CLINICAL TOXICOLOGY
https://doi.org/10.1080/15563650.2020.1842882

Taylor & Francis
Taylor & Francis Group

SHORT COMMUNICATION

Safety and effectiveness concerns of lopinavir/ritonavir in COVID-19 affected patients: a retrospective series

Marc-Antoine Lepage[a,b], Nicholas Rozza[b], Richard Kremer[a,b] and Ami Grunbaum[a,b]

[a]McGill University Health Centre, Montreal, Canada; [b]McGill Faculty of Medecine and Health Sciences, Montreal, Canada

ABSTRACT

Context: Originally developed for treatment of human immunodeficiency virus (HIV), the antiviral combination lopinavir/ritonavir (LPV/r) is being repurposed for treating the novel coronavirus disease (COVID-19) despite minimal experience in this markedly different population and an in-vitro derived EC50 against SARS-CoV-2 several hundred-fold greater than for HIV. We present a case series including a case of severe hyponatremia and a 32-fold overdose raising safety and effectiveness concerns in COVID-19 patients.

Methods: We measured LPV trough concentrations in 12 patients and reviewed their clinical charts for side effects known to occur in HIV patients.

Findings: Compared to established LPV trough concentrations in HIV patients, concentrations in COVID-19 patients were 3-fold greater (19.37 ± 10.12 mcg/mL versus 6.25 mcg/mL). In addition, cholestasis and dyslipidemia toxicity thresholds were exceeded in 12/12 and 11/12 patients respectively. No patients achieved the presumed therapeutic concentration. Side effects included gastrointestinal symptoms (5/12), electrolyte imbalances (4/12), liver enzyme disturbances (5/12) and triglyceride elevations (2/12).

Conclusion: No patients reached presumed therapeutic LPV concentrations despite experiencing side effects and exceeding cholestasis and dyslipidemia toxicity thresholds. This raises concerns for the safety and effectiveness of LPV/r. Clinicians should consider closely monitoring for side effects and not necessarily attribute them to COVID-19.

ARTICLE HISTORY
Received 30 August 2020
Revised 19 October 2020
Accepted 20 October 2020

KEYWORDS
Lopinavir/ritonavir; COVID-19; SARS-CoV-2; safety profile; case series

Context

Lopinavir/ritonavir (LPV/r) is an HIV-protease inhibiting drug combination in which ritonavir's main effect is to potentiate LPV *via* CYP3A4 inhibition. It has gained interest for treating severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) and the associated coronavirus disease (COVID-19). As of July 16, 2020, 59 studies of LPV/r in COVID-19 affected individuals were posted on ClinicalTrials.gov, with accrual goals exceeding 16,000 patients.

The LPV/r monograph [1] reports on five trials, exclusively in the HIV population. Patients were young (mean age 38) and predominantly male (82%).

As of June 20, 2020, our hospital center had hospitalized over 400 COVID-19 patients with a median age of 74.0 (interquartile range 62.– 84.0) and 48% male. Twenty individuals were treated with LPV/r. We report our clinical and laboratory findings in a subset of twelve individuals for whom measured trough drug concentrations were available and extrapolate on the kinetics and effectiveness of LPV/r in this population.

Methods

In the context of a clinical trial (clinicaltrials.gov/ct2/show/NCT04330690), COVID-19 patients were treated with LPV/r 400/100 mg twice daily (as per HIV treatment regimen) for 14 days or until hospital discharge, whichever occurred first.

Unexpected development of severe hyponatremia in one individual initiated investigations in 12 temporally consecutive SARS-CoV-2 infected patients treated with LPV/r to determine LPV troughs and clinical and laboratory abnormalities possibly related to this therapy. Patients treated with LPV/r prior to the index case were not included as LPV measurements were unobtainable.

Plasma trough measurements of total LPV were performed with a validated liquid chromatography tandem mass spectrometry assay. Charts were comprehensively reviewed for all new adverse events known to occur in HIV patients [1] with onset at least 24 h after treatment initiation.

Findings

Trough concentrations (12 h post-dosing) were measured 2–7 days after treatment initiation in 11 of 12 patients (Table 1). In these 11 individuals, mean LPV trough concentrations were higher than concentrations expected in HIV patients (19.37 mcg/mL [standard deviation 10.12] vs 6.25 mcg/mL).

Patient 12 received 160 mL of LPV/r solution (equivalent to 12,800/3,200mg) in a single dose due to a dosing error on

CONTACT Ami Grunbaum ami.grunbaum@mcgill.ca McGill University Health Center – Glen site, 1001 Decarie Blvd, Montreal H4A 3J1, Canada

Table 1. Clinical and Laboratory Findings in COVID-19 Affected Patients Treated with Lopinavir/Ritonavir.

Patient ID	Age (years)	Sex	Duration of Therapy (days)	LPV 12 h Trough Levels (mcg/mL)	Clinical Abnormalities	Laboratory Abnormalities (value; normal range)	Length of Stay (days)	Disposition
1	82	F	5	20.99	Nausea	↑ total bilirubin (29.2; 1.7–18.9 μmol/L)	21	Home
					Bloating	Hyponatremia (115; 133–143 mmol/L)		
					Dizziness	Hyperkalemia (5.8; 3.5–5 mmol/L)		
					Anorexia			
2	28	M	9	10.07	None	↑ ALT (99; 6–45 U/L)	9	Home
3	89	F	6	8.50	None	None	11	Deceased
4	75	F	9	23.19	None	None	8	Home
5	82	F	14	40.64	Diarrhea	None	29	Home
6	31	F	5	13.34	None	↑ total bilirubin (33.5; 1.7–18.9 μmol/L)	15	Deceased
						Hyperkalemia (6.3; 3.5–5 mmol/L)		
7	72	M	6	26.12	Diarrhea	Hypophosphatemia (0.6; 0.8–1.45 mmol/L)	7	Home
8	59	F	8	12.67	None	None	10	Home
9	51	M	5	13.29	Anorexia	Hyponatremia (131; 133–143 mmol/L)	6	Home
					Diarrhea	↑ ALT (57; 6–45 U/L)		
10	81	M	10	31.39	None	↑ TG (2.2; 0.1–1.7 mmol/L)	26	Deceased
11	84	F	2	12.92	Nausea/Vomiting	None	3	Home
12	59	M	4	94.33*	Acalculous cholecystitis	↑ GGT (159; 7–50 U/L)	131	Rehabilitation Centre
					Bradycardia	↑ Lipase (118; 14–45 U/L)		
						↑ TG (7.69; 0.1–1.7 mmol/L)		
Mean (SD)	66 (21)		7 (3)	19.37 (10.12)**			23 (35)	

LPV: lopinavir; ALT: alanine aminotransferase; GGT: gamma glutamyl transferase; TG: triglycerides; SD: standard deviation.
*Extrapolated 12 h trough based on concentrations measured at 34 (34.5 mcg/mL) and 114 (0.89 mcg/mL) hours post overdose.
**Excluding the extrapolated 12 h trough from the overdose (Patient 12).

day 4. At the time, he was receiving enteral nutrition (stopped 4 h post overdose and resumed 12 h later to permit proning). The error was only discovered 24 h later and immediately reported to Poison Control, who recommended against any maneuvers to reduce delayed absorption.

LPV concentrations 34 h (t_1) and 114 h (t_2) post overdose were 34.5 mcg/mL (C_1) and 0.89 mcg/mL (C_2). The calculated half-life (t_2^1) is 15.2 h using $t_2^1 = \ln 2/k = \ln 2/-\ln(C_2/C_1)/(t_2 - t_1)$ where k is the elimination rate constant. We can then extrapolate a 12-hour trough ($C_{t_{12}}$) of 94.3 mcg/mL with $C_{t_{12}} = C_2/e^{-k(t_2-t_{12})} = 0.89/e^{-0.0457(114-12)}$ (Figure 1). Assuming that the overdose follows pharmacokinetics similar to therapeutic dosing of LPV/r in the HIV population with maximum plasma concentration (C_{max}) at 4 h [1], we extrapolate a C_{max} of 136.0 mcg/mL.

6 of 12 patients had clinically relevant adverse events. Gastrointestinal symptoms were observed in 5/12 patients; electrolyte, liver, and triglyceride disturbances in 4/12, 5/12, and 2/12 patients respectively.

All 12 patients exceeded reported cholestasis toxicity thresholds (6.43 mcg/mL) [2]. Patient 12 had an acute acalculous cholecystitis confirmed by ultrasound performed for persistent fevers despite broad-coverage antibiotics for ventilator-associated pneumonia and central line associated bloodstream infection. 11/12 patients exceeded dyslipidemia toxicity thresholds (9.71 mcg/mL) [3]. Patients 10 and 12 were receiving propofol sedation. Patient 12's hypertriglyceridemia necessitated cessation of propofol.

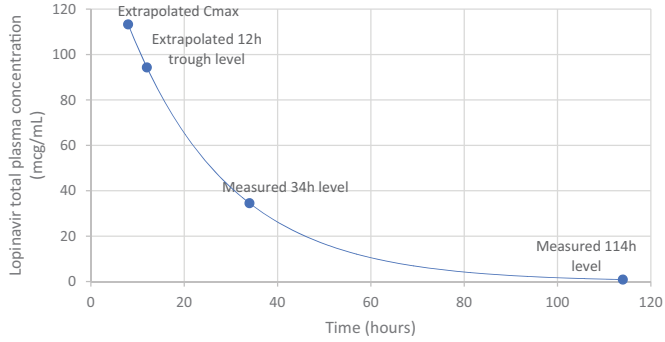

Figure 1. Extrapolated Pharmacokinetics of Lopinavir Overdose in Patient 12. Cmax: maximal plasma concentration reached at 4 hours.

By day 3 of treatment, Patient 1 developed severe hyponatremia (122 mmol/L, baseline 134 mmol/L) with appropriate urinary retention (<15mmol/L), mild metabolic acidosis and hyperkalemia. In the clinical context of acute infection, morning cortisol was inappropriately normal at 285 nmol/L (120–535nmol/L). This was associated with weakness, dizziness, and new onset abdominal pain. Her hyponatremia continued to worsen, nadiring at 115 mmol/L by day 5 despite large doses of NaCl replacement. At this point, she received 50 mg of hydrocortisone before starting fludrocortisone 0.05–0.1 mg daily for 5 days. Electrolyte imbalances and symptoms quickly improved with cortico-therapy, consistent with a diagnosis of adrenal insufficiency. It was later discovered that her usual mometasone was changed to

beclomethasone on day 1 to avoid interactions between CYP3A4 metabolized corticosteroids and LVP/r [4]. However, beclomethasone was initially unavailable and only started on day 4.

In addition to acute acalculous cholecystitis and hypertriglyceridemia, patient 12 experienced sustained bradycardia in the 48 h following the overdose of 50 (IQR 45–55) beats per minute (bpm) from a baseline of 65 (IQR 65–70) bpm. He was chronically on sotalol for atrial fibrillation which had to be discontinued. As sotalol is not metabolized [5], it is unaffected by ritonavir's inhibition of CYP3A4. Renal function was normal at the time. The bradycardia may have been due to either of the drugs or a synergistic effect of both.

Ultimately, 3 patients died and 9 were discharged with an average hospitalization length of 29 days (standard deviation 35).

Discussion

Schoergenhofer [6] and Gregoire [7] have reported that COVID-19 patients treated with LPV/r develop higher LPV concentrations than HIV patients. In these reports, patients were also older than in the LPV/r monograph trials [1]. Older individuals typically receive more medications which may increase drug-drug interactions or lead to the accumulation of side effects. This difference may also be attributed to downregulation of hepatic CYP450 in the acute inflammatory phase [8] and possibly the lower metabolic and clearance rates in this acutely ill and elderly population. Notably, nearly all trough concentrations in our patients exceeded toxicity thresholds for cholestasis [2] and dyslipidemia [3]. High concentrations may also increase frequency and severity of other potential complications, including gastrointestinal symptoms, acute gastritis, and severe anemia as reported by Cao [9].

LPV is 98–99% protein bound. In the absence of human serum (i.e., 100% freely available drug), in-vitro data suggests a half-maximal effective concentration (EC50) for LPV against SARS-CoV-2 of 16.74 mcg/mL [10] compared to 0.006–0.017 mcg/mL for various HIV strains [1]. This concentration is unlikely to be obtained in-vivo without excessive risk, as an accidental overdose of 32 times the intended amount still resulted in sub-therapeutic levels. Even using the calculated C_{max}, peak free LPV concentration is only 1.36–2.72 mcg/mL (1–2% of the total C_{max} calculated above).

The reason(s) why plasma concentrations were not higher in the setting of a 32-fold overdose are unclear. Drug absorption may have been compromised by the effects of critical illness. For such large volume overdose, holding enteral feeds may also have contributed as food increases LVP/r solution absorption [1]. It is also possible that intestinal transporters were saturated [11] and that splanchnic circulation was decreased [12]. Finally, co-administration of dexamethasone, a known CYP inducer, may have contributed to increased metabolization and clearance of LPV [1]. No other potential drug-drug interactions were identified.

Our report has limitations. Although comprehensive, our chart review is at risk of inconsistencies in extraction and clinical documentation. While our findings ($n = 12$) are consistent with those of Schoergenhofer [6] ($n = 8$) and Gregoire [7] ($n = 12$), the three studies are small, limiting external validity to the entire COVID-19 population. Finally, there is no control group, preventing conclusions on efficacy and limiting conclusions on safety as many of LPV/r's side effects may be consequences of COVID-19 itself.

Conclusion

Our report supports previous findings that treating COVID-19 affected patients with standard HIV LPV/r dosages yields approximately 3-fold higher trough LPV concentrations which may result in adverse events [6,7,9]. Despite this risk, the in-vitro derived EC50 for LPV on SARS-CoV-2 was not reached in any of our patients, including the 32-fold overdose. This may in part explain why Cao et al. demonstrated no benefit from LPV/r in hospitalized patients [9]. Consistent with these findings and in light of issues raised by their interim analysis, on July 4, 2020, the World Health Organization reported the Solidarity Trial's recommendation to immediately discontinue the LPV/r treatment arms due to futility and safety concerns [13].

Most importantly, this report will impact on patients enrolled in other trials or who are offered this treatment as part of their clinical care. Our findings should alert clinicians to pay attention for LPV/r side effects and complications. Closer clinical monitoring will enable early detection of toxicity and allow for therapeutic adjustment and possible discontinuation of LPV/r.

Disclosure statement

No potential conflict of interest was reported by the author(s).

ORCID

Marc-Antoine Lepage http://orcid.org/0000-0002-7518-185X
Richard Kremer http://orcid.org/0000-0002-7053-2139
Ami Grunbaum http://orcid.org/0000-0001-8745-0036

References

[1] AbbVie Corporation. Product Monograph. Kaletra. Lopinavir/ritonavir [cited 2019, Sept 27]. Available from: https://www.abbvie.ca/content/dam/abbvie-dotcom/ca/en/documents/products/KALETRA_PM_EN.pdf.
[2] Seminari E, Gentilini G, Galli L, et al. Higher plasma lopinavir concentrations are associated with a moderate rise in cholestasis markers in HIV-infected patients. J Antimicrob Chemother. 2005; 56(4):790–792.
[3] Gutiérrez F, Padilla S, Navarro A, et al. Lopinavir plasma concentrations and changes in lipid levels during salvage therapy with lopinavir/ritonavir-containing regimens. J Acquir Immune Defic Syndr. 2003;33(5):594–600.
[4] Phillips A. Important Safety Information Regarding a Drug Interaction Between Fluticasone Propionate (Flonase/Flovent/Advair) and Ritonavir (Norvir/Kaletra). Health Canada; 2004.
[5] Bayer HealthCare Pharmaceuticals Inc. Betapace (sotalol HCl) 2010. [cited 2019, Sept 27]. Available from: https://www.accessdata.fda.gov/drugsatfda_docs/label/2011/019865s019lbl.pdf.
[6] Schoergenhofer C, Jilma B, Stimpfl T, et al. Pharmacokinetics of lopinavir and ritonavir in patients hospitalized with

Coronavirus Disease 2019 (COVID-19). Ann Intern Med. 2020; 173(8):670–672.

[7] Gregoire M, Le Turnier P, Gaborit BJ, et al. Lopinavir pharmacokinetics in COVID-19 patients. J Antimicrob Chemother. 2020; 75(9):2702–2704.

[8] Morgan ET. Impact of infectious and inflammatory disease on cytochrome P450-mediated drug metabolism and pharmacokinetics. Clin Pharmacol Ther. 2009;85(4):434–438.

[9] Cao B, Wang Y, Wen D, et al. A trial of lopinavir-ritonavir in adults hospitalized with severe Covid-19. N Engl J Med. 2020;382(19): 1787–1799.

[10] Choy K-T, Wong AY-L, Kaewpreedee P, et al. Remdesivir, lopinavir, emetine, and homoharringtonine inhibit SARS-CoV-2 replication in vitro. Antiviral Res. 2020;178:104786.

[11] Holmstock N, Annaert P, Augustijns P. Boosting of HIV protease inhibitors by ritonavir in the intestine: the relative role of cytochrome P450 and P-glycoprotein inhibition based on Caco-2 monolayers versus in situ intestinal perfusion in mice. Drug Metab Dispos. 2012;40(8):1473–1477.

[12] De Paepe P, Belpaire FM, Buylaert WA. Pharmacokinetic and pharmacodynamic considerations when treating patients with sepsis and septic shock. Clin Pharmacokinet. 2002;41(14):1135–1151.

[13] World Health Organization. "Solidarity" clinical trial for COVID-19 treatments 2020. [cited 2020, July 7]. Available from: https://www.who.int/emergencies/diseases/novel-coronavirus-2019/global-research-on-novel-coronavirus-2019-ncov/solidarity-clinical-trial-for-covid-19-treatments.