
Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Clinical Microbiology and Infection 28 (2022) 1657–1658

Contents lists available at ScienceDirect

Clinical Microbiology and Infection

journal homepage: www.clinicalmicrobiologyandinfection.com

Letter to the Editor

# Virological and clinical rebounds of COVID-19 soon after nirmatrelvir/ritonavir discontinuation

Guido Antonelli [1], Daniele Focosi [2,*], Ombretta Turriziani [1], Marco Tuccori [3,4], Rossella Brandi [5], Silvia Fillo [5], Camilla Ajassa [6], Florigio Lista [5], Claudio M. Mastroianni [6]

[1] Department of Molecular Medicine, Sapienza University of Rome, Sapienza University Hospital "Policlinico Umberto I", Rome, Italy
[2] North-Western Tuscany Blood Bank, Pisa University Hospital, Pisa, Italy
[3] Unit of Pharmacology and Pharmacovigilance, Department of Clinical and Experimental Medicine, University of Pisa, Pisa, Italy
[4] Unit of Adverse Drug Reactions Monitoring, Pisa University Hospital, Pisa, Italy
[5] Scientific Department, Army Medical Center, Rome, Italy
[6] Department of Public Health and Infectious Diseases, Sapienza University Hospital "Policlinico Umberto I", Rome, Italy

ARTICLE INFO

*Article history:*
Received 4 May 2022
Received in revised form
9 June 2022
Accepted 28 June 2022
Available online 2 July 2022

Editor: L. Kaiser

*To the Editor,*

We read with interest the recent commentary by Girardin et al. on the pharmacokinetic interactions with nirmatrelvir/ritonavir published online in *Clinical and Microbiological Infections* on 28 March 2022 [1]. Oral small-chemical antivirals have been recently authorized around the world and come with the promise of simplifying the management and reducing hospitalization of COVID-19 outpatients at risk of disease progression. Nirmatrelvir tablets co-packaged with ritonavir tablets (Paxlovid, Pfizer, New York, NY, USA) (300/100 mg dose twice daily for 5 days) was granted a conditional marketing authorization by the European Medicines Agency (EMA) on 29 January 2022 on the basis of a phase 2/3 randomised controlled trial (RCT) in 2246 unvaccinated outpatients (mostly during the wave driven by the Delta variant of concern—July-December 2021) in which treatment at a median of 3 days since onset of symptoms led to an 88.9% reduction in the relative risk of hospitalization (-5.81%) [2]. Retained nirmatrelvir efficacy against variant of concern Omicron BA.1 and BA.2 sublineages has been confirmed *in vitro* [3,4], but there are remaining issues to be explored in details, as demonstrated by the following two case reports.

Case 1 was a 63-year-old male patient vaccinated with 3 doses of BNT162b2 (last dose on October 2021) and exposed to a COVID-19 patient on 28 February 2022. On 5 March, he developed mild pharyngodynia and fever and tested positive for SARS-CoV-2 on nasopharyngeal swab (NPS) at both rapid antigen assay (cutoff/index = 121; COVID-19 Ag FIA-SD BIOSENSOR, Gyeonggi-do, Republic of Korea) and RT-PCR (cycle threshold (Ct) RdRp/N = 21; FTD SARS-CoV-2, Siemens Healthineers, Erlangen, Germany). Being at risk of progression because of age, previous acute myocardial infarction, systemic arterial hypertension, and first-grade obesity, he started nirmatrelvir/ritonavir treatment the same day, which was discontinued on 10 March. On 8 March, symptoms resolved and the antigen assay showed lower reactivity, and on 11 March both antigen and molecular assays were negative (RdRp/N Ct = 37). On 17 March, following sudden rhinorrhea, a repeat RT-PCR was again positive (RdRp/N Ct = 17). On 19 March, RT-PCR in peripheral blood was negative, but on 22 March, RT-PCR was still positive on NPS. On 25 March, the rapid antigen assay turned negative, as well as the RT-PCR on NPS on 29 March (after 24 days from the original onset of the symptoms). Whole-genome sequencing of SARS-CoV-2 at baseline and relapse showed full identity (Omicron BA.2 sublineage), excluding treatment-emergent mutations or reinfection from a different lineage. Serum anti-trimeric Spike IgG (CLIA, Liaison, DiaSorin, Saluggia, Italy) gradually increased from 2750 BAU/mL, after 17 days of infection, to 4690 BAU/mL, 23 days after the onset of the symptoms. Anti-N antibodies (Elecsys Anti-SARS-CoV-2, Roche, Basel, Switzerland) were negative 17 days after infection and became positive 25 days after the onset of the symptoms.

Case 2 was a 64-year-old subject previously vaccinated with 2 doses of ChAdOx1 and 1 dose of BNT162b2 (third dose on July 2021), exposed to a COVID-19 patient on 12 March 2022. On 15

* Corresponding author. D. Focosi, Pisa University Hospital, North-Western Tuscany Blood Bank via Paradisa 2, 56124 Pisa, Toscana, Italy.
*E-mail address:* daniele.focosi@gmail.com (D. Focosi).

https://doi.org/10.1016/j.cmi.2022.06.029

March, he developed headache, rhinorrhea, pharyngitis, low-grade fever, and tested positive at RT-PCR for two SARS-CoV-2 genes (Ct = 35, 36) out of three on the NPS. The following day, he was tested again with RT-PCR (Ct = 22). Being at risk of progression because of age and previous lung cancer, he started nirmatrelvir/ritonavir treatment on 17 March, which was discontinued on 21 March. On 22 March, symptoms resolved and the RT-PCR was negative. On 25 March, following heavy rhinorrhea with no fever, RT-PCR turned back positive (Ct = 22), which was confirmed the following day (Ct = 25) and on 30 March (Ct = 33), before returning negative on 1 April. On 30 March, anti-trimeric Spike IgG reached 1540 BAU/mL, and PRNT50 was 1:1280. Whole-genome sequencing of SARS-CoV-2 at baseline and relapse showed full identity (Omicron BA.2.9 sublineage), again excluding treatment-emergent mutations or reinfection from a different lineage.

Our patients were not screened for different respiratory pathogens during the onset of symptoms, but the persistence of SARS-CoV-2 positivity in RT-PCR suggests a causal association.

A recent preprint discussed from Boston reported a 71-year-old vaccinated and boosted male infected with BA.1 and treated with nirmatrelvir/ritonavir, who similarly relapsed 4 days after completion of the treatment; unfortunately, serology was not assessed at the time of relapse, but both viral load and symptoms remitted at the time anti-Spike IgG was positive [2]. Neither that case nor our two cases attempted viral culturing to prove the occurrence of infectious virions at the time of relapse. More recently Bocau et al. in France showed that 3 out of 7 patients having positive RT-PCR up to day 17 after initial diagnosis and after nirmatrelvir/ritonavir had culturable virus for up to 16 days [3].

We can read in the protocol of the RCT which led to drug approval that nasopharyngeal or nasal swabs were collected on day 1 (baseline) and days 3, 5, 10, and 14, but the peer-reviewed article only reported outcomes at day 5 [4]. A look back at the Center for Drug Evaluation and Research review shows that “Several subjects appeared to have a rebound in SARS-CoV-2 RNA levels around day 10 or day 14 [i.e., days 13 and 17 since onset of first symptoms], although this occurred among subjects with or without potential resistance-associated substitutions detected at day 1 or day 5”, but clinical symptoms were not disclosed at that time and the subset with available samples was very small [5].

Explanations proposed so far for this phenomenon include (more likely) too short schedule discontinued before a protective immune response (which is regularly mounted a few days after completion of drug treatment), insufficient dose in obese patients, pharmacokinetic interactions with concurrent medications lowering plasma levels of nirmatrelvir, or (less likely) failure of the drug to eradicate the virus from some sanctuary tissues. While efficacy at preventing hospitalization remains out of discussion [6], the most urgent questions, regardless of the mechanism, are establishing the frequency of these rebounds at the time of BA.2 in subjects that this time will be fully vaccinated and boosted, and whether they are infectious to contacts during the relapse. Clearly, such RCT in outpatients these days should be no longer run vs. placebo but rather head-to-head (a premiere for COVID-19 antivirals) vs. authorized treatments that have led to similar risk reductions in hospitalization and whose efficacy against BA.2 has been confirmed at least *in vitro* bebtelovimab.

## Transparency declaration

We declare we have no conflict of interest related to this manuscript.

## Author contributions

GA conceived the manuscript. DF wrote the first draft. OT, RB, SF, CA, FL and CMM took care of the patients. MT revised the final version.

## References

[1] Girardin F, Manuel O, Marzolini C, Buclin T. Evaluating the risk of drug-drug interactions with pharmacokinetic boosters: the case of ritonavir-enhanced nirmatrelvir to prevent severe COVID-19. Clin Microbiol Infect 2022 [Epub ahead of print].
[2] Charness M, Gupta K, Stack G, Strymish J, Adams E, Lindy D, et al. Rapid relapse of symptomatic SARS-CoV-2 infection following early suppression with nirmatrelvir/ritonavir. Research Square. 2022. https://doi.org/10.21203/rs.3.rs-1588371/v1.
[3] Boucau J, Uddin R, Marino C, Regan J, Flynn JP, Choudhary MC, et al. Characterization of virologic rebound following nirmatrelvir-ritonavir treatment for COVID-19. Clin Infect Dis 2022 [Epub ahead of print].
[4] 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;386:1397–408.
[5] Center for Drug Evaluation and Research (CDER). Review. Emergency use authorization (EUA) for Paxlovid (nirmatrelvir tables co-packaged with ritonavir tablets). Available at: https://www.fda.gov/media/155194/download. [Accessed 1 May 2022].
[6] 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;386:1475–7.