



Convalescent plasma in outpatients with COVID-19

In *The Lancet Respiratory Medicine*, Andrea Alemany and colleagues¹ report the results of CONV-ERT, a multicentre, double-blind, randomised controlled trial (RCT), administering locally sourced, high-titre COVID-19 convalescent plasma or saline to 376 outpatients in Spain. We commend the investigators for designing and implementing a rigorous outpatient trial during the difficulties of the pandemic. The study was among the few RCTs designed to deliver COVID-19 convalescent plasma to patients with early disease who were mostly seronegative, for whom antiviral therapies are most likely to succeed. Nevertheless, convalescent plasma was not shown to be efficacious.

Only four other studies of COVID-19 convalescent plasma in outpatients have been reported, with mixed results. The first was a double-blind RCT run in Argentina in a population of 160 outpatients aged 75 years or older at risk for disease progression, and COVID-19 convalescent plasma reduced disease progression (16% in those who received convalescent plasma vs 31% in those who received standard care).² The second RCT (CSSC-004 in the USA)³ is the only double-blind RCT in outpatients with COVID-19 that used non-convalescent plasma as the control. In that study, 1181 patients (regardless of risk factors for disease progression) were randomly assigned to receive either high-titre COVID-19 convalescent plasma or placebo control plasma, and COVID-19 convalescent plasma administration led to a reduction in hospitalisation within 28 days (2.9% vs 6.3%; $p=0.004$).³ The third RCT, run in the Netherlands (CoV-Early)⁴ was also double-blind, and included aggregate data together with CONV-ERT because both RCTs participated in the COMPILER project. In the CoV-Early study, 421 (out of the planned 690) outpatients aged 50 years or older at risk for progression were randomly assigned to receive COVID-19 convalescent plasma or fresh frozen plasma. No benefit was detected in the entire cohort, but the effect of convalescent plasma on hospital admission or death was largest in patients with 5 days or less of symptoms; however, this result was not statistically significant (odds ratio 0.658 [95% CI 0.394–1.085]).⁴ Another single-blind RCT in the USA (SIREN-C3PO)⁵ focused on outpatients aged 50 years or older with comorbidities who attended the emergency room. No benefit of convalescent plasma was reported by the authors; however, the outpatients

in this trial possibly represent a subset seeking medical attention because of disease severity, and if one excludes the patients admitted on the index visit from the analysis, there is evidence for convalescent plasma efficacy in reducing hospitalisation from 19.7% to 12.5%, a risk difference of 7.2% (95% CI 0.8–13.0%; $p=0.03$).

So although the results of CONV-ERT raise questions as to the role of convalescent plasma in outpatients with COVID-19, given that some previous trials of COVID-19 convalescent plasma have shown efficacy in this patient group it is worth considering why CONV-ERT did not show any efficacy. One major difference of CONV-ERT compared with the other four outpatient RCTs cited above is the use of pathogen inactivation technologies. The effect of methylene blue on antibody neutralisation in vitro is probably minor, as some studies have shown some reduction in titres and others have not. Methylene blue reduces pathogens by producing oxygen-derived free radicals,⁶ but these can also react with proteins and methylene blue is known to reduce coagulation factor activity in plasma. The region of the antibody that binds antigen is relatively small and possibly less vulnerable to direct oxidative damage. However, other antiviral activities of antibodies are dependent on the Fc region, which is large and requires intact glycosylation for function. Sugars are potentially vulnerable to oxygen-derived free radicals and to reactivity with methylene blue. During the era of serum therapy, methylene blue reportedly deactivated antibody efficacy.⁶ Viral neutralisation in vivo would be followed by formation of antigen–antibody complexes that are then cleared by immune cells using Fc-dependent phagocytosis. Another important antiviral function dependent on Fc integrity is antibody dependent cellular cytotoxicity, which has been strongly implicated in COVID-19 convalescent plasma efficacy.⁷ Because Fc function was not studied in CONV-ERT, whether methylene blue damaged the immunoglobulins in the COVID-19 convalescent plasma used is unknown, but if this happened, it could provide an explanation for the negative results. Of interest, regulatory authorities worldwide are no longer recommending pathogen inactivation for COVID-19 convalescent plasma because there is no evidence for transfusion-transmitted SARS-CoV-2 infection.



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Another potential reason for the negative results of CONV-ERT is the ambitious predefined endpoint, a 50% reduction in hospitalisation, which led to sample size underestimation. Further reducing the sample size, CONV-ERT was terminated at the end of May, 2021, with enrolment of 76% of the target population because more than 85% of the population aged 50 years or older was fully vaccinated in Spain.

The difference in disease progression between the Argentinean, USA, and Dutch RCTs suggests that focusing on outpatients at risk for disease progression can maximise the effectiveness of COVID-19 convalescent plasma, as implemented for monoclonal antibodies. The SARS-CoV-2 omicron (B.1.1.529) variant of concern, which is spreading rapidly around the globe, is unfortunately resistant to most monoclonal antibodies available, and the supplies of recently approved small chemical antivirals are inadequate and not affordable to low-and-middle-income economies. Based on a systematic review of COVID-19 convalescent plasma RCTs,⁸ on Dec 27, 2021, the US Food and Drug Administration expanded its emergency use authorisation for COVID-19 convalescent plasma for outpatients with immune deficiency. Although COVID-19 convalescent plasma collected during former COVID-19 waves is unlikely to be effective against omicron,⁹ collection of convalescent plasma from vaccinated individuals currently represents a promising alternative with a very large pool of regular donors and very high and broad-spectrum neutralising antibody concentrations.¹⁰

CONV-ERT teaches us that even well designed RCTs that test COVID-19 convalescent plasma in conditions where antibody therapies are expected to be effective can have negative results. This implies that there are variables affecting COVID-19 convalescent plasma that we do not understand and the role of convalescent plasma in outpatients with COVID-19 remains unclear. Given that COVID-19 is likely to become endemic and

that convalescent plasma is a relatively inexpensive therapy available even in resource-poor countries, there is a need to understand its potential and limitations. In this regard, additional trials of COVID-19 convalescent plasma that explore the relationship between dose, timing, and clinical status of recipients are needed and welcomed.

DF was an investigator in the TSUNAMI randomised controlled trial of COVID-19 convalescent plasma. AC reports being part of the scientific advisory board of SabTherapeutics, has received personal fees from Ortho Diagnostics, outside of the submitted work, and is an investigator in the CSSC-004 randomised controlled trial.

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- 1 Alemany A, Millat-Martinez P, Corbacho-Monné M, et al. High-titre methylene blue-treated convalescent plasma as an early treatment for outpatients with COVID-19: a randomised, placebo-controlled trial. *Lancet Respir Med* 2022; published online Feb 9. [https://doi.org/10.1016/S2213-2600\(22\)00050-9](https://doi.org/10.1016/S2213-2600(22)00050-9).
- 2 Libster R, Pérez Marc G, Wappner D, et al. Early high-titer plasma therapy to prevent severe COVID-19 in older adults. *N Engl J Med* 2021; **384**: 610–18.
- 3 Sullivan DJ, Gebo KA, Shoham S, et al. Randomized controlled trial of early outpatient COVID-19 treatment with high-titer convalescent plasma. *medRxiv* 2021; published online Dec 21. <https://doi.org/10.1101/2021.12.10.21267485> (preprint).
- 4 Millat-Martinez P, Gharbharan A, Alemany A, et al. Convalescent plasma for outpatients with early COVID-19. *medRxiv* 2021; published online Dec 2. <https://doi.org/10.1101/2021.11.30.21266810> (preprint).
- 5 Korley FK, Durkalski-Mauldin V, Yeatts SD, et al. Early convalescent plasma for high-risk outpatients with COVID-19. *N Engl J Med* 2021; **385**: 1951–60.
- 6 Ross V. Photodynamic action of methylene blue on antipneumococcal serum. *J Immunol* 1938; **35**: 351–69.
- 7 Bégin P, Callum J, Jamula E, et al. Convalescent plasma for hospitalized patients with COVID-19: an open-label, randomized controlled trial. *Nat Med* 2021; **27**: 2012–24.
- 8 Focosi D, Franchini M, Pirofski L-A, et al. COVID-19 convalescent plasma and clinical trials: understanding conflicting outcomes. *medRxiv* 2021; published online Sept 17. <https://doi.org/10.1101/2021.09.07.21263194>.
- 9 Focosi D, Franchini M, Joyner MJ, Casadevall A. Are convalescent plasma stocks collected during former COVID-19 waves still effective against current SARS-CoV-2 variants? *Vox Sang* 2022; published online Jan 12. <https://doi.org/10.1111/vox.13239>.
- 10 Focosi D, Franchini M, Joyner MJ, Casadevall A. Comparative analysis of antibody responses from COVID-19 convalescents receiving various vaccines reveals consistent high neutralizing activity for SARS-CoV-2 variant of concern omicron. *medRxiv* 2021; published online Dec 26. <https://doi.org/10.1101/2021.12.24.21268317>.