

# Paxlovid (Nirmatrelvir/Ritonavir): An Innovative Method for Treating Covid-19?

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## ABSTRACT

Nirmatrelvir Ritonavir 3-C like protease COVID-19 SARS-COV-2 Despite the need for innovative, efficient treatment for the Covid Pandemic, there is currently no curative regimen available, leaving patients to rely on supportive and general treatments. The 3C-like protease (3CLpro) and the main protease (Mpro), two SARS-COV-2 proteins, have been identified as potential targets for antiviral medications. The Mpro might be viable therapeutic target since it plays a significant role in both the pathogenesis of the virus and processing. Nirmatrelvir, an antiviral medication, prevents SARS- CoV-2 from reproducing by blocking Mpro. Another HIV protease inhibitor, ritonavir, was coupled with nirmatrelvir to develop. Ritonavir inhibits the cytochrome P450 3A metabolizing enzyme, extending the half- life of nirmatrelvir and serving as a pharmacological enhancer. Despite major modification of the SARS-COV-2 viral genome, nirmatrelvir still has strong antiviral effectiveness against contemporary coronavirus strains. However, there are still a number of open issues.

The effectiveness of nirmatrelvir and ritonavir in treating SARS-COV-2 infection, as well as their safety and potential adverse effects are discussed in this review of the literature.

## 1. Introduction

The severe acute respiratory Syndrome coronavirus 2 (SARS-COV-2, previously 2019nCov), an enveloped positive sense single-stranded RNA is the causative agent of the coronavirus disease 2019 (COVID 19) pandemic. According to taxonomy, SARS-COV-2 is the member of the Coronaviridae family, which encompasses a wide range of species that may cause moderate to severe human illnesses as well as numerous animal diseases. It also belongs to the Betacoronavirus genus and the Sabercovirus subgenus. Middle East respiratory syndrome (MERS)- COV-1, 229E, NL63, L OC43, HKU1, AND OC43 were the first six human coronaviruses to be found; SARS-COV 2 is now the monoclonal antibodies, different antiviral medications, immunomodulatory medicines, and other therapies for COVID-19 have all been suggested. However, one or more of these issues- low effectiveness, high cost, and hazardous side effects- have prevented these medicines from being widely used.

According to reports, Pfizer's brand-new antiviral medication "Paxlovid" performed well against COVID-19. The medicine is sold under the brand name Paxlovid<sup>TM</sup> and is composed of the generic drugs nirmatrelvir and ritonavir. Nirmatrelvir (300 mg; two 150 mg tablets) and ritonavir (100 mg; one 100 mg tablet) should be taken combined orally once every 12 hours for five days. Nirmatrelvir and ritonavir are

the two antiviral protease inhibitors that make up nirmatrelvir/ ritonavir. Nirmatrelvir is a novel SARS-CoV-2 inhibitor of the 3CLpro enzyme, whereas ritonavir is an HIV protease inhibitor. Ritonavir inhibits the enzyme responsible for Nirmatrelvir metabolism, extending Nirmatrelvir presence in the body and increasing its activity. Adults with COVID-19 who were symptomatic but did not require hospitalization were at risk.

## 2. Nirmatrelvir and variations of the SARS-COV 2

When creating medicinal medicines to combat SARS-CoV-2, two promising compounds to target are the papain-like protease (PLpro) and the 3CLpro (also known as Mpro). Both of these cysteine proteases are crucial for the survival of the SARS-CoV-2. Because mammalian protease enzymes have different substrate specificities, antiviral protease inhibitors may be relatively safe medications for humans. The cleavage and maturation of proteins to render them functional for the viral replication processes need to enzyme 3CLpro (cysteine protease; EC 3.4.22.69) in particular. The enzyme that catalyzes protein cleavage, which is defined by a catalytic dyad of Cys145 and His41, has been the focus of several medicinal chemistry techniques.

Nirmatrelvir is a protease inhibitor that may be used orally and was found in 2021 as a means of creating SARS-CoV-2 3CLpro inhibitors. It binds to the cited dyad of Mpro from its nitrile site. Ritonavir is a tripeptide that binds to the HIV protease's active site to block it. The literature states that the half life of nirmatrelvir was 5.1 hours in rats and 0.8 hours in humans and in primates.

Nirmatrelvir has a moderate bioavailability in rats (34-50%), compared to a relatively low bioavailability in monkeys (8.5%) when administered orally. This is partly because this species engages in oxidative metabolism throughout the gastrointestinal system. Nirmatrelvir generally had modest plasma protein binding, with the average fraction of its free plasma level in rats, monkeys, and humans being 0.314-0.478. Nirmatrelvir was similarly metabolized by hepatocytes of these species, with cytochrome P450 (CYP450) serving as the primary enzyme that oxidized the drug's various functional groups. The primary mediator of Nir was CYP3A4.

Human oxidation of nirmatrelvir (0.99% percent metabolized).

Sandwich-cultured human and animal hepatocytes (between two collagen sheets) suggested that limited excretion of unaltered nirmatrelvir through the kidneys and bile was likely to occur. Studies conducted in vitro showed that nirmatrelvir could both enhance CYP3A activity and reversibly inhibit Mpro in a time dependent manner. When coadministered with ritonavir, which inhibits CYP3A4- the drug's primary metabolizer- the first research in humans revealed rising amounts of nirmatrelvir.

Nirmatrelvir can also effectively block the Mpro proteolytic activity in all seven varieties of human coronavirus, including betacoronaviruses as well as alphacoronaviruses. No inhibitory effect was seen against a number of mammalian proteases, including aspartyl, serine and cysteine proteases, as well as HIV protease, which is a retroviral aspartyl protease (retropepsin), at the highest tested dosage of nirmatrelvir (100M).

In a research investigation by Owen et al. Nirmatrelvir was delivered to human coronavirus-infected A549 and dNHBE cell lines (two cell lines derived from the human respiratory epithelium) in order to test the drug's effectiveness against these strains. Significant antiviral activity was demonstrated by nirmatrelvir against SARS-CoV-2, MERS-CoV, HCoV AND SARS-1 CoV-1; moreover, cytotoxic effects were not seen at dosage lower than 3M.

They also infected a module with SARS-CoV-2 to evaluate the in vivo effectiveness of nirmatrelvir. In

compared to control, nirmatrelvir dramatically reduced both weight loss and fatality rates.

Additionally, the nirmatrelvir group had considerably reduced virus titer levels that were detected in lungs of mice that had been put to death.

Nirmatrelvir was prescribed to high-risk COVID-19 patients in an emergency situation after the FDA authorized it in December 2021. After the illness has persisted for 5 days, nirmatrelvir may be begun, and it must be continued for 5 days straight. The EPIC-HR study evaluated the effectiveness of nirmatrelvir in people who met the specified criteria.

This study was conducted during the variant pandemic, therefore it lacks information on how nirmatrelvir affects adults who have already been exposed for SARS-COV-2 and during the time of omicron pandemic were recently assessed by Arbel et al.

They gave nirmatrelvir to the 3902 people who satisfied the inclusion criteria. Nirmatrelvir caused a significant lower admission and mortality rate of the individuals under 65 age old years when compared to the control, they found.

Nirmatrelvir did not significantly change the course of the illness in younger instances, albeit.

Additionally, Li et al. evaluated the effects of nirmatrelvir on Omicron infected Calu-3-Cells and discovered that even low concentration of the drug was able to suppress Omicron variant replication. Calu-3-Cells were then exposed to either the wild type (WT) or Omicron form of SARS-COV-2 after being treated with serum from covid 19 vaccinated individuals. Wild type COV-2 was unable.

In a separate experiment, Vangeel et al. assured that antiviral efficacy of nirmatrelvir against a number of distinct SARS-COV-2 variants of concern (VOCs) (alpha, beta, gamma, delta, and Omicron). They demonstrated the omicron and all other known VOCs, including demonstrated with the aid of the ORF ab program that the 3CLpro sequence contained two known amino acid alterations (K90R at position 3353 in Beta and P132H at position in Omicron), neither of which involved the 3CLpro active site and were therefore unlikely to affect the sensitivity to nirmatrelvir. Similar to this, they anticipated that nirmatrelvir would continue to be effective against the alpha, beta, gamma, and delta types.

It has been demonstrated that certain missense point mutations in SARS-CoV1 3CLpro, which shares 96% of the amino acid sequence with SARS-CoV-2 3CLpro, may influence the protease activity [20]. These known mutations [21–24] may result in slightly increased (S284, T285, I286) or greatly decreased (F140, R298, N28, G11, N214, S139, E166) catalytic activity. Three missense mutations in the Mpro region of the ORF1a/b gene with > 20% frequency were discovered by lineage comparison [25] of the SARS-CoV-2 genomic material [26]. Additionally, several of these mutations and SARS-CoV-2 variations may be connected, according to reports.

In four consecutive instances from a post-COVID cohort study, Peluso et al. recorded the outcomes following various nirmatrelvir medication regimens [29]. The first patient experienced a clinical relapse and ultimately contracted protracted COVID-19 despite receiving prompt antiviral therapy. Nirmatrelvir was administered to the two more patients 25 and 60 days following the onset of their COVID-19 symptoms, and both participants' conditions improved as a result, according to S.M.R. Hashemian et al. in *Biomedicine &*

*Pharmacotherapy* 162 (2023) 114367 3. The final patient, who had a fresh COVID superimposed on top of a probable lengthy COVID for two years, took nirmatrelvir and saw a considerable reduction in their chronic symptoms. In order to fully understand the impact of prompt nirmatrelvir therapy on the development of acute COVID into long COVID as well as its effects on long COVID itself, more study is necessary. [29].

The S (spike) protein of SARS-CoV-2 serves as a key antigen for the host immune system as a surface protein [30]. The S protein is under strong selection pressure, which causes variant mutations to emerge and reduce the efficacy of vaccines based on this protein [3]. resulting in a reduction in the efficacy of vaccinations based on this protein [31–34]. Mpro and RNAdependent RNA polymerase (RdRp) are two of these proteins that are primarily focused on in the research [35], which are likewise susceptible to potential alterations, and which may thus be better targets for combating this virus.

Fig. 1. Nirmatrelvir can inhibit 3CLpro, an enzyme involved in maturation of proteins, in different variants of SARS-CoV-2, and therefore, suppress their replication.

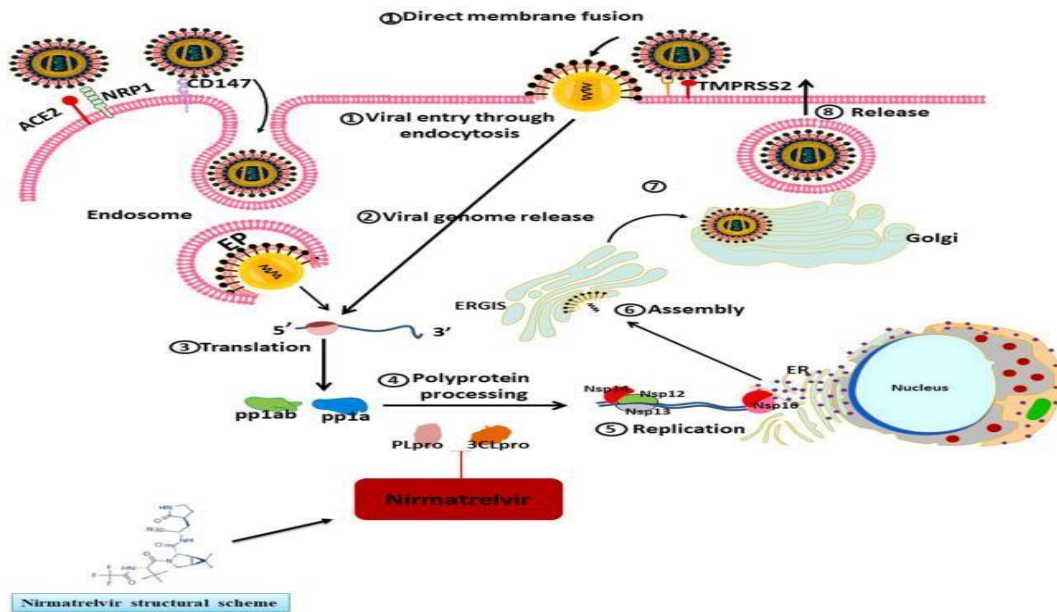
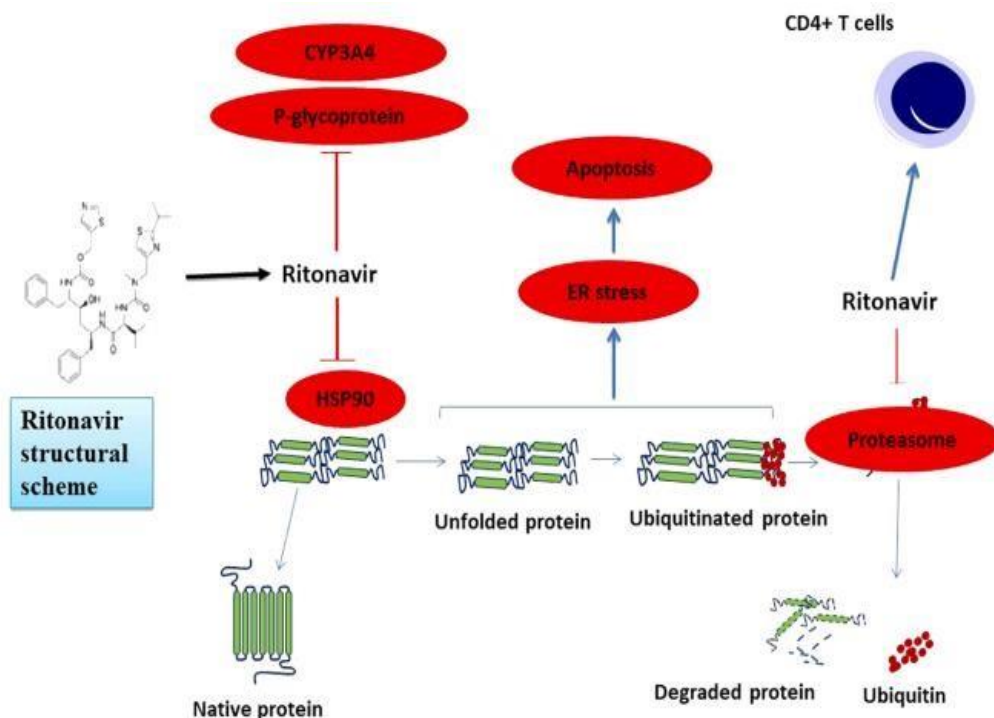


Fig. 2. Cellular processes affected by Ritonavir. Ritonavir may inhibit proteasomes, HSP90 (heat shock protein 90), CYP3A4, as well as P-glycoprotein; it may also modulate the function of immune cells [54].



### 3. COVID-19 therapeutic implications of ritonavir

Ritonavir was the second medication with protease inhibition characteristic that the FDA has approved for use in treating AIDS after saquinavir. Ritonavir can inhibit CYP450-3A4 in addition to the HIV protease, according to studies (Fig. 2). Its primary use nowadays is to boost the bioavailability of concurrently delivered antiretroviral medications (ARVs). When combined with triple antiHIV therapy, it led to higher CD4 + counts and lower HIV RNA levels in people who had never received treatment. [37]

It has been demonstrated that ritonavir improves the efficacy of several ARVs. For instance, in HIV-infected individuals who had received therapy in the past as well as those who had never had it, the combination of lopinavir and ritonavir effectively decreased viral load and improved immunological markers [38,39]. Ritonavir is being explored in conjunction with antineoplastic medicines to treat cancer due to its mechanisms of action. Additionally, the FDA has approved its use in conjunction with dasabuvir, ombitasvir, and paritaprevir for the treatment of HCV genotype 1. Lopinavir/ritonavir was also given FDA approval for HIV treatment.

Protease inhibitors are made more tolerable and effective by the use of pharmacologic augmentation. Ritonavir is the perfect pharmacologic enhancer since it inhibits two crucial metabolic pathways. First-pass metabolism, which happens during absorption, is inhibited by ritonavir. The enterocytes that line the intestine contain both CYP3A4, a crucial cytochrome P450 isoenzyme implicated in drug metabolism, and P-glycoprotein, a drug efflux transporter that pumps medicines out of the gut wall and back into the intestinal lumen [40].

First off, because ritonavir inhibits P-glycoprotein, it might raise the ARV drug's C<sub>max</sub>. Second, by inhibiting CYP3A4, ritonavir prolongs the drug's plasma half-life. Ritonavir may potentially inhibit the P-glycoprotein in CD4 + cells to increase the intracellular half-life of the ARV medication.

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#### **4. COVID-19 and nirmatrelvir/ritonavir treatment**

Najjar-Debbiny et al. enrolled people (18 years) who were experiencing COVID19 for the first time and had risk indicators for development into severe disease and followed them for 28 days during the Omicron pandemic to investigate nirmatrelvir/ritonavir effectiveness against SARS-CoV-2 [55]. In comparison to controls, they found that COVID-19 patients receiving nirmatrelvir/ritonavir had a significantly lower death rate and risk of disease progression into a severe stage. The effects were also more pronounced in patients with immunosuppression, neurological or cardiovascular comorbidities. They conclude that nirmatrelvir/ritonavir has the power to change the course of the disease and the death rate.

Malden et al. planned to assess nirmatrelvir/ritonavir's effectiveness in reducing the risk of the worrisome progression when the FDA authorized it in December 2021 for use in non-severe COVID-19 patients but in risk of advancing into severe illness [56]. They investigated whether patients required to be present in the healthcare system between 5 and 15 days after their nirmatrelvir/ritonavir treatment had ended by gathering the data of 5287 individuals who received it between December 2021 and May 2022. They noted that only 45 of these instances had been reported to the medical system, of which six required hospitalization. Additionally, they noted that around half of these 45 cases had either the risk factor of advanced age (65) or the existence of medical comorbidity.

Taken together, the low rate of further need for medical care found in their research points to nirmatrelvir/ritonavir's capacity to change the course of the illnesses [56].

In a similar retrospective study, Shah et al. examined data from 699,848 Americans diagnosed with COVID-19 in the spring and summer of 2022, and assessed the proportion of them who received nirmatrelvir/ritonavir, as well as how nirmatrelvir/ritonavir altered the course of disease. They found that nirmatrelvir/ritonavir, in general and with different demographic adjustments, was able to significantly lower the rate of hospital admission, which could be translated into its capacity to inhibit the progression toward severe COVID-19; as a result of their findings, they insisted on a more widespread use of this medication in outpatient settings.

#### **5. Nirmatrelvir/ritonavir safety and tolerability**

Nirmatrelvir/ritonavir appears to be effective in battling COVID19, although it is important to consider how it interacts with transplant medications [72]. Immunosuppression and vaccine failure are common in transplant patients, and these concomitant circumstances make them susceptible to the development of severe COVID-19 and mortality [73,74].

Ritonavir inhibits CYP3A, causing concentrations of medicines that are processed by CYP3A to increase by 1.8 to 20 times [75]. The dependency of these medications on CYP3A metabolism explains the abrupt rise in plasma levels of tacrolimus, cyclosporine, calcineurin inhibitors (C The tacrolimus molecule is broken down by intestinal cytochrome CYP3A enzymes, and a rapid increase in its concentration in the circulation might result in posterior reversible encephalopathy syndrome, seizures, renal damage, and deathNIs), or mTOR inhibitors: everolimus, and sirolimus in patients exposed to ritonavir.

In addition to the immunosuppressive medicines mentioned above, other drugs such as statins, calcium channel blockers, and warfarin must be considered while delivering nirmatrelvir/ritonavir. A more extensive list of potential medication interactions may be found in the FDA European Union Authorization document. Nirmatrelvir/ritonavir is a very new drug, therefore nothing is known about its long-term effectiveness or any potential side effects.

Nirmatrelvir/ritonavir, however, has been linked to research that suggest it may also cause typical adverse effects such as headache, emesis, loose stools, and a distorted perception of taste. Muscle pains and high blood pressure were sporadic side effects.

## 6. Conclusion

Nirmatrelvir appears to be an effective antiviral medication for COVID-19, either alone or in combination with ritonavir (Paxlovid), although there are still unsolved problems. To begin, the entire results of comprehensive clinical research have yet to be revealed. Second, it is critical to monitor nirmatrelvir's efficacy against emerging COVID-19 strains in the coming years. The virus's selection pressure can produce further mutations in the protease protein, resulting in a decrease in nirmatrelvir/ritonavir efficiency. Third, keep in mind the interactions with other medicines when considering ritonavir's inhibitory impact on CYP3A4. Despite these factors, available data from randomized studies indicates that nirmatrelvir/ritonavir is efficacious in treating COVID-19 while maintaining a tolerable safety profile, and with the drug's most notable side effects being a decreased risk of developing a serious illness and a higher survival rate. Nirmatrelvir/ritonavir is a helpful weapon in the battle against COVID19, especially for nations with a poor vaccination rate. However, further research is required to demonstrate its effectiveness in treating COVID-19. CRediT authorship contribution statement Hamed Mirzaei involved in conception, design, and drafting of the manuscript. Seyed Mohammad Reza Hashemian, Amirhossein Sheida, Mohammad Taghizadieh, Mohammad Yousef

Memar, Michael R Hamblin, Hossein Bannazadeh Baghi, Javid Sadri Nahand and Zatollah Asemi contributed in data collection and manuscript drafting. All authors approved the final version for submission. Conflict of interest statement The authors have no relevant financial or non-financial interests to disclose

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