Title: Convalescent Plasma Therapy on Patients with Severe or Life-Threatening COVID-19: A Metadata Analysis

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Conflict of Interest Disclosures: No disclosures and conflict of interest were reported.

Funding/Support Information: This work was supported by the Brazilian Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) (INCT-TM/CNPq/FAPESP-465458/2014-9), and PRONEX/FAPERGS (16/2551-0000499-4) that made this research feasible. F.K. received a fellowship from MCT/CNPq (306439/2014-0). We would like to thank Dr. Marcia Triunfol at Publicase® for her assistance in preparing this manuscript.

Authors' Contributions: Prof. Fábio Klamt had full access to all of the data in this study and takes responsibility for the integrity of the data and the accuracy of the data analysis. Francisco Diego Rabelo-da-Ponte: figures, study design, data collection, data analysis, data interpretation, writing. Daiane Silvello: literature search, study design, data collection, data analysis, data interpretation, writing. Juliana Nichterwitz Scherer: study design, data collection. Alejandro Raul Ayala: data analysis, data interpretation, writing. Fabio Klamt: literature search, figures, study design, data collection, data analysis, data interpretation, writing.

Dear Editor, Current therapeutic options to mitigate severe COVID-19 cases remain limited. Prior experience with convalescent plasma (CP) to treat SARS, H1N1, and Ebola patients suggested that passive immunization by plasma transfusion suppresses viremia and improves clinical outcomes, reducing the number of deaths and length of stay in the intensive care unit (ICU) with minimal side effects. These findings are not universal, as Zeng et al. described discouraging effects of convalescent plasma therapy on survival in COVID-19 patients [1], and a recent COVID-19 study stratified by disease severity (n = 103 participants) did not show significant improvement following CP administration, when compared to standard care alone [2]. However, a sub-analysis suggested a potential CP therapeutic benefit in those with advanced disease, including patients with COVID-19 suffering from severe disease (respiratory distress and/or hypoxemia) but not in patients with life-threatening disease (shock, organ failure, or requiring mechanical ventilation) [2].

In order to add more information whether CP administration is effective as a treatment over the continuum of care of COVID-19 patients, we performed a metadata analysis, including random-effects meta-analysis and meta-regression, based on available data [1-9].

The following measures, before and after CP transfusions, were checked: *i*) viral load expressed as rt-PCR cycle threshold (Ct) values (where Ct values ≥ 40 are considered SARS-CoV-2 negatives); *ii*) C-reactive protein levels as a surrogate marker of inflammation resolution; and *iii*) clinical disease severity (WHO 6-point clinical scale) [8] at baseline (date of transfusion) and post-treatment (19.05 ± 10.43 days, mean ± SD) summarized as the Risk Ration (RR) and the Ratio of Mean (ROM) (Figure 1).

Convalescent plasma reduced viral loads (RR 0.13 [95% CI, 0.09 to 0.18], *P* <0.001) (n = 75) as well as C-reactive protein levels (ROM 0.11 [95% CI, 0.01 to 0.86], *P* <0.05) (n = 42), and improved the clinical status of COVID-19 patients, when compared to baseline (ROM

0.53 [95% CI, 0.36 to 0.79], *P* <0.01) (n = 149) (Figure 1). Additionally, we observed that the effect of CP on clinical improvement was not age-dependent (0.02 [95% CI, -0.12 to 0.09], *P* = 0.62), nor did it associate with other concurrently used agents such as antivirals (0.2 [95% CI, -0.82 to 0.93], *P* = 0.8), antibiotics (0.28 [95% CI, -1.31 to 1.17], *P* = 0.82), and hydroxychloroquine (0.02 [95% CI, -0.12 to 0.09], *P* = 0.62). Treatment with CP was associated with a reduction in C-reactive protein levels regardless of the patient's age (0.17 [95% CI, -2.25 to 2.06], *P* = 0.68) and the use of antivirals (0.19 [95% CI, -2.45 to 2.481], *P* = 0.94), antibiotics (0.18 [95% CI, -2.06 to 2.72], *P* = 0.32), and hydroxychloroquine (0.05 [95% CI, -0.70 to 0.79], *P* = 0.56).

Independently, Joyner et al. [10] analyzed key metrics after transfusion in 5,000 ABO-compatible hospitalized adult CP recipients with severe or life-threatening COVID-19 and reported an incidence of adverse events <1%, suggesting that passive immunization is relatively safe.

Given the paucity of donors and our little understanding of COVID-19 infection, the use of CP therapy is currently restricted to severe and life-threatening disease. However, when considering its safety profile and the limited options to treat COVID-19 infection, some have advocated the use of CP beyond the limited scope of life-threatening disease (*i.e.,* critically ill patients). In fact, some studies suggested that CP therapy should be administered earlier in the course of disease [1,2,8,9] since viremia peaks in the first week of infection and a primary immune response develops by days 10–14, allowing for viral clearance.

It is true that not all patients showing the first symptoms of COVID-19 will develop severe disease for which CP transfusion might be indicated; however, biomarkers such as C-reactive protein, interleukin 6 (IL-6), d-dimers, and lactate dehydrogenase, as well as inherent risk factors such as advanced age and multiple comorbidities, may aid in the decision-making process that guides future CP therapy. Large-scale controlled studies that

include a myriad of COVID-19 patients along the continuum of disease severity will help to understand if CP administration earlier in the course of disease can prevent clinical deterioration and improve survival rates. Surrogate inflammatory and cardiovascular markers that correlate with meaningful clinical outcomes such as ICU admission and mortality rates may also help in deciding the most appropriate timing of CP administration.

Conflict of Interest Disclosures: No disclosures and conflict of interest were reported.

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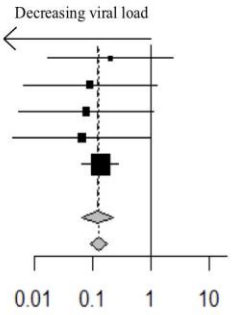
Figure Legend:

Figure 1. Summary of pooled results of convalescent plasma use in COVID-19 patients. The inverse variance weighted method was used to combine summary measures using random-effects models to minimize the effects of heterogeneity between studies [1–9] and sensitivity analyses using fixed-effects models. Negative viral load rates were expressed as the Risk Ratio (RR) and clinical disease severity and C-reactive protein level were expressed as the ratio of mean (ROM). Risk ratios and ratio of mean less than one (1) indicate a positive effect of the intervention.

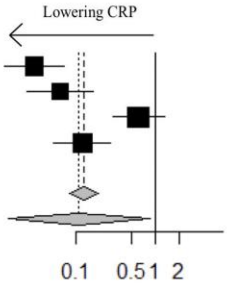
Meta-regression analyses were performed to investigate the influence of other concurrently administered agents and patient age on the outcomes examined. All tests were two-tailed; $P \leq 0.05$ was considered statistically significant. All analyses were performed using the "meta" package in *R* and RStudio software.

Figure 1

Negative viral load rate	No. of patients	No./Total (%) Pre-CP	Post-CP	Risk Ratio (95% CI)	Study weight (%)
Ahn, 2020	2	0/2 (0)	2/2 (100)	0.2 (0.02 to 2.39)	6.4
Shen, 2020	5	0/5 (0)	5/5 (100)	0.09 (0.01 to 1.28)	5.6
Zeng, 2020	6	0/6 (0)	6/6 (100)	0.08 (0.01 to 1.10)	5.5
Duan, 2020	10	0/10 (0)	10/10 (100)	0.07 (0.00 to 1.02)	5.3
Li, 2020	52	0/52 (0)	41/47 (87.2)	0.14 (0.07 to 0.28)	77.1
Fixed effects				0.12 (0.07 to 0.23)	
Random effects				0.13 (0.09 to 0.18)	
$I^2 = 0.0\%$	75				



C-reactive protein level (mg/L)		Mean (SD) Pre-CP	Post-CP	Ratio of mean (95% CI)	Study weight (%)
Ahn, 2020	2	258.3 (121.19)	7.85 (3.04)	0.03 (0.01 to 0.07)	25.0
Shen, 2020	5	160.6 (63.3)	10.32 (10.44)	0.06 (0.02 to 0.17)	24.3
Duan, 2020	10	45.0 (43.94)	27.70 (19.50)	0.62 (0.29 to 1.30)	25.6
Salazar, 2020	25	176.10 (150.4)	21.32 (40.79)	0.12 (0.05 to 0.28)	25.1
Fixed effects				0.13 (0.8 to 0.19)	
Random effects				0.11 (0.01 to 0.86)	
$I^2 = 89\%$	42				



Clinical disease severity (6-point scale)		Mean (SD) Pre-CP	Post-CP	Ratio of mean (95% CI)	Study weight (%)
Ahn, 2020	2	4.0 (0)	1.5 (0.7)	0.38	0
Zhang, 2020	4	4.75 (0.5)	1.25 (0.43)	0.26 (0.18 to 0.37)	13
Shen, 2020	5	4.8 (0.4)	2.6 (2.19)	0.54 (0.26 to 1.14)	8.5
Zeng, 2020	6	4.66 (0.51)	5.11 (2.02)	1.1 (0.79 to 1.52)	13.3
Ye, 2020	6	3.16 (1.47)	1.16 (0.4)	0.37 (0.23 to 0.58)	11.7
Duan, 2020	10	3.80 (1.22)	3.3 (1.76)	0.87 (0.59 to 1.28)	12.6
Salazar, 2020	25	4.12 (0.97)	1.68 (1.49)	0.41 (0.28 to 0.58)	12.9
Liu, 2020	39	3.89 (0.55)	1.79 (1.67)	0.46 (0.34 to 0.62)	13.6
Li, 2020	52	3.92 (0.86)	2.48 (1.75)	0.63 (0.52 to 0.77)	14.5
Fixed effects				0.56 (0.49 to 0.62)	
Random effects				0.53 (0.36 to 0.79)	
$I^2 = 85\%$	149