Drug induced liver injury with licensed antiviral drugs for COVID-19: a narrative review.

Giacomo Casalini[a], Andrea Giacomelli[b] and Spinello Antinori[a,b]

[a] Department of Biomedical and Clinical Sciences, Università degli Studi di Milano, Milan, Italy.

[b] III Division of Infectious Diseases, ASST Fatebenefratelli-Sacco, Luigi Sacco Hospital, Milan, Italy.

Corresponding Author: Prof. Spinello Antinori, Department of Biomedical and Clinical Sciences, Università degli Studi di Milano, Via GB Grassi 74, 20157 Milano, Italy. spinello.antinori@unimi.it.

Keywords: COVID-19, SARS-CoV-2; liver toxicity; remdesivir; molnupiravir; nirmatrelvir/ritonavir; transaminases; non-hospitalized.

Article highlights

- Liver involvement in COVID-19 is multifactorial and drug-induced liver injury plays a significant role.
- This is a narrative review about the hepatic safety profile of the three antivirals licensed for COVID-19 treatment. We used data about hepatobiliary adverse events from English-language randomized clinical trial.
- Remdesivir is potentially hepatotoxic, but liver enzymes abnormalities are usually mild and transient. Liver biochemistry monitoring should be considered especially in patients with pre-existing liver disease and at high risk of liver injury.
- Molnupiravir ha a favourable safety profile and the incidence of liver toxicity with this drug is low in randomized clinical trial.
- Ritonavir-boosted nirmatrelvir can induce liver damage due to the ritonavir component but the only randomized clinical trial didn't reveal any safety issues.
- More studies are needed on special populations (i.e., patients with stable chronic liver disease) and treatment monitoring should be tailored according to individual risk factor for drug-induced liver injury.

Abstract

Introduction. Liver involvement in COVID-19 is usually multifactorial and the three potential mechanisms are direct hepatocyte viral damage, vascular or cellular damage during the cytokine storm of severe COVID-19 and drug-induced liver injury (DILI). To date, three antivirals are licensed for the treatment of COVID-19 by most guidelines: remdesivir, molnupiravir and ritonavir-boosted nirmatrelvir.

Areas covered. We performed a narrative review about the hepatic safety profile of the three antivirals licensed for COVID-19 treatment. We used data about hepatobiliary adverse events from English-language randomized clinical trial.

Expert opinion. Remdesivir was found to be potentially hepatotoxic and abnormalities in liver biochemistry are common (2-34%) but mild and reversible. Molnupiravir exhibits a favourable safety profile and elevations of aminotransferases is usually mild and reversible (up to 11% of patients in one study). Ritonavir-boosted nirmatrelvir is potentially hepatotoxic but in the only phase 3 RCT there were no safety issues and AST/ALT levels increase did not exceed 2.4% of patients. All antivirals have a favourable safety profile, but they are not sufficiently studied in patients with underlying chronic kidney or liver disease. In this special populations, antivirals should be used with caution and careful monitoring during treatment should be pursued on a case-by-case basis.

1. Background

The Coronavirus Disease 2019 (COVID-19) caused by SARS-CoV-2 emerged initially as a primarily pulmonary disease leading to bilateral interstitial pneumonia and potentially to severe respiratory failure. However, it became clear that COVID-19, especially the most severe form, was a systemic rather than an exclusively pulmonary disease, and several studies showed that many organs and systems could be affected, including the liver and the biliary tract [1–3]. Liver involvement in COVID-19 is probably the results of at least three mechanisms: SARS-CoV-2 induced direct damage to hepatocytes and cholangiocytes [4,5], inflammation-induced vascular and cellular damage [6,7] and, finally, drug-induced liver injury (DILI) [8]. The severity spectrum of liver damage in COVID-19 is wide, ranging from mild/moderate and reversible increase in aspartate aminotransferase (AST), alanine aminotransferase (ALT) and bilirubin levels (up to 50% of patients) to uncommon cases of acute liver failure [9]. A pre-existing liver disease is associated with worse outcome from

COVID-19 and the greater the severity of the underlying liver disease, the higher is the risk of mortality [10,11].

Since the beginning of the pandemic, many candidate drugs have been tested in patients with COVID-19 and, most of these drugs (i.e., remdesivir, lopinavir/ritonavir, tocilizumab, interferon, or dexamethasone) were previously used to treat other diseases and their hepatotoxic potential was already known [12]. Studies performed to test candidate drugs in COVID-19 patients report a number of hepatic/biliary adverse event of different degrees, but the causal relationship with the study drug was not always clear. A systematic review and metanalysis from 2020 showed that the pooled incidence of DILI in COVID-19 patients was 25.4% [95% Confidence Interval (CI), 14.2-41.4], ranging from 15.2% (95% CI, 6.4-32; patients treated with remdesivir) to 37.2% (95% CI, 22.7-54.6; patients treated with lopinavir/ritonavir) [13]. Following the publication of randomized clinical trial, the COVID-19 therapeutic armamentarium has been progressively defined and, to date, three antivirals are licensed by the U.S Food and Drug Administration (FDA) for COVID-19 treatment: remdesivir, molnupiravir and ritonavir-boosted nirmatrelvir [14].

Many reviews have been published focusing on liver involvement in COVID-19 and, some of them, also evaluated the impact of drug toxicity. However, most of these studies report data also from non-interventional studies and discuss the hepatic safety profile of candidates drugs that failed to prove their efficacy in COVID-19 patients [8,13]. Furthermore, few studies evaluated the use of antivirals in special populations, such as patients with chronic liver disease.

The scope of this narrative review is to discuss the hepatic safety profile of the three licensed antiviral drugs for COVID-19 treatment focusing on data reported in clinical trials, which represent the highest level of evidence available

2. Methods

We searched the PubMed, Scopus and ClinicalTrials.gov database for clinical trial that investigated the following antivirals in adults subjects: remdesivir, molnupiravir and ritonavir-boosted nirmatrelvir. The following terms were used: remdesivir OR GS-5734 OR molnupiravir OR MK-4482 OR nirmatrelvir OR PF-07321332 OR Paxlovid AND clinical trial OR randomized studies. Studies published between January 1st, 2020, and August 31st, 2022, were considered. We found more than 3000 studies. For the purpose of this review,

we selected only randomized clinical trial which reported data about drug safety, in particular hepatobiliary adverse events. Only article in English language were selected. Retrospective observational studies, case reports or non-randomized studies were excluded. Finally, we also searched the aforementioned databases to find ongoing or scheduled prospective studies about the safety of these antivirals in patients with underlying special medical conditions (in particular chronic liver or kidney disease) which could increase the risk of DILI

3. Remdesivir

3.1 Introduction

Remdesivir is an RNA-dependent RNA polymerase (RdRp) inhibitor which is administered intravenously as a prodrug that is metabolized into an active compound by phosphorylation. Early during the pandemic, the drug showed in-vitro activity against SARS-CoV-2 [15]. Subsequent studies showed that remdesivir is probably active against the Omicron (B.1.1.529) variant of concern and its subvariants BA.1, BA.2, BA.4 and BA.5 [16–18] Remdesivir is FDA-approved for the treatment of non-hospitalized patients at high risk of disease progression without supplemental oxygen (3 days of therapy) and hospitalized patients who require conventional oxygen supplementation (for 5 days or until hospital discharge, whichever comes first) [14]. Several RCTs investigating remdesivir are available and, most of them, report data about DILI. Patients with baseline AST or ALT level of at least 3-5 times the upper reference limit (URL) are usually excluded from RTCs [19–29]; only the trial performed by Wang and colleagues also excluded patients with liver cirrhosis [21].

3.2 DILI from randomized clinical trial

Spinner et al. conducted a multinational, open-label RCT of 10 days or 5 days of remdesivir compared with standard of care in hospitalized patients with moderate COVID-19 in Asia, Europe, and the United States. More than half patients in each group had a baseline cardiovascular disease, most patients (84% in both remdesivir arm) were hospitalized without requiring supplemental oxygen and the most common concomitant medications were steroids (15% in the 10-day arm and 17% in the 5-day arm). Patients in the 10-day group experienced significantly more adverse events (AEs) than the standard group [12.0% difference; 95% CI, 1.6%-21.8%; p = .02], but aminotransferases increase of any grade

was not more common in the remdesivir arms (about one third of patients in each arm). A grade 3 increase (i.e., an increase of 5 to 10 times the URL) was more common for ALT levels than AST levels in patient treated with remdesivir (10/356, 2.8% vs. 5/352, 1,4%) [19]. In the open-label multinational RCT of Goldman and colleagues, a course of 5 days or 10 days of remdesivir was evaluated in patients with severe COVID-19, not on mechanical ventilation or extracorporeal membrane oxygenation. At baseline, about half of patients were hospitalized requiring low-flow supplemental oxygen (56% and 54% in the 5-day and 10-day arm, respectively). The percentages of patients with AEs were similar in the two groups (70% and 74%, respectively). ALT increase occurred in 6% (5-day arm) and 8% (10-day arm) of patients, while the frequency of AST elevation was 5% and 7%, respectively. ALT elevation of at least grade 3 occurred in 12/194 (6.2%, 5-day) and 16/191 (8.4%, 10-day) patients. AST elevation of at least grade 3 occurred in 14/194 (7.2%, 5-day) and 11/190 (5.8%, 10-day) patients. Bilirubin elevation of at least grade 3 (i.e., an increase of 2.6 to 5 times the URL) was uncommon (4 cases in the 10-day arm, 1 case in the 5-day arm). Aminotransferase increase was the reason of discontinuing remdesivir in 2.5% and 3.6% of patients in the 5-day and 10-day groups, respectively [20]. In the placebo-controlled double-blinded RCT conducted on 255 patients by Wang et al., at baseline more than 80% of patients were hospitalized requiring low-flow supplemental oxygen. Seventeen percent of patients in the remdesivir arm and 19% in the placebo arm were treated with lopinavir/ritonavir. The most common liver adverse event was hyperbilirubinemia: 10% in the remdesivir arm (causing drug discontinuation in one case), and 9% in the placebo arm. Median ALT and AST values were comparable at baseline among the study arms. Grade 1-2 AST elevation (i.e., an increase of 1.25 to 5 times the URL) was more common in the placebo group (12% vs. 7%) and led to remdesivir discontinuation in 3 cases. Overall, remdesivir was stopped early because of AEs in 18 (12%) patients versus 4 (5%) patients in the placebo group [21]. In the Adaptive Covid-19 Treatment Trial (ACTT-1), a double-blind, placebo-controlled trial of remdesivir in 1,062 hospitalized patients with COVID-19 in 10 countries, 26.8% of patients were hospitalized receiving mechanical ventilation or extra-corporeal membrane oxygenation (ECMO) at baseline. The combined number of participants with increased AST and ALT was 32 (6%) in the remdesivir arm and 55 (10.7%) in the placebo arm, while hyperbilirubinemia was uncommon (less than 1% of patients in both study groups). Liver-related serious AEs were not reported. Of note, prothrombin time was prolonged in 4.9% of

patients treated with remdesivir (1.6% in the placebo group) [22]. Remdesivir safety was also evaluated in a small, open-label, phase 3 trial in hospitalized patients with moderate COVID-19, remdesivir safety was evaluated patients were randomly assigned to receive remdesivir (5 or 10 days) or standard of care (SoC). Remdesivir-related AEs were 36 in the 5-day arm (19%) and 24 in the 10-day arm (12%). Overall, ALT, AST, and bilirubin median levels at the end of treatment didn't change significantly from the baseline, neither in the 10-day arm or the 5-day arm [AST change: 2.4 U/L (-7.2, 13) in the 5 days arm, 5.1 U/L (-7.8, 12.8) in the 10-day arm, 0.4 U/L (-9.7, 12.8) in the SoC arm; ALT change: 6 U/L (-4, 15) in the 5-day arm, 4 U/L (-6, 9) in the 10-day arm, 1 U/L (-11, 44) in the SoC arm; bilirubin change: 0.1 mg/dL (-0.1, 0.2) in the 5-day arm, 0.1 mg/dL (-0.07, 0.2) in the 10-day arm, 0 mg/dL in the SoC arm]. The elevation of AST plus ALT levels, ALT levels alone and alkaline phosphatase were responsible for remdesivir discontinuation in the 10-day arm in four, one and one patient, respectively (overall, 3% of patients in the 10-day arm). ALT increase was the cause of remdesivir discontinuation in the 5-day arm [30]. The DisCoVeRy study, an open-label, adaptive RCT of remdesivir in hospitalized patients with moderate or severe COVID-19 in Europe, compared remdesivir plus standard of care (SoC) versus SoC alone (SoC included corticosteroids and anticoagulants). Obesity was the most common coexisting condition (34% of patients) and 39% of patients had a severe disease at baseline. AEs or serious AEs were similar among the study groups. Transaminases increase was observed in 11 patients in the remdesivir group (3%, 10 patients with ALT increase) and in 3 patients in the SoC group (1%, ALT in all cases). One patient in the remdesivir arm died because of hepatorenal syndrome and this adverse event was attributed to the study drug by investigators and sponsor's safety team [23]. In the multicentre, open-label, pragmatic RCT of remdesivir in hospitalized patients with COVID-19 in Canada (CATCO), which enrolled 1,282 hospitalized, compared a 10-day regimen of remdesivir with SoC. Baseline renal or hepatic dysfunction were not exclusion criteria. The prevalence of chronic liver disease was 1.7% in the remdesivir arm and 3.9% in the SoC arm. More than 87% of patients in both arms received concomitant therapy with steroids. A new hepatic disfunction, defined as acute liver clinically dysfunction, or ALT levels at day 5 at least twice ALT levels at day 1, occurred in 82 (13.1%) patients in the remdesivir arm and 88 (13.7%) patients in the SoC arm (RR 0.96, C.I. 0.72-1.26). Among patients with new hepatic disfunction, non-survivors were 8 (6.8%) in the remdesivir arm and 21 (14.7%) in the SoC arm (RR 0.47, C.I. 0.21-1.01, *p*

0.04) [31]. Mahajan et al. conducted a small trial comparing remdesivir to SoC in 82 patients over 40 years old with moderate to severe COVID-19. More than two third of patients were receiving low-flow supplemental oxygen at baseline. Mean AST and ALT levels, at baseline and after treatment, were not significantly different (for example, in the remdesivir arm ALT levels at baseline and at the end of treatment were 38.94 ± 13.4 U/L and 39.01 ± 12.03 U/L respectively, *p* 0.67). However, neither the frequency of AST/ALT increase during treatment, nor the degree of increase, are reported [24]. In the double-blind, randomized, placebo-controlled trial conducted by Kalil and colleagues on patients with COVID-19 pneumonia, not on mechanical ventilation, 969 patients were randomly assigned to receive a 10-day course remdesivir alone or in combination with subcutaneous interferon beta-1a. Seventy-seven percent of patients received low-flow supplemental oxygen at baseline. Although this trial was not designed to evaluate remdesivir safety or efficacy, in reports detailed data about liver enzymes trend during treatment. Patients in both arms experienced a mild increase in ALT during treatment course, which peaked at day 5 in the remdesivir arm (mean ALT value of 55.9 U/L, C.I. 49.1-61.9; mean increase of 13.5 U/L from baseline, C.I. 9-18). ALT increase was of a greater magnitude in the remdesivir + interferon arm, as expected by the well-known safety profile of interferon beta-1a. AST levels had an opposite trend, decreasing from baseline of a mean of 5.2 U/L (C.I. -8.6-1.7) in the remdesivir arm at day 5. Serious liver-related adverse event (AST and ALT increase) was reported in only two patients in the remdesivir + interferon arm [32]. The WHO Solidarity trial is one of the largest RCTs which evaluated remdesivir in patients with COVID-19, but elevated liver enzymes were not an exclusion criteria. Unfortunately, data about liver AEs are not reported [33]. In the paper which showed the interim results of this study, the cause of death of patients assigned to the study drug was not liver-related [34]. NOR-Solidarity was an independent, add-on, randomized controlled trial to the WHO Solidarity trial which included additional clinical and biochemistry data collection [27]. Patients were randomised to SoC alone, SoC plus 10-day remdesivir or SoC plus 10-day hydroxychloroquine. Only 4.5% of patients received concomitant systemic steroids and 5.5% of patients were admitted to intensive care unit at baseline. Thirty-four AEs were reported in the remdesivir arm, but none of them was a hepatobiliary disorder [27]. In the ACCT-2 double-blind, randomized, placebo-controlled trial which evaluated baricitinib plus remdesivir in hospitalized adults with COVID-19, 68% of patients had moderate disease at baseline. The number of

patients who had an increase in ALT, AST and bilirubin levels was 6 (1.8%), 16 (3.1%) and 8 (1.6%) in the remdesivir plus placebo arm, and the frequencies were similar in the baricitinib + remdesivir arm (see Table 1 for details). We must note that this trial was not designed to evaluate remdesivir efficacy or safety [28]. NCT04409262 is a phase 3 RCT which will evaluate the efficacy and safety of remdesivir plus tocilizumab compared with remdesivir plus placebo in hospitalized patients with COVID-19 pneumonia; recruitment has terminated, and preliminary results are available. Transaminases increase is reported as an AE in 25 (5.8%) and 10 (4.7%) patients in the remdesivir plus tocilizumab and remdesivir plus placebo arm, respectively. Acute liver failure is reported in one patient per arm [29].

Finally, a 3-day course of remdesivir was evaluated in a randomized, double-blind, placebo-controlled trial on symptomatic, non-hospitalized patients with COVID-19 who were at high risk for disease progression. Median age was 50 years old, 62% of patients had diabetes and only two patients (0.4%) had underlying chronic liver disease (one per arm). A total of 562 patients were randomized to receive remdesivir or placebo within 7 days of COVID-19 symptoms onset. At day 14, the mean change from baseline in ALT levels was minimal and comparable between the study groups (−3.0 ± 21.6 U/L in the remdesivir group and −1.0 ± 27.4 U/L in the placebo group) [26]. See Table 1 for further details.

4. Molnupiravir

4.1 Introduction

Molnupiravir is the oral prodrug of beta-D-N4-hydroxycytidine (NHC), a ribonucleoside that, following phosphorylation by host cell kinases, acts as competitive inhibitor of the viral RdRp. Molnupiravir showed broad antiviral activity against RNA viruses, including SARS-CoV-2 [35]. Despite initial concerns on the mutagenic potential of this drug, molnupiravir was approved by FDA for the treatment of adults with early mild to moderate COVID-19, who are at high risk of progressing to severe disease, and for whom alternative antiviral therapies are not accessible or clinically appropriate [14]. Molnupiravir is expected to be active against the B.1.1.529 (Omicron) variant of concern and its known subvariants [16–18].

4.2 DILI from randomized clinical trial

Molnupiravir was first evaluated in a phase 1 randomized, double-blind, placebo-controlled trial (NCT04392219) which enrolled healthy volunteers. Single and multiple dosing of molnupiravir were found to be well tolerated, liver-related adverse events were not reported and only one patient discontinued treatment with molnupiravir because of a skin rash [36]. In a phase Ib, open-label, dose-escalating, randomized controlled study, molnupiravir was well-tolerated irrespective of the dosage and all the AEs were grade 1 or 2. Among the 4 patients treated with 600 mg twice daily, one patient experienced increase in ALT and gamma glutamyl transferase (GGT) elevation, which it was not observed in the patients who received the dosage of 800 mg or 300 mg twice daily. However, it was not specified if ALT or GGT elevation was grade 1 or 2 [37]. In a phase 2a randomized, double-blind, placebo-controlled trial of molnupiravir in non-hospitalized adults with recently diagnosed COVID-19, at the dosage of 800 mg twice daily AST and ALT increase was uncommon (2 out of 55 patients, 3.6%) with alkaline phosphatase increase observed in only one patient (1.8%) [38]. In the phase 3, double-blind, randomized, placebo-controlled trial MOVe-OUT, which evaluated the efficacy and safety of treatment with molnupiravir in patients with early COVID-19 (within 5 days of symptoms onset), 73.7% of patients were obese and most patients had mild to moderate disease. There were no significant differences between the study groups in the frequency of AEs or severe AEs. For example, the number of patients who experienced at least one AEs related to the assigned regimen was 57 (8%) in the molnupiravir arm and 59 (8.4%) in the placebo arm (estimated difference -0.4, 95% C.I. -3.3 to 2.5). However, data about liver enzymes elevation are not specifically reported [39]. In a more recent, little, randomized, placebo-controlled trial involving hospitalized patients with mild or moderate COVID-19, three AEs were reported in the molnupiravir arm: in 2 cases (2.6%) there was a grade < 3 elevation in ALT levels. Nonetheless, median ALT and AST values at discharge were not significantly different between patients in the molnupiravir and placebo arm [40]. We must note that patients with ALT and/or AST > 3-5 times URL were excluded from some of this studies [37,38]. Molnupiravir was also evaluated in a randomized, placebo-controlled trial which was stopped early because of business reasons. In the first part of the trial (dose-ranging phase 2 study), AST and ALT increase were observed in 8 patients (11.1%) of the 800 mg bid arm, and in one case transaminases increase was reported as a severe AE. Of note, patients with baseline cirrhosis/end-stage liver disease or transaminases increase were excluded [41]. See Table 1 for further details.

5. Ritonavir-boosted nirmatrelvir

5.1 Introduction

Nirmatrelvir is an orally bioavailable protease inhibitor that is active against MPRO, a viral protease that is essential in the viral replication cycle. Nirmatrelvir showed potent pan–human-coronavirus activity in vitro, including against SARS-CoV-2 [42]. Nirmatrelvir is metabolized mainly by the cytochrome P450 (CYP) 3A4. The co-administration of low-dose ritonavir, a strong CYP3A4 inhibitor, is necessary to achieve therapeutic concentration of nirmatrelvir [42]. Nirmatrelvir is expected to be active against the B.1.1.529 (Omicron) variant of concern and its known subvariants [16–18].

5.2 DILI from randomized clinical trial

No safety issues were identified in an accelerated randomized, double-blind, placebo-controlled, phase I study which evaluated safety, tolerability, and pharmacokinetic data of nirmatrelvir with and without ritonavir in healthy subjects. Furthermore, no clinically meaningful changes in laboratory values (≥ grade 2) were observed [43]. In the EPIC-HR study, a phase 2–3 double-blind, randomized, placebo-controlled trial, ritonavir-boosted nirmatrelvir was evaluated in non-hospitalized patients with COVID-19 at high risk for progression to severe disease [44]. Known medical history of active liver disease was an exclusion criteria, while AST or ALT levels did not represent an exclusion criterion. ALT increase was reported in 1.5% of patients treated with ritonavir-boosted nirmatrelvir (vs. 2.4% in the placebo arm), while the frequency of AST elevation was 0.9% and 1.3%, respectively. Grade 3 hyperbilirubinemia and grade 1 cholestasis were each reported in only one patient treated with ritonavir-boosted nirmatrelvir. The rate of study drug discontinuation was lower in ritonavir-boosted nirmatrelvir recipients than placebo recipients (2% vs. 4%) [44]. See Table 1 for further details.

6. Clinical trials in special populations

Most of the previously cited studies evaluated the safety and efficacy of antiviral drugs in patients without chronic kidney or liver disease. The lack of knowledge about the proper management of antivirals in special populations could increase the risk of missing the strategic "treatment window" to reduce the risk of COVID-19 progression, especially in

patients with mild to moderate disease who do not require hospital admission yet. There are some unpublished studies which evaluated the safety of antivirals in patients with stable chronic or liver disease, and we will summarize them below.

In the prospective study NCT04854837 the safety of a 5-day course of remdesivir will be evaluated in haemodialyzed patients requiring hospitalization for COVID-19. Treatment-related AE and increase of transaminases levels will be recorded, recruiting is still active [45]. To our knowledge, there are no clinical trials ongoing about the use of remdesivir in patients with chronic liver disease.

In the phase 1, non-randomized, open label NCT05386589 trial, patients with chronic moderate hepatic impairment with features of cirrhosis (score on the Child-Pugh scale ranging from 7 to 9) will receive a single oral dose of molnupiravir (800 mg) to evaluate the pharmacokinetics of the active compound NHC, together with the rate of AE, compared to healthy matched controls [46]. In a similar study (NCT05386758), pharmacokinetics and safety evaluations will be made on patients with severe renal impairment [Baseline estimated glomerular filtration rate (eGFR) <30 mL/min based on the 2021 CKD-EPI Creatinine equation] compared to healthy matched controls [47].

A phase 1, non-randomized, open-label study, investigated the effect of different degree of renal impairment on the pharmacokinetics and safety of a single oral dose of ritonavir-boosted nirmatrelvir in healthy volunteers. Treatment-related AE were observed only in patients with severe renal impairment and, together with pharmacokinetics parameters, these findings led to a dose reduction recommendation for in patients with moderate renal impairment [48]. Ritonavir-boosted nirmatrelvir safety will be evaluated in haemodialysis patients with COVID-19 (NCT05366192). The drug will be administered at the regular (300 mg) or halved dosage for renal impairment (150 mg) for a total of 5 days. An additional post-dialysis administration equivalent to half the dose received prior to dialysis treatment is scheduled. The primary outcome is the number of patients with a two-fold increase of ALT, AST or total bilirubin from baseline. At the time of writing this paper, results are not available yet [49]. Finally, the safety of ritonavir-boosted nirmatrelvir will be evaluated in healthy patients with sable hepatic impairment (Class B of the Child-Pugh Classification) in the phase 1, non-randomized, open label NCT05005312 trial, which has completed the recruitment. In addition to pharmacokinetics parameter, treatment-related AE will be recorded [50].

Summary and conclusions

Liver and biliary tract damage during COVID-19 is a multifactorial process and, besides SARS-CoV-2 direct and indirect role, DILI is relevant issue. In this narrative review we discussed the hepatic safety profile of the three antivirals actually licensed for COVID-19 treatment, focusing only on clinically relevant and impacting drugs and considering data exclusively from randomized clinical trials, as opposed to previous published reviews. Remdesivir has the potential to induce hepatic damage, but the associated liver enzymes elevations are usually mild and transient. Molnupiravir has a favourable safety profile and the incidence of liver toxicity with this drug is low. Ritonavir-boosted nirmatrelvir can potentially induce liver damage but the only RCT available didn't reveal any safety issues.

Most of the studies considered in our review are about remdesivir. The early 2020 systematic review and meta-analysis, which included randomized and non-randomized studies, found that the incidence of DILI in 208 patients treated with remdesivir was 15.2% (95% CI, 6.4-32) [13]. Considering only data from RCTs, we can conclude that remdesivir is associated with an increase in ALT, AST, and bilirubin levels, as well as moderate prothrombin time prolongation. The frequency and degree of AST and ALT elevation is variable among the studies, ranging from 2% to 34% [20,23]. Transaminases elevation is usually mild to moderate (grade ≤ 3, i.e., less than 5 times the URL) and reversible; elevations of grade 3 or more account for less than 10% of cases. However, not all studies report the degree of transaminases increase [22–27,30,33]. Hyperbilirubinemia is less common than AST/ALT increase, ranging from 1% to 10% of patients, and it is usually mild and reversible [20–22]. Remdesivir discontinuation because of DILI was an uncommon finding, ranging from 2.5% to 12% of patients [20,21,30].

The variability in the frequency and severity of liver enzymes increase may be due to different study design, the threshold of pathological enzymes elevation, baseline clinical characteristics (in particular COVID-19 severity) and concomitant medications. The background of studies from early 2020 differ considerably from the one of the latest publications in terms of concomitant drug use and prevailing circulating SARS-CoV-2 variants. Clinical treatment guidelines of COVID-19 have been consistently updated based on new evidence and, for example, the use of "old drugs" such as lopinavir/ritonavir or hydroxychloroquine was progressively abandoned in favour of systemic steroids and immunomodulatory drugs. As an example, in the CATCO trial 87% of patients received

concomitant systemic steroids while in the trial by Spinner and colleagues from 2020 the percentage does not reach twenty [19,31]. The severity of COVID-19 was found to be associated with the concomitant circulating variant; considering that liver damage is associated with the severity of COVID-19, this may have impacted on the elevations of hepatic enzymes irrespective to the contribution of DILI [51].

Even if many RCTs have been conducted, it is not clear whether the increase of liver function tests during remdesivir treatment is certainly attributable to the drug itself or if it is part of the natural history of COVID-19. In three large, randomized trial (not included in the meta-analysis cited before [13]) conducted on hospitalized patients with COVID-19 of any degree and where remdesivir was compared to placebo or SoC, the frequency of AST and ALT increase did not differ significantly between the study arms [22,23,31].

In contrast to observational studies, in the majority of RCTs patients with ALT/AST levels > 5 times the URL were excluded [19–27]. Even if only one RCT had liver cirrhosis among exclusion criteria, the reported number of patients affected by chronic liver disease was relatively low across all the studies examined [14 (3%) in the study by Beigel, 8 (1.7%) in the CATCO trial, 15 (4%) in the DisCoVeRy trial, 36 (1.3%) in the Solidarity trial and only 1 (0.4%) in the study by Gottlieb], account for less than 1% of patients treated with remdesivir. Therefore, these data are not sufficient to draw firm conclusions about the safety of remdesivir in patients with known liver disease.

[14][14]The number of randomized studies is much lower for the other two antivirals, molnupiravir and ritonavir-boosted nirmatrelvir. In two early phase 1-2 trial with a relatively small number of patients, molnupiravir showed a favourable safety profile and aminotransferases elevation did not exceed 4% of patients [37,38] This safety profile was confirmed in larger phase 3 studies which did not exclude patients with pre-existing increase in ALT or AST levels [39]. In the trial performed by Jayk-Bernal and colleagues, which lead to the Emergency Use Authorization from the FDA, data about liver enzymes elevations are not reported, even if there were no concerning safety signals [39]Liver enzymes elevation is reported in the later trial by Zou and colleagues: the frequency (2 out of 77 patients, 2.6%) is similar to data from earlier studies [40] [14]

Available evidence about safety and efficacy of ritonavir-boosted nirmatrelvir comes from the EPIC-HR study, which excluded patients with known active liver disease. ALT and AST elevations were uncommon and did not exceed 1.5% of patients (most elevations

were of grade 2 or less) and cholestasis was even rarer (two patients) [44]. However, safety concerns about liver toxicity come from the well-established knowledge of ritonavir safety profile, which has been extensively used for the treatment of people living with HIV (PLWH). Ritonavir is usually part of combination therapy together with protease inhibitors and, in this context, in turned out to be associated with elevation of AST/ALT level (about 10% of patients), gamma-glutamyl transferase (up to 20% of patients) and bilirubin (less common, about 1%) [52,53]. Elevation of AST or ALT levels up to 5 times the URL is uncommon (3-4%) and it is observed more frequently in hepatitis co-infected patient [52]. For what concerns patients with a known history of sever hepatic impairment (i.e., Child-Pugh Class C), ritonavir-boosted nirmatrelvir is thus not recommended [14]. [1414]Ritonavir-boosted nirmatrelvir is the antiviral of choice for the treatment of non-hospitalized patients with early COVID-19 at high risk of disease progression [14].

The strength of this review are the focus on clinically relevant antivirals which are approved and endorsed by most guidelines and the use of data from randomized clinical trial. The major limitations are the non-systematic literature search and the absence of a meta-analysis to draw firm conclusions about the global frequency of DILI.

Expert opinion

Antivirals have dramatically changed the management of COVID-19, especially for non-hospitalized subjects, and three of them (remdesivir, molnupiravir and ritonavir-boosted nirmatrelvir) were licensed for COVID-19 treatment after the publication of RCTs. We evaluated the hepatotoxic potential of this three antivirals by analysing data from RCTs and we found that their use is relatively safe, especially for molnupiravir and ritonavir-boosted nirmatrelvir.

We substantially agree with the recommendations issued by the FDA regarding biochemistry screening indications before administering remdesivir [14]. Evaluating liver biochemistry and prothrombin time tests, as well as creatinine, should be considered before starting remdesivir and during treatment, especially if the patient has chronic liver disease or other risk factors for DILI (i.e., concomitant hepatotoxic medications). In case of ALT or AST levels increase to less than 5 times the URL, the decision to initiate treatment should be assessed on a case-by-case basis. Remdesivir discontinuation is warranted if ALT or AST levels increase to 5-10 times the URL; the strength of the recommendations by available

guidelines based on risk/benefit ratio of this drug in hospitalized patients do not justify continuing treatment in this subset of patients [14]. Considering the low risk of drug-to-drug interactions (DDI) of remdesivir, its main clinical use should be the treatment of non-hospitalized patients with early COVID-19 at high risk of disease progression who are not eligible to ritonavir-boosted nirmatrelvir because difficult-to-manage DDI [14].

Even if there is not abundance of data from randomized studies, we can conclude that molnupiravir is associated with a low risk of DILI and, furthermore, no relevant DDI were reported. Monitoring of liver biochemistry is not mandatory, unless clinically indicated on a case-by-case basis. The main concerns about molnupiravir are its mutagenic activity and its lower efficacy compared with ritonavir-boosted nirmatrelvir and remdesivir in the treatment of non-hospitalized patients with early COVID-19 at high risk of disease progression [14].

Ritonavir-boosted nirmatrelvir can induce liver toxicity and most data about its safety profile come from PLWH who are treated for long periods. However, considering the short period of therapy in patients with COVID-19 and the high efficacy of this antiviral, this safety concerns should probably be reconsidered in patients with pre-existing mild liver disease or liver biochemistry abnormalities, in whom treatment should be used with caution and laboratory monitoring is advisable [14]. Main issues about ritonavir-boosted nirmatrelvir use is the high risk of DDI because of the ritonavir component of the combination, a CYP3A and P-glycoprotein inhibitor [14]. Before starting antiviral therapy, clinicians should carefully review concomitant medications to identify any potential interactions.

Finally, we must remember that most of the studies reported in this review excluded patients with stable chronic kidney or liver disease. A few studies are now evaluating antivirals drugs in this special populations, even if most of them are on patients not affected by COVID-19 and the sample size is relatively small. However, the results of these early studies could be useful to draw some initials hints about the handling of antivirals in special populations and, hopefully, they will represent the starting point for setting up larger trials which could definitely establish the safety profile of antivirals in patients with chronic liver or kidney disease. In conclusion, clinicians should pay attention to patients with pre-existing liver disease who require a careful evaluation about the risk-benefit ratio of antiviral treatment, especially with remdesivir or ritonavir-boosted nirmatrelvir.

ORCID Number: Giacomo Casalini 0000-0003-0740-5361; Andrea Giacomelli: 0000-0003-3685-4289; Spinello Antinori: 0000-0003-0569-9407.

Funding: This paper was not funded.

Reviewer disclosures: The authors declare having no conflicts of interest.

References

[1] Wiersinga WJ, Rhodes A, Cheng AC, Peacock SJ, Prescott HC. Pathophysiology, Transmission, Diagnosis, and Treatment of Coronavirus Disease 2019 (COVID-19): A Review. JAMA. 2020;324(8):782-793.

[2] Fan Z, Chen L, Li J, Cheng X, Yang J, Tian C, et al. Clinical Features of COVID-19-Related Liver Functional Abnormality. Clin Gastroenterol Hepatol. 2020 Jun;18(7):1561-1566.

[3] Lei F, Liu YM, Zhou F, Qin JJ, Zhang P, Zhu L, et al. Longitudinal Association Between Markers of Liver Injury and Mortality in COVID-19 in China. Hepatology. 2020;72(2):389-398.

[4] Wang Y, Liu S, Liu H, Li W, Lin F, Jiang L, et al. SARS-CoV-2 infection of the liver directly contributes to hepatic impairment in patients with COVID-19. J Hepatol. 2020;73(4):807-816.

[5] Zhao B, Ni C, Gao R, Wang Y, Yang L, Wei J, et al. Recapitulation of SARS-CoV-2 infection and cholangiocyte damage with human liver ductal organoids. Protein Cell. 2020;11(10):771-775.

[6] Sonzogni A, Previtali G, Seghezzi M, Grazia Alessio M, Gianatti A, Licini L, et al. Liver histopathology in severe COVID 19 respiratory failure is suggestive of vascular alterations. Liver Int. 2020;40(9):2110-2116.

[7] Dufour JF, Marjot T, Becchetti C, Tilg H. COVID-19 and liver disease. Gut. 2022;71(11):2350-2362.

[8] Sodeifian F, Seyedalhosseini ZS, Kian N, Eftekhari M, Najari S, Mirsaeidi M, et al. Drug-Induced Liver Injury in COVID-19 Patients: A Systematic Review. Front Med (Lausanne). 2021;8:731436.
*** This is a systematic review about DILI in COVID-19 until January 2021.**

[9] Mao R, Qiu Y, He JS, Tan JY, Li XH, Liang J, et al. Manifestations and prognosis of gastrointestinal and liver involvement in patients with COVID-19: a systematic review and meta-analysis. Lancet Gastroenterol Hepatol. 2020;5(7):667-678.

[10] Singh S, Khan A. Clinical Characteristics and Outcomes of Coronavirus Disease 2019 Among Patients With Preexisting Liver Disease in the United States: A Multicenter Research Network Study. Gastroenterology. 2020;159(2):768-771.e3.

[11] Moon AM, Webb GJ, Aloman C, Armstrong MJ, Cargill T, Dhanasekaran R, et al. High mortality rates for SARS-CoV-2 infection in patients with pre-existing chronic liver disease and cirrhosis: Preliminary results from an international registry. J Hepatol. 2020;73(3):705-708.

[12] Kariyawasam JC, Jayarajah U, Abeysuriya V, Riza R, Seneviratne SL. Involvement of the Liver in COVID-19: A Systematic Review. American Journal of Tropical Medicine and Hygiene. 2022;106(4):1026-1041.

[13] Kulkarni A v., Kumar P, Tevethia HV, Premkumar M, Arab JP, Candia R, et al. Systematic review with meta-analysis: liver manifestations and outcomes in COVID-19. Aliment Pharmacol Ther. 2020;52(4):584-599.
**** This is a meta-analysis from 2020 about liver involvement in COVID-19.**

[14] COVID-19 Treatment Guidelines Panel. Coronavirus Disease 2019 (COVID-19) Treatment Guidelines. National Institutes of Health. Available at https://www.covid19treatmentguidelines.nih.gov/ Accessed August 31 2022.

[15] Wang M, Cao R, Zhang L, Yang X, Liu J, Xu M, et al. Remdesivir and chloroquine effectively inhibit the recently emerged novel coronavirus (2019-nCoV) in vitro. Cell Res. 2020;30(3):269-271.

[16] Vangeel L, Chiu W, de Jonghe S, Maes P, Slechten B, Raymenants J, et al. Remdesivir, Molnupiravir and Nirmatrelvir remain active against SARS-CoV-2 Omicron and other variants of concern. Antiviral Res. 2022;198.
[17] Takashita E, Kinoshita N, Yamayoshi S, Sakai-Tagawa Y, Fujisaki S, Ito M, et al. Efficacy of Antiviral Agents against the SARS-CoV-2 Omicron Subvariant BA.2. N Engl J Med. 2022;386(15):1475-1477.
[18] Fiaschi L, Dragoni F, Schiaroli E, Bergna A, Rossetti B, Giammarino F, et al. Efficacy of Licensed Monoclonal Antibodies and Antiviral Agents against the SARS-CoV-2 Omicron Sublineages BA.1 and BA.2. Published online 2022.
[19] Spinner CD, Gottlieb RL, Criner GJ, Arribas López JR, Cattelan AM, Soriano Viladomiu A, et al. Effect of Remdesivir vs Standard Care on Clinical Status at 11 Days in Patients With Moderate COVID-19: A Randomized Clinical Trial. JAMA. 2020;324(11):1048-1057.
[20] Goldman JD, Lye DCB, Hui DS, Marks KM, Bruno R, Montejano R, et al. Remdesivir for 5 or 10 Days in Patients with Severe Covid-19. N Engl J Med. 2020;383(19):1827-1837.
[21] Wang Y, Zhang D, Du G, Du R, Zhao J, Jin Y, et al. Remdesivir in adults with severe COVID-19: a randomised, double-blind, placebo-controlled, multicentre trial. The Lancet. 2020;395(10236):1569-1578.
[22] Beigel JH, Tomashek KM, Dodd LE, Mehta AK, Zingman BS, Kalil AC, et al. Remdesivir for the Treatment of Covid-19 — Final Report. N Engl J Med. 2020;383(19):1813-1826.
[23] Ader F, Bouscambert-Duchamp M, Hites M, Peiffer-Smadja N, Poissy J, Belhadi D, et al. Remdesivir plus standard of care versus standard of care alone for the treatment of patients admitted to hospital with COVID-19 (DisCoVeRy): a phase 3, randomised, controlled, open-label trial. Lancet Infect Dis. 2022;22(2):209-221.
[24] Mahajan L, Singh AP, Gifty. Clinical outcomes of using remdesivir in patients with moderate to severe COVID-19: A prospective randomised study. Indian J Anaesth. 2021;65(13):S41-S46.
[25] Kalil AC, Mehta AK, Patterson TF, Erdmann N, Gomez CA, Jain MK, et al. Efficacy of interferon beta-1a plus remdesivir compared with remdesivir alone in hospitalised adults with COVID-19: a double-bind, randomised, placebo-controlled, phase 3 trial. Lancet Respir Med. 2021;9(12):1365-1376.
[26] Gottlieb RL, Vaca CE, Paredes R, Mera J, Webb BJ, Perez G, et al. Early Remdesivir to Prevent Progression to Severe Covid-19 in Outpatients. N Engl J Med. 2022;386(4):305-315.
[27] Barratt-Due A, Olsen IC, Nezvalova-Henriksen K, Kåsine T, Lund-Johansen F, Hoel H, et al. Evaluation of the Effects of Remdesivir and Hydroxychloroquine on Viral Clearance in COVID-19: A Randomized Trial. Ann Intern Med. 2021;174(9):1261-1269.
[28] Kalil AC, Patterson TF, Mehta AK, Tomashek KM, Wolfe CR, Ghazaryan V, et al. Baricitinib plus Remdesivir for Hospitalized Adults with Covid-19. N Engl J Med. 2021;384(9):795-807.
[29] Hoffmann-La Roche. A Phase III, Randomized, Double-Blind, Multicenter Study to Evaluate the Efficacy and Safety of Remdesivir Plus Tocilizumab Compared With Remdesivir Plus Placebo in Hospitalized Patients With Severe COVID-19 Pneumonia. [Accessed 2022 Aug 31]. In: ClinicalTrials.gov [Internet] Bethesda (MD): US National Library of Medicine Available from: http://clinicaltrials.gov/show/NCT04409262 ClinicalTrials.gov Identifier: NCT04409262.
[30] Criner GJ, Criner GJ, Ahn MY, Huhn G, Subramanian A, Lumbreras C, et al. Safety of Remdesivir vs Standard Care in Patients with Moderate Covid-19. Open Forum Infect Dis. 2020;7:S345-S346.
[31] Ali K, Azher T, Baqi M, Binnie A, Borgia S, Carrier FM, et al. Remdesivir for the treatment of patients in hospital with COVID-19 in Canada: a randomized controlled trial. CMAJ. 2022;194(7):E242-E251.
*** One of the latest randomized controlled trial about remdesivir in COVID-19 in which remdesivir did not significantly increase the risk of liver injury.**
[32] Kalil AC, Mehta AK, Patterson TF, Erdmann N, Gomez CA, Jain MK, et al. Efficacy of interferon beta-1a plus remdesivir compared with remdesivir alone in hospitalised adults with COVID-19: a double-bind, randomised, placebo-controlled, phase 3 trial. Lancet Respir Med. 2021;9(12):1365-1376.
[33] Solidarity Trial Consortium W. Remdesivir and three other drugs for hospitalised patients with COVID-19: final results of the WHO Solidarity randomised trial and updated meta-analyses. The Lancet. 2022;399(10339):1941-1953.
[34] WHO Solidarity Trial Consortium. Repurposed Antiviral Drugs for Covid-19 — Interim WHO Solidarity Trial Results. New England Journal of Medicine. 2021;384(6):497-511.

[35] Sheahan TP, Sims AC, Zhou S, Graham RL, Pruijssers AJ, Agostini ML, et al. An orally bioavailable broad-spectrum antiviral inhibits SARS-CoV-2 in human airway epithelial cell cultures and multiple coronaviruses in mice. Sci Transl Med. 2020;12(541).

[36] Painter WP, Holman W, Bush JA, Almazedi F, Malik H, Eraut NCJE, et al. Human safety, tolerability, and pharmacokinetics of molnupiravir, a novel broad-spectrum oral antiviral agent with activity against SARS-CoV-2. Antimicrob Agents Chemother. 2021;65(5).

[37] Khoo SH, Fitzgerald R, Fletcher T, Ewings S, Jaki T, Lyon R, et al. Optimal dose and safety of molnupiravir in patients with early SARS-CoV-2: a Phase I, open-label, dose-escalating, randomized controlled study. J Antimicrob Chemoter. 2021;76(12):3286-3295.

[38] Fischer WA, Eron JJ, Holman W, Cohen MS, Fang L, Szewczyk LJ, et al. A phase 2a clinical trial of molnupiravir in patients with COVID-19 shows accelerated SARS-CoV-2 RNA clearance and elimination of infectious virus. Sci Transl Med. 2022;14(628):7430.

[39] Jayk Bernal A, Gomes da Silva MM, Musungaie DB, Kovalchuk E, Gonzalez A, Delos Reyes V, et al. Molnupiravir for Oral Treatment of Covid-19 in Nonhospitalized Patients. N Engl J Med 2022;386(6):509-520.

*** The randomized clinical trial which led to the approval of molnupiravir for early COVID-19 treatment.**

[40] Zou R, Peng L, Shu D, Zhao L, Lan J, Tan G, et al. Antiviral Efficacy and Safety of Molnupiravir Against Omicron Variant Infection: A Randomized Controlled Clinical Trial. Front Pharmacol. 2022;13.

[41] Merck Sharp & Dohme LLC. A Phase 2/3, Randomized, Placebo-Controlled, Double-Blind Clinical Study to Evaluate the Efficacy, Safety, and Pharmacokinetics of MK-4482 in Hospitalized Adults With COVID-19. [Accessed Aug 31 2022]. In: ClinicalTrials.gov [Internet] Bethesda (MD): US National Library of Medicine Available from: http://clinicaltrials.gov/show/NCT04575584 ClinicalTrials.gov Identifier: NCT04575584.

[42] Owen DR, Allerton CMN, Anderson AS, Aschenbrenner L, Avery M, Berritt S, et al. An oral SARS-CoV-2 Mpro inhibitor clinical candidate for the treatment of COVID-19. Science. 2021;374(6575):1586-1593.

[43] Singh RSP, Toussi SS, Hackman F, Chan PL, Rao R, Allen R, et al. Innovative Randomized Phase I Study and Dosing Regimen Selection to Accelerate and Inform Pivotal COVID-19 Trial of Nirmatrelvir. Clin Pharmacol Ther. 2022;112(1):101-111.

[44] Hammond J, Leister-Tebbe H, Gardner A, Abreu P, Bao W, Wisemandle W, et al. Oral Nirmatrelvir for High-Risk, Nonhospitalized Adults with Covid-19. N Engl J Med. 2022;386(15):1397-1408.

*** The randomized clinical trial which led to the approval of ritonavir-boosted nirmatrelvir for early COVID 19 treatment.**

[45] Istvan Takacs. Safety of Remdesivir Treatment in COVID-19 Patients Requiring Hemodialysis. [Accessed 2022 Aug 31]. In: ClinicalTrials.gov [Internet] Bethesda (MD): US National Library of Medicine Available from: http://clinicaltrials.gov/show/NCT04854837 ClinicalTrials.gov Identifier: NCT04854837.

[46] Merck Sharp & Dohme LLC. An Open-Label, Single-Dose Clinical Study to Evaluate the Pharmacokinetics of Molnupiravir (MK-4482) in Participants With Moderate Hepatic Impairment. [Accessed 31 Aug 2022]. In: ClinicalTrials.gov [Internet] Bethesda (MD): US National Library of Medicine Available from: http://clinicaltrials.gov/show/NCT05386589 ClinicalTrials.gov Identifier: NCT05386589 .

[47] Merck Sharp & Dohme LLC. An Open-Label, Single-Dose Study to Evaluate the Pharmacokinetics of Molnupiravir (MK-4482; MOV) in Participants With Severe Renal Impairment. [Accessed 2022 Aug 31]. In: ClinicalTrials.gov [Internet] Bethesda (MD): US National Library of Medicine Available from: http://clinicaltrials.gov/show/NCT05386758 ClinicalTrials.gov Identifier: NCT05386758.

[48] Toussi SS, Neutel JM, Navarro J, Preston RA, Shi H, Kavetska O, et al. Pharmacokinetics of Oral Nirmatrelvir/Ritonavir, a Protease Inhibitor for Treatment of COVID-19, in Subjects With Renal Impairment. Clin Pharmacol Ther. Published online 2022.

[49] Leyi G. The Safety of Paxlovid (Nirmatrelvir/Ritonavir) in Hemodialysis Patients With SARS-CoV-2 Infection. [Accessed 2022 Aug 31]. In: ClinicalTrials.gov [Internet] Bethesda (MD): US National Library

of Medicine Available from: http://clinicaltrials.gov/show/NCT05366192 ClinicalTrials.gov Identifier: NCT05366192.

[50] Pfizer CT.gov. A Phase 1, non-randomized, open-label study to assess the pharmacokinetics, safety and tolerability of PF-07321332 boosted with ritonavir in adult participants with moderate hepatic impairment and healthy participants with normal hepatic function. [Accessed 2022 Ag 31]. In: ClinicalTrials.gov [Internet] Bethesda (MD): US National Library of Medicine Available from: http://clinicaltrials.gov/show/NCT05005312 ClinicalTrials.gov Identifier: NCT05005312.

[51] Florensa D, Mateo J, Spaimoc R, Miret C, Godoy S, Solsona F, et al. Severity of COVID-19 cases in the months of predominance of the Alpha and Delta variants. Sci Rep. 2022;12(1).

[52] Vogel M, Rockstroh JK. Safety of lopinavir/ritonavir for the treatment of HIV-infection. Expert Opin Drug Saf. 2005;4(3):403-420.

[53] Cooper CL, van Heeswijk RPG, Gallicano K, Cameron DW. A Review of Low-Dose Ritonavir in Protease Inhibitor Combination Therapy. Clin Infect Dis. 2003 Jun 15;36(12):1585-92.