A Systematic Review and Meta-Analysis of an Emerging Therapy against COVID-19: Is Convalescent Plasma a Hidden Gem Not Yet Optimized?

Azza Sarfraz[1,2], Zouina Sarfraz[1,3*], Aminah Abdul Razzack[1,4],
Donald Hathaway III[1], Sindhu Thevuthasan[1,5], Sarabjot Singh-Makkar[1],
Trissa Paul[1], Gaurav Patel[1,6], Muhammad Khawar Sana[1,7], Muzna Sarfraz[1,7],
Franz Richter[1] and Marcos A. Sanchez Gonzalez[1]

[1]*Division of Research & Academic Affairs, Larkin Health System, South Miami, FL, USA.*
[2]*Department of Pediatrics & Child Health, Aga Khan University, Karachi, Pakistan.*
[3]*Department of Research & Publication, Fatima Jinnah Medical University, Lahore, Pakistan.*
[4]*Dr NTR University of Health Sciences, Andhra Pradesh, India.*
[5]*Medical University of the Americas (MUA), Saint Kitts and Nevis, Caribbean.*
[6]*Smt NHL Municipal Medical College, Ahmedabad, Gujarat, India.*
[7]*King Edward Medical University, Lahore, Pakistan.*

Authors' contributions

This work was carried out in collaboration among all authors. Authors AS and ZS designed and directed the project. Authors AS, ZS, AAR, DH, ST, SSM, TP, MKS, MS, FR, MSG contributed to the design and implementation of the research, to the analysis of the results and to the writing of the manuscript. Authors AS and ZS are co-guarantors of the work. All authors read and approved the final manuscript.

Systematic Review Article

Corresponding author: Email: zouinasarfraz@gmail.com;

ABSTRACT

Background: An unprecedented global effort in identifying potentially viable and emerging drugs for effective treatment of the novel coronavirus disease (2019) is being made. Of the most promising candidate therapies, convalescent plasma (CP), albeit controversial, is approved for emergency use authorization (EUA) by the U.S. Food and Drug Administration (FDA). The concept rests on passive immunity, achieved by administering plasma with high titers of neutralizing antibodies to reduce severity of SARS-CoV-2 infection and mortality. The aim of this paper is to assess the clinical improvement, patients' discharge status and all-cause mortality in convalescent plasma versus standard of care COVID-19 patient groups.

Methods: Using PRISMA guidelines, a review was conducted from January, 2020, until October, 2020 employing keywords including "convalescent plasma", "clinical improvement, "mortality", "adverse events", "viral load", "dosing", and survival." Dichotomous data for all-cause mortality, patients' discharge status, and clinical improvement at day 14 of treatment were meta-analyzed applying the Mantel-Haenszel (M-H) random effects model using Review Manager 5.4.

Results: A total of 627 (23.9%) patients in the CP group and 1997 (76.1%) patients in the control group were pooled. The studies were conducted in the United States, China, Netherlands, and Iran. The CP group had a lower association to all-cause mortality as compared to the control group [OR: 0.69; CI: 0.50 to 0.96; P=0.03]. Patients who received CP had higher probability of discharge during the study course [OR: 1.87; CI: 1.1 to 3.18; P=0.02]. Bias was expected in the analysis due to the stratified of study designs included.

Conclusion: Convalescent plasma therapy may be an effective and vital tool with promising historical, current, and expected clinical trial evidence of metrics such as increased safety and reduction of all-cause mortality.

ABBREVIATIONS

ACE-2 RBD: Angiotensin-converting enzyme-2 receptor-binding domain; ACE-2: Angiotensin-converting enzyme 2; CFR: Case-Fatality Rate; CP: Convalescent Plasma; CP: Convalescent Plasma; EBDs: Receptor binding domains; ELISA: Enzyme-linked immunosorbent assay; EUA: Emergency Use Authorization; FDA: Food and Drug Administration; nAbs: Neutralizing antibodies; PRNT: Plaque reduction neutralization test; SAEs: Serious adverse events

1. INTRODUCTION

After being declared a global pandemic on March 11, 2020, the worldwide number of confirmed cases of coronavirus disease 2019 (COVID-19) has reached 26,415,380 including 870,286 deaths, reported by the World Health Organization (WHO) as of 5th September 2020 [1]. The global case-fatality rate (CFR) of COVID-19 is estimated to be 3.3%, slightly higher than that of the United States (US) at 3% on September 5, 2020. Global estimates of age structures across North America and Europe highlight the increased vulnerability of these populations to COVID-19-related deaths. Similarly, in-house transmission due to the household residential patterns in Africa and Asia also predisposes to higher transmission rates. Compounded impacts of both co-residential patterns and age structure in Southern Europe suggest the highest vulnerability across this region [2]. Consequently, treatments and vaccines of proven efficacy are crucial against COVID-19 and are being investigated all over the world in an accelerated manner to combat the advance of the pandemic. On August 23, 2020, the FDA endorsed convalescent plasma as an investigational therapy with potential benefits outweighing the risks. As part of the Expanded Access Program in collaboration with HHS' Biomedical Advanced Research and Development Authority (BARDA) and Mayo Clinic, the FDA has facilitated the delivery of over 70,000 infusions of CP across the United States [3].

Convalescent Plasma (CP), an investigational therapy with the potential to reduce the severity and duration of COVID-19 illness, is classic adaptive immunotherapy that has been used in various epidemics for over one century. In the last two decades, CP therapy has been used in severe acute respiratory disease coronavirus

(SARS-CoV), Middle East respiratory syndrome coronavirus (MERS-CoV), and the 2009 H1N1 pandemic with satisfactory tolerability and efficacy. The lack of efficacy in the Ebola virus disease is likely due to the lack of data of neutralizing antibody titration, suggesting non-optimal dosing of CP therapy [4]. Following the recovery of COVID-19 patients, donors are eligible after 30 days of recovery, on average, expected to contain the highest levels of polyclonal antibodies, whereas donors have typically been selected based on their neutralizing antibody titers in non-emergency settings. Neutralizing monoclonal antibodies, also known as hyperimmune plasma, target the full-length protein along with the angiotensin-converting enzyme-2 receptor-binding domain (ACE-2 RBD) of the S protein and thereby inhibit viral amplification, elaborated in Figure 1. [4] The plaque reduction neutralization test (PRNT),

required to quantify the neutralizing antibodies, is not feasible in resource-limited and time-sensitive circumstances thereby resulting in the implementation of the enzyme-linked immunosorbent assay (ELISA) as a proxy which targets the recombinant receptor binding domains (RBDs) of the viral anti receptor.

The pandemic caused by the SARS-CoV-2 coronavirus is the primary candidate for both traditional prevention using vaccines and passive immunity approaches using convalescent plasma therapy. Given the logistical feasibility and evidence from previous infectious outbreaks, recovered COVID-19 patients with high neutralizing antibody titers serve as a viable source of CP. We conducted a systematic review and meta-analysis to synthesize the available evidence concerning the effectiveness of CP administration in COVID-19 patients.
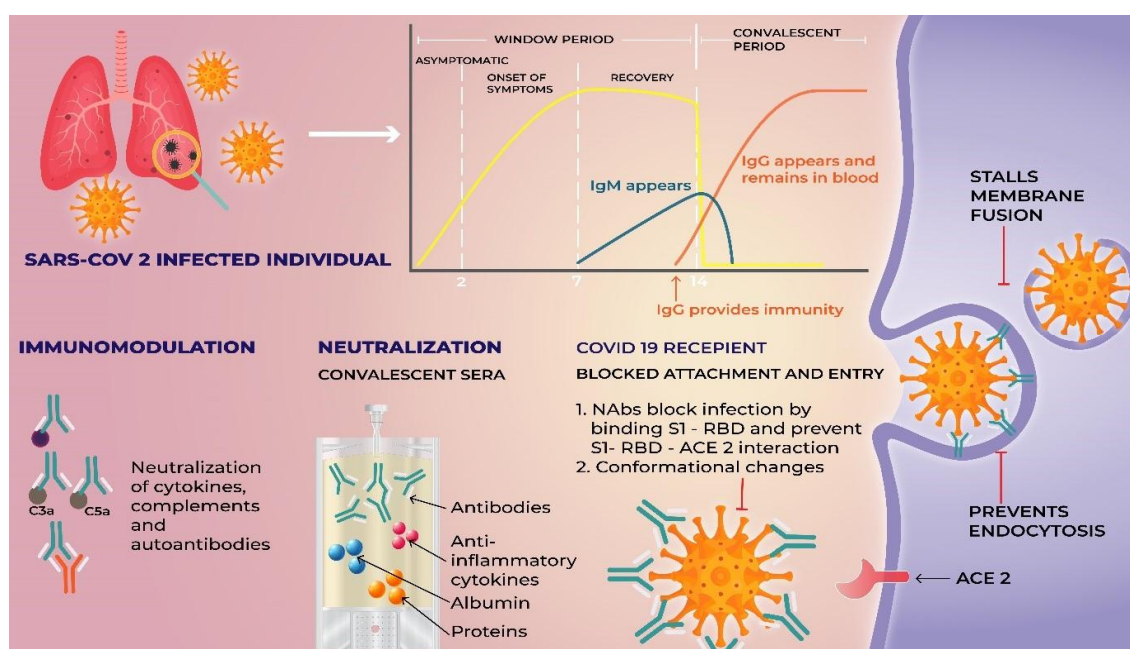
Figure 1. Effects of convalescent plasma on SARS-CoV-2

A patient infected with SARS-CoV-2 develops IgM antibodies by day 7 which lasts for a week. IgG antibody production begins at day 14 which lasts for longer time. For convalescent plasma therapy, plasma rich in IgG antibodies is collected between day 14 and day 21. This is when there is significant rise in IgG antibodies. The plasma also contains many anti-inflammatory cytokines and autoantibodies which block complement activation, and control cytokine storm further enhancing its immunomodulatory effects. Neutralizing antibodies (NAbs) provide passive immunity and are crucial for viral clearance through neutralization The NAbs compete with RBD of S1 spike protein for binding to ACE-2 and prevent viral entry and attachment. This can be done by (i) either direct neutralization or (ii) structural conformational changes. NAbs can also hinder the internalization of virion into endosome and stall membrane fusion and thereby release of genetic material and further viral amplification

The SARS-CoV-2 virion binds the angiotensin-converting enzyme 2 (ACE-2) receptor through the receptor-binding domain (RBD) present in the S1 subunit of S spike protein. Neutralizing antibodies (nAbs) block the virus-host attachment against either human cell ACE2 or the viral RBD present in the spike protein. This dampens viral internalization leading to degradation of the virus and further viral clearance in the host cell. Effects of convalescent plasma on SARS-CoV-2 are illustrated in Figure 1. The aim of this paper is to meta-analyze the clinical improvement, patients' discharge status and all-cause mortality in convalescent plasma versus standard of care COVID-19 patient groups.

2. METHODS

The rationale for this paper is to provide an information source for healthcare providers, researchers, and policy makers by integrating existing information pertaining to convalescent plasma therapy for COVID-19, present critical evidence from previous infectious outbreaks, and discuss enrollment and implementation in clinical trials. Using explicit methods in our paper, we limit biases, and intend to improve reliability and accuracy of literature pertaining to convalescent plasma during the COVID-19 pandemic.

2.1 Search Strategy and Selection Criteria

The systematic review and meta-analysis adhere to the PRISMA guidelines. Three online bibliographic databases in addition to grey literature sources were searched from January 1, 2020, to October 7, 2020: PubMed, CINAHL Plus, COCHRANE Central, and medRxiv. We identified an empirical approach to derive the search strategy for identifying clinical studies of convalescent plasma administered to interventional and control groups (three on COVID-19 and three on previous coronaviruses). The strategy was developed in PubMed using the identified keywords and the article identification numbers. From the six studies located during the test run the following key terms were identified, *convalescent plasma, clinical improvement, mortality, adverse events, viral load, dosing, and survival*. We manually examined all preliminary search results and built our strategy in an attempt to identify all relevant studies in the bibliographic databases and grey literature. The method helped in achieving maximum sensitivity for our potential search results. The key search terms of interest following the initial test sets were a combination of the following: *coronavirus, convalescent plasma, and clinical trial* (Figure 2).

Eligible studies were any clinical studies, involving treatment with convalescent plasma, or placebo comparison studies involving other COVID-19 therapies. Non-control group studies were excluded to ensure comparability of the effect, and three studies were excluded on this basis.

2.2 Quality Assessment

The Cochrane Risk of Bias tool for randomized controlled trials (RCTs), the Risk of Bias in Non-randomized Studies of Interventions-I (ROBINS-I) tool for controlled non-randomized studies of interventions (NRSIs), and the Newcastle Ottawa Scale (NOS) was used for cohort studies. The appraisal was conducted independently by two authors (AS and ZS), and the consensus was achieved through discussion. The study was not registered due to the time-sensitive nature of the topic.

2.3 Data Analysis

Two authors removed the duplicates (AS and ZS), early-to-mid career researchers trained in this method, screened the titles and abstracts independently and then extracted the full-text articles based on relevance. All discrepancies were resolved through discussion. A third author verified the eligibility of included studies (MSG). Two authors (AS and ZS) extracted the data using a custom spreadsheet to record the study design, registration number, country, setting, characteristics of groups, age, and gender of participants, CP administration day, clinical improvement, discharged patients, severe adverse events, all-cause mortality, length of stay, CP dosage, and concomitant therapies (Table 1). Quantitative data were analyzed using the Cochran Mantel Haenszel random-effects model on Review Manager (RevMan) 5.4. The effect size was computed via Odds Ratio (OR) statistics whereas the quantification of dispersion of the effect sizes was measured using the I^2 index. The analysis included three a priori outcomes: all-cause mortality (the number of patients that died by throughout the study), clinical improvements at day 14 (select studies were included due to limitation of data), and the number of patients that were discharged.

2.4 Funding Source

No funding was obtained for the study.
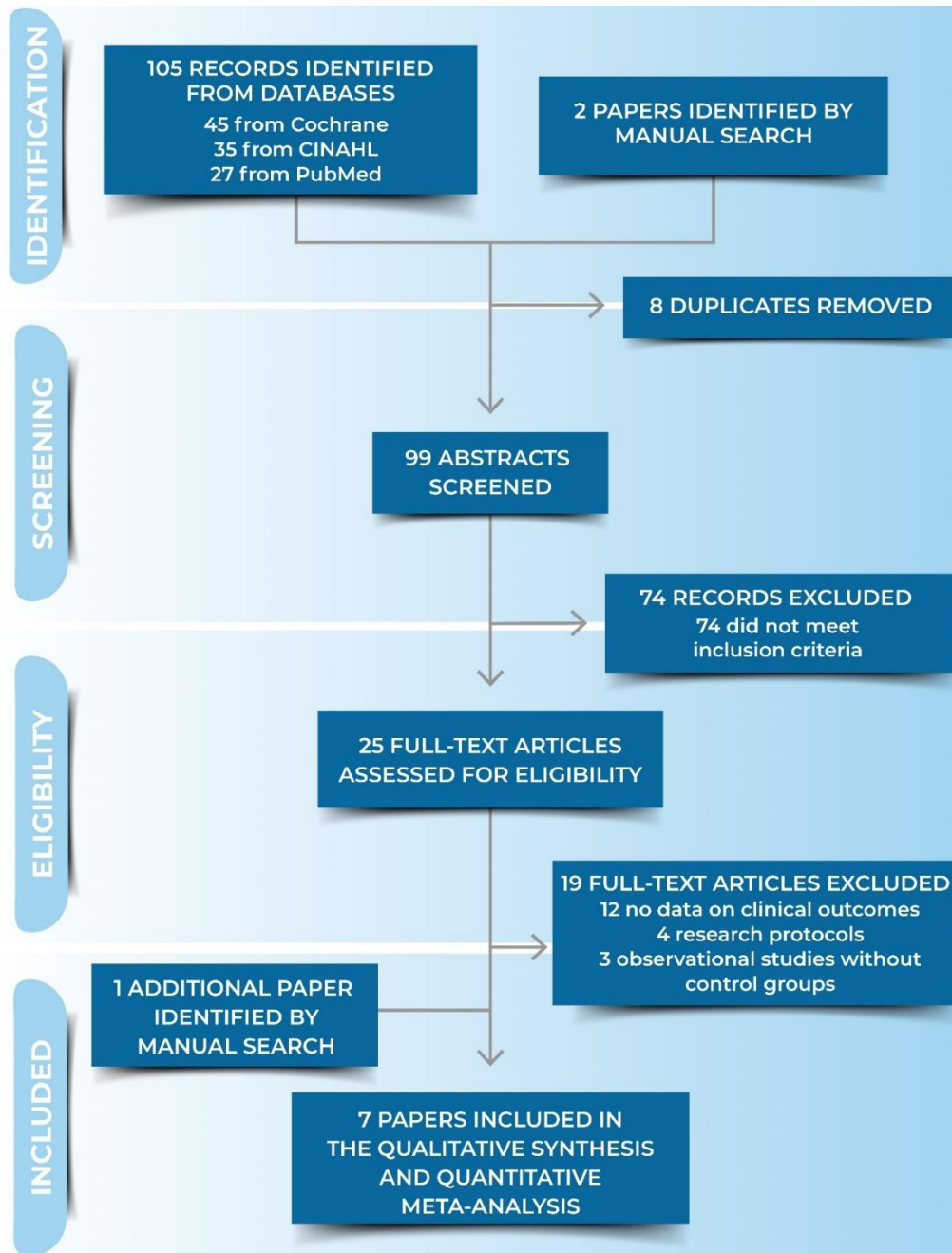
Figure 2. PRISMA flowchart

3. RESULTS

The literature search yielded 108 articles (including manual searching and lateral entries). After removing duplicates and excluding studies based on screening abstracts or through full-text examination, 7 studies were identified as eligible for inclusion (Table 1). The 7 papers consisted of 627 (23.9%) patients in the convalescent plasma group and 1997 (76.1%) patients in the control group. The studies were conducted in the United States, China, Netherlands, India, and Iran. All 7 studies (100%) reported morality, CP dosage, and concomitant medication data. Only 5 studies (71.4%) noted the number of patients that were discharged. A total of 4 studies (57.1%) reported the number of patients that clinical improved at day 14 of treatment, whereas only 2 studies

(28.6%) documented clinical improvement at day 7 and 28, and the time to improve for the treatment and control group.

3.1 Clinical Improvement at Day 14

Three of the 7 studies presented data on clinical improvement at day 14 (Figure 3). The CP group had a higher likelihood of showing clinical improvement at day 14 as compared to the control group (OR: 1.46; CI: 0.88 to 2.45; P=0.15). There was no heterogeneity between the studies (I^2=0%).

3.2 All-Cause Mortality

All studies documented mortality rates between the two groups by the end of the study duration (Figure 3). The CP group had a lower mortality risk as compared to the control group (OR: 0.69; CI: 0.50 to 0.96; P=0.03). Between all seven studies, no heterogeneity was found with an I^2 value of 0%.

3.3 Patients' Discharge Status

Three of the 7 studies were utilized to assess the patients' discharge status (Figure 4). The CP group had a higher probability of discharge during the course of the study as compared to the control group (OR: 1.87; CI: 1.1 to 3.18; P=0.02). There was no heterogeneity between the studies (I^2=0%).

3.4 Quality Appraisal of Included Studies

Overall, the risk of bias of all included studies was high due to the study design, the type of participants, and the previous or concomitant treatments. Agarwal et al.'s trial had a low risk of bias, whereas the evidence from the other included RCTs was either unclear for mortality outcomes or had a high risk of bias for safety outcomes. There was a critical risk of bias in the included non-randomized clinical trials. The risk of bias in outcomes measurement was low for Zeng, 2020, and it was moderate for Xia, 2020, and Liu, 2020. Since publication bias could influence all analyses and conclusions formed, all relevant trial registries were also searched to detect ongoing trials and completed studies.

4. DISCUSSION

Our findings must be used with caution, while major efforts are being made to identify evidence for safety and survival of convalescent plasma for COVID-19. The existing studies provide evidence and insight to support the benefits of CP therapy among COVID-19 patients. We report beneficial outcomes of CP therapy in hospitalized COVID-19 patients with reduced all-cause mortality and clinical improvement at day 28 when compared with control groups. In the first few months of the COVID-19 pandemic, CP therapy gained attention as a potentially efficacious drug followed by minimal evidence of efficacy due to the lack of high-quality evidence from randomized clinical trials. Ever since recent studies have supported the benefits of administering humoral immunity conferred by CP therapy. Similarly, our findings support the use of CP as an effective therapy in COVID-19 illness with prior studies reporting minimal adverse events.

A Cochrane living review has insofar included 20 studies, of which 1 RCT, 3 controlled NRSIs, and 16 non-controlled NRSIs were assessed for benefits in clinical improvement or mortality [5]. However, a high risk of bias was noted due to the laxity of evaluating convalescent plasma irrespective of study design, disease severity, type of participants and previous or concurrent treatments [5]. Results from the PLACID trial indicated that no benefits in all-cause mortality at day 28 or the progression to severe disease among moderately-ill patients were seen post convalescent plasma treatment [6]. The progression to severe disease in the convalescent plasma arm and the best standard of care arm was 7.2% and 7.4% respectively, suggesting no survival benefits in the intervention group [6]. Moreover, the trial conducted among 464 patients found that no differences were found in the WHO ordinal scale for clinical improvement at any time points for observation [6]. Zeng et al. state that the clinical efficacy of treatment is associated with the CP transfusion time, the transfused dose, and CP neutralizing antibody level, with benefits findable in COVID-19 patients with severe disease [7]. A single center open label phase II randomized control trial assessed the pathogen and host-intrinsic factors and the immunological outcomes post convalescent plasma therapy in 80 patients (N=40 in both SOC and CPT arms) [8]. While no statistical significance was found for clinical improvement of patients in the CPT arm, improvements were noted in 1) immediately mitigating hypoxia, 2) reducing length of hospital stay, and 3) improving survival in severe COVID-19 patients [8]. The trial ascertains that precise therapeutic therapies are necessitated for moderate to severely infected COVID-19 patients [8].
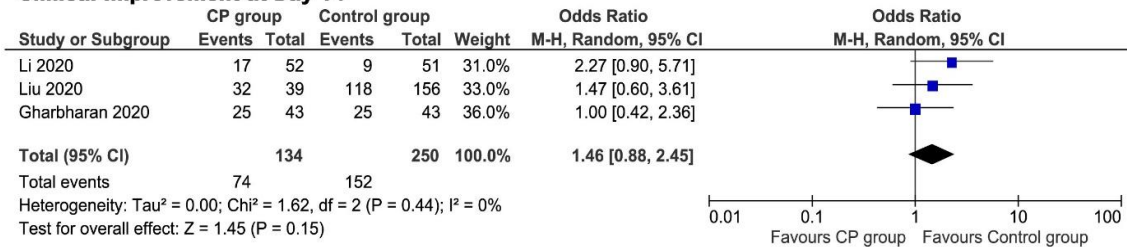
Table 1. Characteristics of included studies

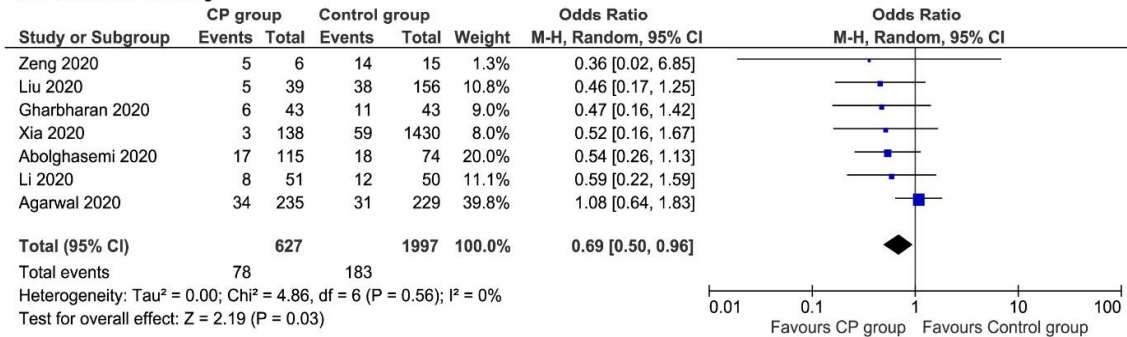| Author, year	Design (registration)	Time between symptom onset and CP administration	Duration after symptom resolution of CP donors	CP dosage	Concomitant therapies
Li et al, 2020	Randomized Clinical Trial (ChiCTR2000029757).	Median (IQR): 27d (22d-39d) for the onset of CP administration, with 3/49 (6.1%) ≤14d and 46/49 (93.9%) >14d.	Previously diagnosed patients who fully recovered (2 negative PCR tests), and were discharged from the hospital for more than 2 weeks.	Plasmapheresis. 96% or more were given 1 dose (median dosage of 200 mL, IQR 200-300 mL)	Both groups received: 1. Antivirals, 2. Interferon, 3. Chinese herbal medicine, 4. Antibacterial, 5. Antifungal, 6. Steroids, and 7. Human immunoglobulin.
Gharbharan et al, 2020	Randomized Clinical Trial (NCT04342182).	Median (IQR): 9d (7d – 13d) for the onset of CP administration.	Median (IQR): 20d (15d – 25d) after the symptoms subsided of 19 donors.	Plasmapheresis. 1 dose (300ml of plasma with anti-SARSCoV-2 neutralizing antibody titers of at least 1:80).	Both groups were allowed off-label drugs administration: 1. azithromycin, 2. chloroquine, 3. tocilizumab, 4. lopinavir/ritonavir, and 5. anakinra.
Abolghasemi et al, 2020	Non-Randomized Clinical Trial (IRCT20200325046860N1).	Only patients with ≤7 days since illness onset were included.	Donors had no remaining symptoms of COVID-19 infection for a minimum of 14 days before donation	CCP units. Most received a total of 500 mL of plasma.	Both groups received 1. Lopinavir/Ritonavir, 2. Hydroxychloroquine and 3. Anti-inflammatory agents.
Xia et al, 2020	Retrospective Cohort Study.	Median (IQR): 45d (39d-54d) for the onset of CP therapy.	Blood from donors was obtained at least three weeks after the onset of symptoms.	Eligible CCP units. Most patients received 1-2 units (200-400ml) of plasma.	All patients received 1. Traditional Chinese medicine, and 2. Anti-viral therapy.
Liu et al, 2020	Retrospective Matched Control Study.	Median (IQR): 7d (0d-14d) symptoms prior to initial presentation, and 4d (1d-7d) between admission and	Not reported.	Plasmapheresis ABO-matched. 2 doses (250 mL of each dose).	Both groups received 1. Broad-spectrum antibiotics, 2. Azithromycin, 3. Therapeutic

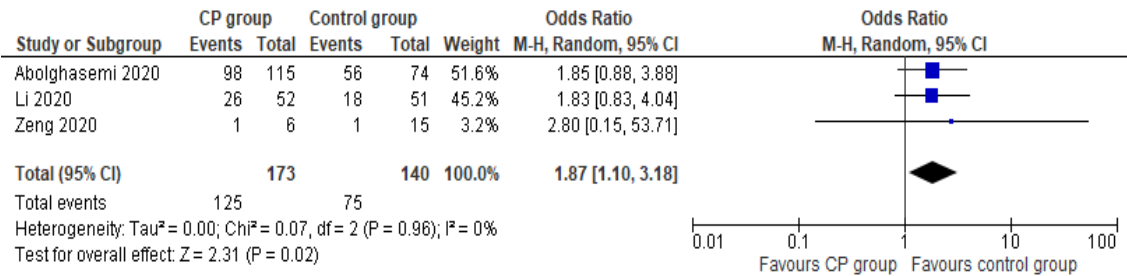| Author, year	Design (registration)	Time between symptom onset and CP administration	Duration after symptom resolution of CP donors	CP dosage	Concomitant therapies
		transfusion.			anticoagulants, 4. Hydroxychloroquine, 5. Corticosteroids, 6. Stem cells, 7. Directly acting antivirals, 8. Interleukin 1, and 9. Interleukin 6 inhibitors.
Zeng et al, 2020	Retrospective Matched Control Study.	Median (IQR): 21.5d (17.8d–23d) duration between viral shedding and treatment.	Young adults who recovered from COVID-19 for 1–2 weeks.	1-2 doses (300 mL of each dose, ranging from 200 mL to 600 mL).	Both groups were given 1. Antibiotics, 2. Antiviral therapy, 3. Traditional Chinese medicine, 4. Steroid therapy, and 5. Intravenous immunoglobulins.
Agarwal et al, 2020	Open label, parallel arm, phase II, multicentre, randomised controlled trial.	Median (IQR): 8d (6d-11d) symptoms onset to enrolment in the intervention and control arm.	Men or nulliparous women aged 18 to 65, with completely resolved symptoms for 28 days before CP donation or a period of 14 days before donation with two negative RT-PCR test results.	2 doses (200 mL of convalescent plasma per dose).	Both arms were administered 1. Hydroxychloroquine, 2. Lopinavir/Ritonavir, 3. Methylprednisolone, 4. Dexamethasone, 5. Hydrocortisone, 6. Tocilizumab, 7. Heparin (UFH/LMWH), 8. Azithromycin, 9. Intravenous immunoglobulin, 10. Other antibiotics.

Clinical Improvement at Day 14

Study or Subgroup	CP group Events	Total	Control group Events	Total	Weight	Odds Ratio M-H, Random, 95% CI
Li 2020	17	52	9	51	31.0%	2.27 [0.90, 5.71]
Liu 2020	32	39	118	156	33.0%	1.47 [0.60, 3.61]
Gharbharan 2020	25	43	25	43	36.0%	1.00 [0.42, 2.36]
Total (95% CI)		134		250	100.0%	1.46 [0.88, 2.45]
Total events	74		152			

Heterogeneity: Tau² = 0.00; Chi² = 1.62, df = 2 (P = 0.44); I² = 0%
Test for overall effect: Z = 1.45 (P = 0.15)

All-cause mortality

Study or Subgroup	CP group Events	Total	Control group Events	Total	Weight	Odds Ratio M-H, Random, 95% CI
Zeng 2020	5	6	14	15	1.3%	0.36 [0.02, 6.85]
Liu 2020	5	39	38	156	10.8%	0.46 [0.17, 1.25]
Gharbharan 2020	6	43	11	43	9.0%	0.47 [0.16, 1.42]
Xia 2020	3	138	59	1430	8.0%	0.52 [0.16, 1.67]
Abolghasemi 2020	17	115	18	74	20.0%	0.54 [0.26, 1.13]
Li 2020	8	51	12	50	11.1%	0.59 [0.22, 1.59]
Agarwal 2020	34	235	31	229	39.8%	1.08 [0.64, 1.83]
Total (95% CI)		627		1997	100.0%	0.69 [0.50, 0.96]
Total events	78		183			

Heterogeneity: Tau² = 0.00; Chi² = 4.86, df = 6 (P = 0.56); I² = 0%
Test for overall effect: Z = 2.19 (P = 0.03)

Figure 3. Forrest plots of clinical improvement at day 14 and all-cause mortality in CP and control groups

Study or Subgroup	CP group Events	Total	Control group Events	Total	Weight	Odds Ratio M-H, Random, 95% CI
Abolghasemi 2020	98	115	56	74	51.6%	1.85 [0.88, 3.88]
Li 2020	26	52	18	51	45.2%	1.83 [0.83, 4.04]
Zeng 2020	1	6	1	15	3.2%	2.80 [0.15, 53.71]
Total (95% CI)		173		140	100.0%	1.87 [1.10, 3.18]
Total events	125		75			

Heterogeneity: Tau² = 0.00; Chi² = 0.07, df = 2 (P = 0.96); I² = 0%
Test for overall effect: Z = 2.31 (P = 0.02)

Figure 4. Forrest plots of patients' discharge status in CP and control groups

A study conducted in the United States looked at 5,000 patients who were given convalescent plasma treatment; 81% of these patients had severe or critical COVID-19 illness [9]. The overall incidence of serious adverse events (SAEs) was less than 1% in the first four hours following plasma transfusion, and the 7-day mortality rate was 14.9% (95% CI, 13.8%, 16%). In another study conducted in Eastern Anatolia, 26 COVID-19 patients received convalescent plasma and had no severe adverse events after transfusion [10]. The findings suggested statistically significant contributors to mortality including age and lymphocyte counts. Patients who died were older (74.6 years vs. 61.85 years, p=0.018) and had more severe lymphopenia (0.47 vs. 1.18, p=0.001). Patients with COVID-19 in the early stages of disease who received convalescent plasma improved with treatment [10].

As part of the Expanded Access program, the Mayo Clinic assessed the safety and efficacy along with 7-day and 30-day mortality in 35,322 hospitalized patients suffering from COVID-19 [11]. The 7-day mortality rate was lower in patients transfused within 3-day time points (8.7%) in contrast to patient who were transfused ≥ 4 days of COVID-19 diagnosis (11.9%). The 30-day mortality also yielded similar results (21.6% vs. 26.7%, p<0.0001). Furthermore, the IgG antibody titers corresponded to the mortality gradient with patients who received plasma with high IgG antibody levels had a lower 7-day

mortality rate (8.9%) contrary to the much higher 7-day mortality rate (13.7%) observed in patients transfused with low IgG antibody levels. Similar IgG dose-response ratio was also observed in the 30-day mortality rate. The pooled relative risk (RR) among patients who received high IgG plasma levels was 0.65 for 7-day mortality rates and 0.77 for 30-day mortality rates when compared to patients with low IgG titers. The findings suggested the efficacy of high IgG titers and the dire need for early transfusion of antibodies to achieve a greater reduction in mortality rates.

4.1 Evidence from Previous Infectious Outbreaks

The benefits of convalescent plasma as a therapeutic tool have been witnessed since the SARS-CoV outbreak dating back to 2003 [12]. In a cohort of 80 SARS-CoV patients, it was found that patients were administered convalescent plasma at an average time of 2 weeks post symptom onset. Additionally, 48 of 80 patients who received convalescent plasma therapy before the two-week time point had a higher likelihood of improvement as opposed to those who were given therapy post the two-week time point (58.3% vs. 15.6%, p<0.001) [13]. Notably, the mortality rate was lower in the group that received plasma earlier (6.3% vs. 21.9%, p=0.08). A systematic review and meta-analysis conducted in 2013 that looked at SARS-CoV and severe influenza found that there was a significant decrease in the odds of mortality for patients given convalescent plasma therapy (OR=0.25, 95% CI: 0.14-0.45, I^2=0%) [14]. However, the included studies did not record the concentration of neutralizing antibodies in the plasma or the standard doses administered use [13,14]. Evidence from the SARS epidemic did not delineate the correlation between plasma volumes given or donor coronavirus antibody titers and clinical outcome; this could be misleading as physicians administered multiple unstandardized infusions from various donors in attempts to stop the patients from deteriorating [13].

Convalescent plasma taken from patients who recovered from the 2009 pandemic influenza A (H1N1) infection was enriched into hyperimmune IV immunoglobulin (H-IVIG), which has higher pathogen-specific antibody levels and prepared from pooled plasma in a cohort. The findings reported an independent reduction in post-treatment viral loads among the sub-group of 17 patients receiving H-IVIG relative to 18 patients in the comparator group receiving IV immunoglobulin (p=0.04 vs p=0.02); H-IVIG treatment independently reduced mortality in patients infected with H1N1 when initiated within 5 days of symptom onset (OR: 0.14; 95% CI: 0.02-0.92; p=0.04) [15]. A meta-analysis of eight studies between 1918 to 1925 analyzed outcomes among 1,703 patients infected with the Spanish flu with a significant difference in mortality between the treatment (n=336) and control group (n=1219) being 8% to 26% (pooled risk difference: 21%, 95% CI; 15% to 27%) [16].The absolute risk differences in mortality between the early treatment group (<4 days of symptom onset) and the late treatment group (>/=4 days of symptoms onset) was 26% to 50% (pooled risk difference: 41%, 95% CI: 29% to 54%).

4.2 Enrollment and Implementation in Clinical Trials

In 2013, the WHO declared convalescent plasma as one of the most promising therapies against the MERS epidemic at the International Severe Acute Respiratory and Emerging Infection Consortium (ISARIC) MERS-CoV Outbreak Readiness Workshop [17]. A research protocol of convalescent plasma therapy for patients suffering from middle east respiratory syndrome (MERS) was formulated by Arabi et al. in Saudi Arabia whereby individuals with antibody titers >1:160, and no clinical or laboratory evidence of infection would be screened for plasma donation eligibility. The potential donors would be tested for the presence of antibodies by ELISA and immunofluorescence assay (IFA) along with the fulfillment of standard donation criteria. The protocol stated that critically-ill hospitalized patients would receive 2 units of convalescent plasma and be monitored for outcomes based on viral load clearance [18]. Consequently, Arabi et al. recruited 443 plasma donors per the protocol; however, only 12 subjects had a positive ELISA result, and 9 out of the 12 participants had positive IFA implicating the low rates of seroconversion for MERS-CoV [19]. These findings reiterate the challenge of standardized donation criteria of CP, especially during emergency settings.

Clinical trials investigating the efficacy of CP therapy against the Ebola Virus Disease (EVD), conducted between 2014 to 2015, were plagued with many limitations such as the patients in the control group having various comorbidities which

affected mortality, lack of quantification of the anti-Ebola antibody (EBOV) and neutralizing antibody when the transfusion was taking place, and using PCR instead of measuring viral load [20]. Findings demonstrated that the patients who were given higher doses of plasma anti-EBOV IgG resulted in reduced viral load with no notable side effects of CP therapy identified. The feasibility of the trial designs was limited by the lack of adequate resources in West African countries during the EVD outbreak. EVD trials were unable to recruit adequate participants in randomized designs [20]. For instance, the Ebola-Tx trial enrolled controls if compatible plasma was unavailable; however, the supply of CP exceeded the number of enrolled patients resulting in the administration of CP therapy to all the participants [21].

5. LIMITATIONS

Our findings were limited by the lack of comparable efficacy of CP in either severe or moderate patients in the randomized clinical trials. The time-points at which CP was administered in COVID-19 were not provided by all the studies in our analysis. The viral load of the patients was not monitored in a few studies, accounting for the lack of adequate insight into the biological mechanisms of CP. Furthermore, the patients in the control groups were not as severely-ill as in the interventional groups in non-randomized studies, possibly distorting the effect of the study intervention. Some studies ended prematurely due to the varying guidance from governing health bodies. Consequently, the determination of definite outcomes concerning the clinical benefits of CP was not ascertained. Additionally, the open-label design of the randomized clinical trials in our analysis may have introduced conduct bias. While randomization in the clinical trials contributes to bias reduction, the ascertainment bias, referring to the knowledge of intervention the patient is receiving, may result in reduced adherence or increased drop-out rates in open-label designs.

6. RECOMMENDATIONS

- First, CP and its efficacy ought to be thoroughly monitored by healthcare workers (HCWs) keeping records of the dosage, harvesting procedures, storage of specific plasma volumes, use in specific risk groups, and adverse events.
- Second, recipients of CP must potentially receive the therapy as part of a clinical trial

in an approved health facility only after screening against the eligibility criteria.

- Third, potential donors may be similarly screened as recipients to identify their eligibility. Donors should be considered to be eligible if they have recovered from COVID-19 and are asymptomatic for at least 2 weeks.
- Fourth, while the specific neutralizing antibodies in CP have been examined in recent literature, CP may be administered after determining the levels of antibody titers alone. However, trials may analyze the specific neutralizing antibodies within CP to identify trends and promote the development of a rigid protocol to ensure optimized therapy.
- Fifth, trials are necessary to ascertain the relevant clinical manifestations for which COVID-19 patients require CP transfusions. Findings from multiple-arm trials can identify the benefits of administering CP at different stages of COVID-19 disease severity.

7. CONCLUSION

Our findings support the practicality, feasibility, and safety of convalescent plasma as COVID-19 therapy. Scientists are currently exploring the effectivity of early convalescent plasma therapy and monoclonal antibodies. While a large number of observational studies and trials demonstrate the administration of convalescent plasma to patients with severe COVID-19 disease, it must be noted that the mechanism of neutralizing antibodies is intended to prevent the initial binding of the virus during the early phase of illness. While our study suggests that there is effectiveness in hospitalized patients, keeping in mind theoretical considerations, the best time of use may be in the outpatient department, or for mild-moderate COVID-19 infections within 7 days of symptoms onset. Ongoing clinical trials must refine current COVID-19 therapies and must ensure easy administration and feasibility. Multicenter studies are required to solidify the potential of clinical improvement, survival rates of early-phase infusion of CP in COVID-19 patients, in addition to determining a standardized protocol of dosing, antibody titers, and timeline of plasma donation and collection.

8. FUNDING

CONSENT

It is not applicable.

ETHICAL APPROVAL

No ethical approval was required as no patient data was collected.

ACKNOWLEDGMENTS

All authors thank Jack Michel MD for his efforts in boosting scholarly activities at the Larkin Health System, South Miami, FL, USA.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. WHO Coronavirus Disease (COVID-19) Dashboard.
2. Esteve A, Permanyer I, Boertien D, Vaupel JW. National age and coresidence patterns shape COVID-19 vulnerability. Proc Natl Acad Sci U S A. 2020;117(28):16118–20.
3. fda issues emergency use authorization for convalescent plasma as potential promising COVID–19 treatment, another achievement in administration's fight against pandemic.
4. Marano G, Vaglio S, Pupella S, Facco G, Catalano L, Liumbruno GM, et al. Convalescent plasma: New evidence for an old therapeutic tool? Blood Transfusion. 2016;14:152–7.
5. Piechotta V, Chai KL, Valk SJ, Doree C, Monsef I, Wood EM, et al. Convalescent plasma or hyperimmune immunoglobulin for people with COVID-19: A living systematic review. Cochrane Database of Systematic Reviews; 2020.
6. Agarwal A, Mukherjee A, Kumar G, Chatterjee P, Bhatnagar T. Convalescent plasma in the management of moderate COVID-19 in India: An open-label parallel-arm phase II multicentre randomized controlled trial (PLACID Trial). medRxiv; 2020.
7. Zeng H, Wang D, Nie J, Liang H, Gu J, Zhao A, et al. The efficacy assessment of convalescent plasma therapy for COVID-19 patients: A multi-center case series. Signal Transduct Target Ther; 2020.
8. Ray Y, Paul SR, Bandopadhyay P, Rozario R, Sarif J, Lahiri A, et al. Clinical and immunological benefits of convalescent plasma therapy in severe COVID-19: Insights from a single center open label randomised control trial. medRxiv; 2020.
9. Joyner MJ, Wright RS, Fairweather D, Senefeld JW, Bruno KA, Klassen SA, et al. Early safety indicators of COVID-19 convalescent plasm in 5000 patients. J Clin Invest; 2020.
10. Erkurt MA, Sarici A, Berber İ, Kuku İ, Kaya E, Özgül M. Life-saving effect of convalescent plasma treatment in covid-19 disease: Clinical trial from eastern Anatolia. Transfus Apher Sci; 2020.
11. Joyner MJ, Senefeld JW, Klassen SA, Mills JR, Johnson PW, Theel ES, et al. Effect of convalescent plasma on mortality among hospitalized patients with COVID-19: Initial Three-2 Month Experience. medRxiv; 2020.
12. Wooding DJ, Bach H. Treatment of COVID-19 with convalescent plasma: Lessons from past coronavirus outbreaks. Clin Microbiol Infect; 2020.
13. Cheng Y, Wong R, Soo YOY, Wong WS, Lee CK, Ng MHL, et al. Use of convalescent plasma therapy in SARS patients in Hong Kong. Eur J Clin Microbiol Infect Dis. 2005;24(1):44–6.
14. Mair-Jenkins J, Saavedra-Campos M, Baillie JK, Cleary P, Khaw FM, Lim WS, et al. The effectiveness of convalescent plasma and hyperimmune immunoglobulin for the treatment of severe acute respiratory infections of viral etiology: A systematic review and exploratory meta-analysis. J Infect Dis. 2015;211(1):80–90.
15. Hung IFN, To KKW, Lee CK, Lee KL, Yan WW, Chan K, et al. Hyperimmune IV immunoglobulin treatment: A multicenter double-blind randomized controlled trial for patients with severe 2009 influenza A(H1N1) infection. Chest; 2013.
16. Luke TC, Kilbane EM, Jackson JL, Hoffman SL. Meta-analysis: Convalescent blood products for Spanish influenza pneumonia: A future H5N1 treatment? Vol. 145, Annals of Internal Medicine. 2006; 599–609.
17. WHO-ISARIC joint MERS-CoV Outbreak Readiness Workshop: Clinical management and potential use of convalescent plasma; 2013.
18. Arabi Y, Balkhy H, Hajeer AH, Bouchama A, Hayden FG, Al-Omari A, et al.

Feasibility, safety, clinical, and laboratory effects of convalescent plasma therapy for patients with Middle East respiratory syndrome coronavirus infection: A study protocol. Springerplus. 2015;4(1):1–8.

19. Arabi YM, Hajeer AH, Luke T, Raviprakash K, Balkhy H, Johani S, et al. Feasibility of using convalescent plasma immunotherapy for MERS-CoV infection, Saudi Arabia. Emerg Infect Dis. 2016; 22(9):1554–61.

20. Adebamowo C, Bah-Sow O, Binka F, Bruzzone R, Caplan A, Delfraissy JF, et al. Randomised controlled trials for Ebola: Practical and ethical issues. The Lancet. 2014;1423–4:384.

21. Van Griensven J, De Weiggheleire A, Delamou A, Smith PG, Edwards T, Vandekerckhove P, et al. The use of ebola convalescent plasma to treat ebola virus disease in resource-constrained settings: A perspective from the field. Clinical Infectious Diseases. 2016;62:69–74.