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Editorial

Impact of pathogen-reduction technologies on COVID-19 convalescent plasma potency

**A R T I C L E  I N F O**

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- Pathogen reduction technologies
- COVID-19 convalescent plasma
- Neutralizing antibodies
- Efficacy
- IgG subclasses

**A B S T R A C T**

Pathogen reduction technologies (PRT) have been recommended by many regulatory authorities to minimize the residual risk of transfusion-transmitted infections associated with COVID-19 convalescent plasma. While its impact on safety and its cost-effectiveness are nowadays well proven, there is theoretical concern that PRT could impact efficacy of convalescent plasma by altering concentration and/or function of the neutralizing antibodies (nAb). We review here the evidence supporting a lack of significant detrimental effect from PRTs on nAbs.

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Several pathogen reduction technologies (PRT) have been approved and are currently marketed. In recent years, photo-inactivation in the presence of a photosensitizer has become the standard for single donation inactivation: approved technologies include combination of methylene blue + visible light [1] (Theraflex®, amotosalen (S-59) + ultraviolet A [2] (Intercept®), and riboflavin + ultraviolet B [3] (Mirasol®).

Although neither the US Food and Drug Agency (FDA) [4] nor the European Center for Disease Control (ECDC) [5] are mandating PRT for COVID19 convalescent plasma, the European Commission indicates that “The processing that is routinely applied in the country or blood establishment for the preparation of plasma for transfusion should be applied. Thus, PRTon should be applied (to convalescent plasma) if it has been the normal practice in the blood establishment and should not be introduced for this particular blood component if not normally applied for plasma for transfusion” [6]. The Working Party on Global Blood Safety of the International Society for Blood Transfusion (ISBT) initially recommended it as “highly desirable” [7], but this position was later tempered as “the benefit of PRT in the context of SARS-CoV-2 is unclear”, and “most countries have elected not to implement PRT specifically for convalescent plasma” [8].

Many national authorities consider that under emergency settings the donor screening and conventional viral nucleic acid testing (NAT) (i.e. HIV, HCV and HBV NAT) would not be enough to ensure convalescent plasma safety [9]: most convalescent plasma donors have no donation history, and their donations should be considered at higher risk. Under this scenario, the introduction of additional virological testing (e.g. HAV, HEV, and B19) and PRT would approximately double the final cost of the therapeutic convalescent plasma dose. Since these additional nonenveloped viruses are nevertheless inactivated by PRT, PRT has finally remained the sufficient precautionary measure [10].

Viral SARS-CoV-2 RNA is detectable at low viral loads in a minority of serum samples collected from patient with acute infection, but it is not associated with infectious virions [11]. Some PRT have nevertheless been shown to inactivate SARS-CoV-2 RNA in plasma, with very similar mean log reduction of > 3.32 for Intercept® [12] and up to ≥ 4.79 for Mirasol® [13,14]. No SARS-CoV-2 inactivation data have been published for methylene blue technology (Theraflex® Methylene Blue, MacoPharma) as of now. The demonstration of the maximum inactivation capacities is limited by the maximum viral titers of the clinical isolates available: so an even higher overall inactivation capacity is to be expected (the Intercept® inactivation capacity for SARS-CoV was demonstrated to be as high as ≥ 5.5 log [2]).

Given the increased risk profile of convalescent plasma donors and the highly vulnerable condition of recipients suffering from COVID19, the inactivation capacity a broad range of pathogens should be taken into account when aiming at making the convalescent plasma as safe as possible. Also in this regard large differences are apparent for the different technologies with Intercept® having the largest dataset [15].

Last but not least, the clinical experience with PRT-treated plasma should be taken into account. While Intercept® treated plasma has been shown to be safe and efficacious to treat acquired or inherited coagulopathies and in therapeutic plasma exchange [16–21], and also for Theraflex® Methylene Blue a series of studies have been performed, we did not find data from randomized, controlled trials for Mirasol® treated plasma.

The potency of convalescent plasma can be affected by several variables, whose impact need to be formally studied. For example the type of collection (apheresis vs. recovered plasma) or storage temperature [22] have been shown not to affect the antibody content. PRT treatment is one of the main variables which needs to be assessed when manufacturing convalescent plasma.
On December 7th, we searched both peer-reviewed (PubMed and Google Scholar) and not peer-reviewed (medRxiv and bioRxiv) online repositories using the query: “[pathogen inactivation” OR “pathogen reduction technologies”) AND (“immunoglobulins” or “neutralizing antibodies”). Table 1 summarizes the evidence of moderate to no detrimental impact of PRT treatment on overall immunoglobulin content or anti-SARS-CoV-2–specific immunoglobulins (measured with either high-throughput serology or viral neutralization tests) in plasma treated with PRT. While high-throughput serology detects drops in IgG1 and IgG3 sub-classes, it does not investigate a theoretical detrimental impact on IgA and IgM, which have a fundamental role in SARS-CoV-2 neutralization [23,24]. Viral neutralization tests instead account for all immunoglobulin classes.

While most studies found no significant decline [25–27], in the largest head-to-head comparison study to date, PRT with methylene blue or with amotosalen provided the greater likelihood of preserving nAb in convalescent plasma compared to riboflavin [28]. Nevertheless, this study suffers from major methodological limitations. It is worth noting though that studies showing decrease in levels of plasma proteins, including antibodies as well as complement after PRT treatment, clearly demonstrated that remaining levels were maintained within reference levels [29,30].

In addition to the specific antibodies, the overall retention of coagulation factors and anti-thrombotic factors is of highest interest also for the treatment of COVID19 patients. Convalescent plasma can include different soluble factors expected to be of special benefit such as antithrombin III [31], decay receptors (e.g. ACE2* exosomes [32]), anti-inflammatory cytokines, complement factors, or, in partially ABO-matched units, anti-A isoagglutinins (expected to inhibit SARS-CoV-2 entry [11]), for which effect from PRT remains to be investigated [33].

PRT have been successfully used to prepare convalescent plasma which has been transfused in many clinical trials to date without any detrimental effect (namely Intercept®[27,34,35], and Mirasol® ([36], NCT04385186)). One advantage of Mirasol® is the lower minimum required volume of convalescent plasma to be treated, namely 150 mL (vs. 385 mL in Intercept®), which makes PRT of recovered plasma or thawed apheresis aliquots possible without pooling. On the other hand, Intercept allows generation of up to 3 aliquots in the same inactivation kit, thus saving time and resources. Additionally, in-hospital-made methylene blue and light treatment has been used in a Chinese trial [36].

In conclusion, while the choice to mandate or not PRT for convalescent plasma in individual countries largely depend on pharmacoeconomics, there are enough evidences to conclude that the approach preserves potency. Further studies are needed to assess whether PRT improves the safety of convalescent plasma in regard to reduced transfusion-transmitted infections.

Disclosure of interest

The authors declare that they have no competing interest.

References

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**Table 1**

Impact of different PRT on overall immunoglobulin levels and/or anti-SARS-CoV-2 neutralizing antibodies.

| PRT brand | PRT effect on IgG or nAb levels | # of treated plasma units | Ref |
|-----------|----------------------------------|---------------------------|-----|
| Intercept® | No significative difference in EBOV nAb titer | 10 | [37] |
| | – 2% to –4% EBOV IgG | 1 | [38] |
| | – 3.8% anti-SARS-CoV-2 N-protein IgG | 5 | [25] |
| | No difference in SARS-CoV-2 nAb titer and N-protein antibodies | 48 | [39] |
| | No significative difference in SARS-CoV-2 nAb titer | 110 | [23] |
| | Among units with the initial SARS-CoV-2 nAb titre ≥ 80: 60% (47%–73%, CI) were unchanged | 30 | [27] |
| | 40% decreased by one dilution | 140 | [28] |

Mirasol®

| – 13% to –22% total IgG after 69 week storage at –30 °C | 6 | [40] |
| – 17.1% total IgG and –23.6% IgG1 (significant) | 6 | [29] |
| – 16.6% total IgG and –32.3% IgG1 (significant), but no significant difference in Tetanus, Diphtheria and Pneumococcal protective antibody titers | 6 | [30] |
| – 12.7% anti-SARS-CoV-2 N-protein IgG | 20 | [31] |
| Among units with the initial SARS-CoV-2 nAb titre ≥ 80: 43% (26%–61%, CI) were unchanged | 140 | [28] |
| 50% had a one-dilution decrease | 22 | [31] |
| 7% had a two-dilution decrease | 1401 | [28] |

Mirasol®

| – 4.8% anti-SARS-CoV-2 N-protein IgG | 22 | [31] |
| Among units with the initial SARS-CoV-2 nAb titre ≥ 80: 81% (71%–91%, CI) of units remained unchanged | 1401 | [28] |
| 19% decreased by one dilution |
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D. Focosi
North-Western Tuscany Blood Bank, Pisa University Hospital, via Paradisa 2, 56124 Pisa, Italy

M. Franchini
Department of Hematology and Transfusion Medicine, Carlo Poma Hospital, Mantua, Italy

* Corresponding author.
E-mail address: daniele.focosi@gmail.com
(D. Focosi)

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