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Original Article

Real world effectiveness of early ensitrelvir treatment in patients with SARS-CoV-2, a retrospective case series

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ABSTRACT

Background: Ensitrelvir, a 3C-like protease inhibitor, received emergency approval in Japan in November 2022 for treating non-hospitalized patients with mild-to-moderate COVID-19. However, confirmation of its real-world clinical effectiveness is limited.

Methods: This retrospective study evaluated 18 vaccinated outpatients (15 men; median age, 39.5 years; range, 26–56), treated with a 5-day oral ensitrelvir regimen (375 mg loading dose, followed by 125 mg daily) between December 1, 2022, and January 31, 2023. Nasal swabs were collected on days 0, 3, 6, and 9 for RT-qPCR to assess viral load. Variants were identified by Sanger sequencing, and outcomes were compared to historical controls. Patients were followed for 60 days to monitor for post-acute sequelae of COVID-19 (PASC).

Results: Symptoms such as mild fever and sore throat improved rapidly after one day of ensitrelvir treatment, with 66 % of patients recovering within six days. All individuals were infected with the BA.5 Omicron variant. Viral loads, as measured by Ct values, increased significantly from 21.82 at symptom onset to 37.65 b y day 6, with SARS-CoV-2 RNA undetectable in most patients by day 9. Those treated within 48 h of symptom onset showed the viral load reduction. Compared to historical controls, where symptom resolution took 8.5 days, ensitrelvir shortened recovery time to as little as 1.4 days for over 66 % of patients.

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Conclusion: Ensitrelvir treatment resulted in rapid symptom relief and significant viral load reduction, with no adverse events, viral rebound, or PASC symptoms, demonstrating its potential efficacy and safety. Larger studies are needed for further confirmation.

1. Introduction

During the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) pandemic, various drugs have been repurposed or developed for treating patients with SARS-CoV-2 infection [1–4]. The risk of SARS-CoV-2 poses a particular risk to people living with preexisting conditions that impair immune response or amplify pro-inflammatory response and risk of development of post-COVID syndrome (long COVID) [5,6]. While vaccination has lowered the risk of severe illness, waves of infection driven by new immune-evasive variants continue to challenge healthcare systems globally [7–11]. This issue is especially pressing in low-income countries, where limited access to mRNA vaccines and the high cost of new antiviral treatments underscore the urgent need for affordable, safe, and widely accessible therapeutic options [7,8,10–12]. Ensitrelvir, a novel oral SARS-CoV-2 3C-like protease inhibitor, was granted emergency use authorization by the Japanese Ministry of Health, Labour and Welfare (November 2022) for the treatment of non-hospitalized patients with mild-to-moderate SARS-CoV-2 infection during the pandemic [13–15]. The drug was intended to lower the risk of hospitalization, minimize hyperinflammation, prevent death or post-COVID-19 conditions in these patients, and reduce the burden on healthcare systems [16–18]. Previous trials have shown antiviral efficacy with a 5-day oral treatment regimen of ensitrelvir against different SARS-CoV-2 variants [14,15,19,20]. However, there is limited or non-real-world evidence available outside of these investigations. This retrospective study aimed to evaluate the treatment patterns, patient characteristics, and outcomes of individuals treated with ensitrelvir following its emergency approval during the Omicron-dominant period in Japan.

2. Methods

This retrospective case series presents data on the use of ensitrelvir in treating 18 symptomatic, vaccinated outpatients with mild-to-moderate SARS-CoV-2 infection, following its emergency approval between December 1, 2022, and January 31, 2023. Individuals (15 men; median age, 39.5 years; range, 26–56), were treated with a 5-day oral regimen of ensitrelvir consisting of three tablets (each tablet 125 mg) every 8 h on the first day and one tablet for the following four days.

Nasal swab samples were collected from each patient at days 0, 3, 6, and 9 for viral load assessment using reverse transcription quantitative real-time polymerase chain reaction (RT qPCR) using Gene Xpert Xpress

Table 1
Demographics and baseline disease characteristics of patients (n = 18) infected with SARS-CoV-2.

Patients ID	Age (years)	Sex	Duration of symptoms onset (days)[a]	Fever[b]	Symptoms on set	Underlying diseases	Duration until symptoms relieved (days)	Duration until symptoms disappeared (days)	Post-acute sequelae of COVID-19 (PASC) symptoms[c]	Adverse Events
XO1	56	Male	2	high	Headache, Cough, Sputum	–	1	2	non	non
XO2	55	Male	2	high	Chills, Headache, Nasal congestion	–	1	2	non	non
XO3	34	Male	2	none	Sore throat, Hoarseness	–	1	2	non	non
XO4	31	Female	0	mild	Tiredness	–	2	3	non	non
XO5	51	Male	0	mild	Sore throat	–	1	2	non	non
XO6	26	Male	0	none	Sore throat, Hoarseness, Sputum	–	1	1	non	non
XO7	49	Female	2	high	Sore throat, Sputum	–	1	6[c]	non	non
XO8	49	Male	0	mild	Sore throat	–	1	6[c]	non	non
XO9	36	Male	1	none	Sore throat	–	1	6[c]	non	non
XO10	47	Male	0	none	Sore throat, Rhinny nose	Chronic sinusitis	1	3	non	non
XO11	41	Male	1	mild	Sore throat, Rhinny nose, Cough	Allergic rhinitis, Bronchial asthma	2	6[d]	non	non
XO12	30	Male	0	mild	Sore throat	–	1	6	non	non
XO13	53	Male	0	high	Sore throat	Allergic rhinitis	1	6[c]	non	non
XO14	38	Male	1	none	Sore throat, Hoarseness	–	1	3	non	non
XO15	49	Male	1	mild	Cough	–	1	2	non	non
XO16	33	Male	1	mild	Sore throat	–	1	4	non	non
XO17	30	Male	2	high	Sore throat	–	1	5	non	non
XO18	28	Female	2	high	Sore throat, Cough, Joint pain, Back pain	–	1	6	non	non

Ct; cycle threshold.
[a] 0 (≤24 Hours), 1 (>24 - ≤48 Hours), 2 (>48 - ≤72 Hours).
[b] Mild: 37.1 °C - 38.0 °C; High: ≥38.1 °C.
[c] Mild symptoms remained on day 6: mild sore throat.
[d] Mild symptoms remained on day 6: mild cough.
[e] Post-acute sequelae of COVID-19 (PASC) symptoms were followed up for 60 days.

SARS-CoV-2. Variants were identified through Sanger sequencing, as described previously [7,8,10,12,21]. Results were analyzed in relation to the days of onset of symptoms and compared to our historical control data to assess treatment effects. Individuals were followed for up to 60 days to monitor for any symptoms related to post-acute sequelae of COVID-19 (PASC).

3. Results

Patient demographics and clinical characteristics are summarized in Table 1. Most common symptoms were mild fever (37.1 °C - 38.0 °C) and sore throat, which were reduced after one day of ensitrelvir treatment. Of significance, 33 % of patients had their symptoms improve within two days and another 33 % had their symptoms improve between days 3 and 6 of starting ensitrelvir treatment (Table 1). One patient had a mild cough and five others had mild sore throat for over six days. The mean cycle threshold value (Ct) for the onset of symptoms was 21.82, and it was increased to 37.65 on day six (Figs. 1 and 2). However, 10 patients Ct value over 35 within six days of ensitrelvir treatment (Figs. 1 and 3). One patient's SARS-CoV-2 RNA became undetectable on day 3, two patients on day 6, and twelve patients on day 9 (Fig. 1). Interestingly, there was a significant ($p < 0.5$) Ct value difference between day 0 and day 3 (delta Ct) of ensitrelvir treatment in patients with symptoms onset ≤24 h (0 days) and >24 - ≤ 48 h (1 day) (Fig. 3). Also, there were significant ($p < 0.5$) Ct value differences between day 0 and day 3 of ensitrelvir treatment in patients with symptoms onset ≤24 h (0 days) and >24 (1 and 2 days). As these patients were treated as part of the routine standard care, we do not have an existing control arm. However, we compared our results with our historical control data to assess the treatment effects. For mild symptomatic omicron variant outpatients without any treatment, median times to resolution of symptoms were 8.5 days (95 % CI 6−11 days) (data not shown). However, the majority of (>66 %) patients had their symptoms resolved for 1.4 days (95 % CI 1−6 days) following ensitelvir treatment. All individuals were infected with the BA.5 Omicron variant. We did not observe any increase in the Ct value (lower Ct value), which would indicate a higher viral load, nor did any patients experience a recurrence of symptoms or the emergence of new adverse events. Additionally, none of the patients showed signs or symptoms associated with post-acute sequelae of COVID-19 (PASC) during the follow-up period.

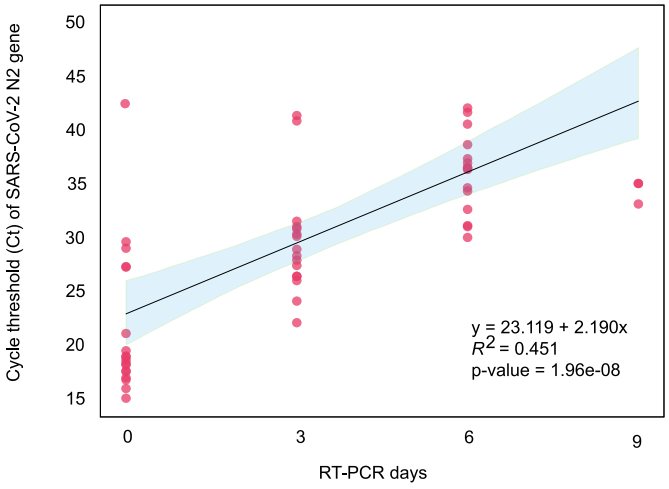

Fig. 1. Viral load detected in nasal swabs obtained from patients (n = 18) infected with SARS-CoV-2 in day 0,3,6 and 9 (Cycle threshold (Ct) values of N2 gene on RT-PCR assay). The line within the box corresponds to the median and the cross to the mean of the distribution, while the whiskers indicate the highest and the lowest values. *p* (two-tail) vale was calculated form paired *t*-test and $p < 0.5$ consider as significant.

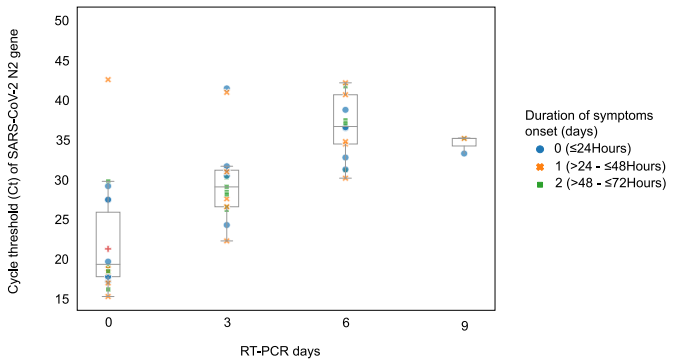

Fig. 2. Duration of symptoms onset (days) and viral load detected in nasal swabs obtained from patients (n = 18) infected with SARS-CoV-2. The line within the box corresponds to the median and the cross to the mean of the distribution, while the whiskers indicate the highest and the lowest values. *p* (two-tail) vale was calculated form paired *t*-test and $p < 0.5$ consider as significant.

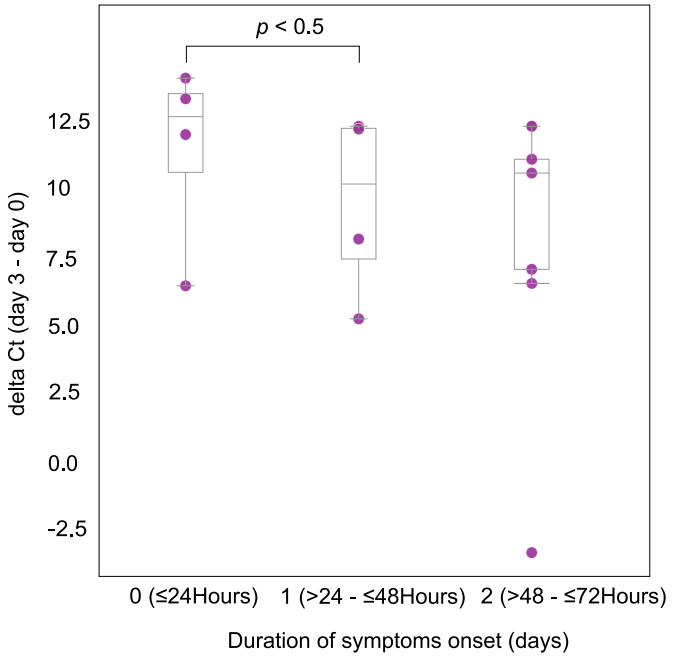

Fig. 3. Duration of symptoms onset (days) and viral load difference between day 0 and 3 (delta Ct). The line within the box corresponds to the median and the cross to the mean of the distribution, while the whiskers indicate the highest and the lowest values. *p* (two-tail) vale was calculated form paired *t*-test and $p < 0.5$ consider as significant.

4. Discussion

This is the first time that real-world clinical data on the reduction of SARS-CoV-2 viral load using ensitrelvir is being presented. One advantage of this study is that it was conducted during the pandemic when the BA.5 Omicron subvariant caused most infections. The incidence of viral load reduction observed in this study was similar to or greater than that seen in prior clinical trials [14,15,19]. This study also found that when given within 24 h of symptom onset, ensitrelvir substantially affects viral clearance and symptom relief. These results reemphasize that drugs that target viral replication should be most effective if administered early in the viremic phase [1,2]. Early antiviral responses may lower the risk of severe illness, development of immunological complications, and long COVID symptoms [1,2,22]. Our study also did not observe any viral load rebound, adverse events, recurrence of moderate-to-severe

symptoms, or development of resistance to ensitrelvir among patients and symptoms of post-acute sequelae of COVID-19 (PASC). As all individuals were infected with the BA.5 Omicron variant, the data clearly demonstrate that ensitrelvir had a beneficial effect not only on symptom resolution but also in preventing the development of post-acute sequelae of COVID-19 (PASC).

Additionally, patients' treatment with ensitrelvir was generally well tolerated. This study was limited by its relatively small cohort size; however, it was still able to generate important preliminary findings. Furthermore, the use of qPCR in this study to determine viral load does not reflect infectious viruses, which limited some interpretations. However, it was found that early treatment with ensitrelvir could significantly change the clinical trajectory of individuals towards fewer hospitalizations related to SARS-CoV-2 infection and reduce the risk of adverse events. Our study also suggests that compared to currently approved antiviral drugs used in this setting, ensitrelvir shows promising efficacy in rapid viral titer and viral RNA reduction, as well as favorable clinical outcomes, including earlier symptom relief or prevention of severe SARS-CoV-2 infections [23,24] and prevention of symptoms of post-acute sequelae of COVID-19 (PASC). As a preliminary retrospective study, the authors note that larger clinical data is needed to confirm observed findings.

5. Conclusion

Ensitrelvir treatment led to rapid symptom improvement in most BA.5 Omicron variant patients, with over 66 % recovering within six days. Viral load decreased significantly, especially in those treated within 48 h of symptom onset. No patients experienced symptom recurrence, adverse events, or post-acute sequelae, indicating its potential efficacy and safety.

CRediT authorship contribution statement

Shuichi Abe: Conceptualization, Data curation, Formal analysis, Funding acquisition, Investigation, Methodology, Project administration, Writing – review & editing. **Dhammika Leshan Wannigama:** Conceptualization, Data curation, Formal analysis, Funding acquisition, Investigation, Methodology, Project administration, Resources, Software, Supervision, Validation, Writing – original draft. **Yu Suzuki:** Data curation, Formal analysis, Writing – review & editing. **Daisuke Akaneya:** Data curation, Formal analysis, Writing – review & editing. **Junko Igarashi:** Data curation, Methodology, Writing – review & editing. **Mayu Suto:** Data curation, Methodology. **Kazunori Moriya:** Data curation, Formal analysis, Resources, Writing – review & editing. **Daisuke Ishizawa:** Data curation, Formal analysis, Resources. **Yoshikazu Okuma:** Conceptualization, Data curation, Validation, Writing – review & editing. **Parichart Hongsing:** Conceptualization, Data curation, Formal analysis, Funding acquisition, Investigation, Writing – original draft, Writing – review & editing. **Cameron Hurst:** Data curation, Formal analysis, Supervision, Writing – review & editing. **Thammakorn Saethang:** Data curation, Formal analysis, Resources. **Paul G. Higgins:** Conceptualization, Data curation, Formal analysis, Investigation, Supervision, Writing – review & editing. **Stephen M. Stick:** Supervision, Writing – review & editing. **Anthony Kicic:** Conceptualization, Data curation, Formal analysis, Resources, Software, Supervision, Writing – review & editing.

Ethical approval

This study was conducted in accordance with the principles of the Declaration of Helsinki, Good Clinical Practice guidelines, and other applicable laws and regulations. The study was reviewed and approved by the institutional review board at Yamagata Prefectural Central Hospital, Yamagata, Japan (12/2022). All patients or their legally acceptable representatives provided written informed consent.

Data availability statement

The authors confirm that the data supporting the findings of this study are available within the article.

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Declaration of competing interest

No author declares any potential conflict of interest or competing financial or non-financial interest in relation to the manuscript.

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References

[1] Murakami N, Hayden R, Hills T, Al-Samkari H, Casey J, Del Sorbo L, et al. Therapeutic advances in COVID-19. Nat Rev Nephrol 2023;19(1):38–52.
[2] Ning Q, Wu D, Wang X, Xi D, Chen T, Chen G, et al. The mechanism underlying extrapulmonary complications of the coronavirus disease 2019 and its therapeutic implication. Signal Transduct Targeted Ther 2022;7(1):57.
[3] Wannigama DL, Hurst C, Phattharapornjaroen P, Hongsing P, Sirichumroonwit N, Chanpiwat K, et al. Early treatment with fluvoxamine, bromhexine, cyproheptadine, and niclosamide to prevent clinical deterioration in patients with symptomatic COVID-19: a randomized clinical trial. eClinicalMedicine 2024;70.
[4] Imsuwansri T, Jongthitinon T, Pojdoung N, Meesiripan N, Sakarin S, Boonkrai C, et al. Assessment of safety and intranasal neutralizing antibodies of HPMC-based human anti-SARS-CoV-2 IgG1 nasal spray in healthy volunteers. Sci Rep 2023;13(1):15648.
[5] Davis HE, McCorkell L, Vogel JM, Topol EJ. Long COVID: major findings, mechanisms and recommendations. Nat Rev Microbiol 2023.
[6] Wannigama DL, Amarasiri M, Phattharapornjaroen P, Hurst C, Modchang C, Chadsuthi S, et al. Increased faecal shedding in SARS-CoV-2 variants BA.2.86 and JN.1. Lancet Infect Dis 2024;24(6):e348–50.
[7] Wannigama DL, Amarasiri M, Phattharapornjaroen P, Hurst C, Modchang C, Chadsuthi S, et al. Wastewater-based epidemiological surveillance of SARS-CoV-2 new variants BA.2.86 and offspring JN.1 in south and Southeast Asia. J Trav Med 2024.
[8] Wannigama DL, Amarasiri M, Hongsing P, Hurst C, Modchang C, Chadsuthi S, et al. COVID-19 monitoring with sparse sampling of sewered and non-sewered wastewater in urban and rural communities. iScience 2023:107019.
[9] Rad SMAH, Wannigama DL, Hirankarn N, McLellan AD. The impact of non-synonymous mutations on miRNA binding sites within the SARS-CoV-2 NSP3 and NSP4 genes. Sci Rep 2023;13(1):16945.
[10] Wannigama DL, Amarasiri M, Phattharapornjaroen P, Hurst C, Modchang C, Chadsuthi S, et al. Tracing the new SARS-CoV-2 variant BA.2.86 in the community through wastewater surveillance in Bangkok, Thailand. Lancet Infect Dis 2023;23(11):e464–6.
[11] Anupong S, Chantanasaro T, Wilasang C, Jitsuk NC, Sararat C, Sornbundit K, et al. Modeling vaccination strategies with limited early COVID-19 vaccine access in low- and middle-income countries: a case study of Thailand. Infectious disease modelling. 2023.
[12] Anupong S, Chadsuthi S, Hongsing P, Hurst C, Phattharapornjaroen P, Ali Hosseini RSM, et al. Exploring indoor and outdoor dust as a potential tool for detection and monitoring of COVID-19 transmission. iScience 2024:109043.
[13] Sasaki M, Tabata K, Kishimoto M, Itakura Y, Kobayashi H, Ariizumi T, et al. S-217622, a SARS-CoV-2 main protease inhibitor, decreases viral load and ameliorates COVID-19 severity in hamsters. Sci Transl Med 2023;15(679):eabq4064.
[14] Mukae H, Yotsuyanagi H, Ohmagari N, Doi Y, Imamura T, Sonoyama T, et al. A randomized phase 2/3 study of ensitrelvir, a novel oral SARS-CoV-2 3C-like protease inhibitor, in Japanese patients with mild-to-moderate COVID-19 or asymptomatic SARS-CoV-2 infection: results of the phase 2a part. Antimicrob Agents Chemother 2022;66(10):e00697. 22.
[15] Shimizu R, Sonoyama T, Fukuhara T, Kuwata A, Matsuo Y, Kubota R. Safety, tolerability, and pharmacokinetics of the novel antiviral agent ensitrelvir fumaric acid, a SARS-CoV-2 3CL protease inhibitor, in healthy adults. Antimicrob Agents Chemother 2022;66(10):e0063222.
[16] Yamato M, Kinoshita M, Miyazawa S, Seki M, Mizuno T, Sonoyama T. Ensitrelvir in patients with SARS-CoV-2: a retrospective chart review. J Infect Chemother 2024;30(9):946–50.

[17] Lin M, Zeng X, Duan Y, Yang Z, Ma Y, Yang H, et al. Molecular mechanism of ensitrelvir inhibiting SARS-CoV-2 main protease and its variants. Commun Biol 2023;6(1):694.
[18] Yotsuyanagi H, Ohmagari N, Doi Y, Yamato M, Fukushi A, Imamura T, et al. Prevention of post COVID-19 condition by early treatment with ensitrelvir in the phase 3 SCORPIO-SR trial. Antivir Res 2024;229:105958.
[19] Mukae H, Yotsuyanagi H, Ohmagari N, Doi Y, Sakaguchi H, Sonoyama T, et al. Efficacy and safety of ensitrelvir in patients with mild-to-moderate COVID-19: the phase 2b part of a randomized, placebo-controlled, phase 2/3 study. Clin Infect Dis 2022.
[20] Nobori H, Fukao K, Kuroda T, Anan N, Tashima R, Nakashima M, et al. Efficacy of ensitrelvir against SARS-CoV-2 in a delayed-treatment mouse model. J Antimicrob Chemother 2022;77(11):2984–91.
[21] Wannigama DL, Amarasiri M, Hurst C, Phattharapornjaroen P, Abe S, Hongsing P, et al. Tracking COVID-19 with wastewater to understand asymptomatic transmission. Int J Infect Dis 2021;108:296–9.
[22] Hu B, Guo H, Zhou P, Shi Z-L. Characteristics of SARS-CoV-2 and COVID-19. Nat Rev Microbiol 2021;19(3):141–54.
[23] Wong CKH, Au ICH, Lau KTK, Lau EHY, Cowling BJ, Leung GM. Real-world effectiveness of early molnupiravir or nirmatrelvir–ritonavir in hospitalised patients with COVID-19 without supplemental oxygen requirement on admission during Hong Kong's omicron BA.2 wave: a retrospective cohort study. Lancet Infect Dis 2022;22(12):1681. 93.
[24] Vegivinti CTR, Evanson KW, Lyons H, Akosman I, Barrett A, Hardy N, et al. Efficacy of antiviral therapies for COVID-19: a systematic review of randomized controlled trials. BMC Infect Dis 2022;22(1):107.