Liver Injury in Treatment of COVID-19 Patients: Lopinavir/Ritonavir vs Atazanavir/Ritonavir

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Abstract

Introduction: Protease inhibitors (PI) were identified as a promising treatment option for COVID-19 during the early phase of the pandemic. Incidences of elevated serum liver biochemistries have been observed in 14-53% hospitalised COVID-19 patients.

Objective: The objective of this study was to compare the proportion of liver injury between COVID-19 patients receiving lopinavir/ritonavir (LPV/r) and atazanavir/ritonavir (ATZ/r), to determine the time to onset to, factors associated with and clinical outcomes of grade 3-4 liver injury.

Method: A retrospective cross-sectional study was conducted among adult COVID-19 patients admitted to Sungai Buloh Hospital from 15th February to 30th April 2020, who received either LPV/r or ATZ/r as part of their treatment. Data were collected from patients' medical records in the internal Electronic Hospital Information System.

Results: This study involved 212 patients with 106 patients in each group. A statistically significant relationship between the two PIs and the severity of liver injury was found (p=<0.001). The number of patients who developed grade 3-4 liver injury in ATZ/r and LPV/r group were 27 (25.5%) and 73 (68.9% respectively. The median time to onset of grade 3-4 liver injury in ATZ/r and LPV/r group were 4 days (IQR 3) and 6 days (IQR 9) respectively. There was statistically significant associations between the type of PI used and duration of PI treatment with development of grade 3-4 liver injury in COVID-19 patients (p=0.070, Nagelkerke R^2=0.368). Among the grade 3-4 liver injury patients, 90 (90.0%) were discharged, 6 (6.0%) remained hospitalised at the end of data collection period and 4 (4.0%) deceased.

Conclusion: Liver injury was observed in COVID-19 patients receiving LPV/r or ATZ/r. More grade 3-4 liver injury were seen in the ATZ/r group.

Keywords: COVID-19, liver injury, protease inhibitor

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Introduction

In March 2020, the World Health Organization (WHO) declared the global outbreak of infectious disease caused by a novel coronavirus termed severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) as the COVID-19 pandemic. By May 2023, COVID-19 was no longer an international public health emergency and in transition to becoming endemic (1). In the initial stages of the pandemic, when effective treatments were limited, commercially available antiretroviral agents such as lopinavir/ritonavir (LPV/r) and atazanavir were identified as one of the promising treatment options for COVID-19. In vitro studies suggest that these protease inhibitors (PI) have activity against SARS-CoV-2 via inhibition of 3-chymotrypsin-like protease enzyme (2-4). At Sungai Buloh Hospital in the state of Selangor, Malaysia, LPV/r and atazanavir/ritonavir (ATZ/r) were initially used for the treatment of COVID-19 adult patients who were in stage 3A or higher. The prescribed regimen included lopinavir 400mg/ritonavir 100mg twice daily or atazanavir 300mg/ritonavir 100mg daily for a duration of up to 10 days (5).

The use of PI-based treatments in HIV patients has been associated with hepatotoxicity, which is characterised by a significant increase in serum alanine aminotransferase (ALT) and/or aspartate aminotransferase (AST) levels, as well as unconjugated hyperbilirubinemia. The liver injury typically occurs within one to 8 weeks after initiating treatment (6, 7). The mechanisms behind drug-induced liver injury

(DILI) in these patients are not fully understood due to multiple factors (6). In hospitalised COVID-19 patients, elevated serum liver biochemistries (ALT/AST and/or total bilirubin (TB)) have been observed in 14-53% of cases, with a higher incidence in severe COVID-19 patients (8). Fan et al. reported that more than one third of COVID-19 patients in a hospital in Shanghai, China developed abnormal liver function. A high proportion of these patients had received LPV/r as COVID-19 treatment (9). Studies have indicated that liver damage is more common in patients with severe pneumonia, possibly due to inflammatory cytokines released during cytokine release syndrome (CRS) (10, 11). Inflammatory markers serve as an early alarm indicators of potential liver impairment in COVID-19 patients (12).

It remains unclear if elevated liver biochemistries are caused by the SARS-CoV-2 infection itself, CRS, ischemia/hypotension or due to DILI since some pharmacological agents used may be hepatotoxic. Hence, regular monitoring of liver biochemistries is recommended in these patients, regardless of the baseline values (8). At the time this study was conducted, PI was widely used for COVID-19 treatment. The increasing reporting of liver injury, especially in patients with severe COVID-19 stages, warrants further investigation on the possibility of DILI and other potential risk factors to liver injury. While there were many studies on PI-related liver injury among HIV patients, limited studies were done in the COVID-19 patient population. In addition, although studies have reported DILI among COVID-19 patients treated with LPV/r, no studies have compared liver injury in LPV/r and ATZ/r treated COVID-19 patients.

Therefore, this study was conducted to investigate PI related liver injury in the COVID-19 patient population and to serve as a reference for future studies. The objectives of this study were to compare the proportion of liver injury between COVID-19 patients receiving LPV/r and ATZ/r, to determine the time to onset of grade 3-4 liver injury in COVID-19 patients receiving LPV/r and ATZ/r, to determine the factors associated with development of grade 3-4 liver injury in COVID-19 patients receiving LPV/r and ATZ/r, and to describe the clinical outcomes of COVID-19 patients who developed grade 3-4 liver injury after initiation of LPV/r and ATZ/r.

Method

Study design

This retrospective cross-sectional study was conducted among adult COVID-19 patients admitted to Sungai Buloh Hospital from 15th February 2020 to 30th April 2020, receiving either LPV/r or ATZ/r as part of their treatment. Data were collected from patients' medical records in the internal Electronic Hospital Information System (eHIS). This study was registered with the National Medical Research Register (NMRR-20-900-54895). Ethical approval was obtained from the Medical Research & Ethics Committee, Ministry of Health Malaysia as well (KKM/NIHSEC/P20-1078(6)).

Study population

The inclusion criteria in this study were adults above 18 years old with laboratory-confirmed 2019-nCoV infection through real-time reverse transcription-polymerase chain reaction, receiving LPV/r or ATZ/r as part of COVID-19 treatment and are PI treatment naive. Patients who received only 1 day of treatment with LPV/r or ATZ/r without available baseline liver function test results, as well as those with missing data in the eHIS records, were excluded. Due to a change in prescribing practice during the study period, where alternative treatment options for COVID-19 were discovered, all adult patients on ATZ/r were included and matched with an equal number of patients on LPV/r. All patients were enrolled using convenience sampling. The selection of patients on LPV/r was randomized using Microsoft Excel randomization function.

Outcome measures

Primary outcome was the difference in the proportion of liver injury between COVID-19 patients receiving LPV/r and ATZ/r. The elevation of serum ALT, AST, and TB levels after the initiation of LPV/r or ATZ/r was documented and graded based on severity. Secondary outcome measures included time to onset of grade 3-4 liver injury, factors associated with development of grade 3-4 liver injury and clinical outcome of patients who developed grade 3-4 liver injury. This study focused on grade 3-4 liver injury (severe/life threatening) as these grades of liver injury were considered significant and may require intervention(s).

Definition of liver injury

Severity grading of DILI used in this study is based on the grading system developed by the Acquired Immune Deficiency Syndrome Clinical Trials Group (ACTG) as shown in Table 1. The grading system assesses the severity based on multiples of the upper limit of the normal range for ALT, AST, and TB levels. In patients with underlying liver disease, such as hepatitis B or C, the grading of liver biochemistries is adjusted to baseline values (13). In cases where there were discordant grades for ALT/AST/bilirubin, the higher grade was used for classification.

Table 1: Severity grading of drug induced liver injury

Feature	Grade 0 (Normal)	Grade 1 (Mild)	Grade 2 (Moderate)	Grade 3 (Severe)	Grade 4 (Life threatening)
ALT	<1.25	1.25 – 2.5	>2.5 – 5.0	>5.0 - 10	>10
AST	<1.25	1.25 – 2.5	>2.5 – 5.0	>5.0 - 10	>10
TB	Normal	>1.0 – 1.5	>1.5 – 2.5	>2.5 - 5	>5

Normal laboratory range in Sungai Buloh Hospital:
ALT(10 - 49 U/L); AST(0 - 34 U/L); TB(>5 days-60 years: 5 - 21 umol/L, 60-90 years: 3 - 19 umol/L, >90 years: 3 - 15 umol/L)
Abbreviations: ALT = Alanine aminotransferase; AST = Aspartate aminotransferase; TB = Total Bilirubin

Definition of factors associated with liver injury

A list of factors associated with liver injury in patients on PI was ascertained through review of literature: underlying hepatitis B or C, other new concomitant hepatotoxic drug use, abnormal transaminases on admission, COVID-19 disease severity, presence of CRS, age and gender.

Definition of hepatotoxic drug

A list of hepatotoxic drugs was ascertained through literature review but not limited to the following: antibiotic (penicillin/cephalosporin/azithromycin), antifungal, antituberculosis drug, paracetamol, nonsteroidal anti-inflammatory drugs, valproic acid, amiodarone, tamoxifen, methotrexate, statin, glucocorticoids and traditional medication.

Definition of COVID-19 severity staging

Following guidelines provided by the Ministry of Health Malaysia, COVID-19 severity staging is classified as follow.

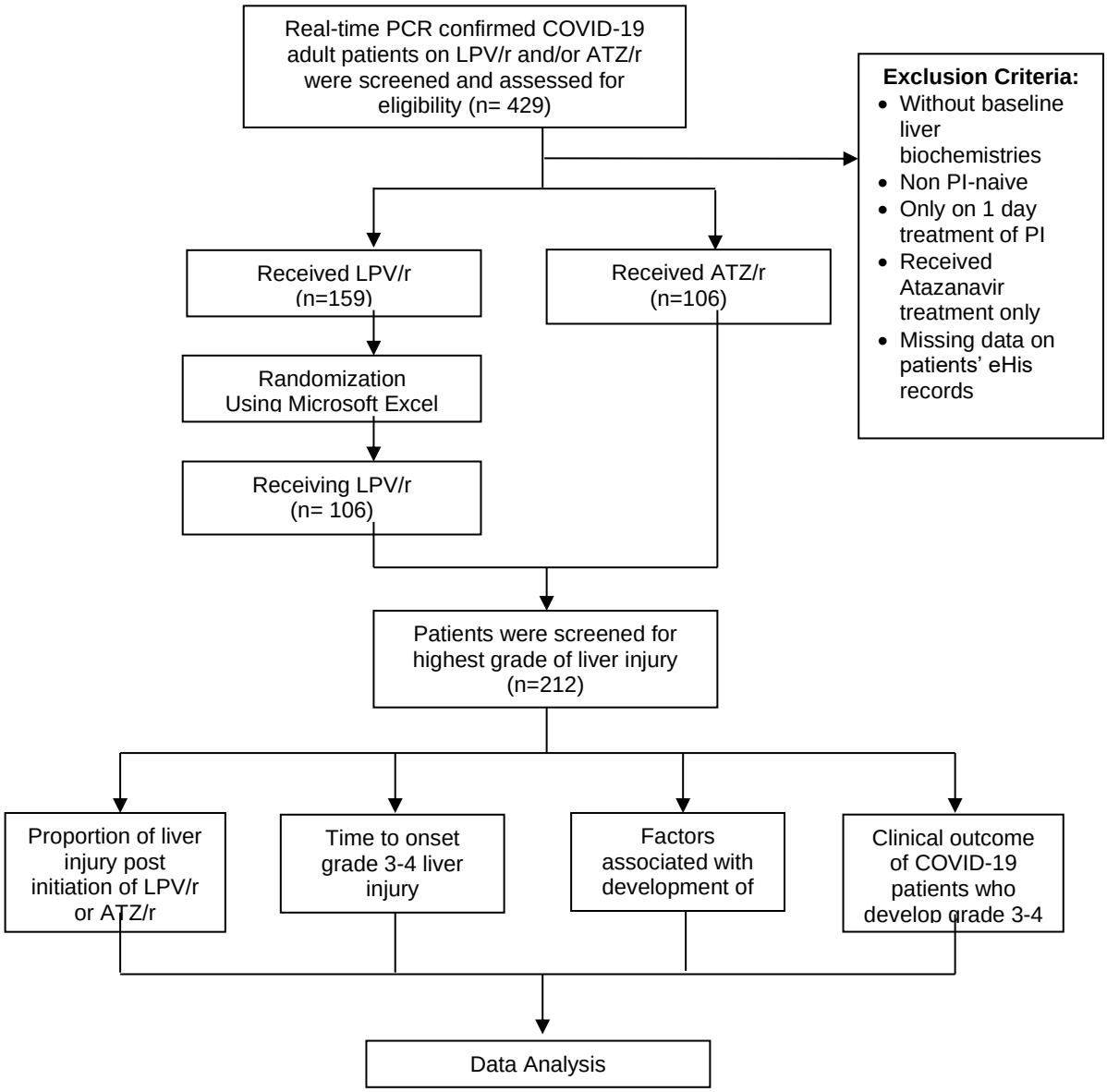
Stage 1 : Asymptomatic
Stage 2A : Symptomatic but without pneumonia without fever
Stage 2B : Symptomatic but without pneumonia with fever
Stage 3A : Pneumonia but not requiring oxygen without fever
Stage 3B : Pneumonia but not requiring oxygen with fever
Stage 4A : Pneumonia but requiring oxygen without fever
Stage 4B : Pneumonia but requiring oxygen with fever
Stage 5 : Critically ill

Statistical analysis

Categorical variables were expressed as frequency and percentages, while continuous variables were presented as medians with interquartile range (IQR), as appropriate. Comparison of categorical variables was done using Chi-square (χ^2) test. Comparison of non-parametric continuous variables was done using Mann-Whitney U test. Binary logistic regression was used to determine the factors associated with development of grade 3-4 liver injury. All statistical analyses were performed using the Statistical Package for Social Sciences (SPSS) version 27. A two tailed P value of <0.05 was considered statistically significant.

Results

In this study, 212 patients were included, with 106 patients in each group. Study flowchart was displayed in Figure 1. The demographic and baseline characteristics were presented in Table 2. Majority of the patients in both the LPV/r and ATZ/r groups were in the age range of 51 to 70 years old. There was only one patient in the study population with underlying chronic liver disease, which was in the ATZ/r group. Among the patients, 61(57.5%) in the LPV/r group and 48(45.3%) in the ATZ/r group had abnormal liver test results upon admission. There was no statistically significant difference in demographics and baseline characteristics between LPV/r group and ATZ/r group, except for comorbidity of respiratory disease (p=0.045).



Abbreviation: PCR = Polymerase chain reaction; LPV/r = Lopinavir/Ritonavir; ATZ/r = Atazanavir/Ritonavir; PI = Protease inhibitor; eHis = Electronic Hospital Information System; ALT = Alanine aminotransferase; AST = Aspartate aminotransferase; TB = Total Bilirubin

Figure 1: Study flowchart

Table 2: Demographics and baseline characteristics of study population (n=212)

Characteristics	LPV/r (n=106)	ATZ/r (n=106)	*p*-value [a]
Age (years), n (%)			
18 – 30	4 (3.8)	9 (8.5)	0.235
31 – 50	30 (28.3)	38 (35.8)	
51 – 70	67 (63.2)	54 (50.9)	
> 70	5 (4.7)	5 (4.7)	
Gender, n (%)			
Male	79 (74.5)	66 (62.3)	0.055
Female	27 (25.5)	40 (37.7)	
Ethnicity, n (%)			
Malay	81 (76.4)	82 (77.4)	0.269
Chinese	13 (12.3)	9 (8.5)	
Indian	2 (1.9)	7 (6.6)	
Others	0 (0)	1 (0.9)	
Foreigner	10 (9.4)	7 (6.6)	
Comorbidity, n (%)			
Cardio/cerebrovascular disease	48 (45.3)	57 (53.8)	0.216
Respiratory disease	11 (10.4)	5 (4.7)	0.045
Chronic kidney disease	7 (6.6)	14 (13.2)	0.108
Chronic liver disease	0 (0)	1 (0.9)	-
Endocrine	34 (32.1)	39 (36.8)	0.470
Immunodeficient/malignancy	2 (1.9)	2 (1.9)	0.651
Others	9 (8.5)	18 (17)	0.064
No known medical illness	39 (36.8)	34 (32)	0.470
COVID-19 stages, n (%)			
Mild (Stage 1-3)	46 (43.4)	50 (47.2)	0.581
Severe (Stage 4-5)	60 (56.6)	56 (52.8)	
Baseline ALT/ AST on admission, n (%)			
Normal (ALT 10 - 49 U/L; AST 0 - 34 U/L)	45 (42.5)	58 (54.7)	0.074
Abnormal (ALT > 49 U/L; AST > 34 U/L)	61 (57.5)	48 (45.3)	
Newly added concomitant hepatotoxic drugs (other than PI)	51 (48.1)	44 (41.5)	0.334

[a] p-value derived from Chi-square (X^2) analysis

Abbreviation: LPV/r = Lopinavir/Ritonavir; ATZ/r = Atazanavir/Ritonavir; ALT = Alanine aminotransferase; AST = Aspartate aminotransferase; PI = Protease inhibitor

Table 3 demonstrated the statistically significant relationship between the two PIs and severity of liver injury ($p<0.001$), except in Grade 2. Most patients who developed Grade 3 [52(49.1%)] and Grade 4 [21(19.8%)] liver injury belonged to the ATZ/r group. In this group, TB is the predominant liver biochemistry causing liver injury across all the grades. On the contrary, in the LPV/r group, many patients fall into Grade 1 liver injury [37(34.9%)]. Although in the LPV/r treatment group, the proportion of patients developing liver injury decreases as we go into more severe stages, almost all patients developed abnormal elevation in all the three liver biochemistries in Grade 4.

Table 3: Proportion of liver injury in COVID-19 patients receiving LPV/r and ATZ/r

Drug	Liver Injury Grading					$X^2(df)$	*p*-value[b]
	Normal n (%)	Grade 1 n (%)	Grade 2 n (%)	Grade 3 n (%)	Grade 4 n (%)		
LPV/r (n=106)	24 (22.6)	37 (34.9)	18 (17.0)	18 (17.0)	9 (8.5)	46.939 (4)	<0.001
ATZ/r (n=106)	9 (8.5)	8 (7.5)	16 (15.1)	52 (49.1)	21 (19.8)		

[b] p-value derived from Chi-square (χ^2) analysis
Abbreviation: LPV/r = Lopinavir/Ritonavir; ATZ/r = Atazanavir/Ritonavir

There was a statistically significant difference (p<0.001) between the two groups in terms of the median peak value of TB (Table 4). The ATZ/r group had a higher median peak value [Median 68 µmol/L (IQR 46-88)] compared to LPV/r group [Median 25.7 µmol/L (IQR 17-48.3)], despite a similar baseline median for TB between the two groups. Among the 27 patients in the LPV/r group with grade 3-4 liver injury, 20(71%) of patients were continued on LPV/r while only one patient had their drug discontinued. On the other hand, 42(57.5%) out of 73 patients in the ATZ/r group who developed grade 3-4 liver injury were continued on ATZ/r; 23(31.5%) had their drug discontinued; 8(34.8%) had their drug switched to LPV/r. Majority of them in both groups were asymptomatic and their liver biochemistries returned to normal levels upon discontinuation or completion of treatment.

Table 4: Liver biochemistries of COVID-19 patients receiving LPV/r and ATZ/r

Lab Values		LPV/r (n=106) Median (IQR)	ATZ/r (n=106) Median (IQR)	*p*-value [c]
ALT (U/L)	Baseline	34 (22.0-55.3)	29.5 (16.8-48.3)	0.083
	Peak	69 (33.0-106.6)	48.5 (25.3-107.3)	
AST (U/L)	Baseline	38.5 (26.6-58.3)	30 (22-49)	0.979
	Peak	52 (35-102)	41 (28-76)	
TB (umol/L)	Baseline	9.7 (7.0-14.0)	9.8 (7.0-13.0)	<0.001
	Peak	25.7 (17.0-48.3)	68 (46.0-88.0)	

[c] p-value derived from Mann-Whitney U analysis
Abbreviation: LPV/r = Lopinavir/Ritonavir; ATZ/r = Atazanavir/Ritonavir; ALT = Alanine aminotransferase; AST = Aspartate aminotransferase; TB = Total Bilirubin

Time to onset of grade 3-4 liver injury was shorter in the ATZ/r group than in the LPV/r group [median 4 days (IQR 3 days) vs median 6 days (IQR 9 days)]. Whereas, the duration of PI treatment in the ATZ/r group were prescribed a shorter course [(median 5 days (IQR 2 days)] as compared to those in the LPV/r group [median 7 days (IQR 4 days)].

Logistic regression analysis was conducted to determine factors associated with the development of grade 3-4 liver injury among COVID-19 patients receiving LPV/r or ATZ/r as part of their treatment (Table 5). Univariate logistic regression showed that there were statistically significant associations between the type of PI used (p<0.001) and presence of CRS (p<0.001) with the development of grade 3-4 liver injury. The binary logistic regression model indicated statistically significant associations between type of PI used and duration of PI treatment (p=0.070, NagelkerkeR^2=0.368) with development of grade 3-4 liver injury. The use of ATZ/r was associated with higher odds of developing grade 3-4 liver injury (p<0.001). Underlying chronic liver disease was not included in the analysis as there was only one patient.

Table 5: Association between potential factors with development of grade 3-4 liver injury

Variable	Univariate Logistic Regression			Binary Logistic Regression		
	(b)	Crude Odds Ratio (95% CIs)	*p*-value[d]	(b)	Adjusted Odds Ratio (95% CIs)	*p*-value[d]
Age						
18-30 years old	0	1.00		0	1.00	
31-50 years old	-0.53	0.59 (0.18-1.99)	0.393	-0.61	0.55 (0.14-2.20)	0.394
51-70 years old	-0.79	0.46 (0.14-1.47)	0.189	-0.68	0.51 (0.13-2.00)	0.331
> 70 years old	0.92	2.50 (0.37-16.89)	0.347	1.43	4.16 (0.48-36.33)	0.197
Gender						
Male	0	1.00		0	1.00	
Female	0.47	1.61 (0.90-2.88)	0.111	0.40	1.49 (0.74-3.00)	0.261
Type of PI used						
Lopinavir/Ritonavir	0	1.00		0	1.00	
Atazanavir/Ritonavir	1.87	6.47 (3.55-11.79)	<0.001	2.39	10.94 (4.93-24.26)	<0.001
New concomitant hepatotoxic drug use						
No	0	1.00		0	1.00	
Yes	0.32	1.38 (0.80-2.37)	0.247	0.12	1.13 (0.52-2.47)	0.758
Presence of CRS						
No	0	1.00		0	1.00	
Yes	1.85	6.33 (2.08-19.33)	<0.001	0.92	2.51 (0.65-9.72)	0.184
COVID-19 staging						
Mild (Stage 1,2,3)	0	1.00		0	1.00	
Severe (Stage 4,5)	0.48	1.62 (0.94-2.80)	0.083	0.23	1.26 (0.58-2.75)	0.559
Abnormal ALT/AST level on admission						
No	0	1.00		0	1.00	
Yes	0.20	1.22 (0.71-2.09)	0.477	0.43	1.54 (0.74-3.19)	0.248
Duration of PI treatment, day	0.06	1.06 (0.97-1.17)	0.210	0.18	1.20 (1.05-1.37)	0.009

[d] Data analyzed using binary logistic regression (Enter method)

Constant -2.705

Hosmer and Lemeshow Test $X^2(8)=14.471$, p = 0.070; Classification table 74.1%; Area under ROC curve 80.9%

Abbreviation: PI = Protease inhibitor; CRS = Cytokine release syndrome; ALT = Alanine aminotransferase; AST = Aspartate aminotransferase

Twenty seven (25.5%) patients in LPV/r group and 73(68.9%) patients in ATZ/r group developed grade 3-4 liver injury. Among the 27 patients who received LPV/r, 19(70.4%) were discharged, 5(18.5%) remained hospitalised at the end of data collection period and 3(11.1%) deceased. Among the 73 patients who received ATZ/r, 71(97.3%) were discharged, one (1.4%) remained hospitalised and one (1.4%) deceased. Among the discharged patients in the ATZ/r group, two were discharged with jaundice, two with elevated liver function and three with hyperbilirubinemia.

Discussion

This study was conducted to compare the proportion of liver injury between COVID-19 patients receiving LPV/r and ATZ/r as part of their COVID-19 treatment. We found that the proportion of grade 3-4 liver injury in the ATZ/r group was higher than LPV/r, which was predominantly presented as hyperbilirubinemia. Consistent with a previous study by McDonald et al. (2012), TB elevation was predominantly observed in ATZ/r patients across all grades. During this 96-week randomised controlled trial, approximately 44% of HIV patients receiving ATZ/r experienced hyperbilirubinemia (grade 3–4) (16).

Numerous studies have suggested the potential risk of hepatotoxicity in HIV patients receiving PI. The incidence of hepatotoxicity in registration trials for selected PIs were reported to be 2.2-9.5 cases in every 100 patients exposed to LPV/r,, 2-7 cases in every 100 patients exposed to atazanavir, 5.3-9.5 cases in every 100 patients exposed to ritonavir (6). During the recent COVID-19 pandemic, several studies have also reported DILI in COVID-19 patients after initiation of LPV/r treatments (9, 17, 18). These patients developed liver injury and were more likely to progress to severe disease, especially among those with abnormal liver test results which was associated with prolonged hospital stay. In our study, approximately half of the patients presented abnormal liver biochemistries upon admission. The development of grade 3-4 liver injury was observed in both ATZ/r and LPV/r group, with higher proportion in ATZ/r group.

Nonetheless, the severity of DILI could be overestimated as it was based on the elevation of liver biochemistry without clinical symptoms such as jaundice (13). Although there are studies that reported strong association of drugs used for treatment of COVID-19 with liver damage, these studies focused on the magnitude of liver enzyme elevations and not on the time to the onset of liver damage. LiverTox described that PI caused an elevation of liver enzyme within one to 12 weeks of starting therapy and the pattern of serum enzyme elevations has varied from hepatocellular to mixed to cholestatic type of liver injury (7). Wang et al. reported that ALT elevations occurred between days 4 and 17 of hospitalisation, with the mean of 7.3 days in severe cases and mean of 10.7 days in mild cases (19). In our study, grade 3-4 elevation of ALT, AST and/or TB was observed on a median of 4 days and 6 days after the initiation of ATZ/r and LPV/r respectively. Liver injury was observed even in patients on a short duration of treatment, suggesting that liver injury can occur in less than one week with PI. Considering the risk of hepatotoxicity of LPV/r, China's guidelines for the diagnosis and treatment of 2019 novel coronavirus (2019-nCoV)-infected pneumonia have recommended that the treatment with LPV/r should be less than 10 days (20).

Some of the predisposing factors that contribute to abnormal liver biochemistries in COVID- 19 patients include direct viral damage, DILI, systemic inflammatory response syndrome and cytokine storm, hypoxia-reperfusion dysfunction and reactivation of pre-existing liver disease (21-23). Two studies demonstrated that abnormal liver function was more likely to occur in severe cases and indicated that these injuries could be attributed to DILI or associated with changes in disease status (17, 19). A liver biopsy from a deceased COVID-19 patient who had elevated liver biochemistries showed moderate microvesicular steatosis, mild lobular and portal activity, indicating that the injury could be induced by the viral infection or hepatotoxic drug use (24). On the contrary, the association of COVID-19 severity, other new concomitant hepatotoxic drug use and presence of CRS with grade 3-4 liver injury were not demonstrated in this study.

Chronic obstructive pulmonary disease (COPD), among the comorbidities, was most likely to be associated with severe COVID-19, intensive care unit (ICU) admission and liver injury in COVID-19 patients (14). Liver injury associated with acute respiratory distress syndrome is proposed to be the result of paracrine action of cytokines and other pro-inflammatory mediators along with hypoxemia, oxidative stress, toxins and hypoperfusion (15). In this study, however, there were significantly more patients with comorbidity of respiratory disease in the LPV/r group but most patients who developed Grade 3 and 4 liver injury belonged to the ATZ/r group instead. More evidence is needed to determine the relationship between respiratory disease and liver injury.

Liver injury in mild cases of COVID-19 is often transient and can return to normal without requiring treatment (19, 25). In this study, majority of patients who developed grade 3-4 liver injury were discharged. Although there were 4 deaths in this study, no evidence showed that DILI was the cause of death. Fan et al. reported that LPV/r caused liver function test elevations that contributed to prolonged length of hospitalisation (9). In our study, prolonged hospitalizations for monitoring of liver function were observed in some patients, while few were discharged with abnormal liver function with appointment to monitor liver function at primary care clinics. In this study, ATZ/r-induced hyperbilirubinemia patients were mostly asymptomatic and bilirubin levels returned to normal after completion or discontinuation of treatment.

There were several limitations in this study. This study was initially designed as a pilot study. Therefore, sample size calculation was not performed. Additionally, this study was conducted in a single center in Malaysia, limiting the generalisability of the findings to other regions with varying epidemiological characteristics. There is also a possible source of bias in the patient selection method due to change in the prescribing practice during the study period. The complete history on hepatotoxic drug use, alcohol intake and traditional supplement use prior to admission were not collected. The serum liver biochemistries were also not taken at fixed intervals. There was no standardised protocol on the duration of PI treatment and recommended management upon development of liver injury in COVID-19 patients. The management of patient was based on the physician's clinical judgment. At the point of producing this report, LPV/r and ATZ/r were no longer included in the latest version of COVID-19 Treatment Guide in Adult Patients at Sungai Buloh Hospital as newer antivirals and immunomodulatory agents have been developed for the treatment of COVID-19 (26). Nonetheless, liver function monitoring is still advocated in COVID-19 patients, including those who are treated with the newer PI, nirmatrelvir/ritonavir.

Conclusion

Liver injury was observed in both groups of COVID-19 patients receiving LPV/r or ATZ/r. There is significantly more grade 3-4 liver injury seen in the ATZ/r group which presented predominantly as hyperbilirubinemia and occurred at a shorter time of onset as compared to LPV/r group. The predisposing factors that significantly contributed to liver injury include type of PI used and the duration of PI treatment. Future research could look into clinically significant liver injury in COVID-19 patients and also to develop guidelines and treatment protocol to improve hepatotoxicity management in this population.

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Conflict of Interest Statement

The authors declare that there is no conflict of interest.

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