proinflammatory factor NF- κ B activity. To test for any possible systemic toxicity, the researchers gave high doses of intravenous CDC (10 mg/kg twice daily) to dogs for 14 days and found no adverse effect. This suggests that CDC is safer for use in ARDS unlike systemic anti-inflammatory agents such as corticosteroids.^[74] More recently, Zhang *et al.* studied how pulmonary administration of a novel water-soluble CDC affects the lungs of mice infected with *Klebsiella pneumoniae*.^[76] These gram-negative bacteria are one of the most common pathogens that lead to ARDS. The study found that the intervention was significantly correlated with positive outcomes in the lungs such as reduced bacterial presence, lung injury, and oxidative stress. There was reduced expression of *Klebsiella* virulence factor (hemolysin), NF- κ B, hypoxia-inducible factors, and other inflammatory markers after CDC treatment. Lung compliance and epithelial lining were also better preserved in mice given pulmonary administration of CDC.^[76]

Another type of inhaled curcumin nanoformulation was studied by Hu *et al.* in mice models induced with idiopathic pulmonary fibrosis, which may share similar pathogenesis with post-COVID-19 pulmonary fibrosis.^[77] The researchers prepared large porous microparticles (LPMPs) loaded with curcumin and poly(lactic-co-glycolic) acid (PLGA) using evaporation/emulsion method. With a large mean geometric diameter (10 μ m) and small aerodynamic diameter (3.12 μ m), the LPMPs were big enough to escape alveolar macrophages but small enough to be abundantly deposited in the lung tissue. The findings showed that curcumin-PLGA LPMPs had higher antifibrotic activity, faster drug release, higher

distribution into lungs, and higher collagen inhibition compared to curcumin powder alone. Moreover, downstream signaling through NF- κ B and TGF- β 1 pathways was successfully suppressed by the LPMPs, strengthening evidence for its usefulness in pulmonary fibrosis management.^[77]

Up to present, however, there is no clear guideline regarding the recommended dosage of aerosol, choice of inhalation device, and timing of delivery.^[75] The safety profile for using inhaled curcumin nanoformulations in the treatment of pulmonary fibrosis models has also not been clearly defined. Since fibrosis is a chronic and progressive disease, future studies should investigate long-term effects and any potential toxicity of inhaled curcumin nanoformulations in the body. It is also important to note that despite all the promising findings from oral, inhaled, and other forms of curcumin nanoformulations for various diseases, most of the studies were done in pre-clinical models such as animals. There is no record of any clinical trial involving the use of inhaled curcumin nanoformulations, although clinical trials using oral or other forms of curcumin nanoformulations have been conducted in several countries including India and Iran.^[67] However, the number of patients included is still limited and none has been specifically intended for targeting TGF- β in pulmonary fibrosis. Therefore, at present, it is still too early to claim the superiority of inhaled curcumin nanoformulations over more conventional drugs for the targeted treatment of human diseases including post-COVID-19 fibrosis. Nevertheless, this review helps provide an understanding of how conjugation of curcumin with nanoparticles can be considered for alternative treatment for post-COVID-19 fibrosis, especially when conventional drugs fail.

CONCLUSION

In conclusion, a substantial portion of severe COVID-19 patients will develop ARDS, which is a predisposing factor for pulmonary fibrosis. ARDS is caused by cytokine storm as a result of immune system dysregulation. Following persistent inflammation, fibroproliferation activity will be increased. Among the various profibrotic pathways, TGF- β is the most important mediator and is actually up-regulated in the lungs following SARS-CoV-2 infection. Recent studies have suggested the use of corticosteroid, nintedanib, or pirfenidone for post-COVID-19 fibrosis, but there are limitations regarding either their cost, safety, accessibility, or route of administration. Curcumin should therefore be explored as an alternative treatment for the condition given its wide acceptability, low cost, and suitable pharmacodynamic profile, particularly its antifibrotic properties specific to TGF- β . Encapsulating curcumin in nanoformulations such as cyclodextrin in animal and cell line models of ARDS and pulmonary fibrosis have been shown to be safe and actually increases the active compound's bioavailability and efficacy. Since curcumin nanoformulations can be administered via aerosol inhalation, they may enable severe COVID-19 patients to receive timely antifibrotic therapy despite being mechanically ventilated. We suggest further research, especially in clinical settings, regarding efficacy and safety studies, recommended dosing and timing of aerosol therapy, as well as long-term effects of inhaled curcumin nanoformulations in the body.

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CONFLICT OF INTEREST

We declare there is no conflict of interest.

Authors contributions

ECY, MMK, and APK conceptualized the idea for the review, performed the literature search, and drafted the manuscript. ML and WA reviewed the manuscript, suggested changes, and helped edit the manuscript for final submission. All authors approved of the final manuscript for publication. All authors have made substantive contribution to this study and/or manuscript, and all have reviewed the final paper.

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