

Review Article

Indian J Med Res 153, January & February 2021, pp 64-85
DOI: 10.4103/ijmr.IJMR_3092_20



Use of convalescent plasma for COVID-19 in India: A review & practical guidelines

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Received July 19, 2020

Convalescent plasma (CP) therapy is one of the promising therapies being tried for COVID-19 patients. This passive immunity mode involves separating preformed antibodies against SARS-CoV-2 from a recently recovered COVID-19 patient and infusing it into a patient with active disease or an exposed individual for prophylaxis. Its advantages include ease of production, rapid deployment, specificity against the target infectious agent, and scalability. In the current pandemic, it has been used on a large scale across the globe and also in India. However, unequivocal proof of efficacy and effectiveness in COVID-19 is still not available. Various CP therapy parameters such as donor selection, antibody quantification, timing of use, and dosing need to be considered before its use. The current review attempts to summarize the available evidence and provide recommendations for setting up CP protocols in clinical and research settings.

Key words Antibody-dependent enhancement - anti-SARS-CoV-2 antibodies - convalescent plasma - COVID-19 - donor selection - neutralizing antibodies - passive immunization

COVID-19 in India

The cases of COVID-19 in India, the pandemic caused by SARS-CoV-2, has reached to 10,582,647 and total deaths above 152,000 on January 19, 2021¹. There is currently no proven specific therapy, and multiple novel and repurposed molecules are being used on an experimental basis. The lack of effective therapeutic options has been a major hurdle in our pandemic mitigation measures.

Convalescent plasma (CP), an overview

Convalescent plasma (CP) is a mode of passive immunization wherein preformed antibodies against an infectious agent are infused into a susceptible host with the aim of either preventing or treating the infection² (Fig. 1). CP was widely used in the past to treat several bacterial (diphtheria, tetanus, pneumococcal pneumonia and meningococemia) and viral infections (rabies, poliomyelitis and

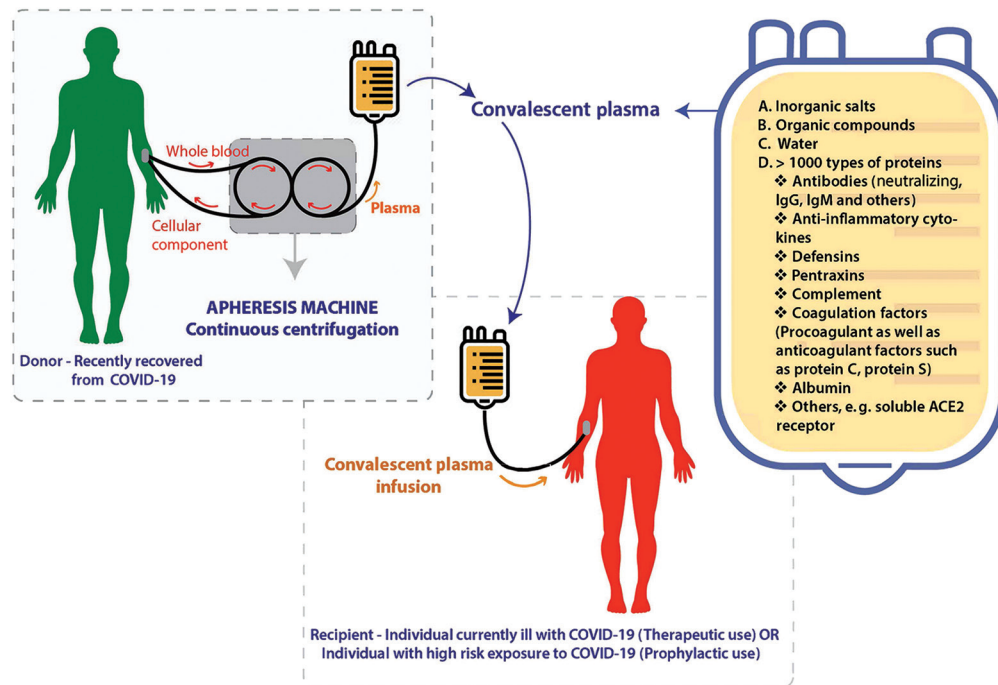


Fig. 1. Overview of convalescent plasma (CP) therapy. The Figure shows the process of CP donation and its use in a patient with COVID-19. CP contains over a thousand types of proteins. Apart from the antibodies against SARS-CoV-2, various other proteins may contribute to the beneficial effects of CP administration, including anti-inflammatory cytokines, complement and clotting factors.

measles) in the pre-antibiotic era³⁻⁵. Safer and more standardized modalities such as hyperimmune globulins and monoclonal antibodies are currently available against diseases caused by agents such as *Clostridium tetani*, hepatitis A and B, rabies and respiratory syncytial virus⁴ (Fig. 2). CP still retains an important place in the management of infections caused by novel pathogens because of the rapidity and ease with which it can be put to clinical use (Table I). Hence, it has been widely used during almost all recent epidemics, including H1N1 influenza^{6,7}, SARS^{8,9}, Middle East respiratory syndrome (MERS)^{10,11} and Ebola^{12,13}. Most of the literature from these epidemics is in the form of case reports or series. There are very few randomized control trials (RCTs)^{14,15} or systematic reviews and meta-analyses^{16,17}. Past experience unequivocally attests to CP's safety with very few adverse events reported^{17,18}. Pooled analyses suggest the benefit of CP use in diseases such as H1N1, SARS and Argentine haemorrhagic fever, especially when used early in the illness^{16,17}. Data from across the globe regarding various aspects of CP use are accumulating at a fast pace. The current review aims to summarize the available evidence in COVID-19

disease and provide the recommendations for setting up CP protocols in clinical and research settings.

Immunological aspects of convalescent plasma therapy in COVID-19

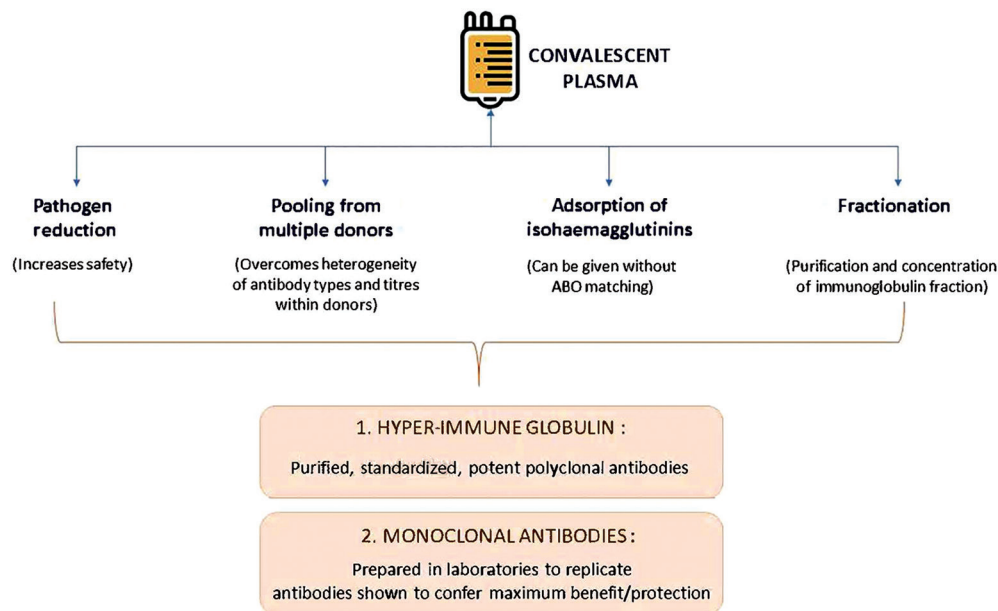
CP therapy involves collecting plasma from an individual who has recently recovered from infection and infusing it into an at-risk individual^{2,19} (Fig. 1). Such plasma contains several humoral factors capable of providing immunity, but the most important one is believed to be the polyclonal antibodies against the target agent¹⁸. CP confers immune-protection by numerous mechanisms (Fig. 3). Direct viral neutralization by neutralizing antibodies (NAbs) and the immunomodulatory actions that limit host damage are important mechanisms in COVID-19¹⁸.

Immunological studies have demonstrated a diverse antibody repertoire in the serum of recently recovered patients²⁰. Antibodies directed against the receptor-binding domain of the viral spike protein (S-RBD) bind them, limit cell entry, and inhibit the virus amplification. Laboratory studies have shown potent neutralizing activity by such antibodies repertoire against the live virus²¹⁻²⁵. The majority of patients achieve seroconversion within two weeks of symptom onset^{21,26-28} (Fig. 4). While data regarding the

Table I. Suitability of convalescent plasma (CP) use during outbreaks by novel pathogens

Rapidity of deployment	CP can be put to clinical use as soon as a pool of individuals recovered from the illness is available
Ease of production and use	Plasma collection and infusion are relatively straightforward and commonly performed procedures. Most healthcare workers will be familiar with the procedure and will require no extra training. In resource-constraint settings, convalescent whole blood can also be used instead of plasma
Accessibility	The expertise and equipment to perform plasmapheresis, storage and deployment are commonly available in most tertiary care centres around the globe
Specificity	The CP, by its very nature, is specific against the targeted infectious agent. In a few instances, such as the 1918 Spanish flu, CP was used even before the identification of the pathogenic agent. The use of CP locally will help prepare a product with specific action against the particular antigenic variant prevalent in that particular geographical area
Immediate onset of action	Helps confer immediate protection to individuals already suffering from the disease
Safety	Risks of CP infusion are minimal, and it is a safe therapy
Scalability	Can be easily scaled up to the level of a large population

Source: Ref. 2

**Fig. 2.** Post-donation modifications of convalescent plasma (CP). The Figure shows the possible post-donation modifications to CP to create safer, more potent, standardized passive immunization products such as hyperimmune globulins and monoclonal antibodies against SARS-CoV-2.

long-term duration of this humoral response in COVID-19 are still evolving, studies conducted during the SARS epidemic have shown that this antibody response is short-lasting, with peak levels at around 3–4 months followed by gradual waning over the next two years²⁹.

Measurement of antiviral activity of convalescent plasma: Neutralizing antibody titers

The neutralizing activity of CP directly quantifies the plasma's protective activity and has been used as a gold standard for the quantification of its efficacy. These are functional assays that involve incubating

serial dilutions of the plasma with a standard dose of the live virus and inoculation of this mixture in a culture medium. The dilution of the plasma that prevents the culture of the virus or its cytopathic effect is defined as the neutralizing titre of the plasma³⁰. However, these assays are not available on a large scale for CP therapy because of accessibility and standardization problems.

The use of commercially available serological assays as alternatives to neutralizing assays is an attractive option. A few recent publications have demonstrated a good correlation of several such

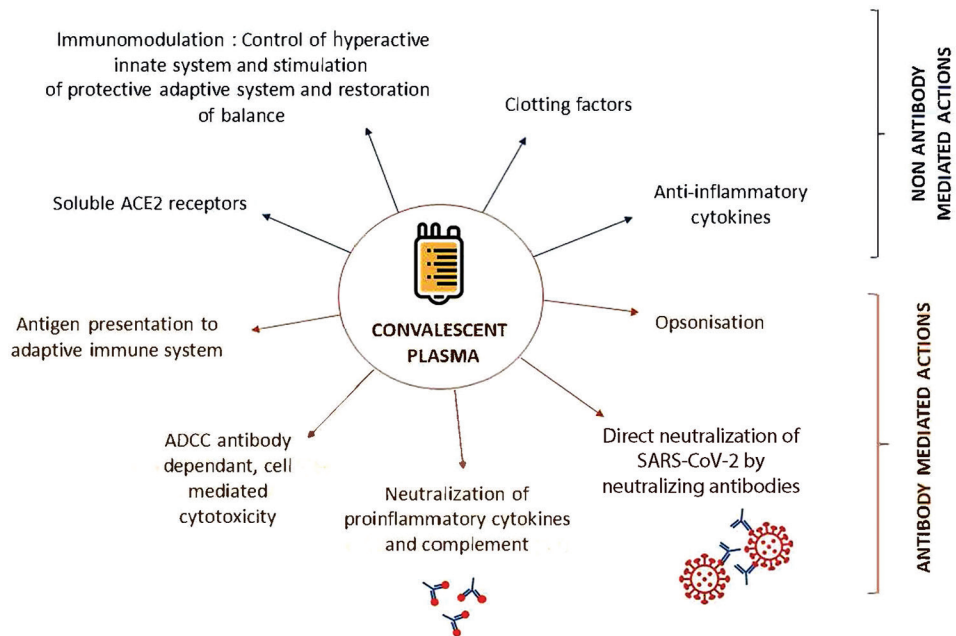


Fig. 3. Mechanisms of action of convalescent plasma (CP). The Figure depicts the multiple possible mechanisms of action of CP. Of note, there are multiple non-antibody-based as well as non-viral neutralization-based mechanisms of action. Hence, CP with low or absent anti-SARS-CoV-2 antibodies can theoretically provide beneficial effect when administered to COVID-19 patients. ADCC, antibody dependent cellular cytotoxicity.

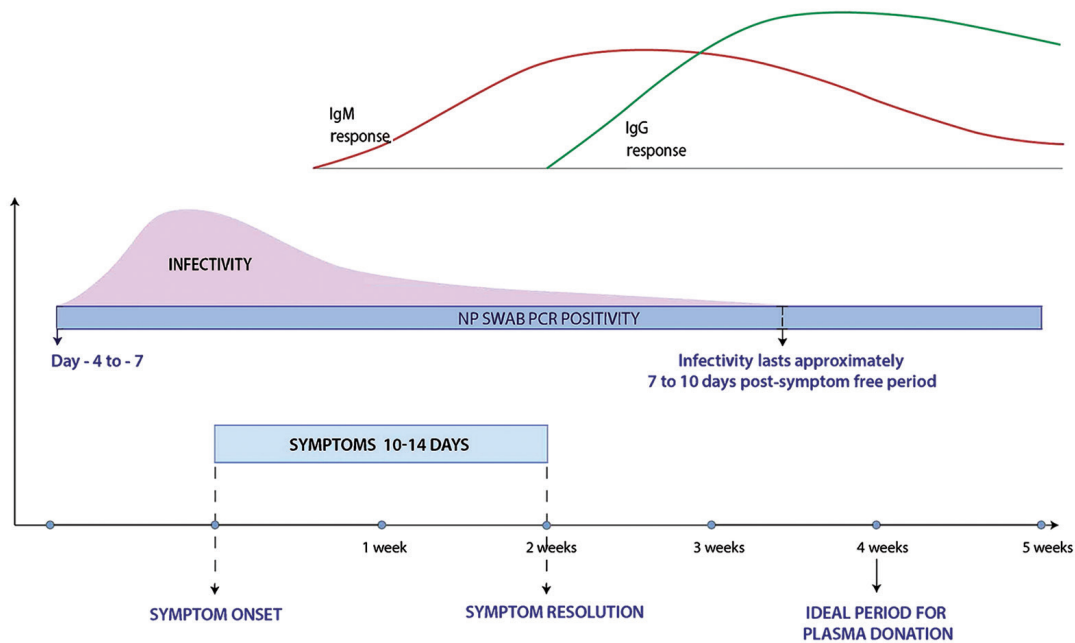


Fig. 4. Current understanding of the relevant immunological concepts in COVID-19. The Figure explains the current concepts in the humoral immune response of COVID-19. (i) IgM antibody production starts at around 4-7 days, peaks by approximate week three and wanes gradually by 3-4 months from symptom onset^{20,21,28}. (ii) IgG antibody production starts at around 10-14 days. The disappearance of IgG response is not exactly known; however, based on data during the earlier SARS epidemic, it is expected to be short-lasting and be undetectable in most individuals by 24-36 months²⁹. (iii) Period of infectivity peaks a couple of days before symptom onset.

serological titres, especially assays targeting the S-RBD domain and neutralizing assays^{23,24,31-33}. The New York

Blood Centre (NYBC) found that two commercial assays (by Abbott and ORTHOS) showed a good

Table II. Challenges in the development of assays for antibody titre quantification for convalescent plasma (CP) therapy

Problems in the availability of neutralizing assays	Suggested solutions
Requires handling of live virus. Requires biosafety level 3 laboratory. Complex methodology. High turnaround time. Not amenable for automation. Not amenable for high throughput testing. Currently being done in India in NIV, Pune, on a research basis only.	Development of safer alternatives to neutralization assays such as using a non-infective pseudo-typed virus ^{24,32} . Use of ELISA and other serological assays to quantify antibodies in CP.
Problems in the standardization of antibody assays	Suggested solutions
Variability in reported assays in literature with respect to (i) Methodology and reagents. (ii) Platforms used (neutralizing assays, ELISA, CLIA, high throughput serological assays, lateral flow assays, etc.). (iii) Target antigen (S, N, others), target isotype (IgA, IgM, IgG1, IgG2, IgG3). (iv) Cut-offs used to define high titre antibodies. For instance, USFDA recommends CP with neutralizing titres >1:160 ³⁸ while the European Commission recommends >1:320 by neutralizing assay ³⁹ . The RCT from Wuhan, China, used donors with antibody titres >1:640 measured by an IgG ELISA detecting S-RBD domain, corresponding to 1:20 by neutralizing assays by their laboratory analysis ³³ . The recent EUA by the USFDA uses S/CO ratio >12 by the ORTHO-VITROS assay to define high titre plasma ⁴⁰ .	Accumulating data from RCTs will help determine what cut-offs should be used for high antibody titre CP. Comparative studies between neutralization assays and various types of serological assays are needed, and several are currently underway. Ideally, these surrogates of neutralization assays need to be tested for efficacy in an RCT setting before widespread use.
RCTs, randomized controlled trials; EUA, Emergency Use Authorization; ELISA, enzyme-linked immunosorbent assay; CLIA, chemiluminescent immunoassay; NIV, National Institute of Virology; USFDA, U.S. Food and Drug Administration	

correlation with NAb titres in a study of serum from 370 CP donors³⁴. The ConCOVID group investigators from the Netherlands found a good correlation of a commercially available ELISA with neutralization assays^{32,35}. The assay manufactured by EuroImmun AG has also been shown to correlate with neutralization assays³⁶ and used in a RCT from Spain³⁷. Another issue in the antibody titre measurement of CP is the wide variability in the methodology and cut-offs used in the literature (Table II). Ideally, these parameters need to be standardized and validated in clinical trials before clinical use.

Ideal donor

A potential CP donor for COVID-19 is a healthy adult who has recently recovered from COVID-19 infection and remained asymptomatic for a minimum of 14 days. This period is based on the estimates of antibody kinetics and period of infectivity (Fig. 4). Documented nasopharyngeal swab PCR negativity before CP donation is no longer mandatory in most CP protocols across the globe⁴¹. Based on the available antibody kinetics data, the ideal window for donation is 3-4 months after symptom onset²⁹.

There is a wide variability in the titres of anti-SARS-CoV-2 antibodies within donors⁴². Initial data from China showed that a high proportion of donors

(39/40) possessed NAb titres >1:160⁴³. However, recent reports have shown more variations. In the NYBC report, 55 per cent of the screened 370 CP donors did not have high titre antibodies, while 10 per cent had very high levels³⁴. The ConCOVID group reported similar findings in a cohort of 115 plasma donors, with 43 per cent screened donors having neutralizing titres >1:320 and 10 per cent with titres >1:1280³⁵. In other studies, the proportion of donors found lacking in high titre antibodies ranged from 25 to 50 per cent^{23,44,45}, with about 10-20 per cent having very high antibody levels of more than 20 times the cut-off limits, the so-called super-donor phenotype⁴⁵. Older age, male gender and clinical features such as moderate-to-severe disease²⁷, high C-reactive protein (CRP) and lymphocytopenia^{23,46} have been found to be associated with higher NAb titres. Notably, asymptomatic individuals have been found to mount a very transient and low titre antibody response⁴⁷. These clinical and demographic factors may help guide the selection of suitable donors.

Ideally, donor selection should be based on their NAb titres, but such an approach is currently not feasible in India (Table II). Due to the unavailability of such assays, most clinical and trial protocols are proceeding with CP therapy without quantification of antibody titres. Of note, the compassionate use programmes

in the USA and Israel followed this approach and determined antibody titres of the infused plasma units *post hoc*, and their findings have been recently published^{36,48}. The Indian Council of Medical Research (ICMR)-sponsored PLACID trial also followed this strategy. The antibody titres in this study were found to be low, with a median neutralization titre of 1:40 [interquartile range (IQR) 1:30-1:80]⁴⁹. These findings have highlighted the hazards of proceeding with CP therapy without appropriate antibody quantification in real time. For now, it seems to be a reasonable strategy to screen potential CP donors for the presence of anti-SARS-CoV-2 antibodies using commercially available serological tests to exclude donors with low anti-SARS-CoV-2 antibodies.

Process of plasma donation

A prospective donor should fulfil the standard donor screening criteria, as per the Drugs and Cosmetics Rules, 1945⁵⁰, and the COVID-19 advisory from National Blood Transfusion Council (NBTC)⁵¹. Plasma donation is a voluntary exercise and requires informed consent. Although whole blood can be used in resource-limited settings, plasmapheresis remains the preferred method of CP donation because of the larger collection volume of plasma, the feasibility of repeated collections and the minimal impact on the donor's haemoglobin. Up to 15 per cent of total blood volume (400-800 ml per session) can be safely donated in a single sitting^{41,52-54}. Recommendations allow for serial plasma donations by a single donor with a minimum gap of seven days between consecutive plasmapheresis sessions^{54,55}.

Donated plasma is frozen at less than -30°C (preferably -40 to -80°C) within 8-24 h of collection and can be stored for up to 12 months. Pathogen inactivation by techniques such as photochemical inactivation or solvent detergent treatment and pooling from multiple donors are other post-donation processing options to improve the safety and quality (Fig. 2).

Clinical use of convalescent plasma

Despite the widespread use and publicity of CP, it remains an experimental therapy. Conclusive proof of its efficacy and the parameters of its effective use remain to be firmly established. In such a situation, it is advisable to use CP only under a research protocol after discussing the experimental nature and potential risks of CP use with the recipient.

The use of CP can be envisioned primarily in four clinical settings. The factors related to the dosing of CP in different settings are summarized in Tables III and IV. A cardinal principle is that earlier use in the course of illness is expected to be more beneficial^{2,4}. Early in the course of the disease, the viral burden is less, and the infection is not yet firmly established. The hypothesized mechanism of action of CP is by prevention or delay in the establishment of infection long enough to allow for the host immune response to clear the infection. In the latter stages of illness, the hyper-inflammatory state is the major driver of morbidity. The ongoing tissue damage maintains the inflammatory state as a vicious cycle, and control of viral replication may have a minimal effect on disease course. As a corollary, CP may be expected to be an effective modality for post-exposure prophylaxis.

Table III. Clinical use and dosing of convalescent plasma use

Setting	Description	Suggested dosing
Post-exposure prophylaxis	High risk contacts such as healthcare workers or contacts of COVID-19 patients who have high risk comorbidities	One unit of 50-100 ml plasma
Asymptomatic-to-mild illness	These patients are asymptomatic to mildly symptomatic with no organ dysfunction. Treatment is with an intent to prevent organ dysfunction. Further sub-classification in this category includes patents with high risk comorbidities such as age, obesity, hypertension and T2DM	1-2 units of standard volume (200-250 ml) plasma
Moderate-to-severe illness	Patients with organ dysfunction, hypoxia, ARDS, requiring oxygen supplementation	2-4 units of standard volume (200-250 ml) plasma
Critically ill	Patients on life support such as mechanical ventilator and ECMO	Higher doses up to 7 units of standard volume plasma, administered once daily

Note: The hypothesized dose is based on the estimate of viral load in the patient. Hence, the doses to be used are lower in mild illness and higher in severe illness. At the same time, the expected benefit of CP in severe and critically ill patients is thought to be minimal. T2DM, type 2 diabetes mellitus; ARDS, acute respiratory distress syndrome; ECMO, extracorporeal membrane oxygenation
Source: Ref. 2

Table IV. Factors to be considered to determine the accurate dose of convalescent plasma (CP)

Variables required to decide the exact dose of CP
Exact antibody titre of the infused unit of CP The clinical setting for which CP is being administered (Table III) Desired levels of antibody in the recipient known to confer protection in the particular setting* Total body plasma volume of the recipient (based on height and weight) <i>In vivo</i> pharmacokinetics of the infused antibodies*
Hypothetical calculation of CP dose
CP infusion has been planned for an 80 kg patient with moderate disease. The CP available has antibody titres of 1:160. The hypothetical target antibody titre for protection in this setting of moderate disease is 1:20. The total plasma volume of the patient at 40 ml/kg would be 3200 ml. Infusing plasma of volume x would give a final concentration of $(1/160) \times (x/3200+x)=1/20$. Completing the calculation, the dose of CP needed to be administered would be approximately 450 ml or 6 ml/kg. *Parameters which are not currently known <i>Source:</i> Ref. 2

Table IV shows the suggested dosing for use of CP in different clinical settings.

Accumulating evidence suggests that any potential benefit of CP use may be found in mild and moderate illness rather than the critically ill^{33,43}. Studies during the SARS epidemic showed CP to be more useful in symptomatic patients who were seronegative and PCR positive⁸. Results from the RCT from the Netherlands suggest the need to choose such seronegative COVID-19 patients early in the course of illness³⁵. However, the shortages of high antibody titre donors preclude the use in prophylactic or mild illness settings on a large scale. Thus, in the current scenario, the potential use of CP may be prioritized in the following settings: (i) Post-exposure prophylaxis for healthcare workers; (ii) Post-exposure prophylaxis of individuals with comorbidities at higher risk of severe illness; and (iii) Treatment of symptomatic individuals with moderate disease, especially in the first week of illness.

Risks of convalescent plasma use

Despite the use of guideline-based screening strategies, CP carries a non-zero risk of transfusion-associated infections. Other complications include allergic reactions, transfusion-related acute lung injury (TRALI) and transfusion-associated circulatory overload (TACO). COVID-19 patients with their compromised respiratory reserve are especially vulnerable to TRALI/TACO, which may be difficult to distinguish from the progression of COVID-19. In the periodically published safety analyses from the Expanded Access Program (EAP) in the USA where the safety data of the first 5000⁵⁶ and later 20,000 patients⁵⁷ have been reported, the incidence of these

reactions was very low (<1%), and only two serious adverse events could be conclusively attributed to plasma therapy. Another theoretical concern is the antibody-mediated enhancement of infectivity, a phenomenon described with several viral infections such as dengue^{58,59}. It is believed to occur when an individual harbours pre-existing antibodies against a closely related strain of the virus (such as the SARS-CoV-1, MERS or other coronaviruses). It is hypothesized that these pre-existing antibodies are sub-neutralizing in nature and mask the viral epitopes from immune recognition, thus facilitating intracellular entry and survival, leading to a paradoxical worsening of the illness⁵⁸. There is no evidence available to suggest the role of antibody-mediated enhancement in COVID-19.

Worldwide use of convalescent plasma in COVID-19

The earliest reports of CP use came from China. These and other initial reports have been summarized in Table V. By early March, CP therapy was adopted in the USA, Italy, Israel, Spain and several other countries on compassionate grounds in severely ill patients or as part of clinical trials. Several large single-arm studies from these centres (Table VI) provided a proof of concept and established the feasibility and safety of CP in COVID-19. These have reinforced the general theme that earlier administration is associated with better outcomes. The limitations of these studies included small sample sizes, absent or non-randomized control arms, simultaneous use of multiple experimental therapies and a non-uniform study design and donor selection criteria. Most of these studies were in severe and critically ill patients.

Table V. Initial case reports and case series in the use of convalescent plasma (CP) in COVID-19

Study	Study design	Convalescent plasma characteristics	Results
Shen <i>et al</i> ⁶⁰ (Shenzhen, China) published on March 27, 2020	Study type: Case series n=5 Study population: Critically ill, all on mechanical ventilation, one on ECMO, all with high viral load at the time of infusion of plasma. Control arm: Nil Blinding: Nil Randomization: Nil Other treatments: methylprednisolone, antivirals (favipiravir, lopinavir/ritonavir, darunavir, arbidol, interferon-alpha 1b)	Donor characteristics: Age 18-60 yr, asymptomatic for at least 10 days. Severity of COVID not mentioned. Donor antibody levels: ELISA for anti-SARS-CoV-2 antibody titre > 1:1000 and neutralization antibody titre > 1:40. Plasma dose: 2 units of 200-250 ml each, the same day as the donation. Plasma infusion timing: Median 22 days (14-24) after symptoms	Improvement in clinical features (fever, SOFA score), laboratory parameters (CRP, IL-6, PCT), radiological clearing on CT scan and viral shedding measured by PCR. Three patients weaned off the ventilator and discharged, the other two stables on ventilator with one weaned off ECMO until time of reporting
Zhang <i>et al</i> ⁶¹ (Guangdong, China) published on March 31, 2020	Study type: Case series n=4 Study population: Critically ill, 1 NIV, three on a mechanical ventilator, 2 ECMO, 2 on CRRT Control arm: Nil Blinding: Nil Randomization: Nil Other treatments: methylprednisolone, antivirals (a combination of antivirals in all)	Donor characteristics: Not mentioned. Donor antibody levels: Not done pre-transfusion. Plasma dose: 1-8 units per patient (200, 300, 900 and 2400 ml in each patient). Plasma infusion timing: Median 18 days (16-19) after symptoms.	Clinical and radiological improvement in all, three being discharged to home and one discharged to a step-down facility. In one patient assessed for viral load by PCR, there was rapid viral clearance.
Ahn <i>et al</i> ⁶² (Seoul, Korea) published on April 2, 2020	Study type: Case series n=2 Study population: Critically ill, on mechanical ventilation Control arm: Nil Blinding: Nil Randomization: Nil Other treatments: methylprednisolone, lopinavir/ritonavir	Donor characteristics: Male donor in 20s with symptomatic COVID. Time since symptom resolution not mentioned. Donor antibody levels: Positive for anti-SARS-CoV-2 by IgG ELISA. Plasma dose: Total 500 ml given in two divided doses. Plasma infusion timing: Day 22 and 6 after symptom onset.	Rapid clinical improvement, including improvement in fever, oxygenation status and viral PCR. One could be extubated and discharged from the hospital; the other required tracheostomy but could be weaned off from ventilator.
Duan <i>et al</i> ⁴³ (Wuhan, China) published on April 6, 2020	Study type: Case series n=10 Study population: Critically ill patients, 3 on mechanical ventilator, 3 on HFNC, 2 on nasal prongs, 1 pregnant patient Control arm: Nil Blinding: Nil Randomization: Nil Other treatments: methylprednisolone, multiple lines of antivirals	Donor characteristics: Not mentioned. Donor antibody levels: Neutralization antibody titre > 1:640. Plasma dose: 1 unit of 200 ml plasma, treated with methylene blue photochemistry for pathogen inactivation. Plasma infusion timing: Median 16.5 days (11-20) after symptoms	Improvement in clinical features, laboratory parameters (lymphocyte count, CRP), variable radiological clearing. Clearance of viral shedding in 7/10. Mechanical ventilation and HFNC weaned from 2 and 1 patient, respectively. Overall, 3 discharged, 7 improved. Better outcomes in those treated before day 14. Significant benefit in outcomes as compared to a historical cohort (3 deaths, 6 stable, 1 improved).

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Study	Study design	Convalescent plasma characteristics	Results
Ye <i>et al</i> ⁶³ (Wuhan Huoshenshan Hospital- the newly built dedicated COVID hospital in Wuhan, China) published on April 15, 2020	Study type: Case series n=6 Study population: Mild to moderately ill patients, including one asymptomatic patient. 4/6 requiring supplemental oxygen, 0/6 in ICU or mechanical ventilator. Control arm: Nil Blinding: Nil Randomization: Nil Other treatments: Antivirals such as arbidol and corticosteroids	Donor characteristics: Donation after at least three weeks of asymptomatic period. Donor antibody levels: Not done. Plasma dose: 1-3 units per patient. Plasma infusion timing: Relatively late as compared to other studies. Median 32 days (32-39) from symptom onset.	Symptomatic and radiological improvement in all symptomatic patients. No ICU admission, No deaths. Three patients were PCR negative seen before plasma infusion, decision to infuse because of persistent symptoms and radiological findings, and one patient was asymptomatic PCR-positive patient
Zeng <i>et al</i> ⁶⁴ (Zhengzhou, China) published on April 29, 2020	Study type: Retrospective observational study. n=6 Study population: Critically ill, with respiratory failure, 5 on ventilator, 4 on ECMO. Control arm: Contemporary matched cohort, n=15 Blinding: Nil Randomization: Nil Other treatments: methylprednisolone 4/6, IVIG 5/6, other details not mentioned	Donor characteristics: Asymptomatic for two weeks, rest details not mentioned. Donor antibody levels: positive for IgG, however, no quantification of titre available. Plasma dose: 1-2 units of plasma, median volume 300 ml. Plasma infusion timing: Median 21.5 days (17-23) after diagnosis	100 per cent viral clearance in plasma group versus 21 per cent in comparator group. Mortality of 5/6 and 14/15 in the two groups
Rajendran <i>et al</i> ⁶⁵ , systematic review published on May 1, 2020	Study type: Systematic review of CP studies until April 19, 2020. n=27 participants from 5 studies. Study population: Not applicable. Blinding: None in all 5 studies Randomization: None in all 5 studies. Other treatments: Not applicable.	The review contains details of the CP preparations and dosing used in individual studies	A review of these five uncontrolled case series suggests that CP use may be beneficial in COVID-19. The studies could demonstrate rise in neutralizing antibody titres as well as viral clearance

SARS-CoV-2, severe acute respiratory syndrome coronavirus 2; ICU, intensive care unit; CRP, C-reactive protein; PCR, polymerase chain reaction; CT, computed tomography; PCT, procalcitonin; IVIG, intravenous immunoglobulin; CRRT, continuous renal replacement therapy; HFNC, high flow nasal cannula; SOFA, sequential organ failure assessment

Table VI. Large cohort studies and randomized control trials in COVID-19

S. No.	Study	Study design	Connalescent plasma characteristics	Results
1	Salazar <i>et al</i> ⁴⁴ (Houston, USA) published on May 31, 2020	<p>Study type: Single-arm intervention study. n=25</p> <p>Study population: Severe or life-threatening COVID.</p> <p>12 on mechanical ventilator, 1 on ECMO.</p> <p>Control arm: Nil</p> <p>Blinding: Nil</p> <p>Randomization: Nil</p> <p>Other treatments:</p> <p>Anti-inflammatory (corticosteroids, tocilizumab) in 72 per cent, HCQ + azithromycin in 100 per cent, remdesivir, lopinavir/ritonavir, ribavirin</p>	<p>Donor characteristics: n=9. Age 23-67 yr. All had symptomatic COVID, one requiring hospitalization. Asymptomatic for at least 14 days before donation.</p> <p>Donor antibody levels: ELISA for anti-SARS-CoV-2 antibody titre in CP ranged from 0 to 1350</p> <p>Plasma dose: One transfusion of 300 ml plasma. A single patient received second transfusion after six days.</p> <p>Plasma infusion timing: median 10 days (7.5-12.5 IQR) after symptoms</p>	<p>At day 14, 19 patients had at least 1-point improvement of clinical status (WHO 6-point ordinal scale), 11 were discharged, 3 deteriorated, 1 death. No AE reported.</p> <p>One patient had skin rash and three patients had thrombotic events during follow up.</p> <p>No clear correlation between ELISA IgG titre and outcomes. On long-term follow up, 20 discharged, only two remained intubated, none on ECMO. Of the 26 units of CP transfused, 5 units were of low titre (arbitrarily defined as <150 with 2 units below 50) whereas 7 were of titres 1350 or more</p>
2	Joyner <i>et al</i> ⁶⁶ (USFDA expanded access programme for convalescent plasma with patients from >2000 centres across USA) published as a pre-print on May 14, 2020	<p>Study type: Single-arm intervention study n=5000</p> <p>Study population: Severe or life-threatening illness, 66 per cent in ICU, 72 per cent in respiratory failure.</p> <p>Control arm: Nil</p> <p>Blinding: Nil</p> <p>Randomization: Nil</p> <p>Other treatments: Not mentioned</p>	<p>Donor characteristics: Not mentioned</p> <p>Donor antibody levels: Not mentioned, (samples archived for antibody titre analysis in future)</p> <p>Plasma dose: 1-2 units (200-500 ml) per recipient</p> <p>Plasma infusion timing: Not mentioned</p>	<p>Within first four hours of infusion, SAE n=36, incidence <1 per cent, mortality 0.3 per cent. 25/36 reported SAE deemed related to transfusion (mortality=4, TACO=7, TRALI=11, severe allergic reaction=3). Only 2/36 SAE deemed definitely related to infusion. 4/15 deaths deemed related to CP (3 possibly related, 1 probably related, none definitely related) seven days mortality whole cohort 14.9 per cent. No report of thrombosis associated SAE.</p>
3	Liu <i>et al</i> ⁶⁶ (New York, USA) published as a pre-print on May 22, 2020	<p>Study type: Single-arm intervention study n=39</p> <p>Study population: Severe or life-threatening illness, 4 on mechanical ventilator</p> <p>Control arm: Cohort of controls identified by propensity score-based matching from a pool of 4152 contemporary patients (n=78)</p> <p>Blinding: Nil</p> <p>Randomization: Nil</p> <p>Other treatments: Corticosteroids, HCQ, antivirals, stem cell therapy, IL6 and IL1 inhibitors</p>	<p>Donor characteristics: Not mentioned</p> <p>Donor antibody levels: Anti-spike antibody titres >1:320</p> <p>Plasma dose: 2 units of approximately 250 ml each</p> <p>Plasma infusion timing: Median four days (1-7) days from admission to transfusion</p>	<p>Significantly improved clinical outcomes as compared with control arm. Significant mortality benefit as compared to control arm in non-intubated patients, but not in intubated patients (HR 0.19). Overall, in median follow up of 11 days (1-28) plasma arm, 71.8 per cent were discharged and 12.8 per cent had died.</p>

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S. No.	Study	Study design	Convallescent plasma characteristics	Results
4	Perotti <i>et al</i> ⁶¹ (Italy) published on May 26, 2020	<p>Study type: Single-arm intervention study n=46</p> <p>Study population: Moderate-to-severe ARDS, with high CRP. 30 on CPAP and 7 on MV.</p> <p>Control arm: Cohort of controls identified by propensity score-based matching from a pool of 4152 contemporary patients (n=78)</p> <p>Blinding: Nil</p> <p>Randomization: Nil</p> <p>Other treatments: Antibiotics, HCQ, anticoagulation</p>	<p>Donor characteristics: Not mentioned</p> <p>Donor antibody levels: Neutralizing antibody titres > 1:80</p> <p>Plasma dose: 1-3 units of CP 250-300 ml each</p> <p>Plasma infusion timing: Mean time of symptom onset 14 days</p>	<p>3 deaths (6.5%) within seven days of CP administration. Mortality in a concurrent cohort with same eligibility criteria, mortality was 30 per cent. 26 out of 30 patients weaned off CPAP in one week and 3 of 7 were extubated. Two patients required ECMO after enrolment. Two serious AEs possibly related to CP. Plasma infusion was interrupted in one case</p>
5	Li <i>et al</i> ⁶³ (Wuhan, China) published on June 3, 2020	<p>Study type: Open-label, multicentre randomized control trial n=103</p> <p>Study population: Severe or life-threatening illness</p> <p>Blinding: Nil</p> <p>Randomization: Yes</p> <p>Other treatments: Antivirals, antibacterials, steroids, IVIG, Chinese herbal medicine</p>	<p>Donor characteristics: Age 18-55 yr, discharged at least two weeks ago, 2 nasopharyngeal swabs negative by PCR.</p> <p>Donor antibody levels: Antibody titres in CP products determined by neutralization antibody titres as well as ELISA targeting IgG antibody against S-RBD domain.</p> <p>Positive correlation between the two assays found. CP units with ELISA titres above 1:640 only were selected for infusion</p> <p>Plasma dose: 4-13 ml/kg plasma given per patient. 96 per cent received single unit of plasma 200 ml</p> <p>Plasma infusion timing: Symptom onset to randomization time, median 30 days (20-39 IQR)</p>	<p>Study stopped after 103 of the planned 200 patients enrolled. Primary outcome of clinical improvement (discharge or improvement in 2 points on a 6-point scale) achieved in 59 per cent of study group versus 43 per cent in control arm (HR 1.4). Significant difference in severe disease cohort (91 vs. 68% HR 2.15 P=0.03) as compared to critically ill (20.7% vs. 24.7% HR 0.88 P=0.83). No difference in day 28 mortality, time to discharge. Early virological clearance in Study group seen. (Neg PCR at 72 h in 87% vs. 37.5%) Two post-transfusion AEs that improved with supportive care</p>
6	Hegerova <i>et al</i> ⁶¹ (Seattle, USA) published on May 19, 2020	<p>Study type: Single-arm intervention study. n=20</p> <p>Study population: Severe and critically ill patients, one-third on mechanical ventilator.</p> <p>Control arm: Age, co-morbidities, WHO and SOFA score-matched control group.</p> <p>Blinding: Nil</p> <p>Randomization: Nil</p> <p>Other treatments: Azithromycin, HCQ. Half of control group received remdesivir</p>	<p>Donor characteristics: 8 donors, all symptomatic with mild illness, not requiring hospitalizations. CP donation after 28 days of symptom-free period</p> <p>Donor antibody levels: 7/8 positive for IgG and high positive in 1 and moderate positive in 3 by EuroImmune[®]</p> <p>Plasma dose: 1 unit of CP</p> <p>Plasma infusion timing: Median four days (1-7) days from admission to transfusion</p>	<p>10 per cent deaths and 25 per cent discharges by day 7. No deaths if CP received within 7 days of hospitalization. In control group, 25 per cent death and 35 per cent discharge at day 7. 20 per cent incidence of VTE in both groups. Significant difference in remdesivir use between two groups.</p>

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S. No.	Study	Study design	Convalescent plasma characteristics	Results
7	Hartman <i>et al</i> ⁶⁸ (University of Wisconsin, USA) published as a pre-print on May 22, 2020	<p>Study type: Single-arm intervention study n=31</p> <p>Study population: Severe or life-threatening illness</p> <p>Blinding: Nil</p> <p>Randomization: Nil</p> <p>Control arm: Nil</p> <p>Other treatments: Not mentioned</p>	<p>Donor characteristics: Not mentioned</p> <p>Donor antibody levels: Not available for screening</p> <p>Plasma dose: Not mentioned</p> <p>Plasma infusion timing: Not mentioned</p>	<p>Four patients died all deaths in life threatening disease cohort. 15 severely ill patients avoided ICU care or mechanical ventilation. 10 patients with life-threatening illness could be extubated. Most patients had significant decrease in respiratory support requirement by day 7 of CP administration. Although this study provides evidence for use of CP even without antibody titre determination, key information regarding CP use such as donor characteristics, dose given are not mentioned</p>
8	Rasheed <i>et al</i> ⁶⁹ (Baghdad, Iraq) published as a pre-print on September 1, 2020	<p>Study type: Parallel arm intervention study n=49 (21 received CP, 28 in observation arm)</p> <p>Study population: COVID-19 patients with hypoxia. Overall critically ill population with >50 per cent on mechanical ventilator</p> <p>Blinding: Nil</p> <p>Randomization: Nil. Patients allotted to study arm based on availability of ABO compatible CP. Exact allocation methodology not mentioned</p> <p>Control arm: Nil</p> <p>Other treatments: Not mentioned</p>	<p>Donor characteristics: Not mentioned</p> <p>Donor antibody levels: Plasma tested by anti SARS-CoV-2 IgG ELISA and those with IgG index >1.25 selected for donation</p> <p>Plasma dose: Not mentioned</p> <p>Plasma infusion timing: Mean duration of symptoms of whole cohort 15.7 days</p>	<p>Mortality in CP group 4.8 per cent versus control arm 28.5 per cent. Around 15 per cent of patients were seropositive on day 0 of enrolment; however, these patients also showed benefit from CP administration. No significant AEs related to CP</p>
9	Gharbharan <i>et al</i> ⁶⁵ (14 centres across Netherlands) published as a pre-print on June 3, 2020	<p>Study type: Open-label, multicentre randomized control trial n=86</p> <p>Study population: At enrolment 13 per cent ICU or mechanically ventilated patients</p> <p>Blinding: Nil</p> <p>Randomization: Yes</p> <p>Other treatments: Not mentioned</p>	<p>Donor characteristics: Detailed analysis of 115 screened donor characteristics and their antibody titres have been described.</p> <p>Donor antibody levels: Neutralizing antibody assay by plaque reduction neutralizing test was performed in 115 potential donors. CP from 9 donors was infused in study arm with median titre of 1:640 (IQR 1:320-1:1280)</p> <p>Plasma dose: Single dose of 300 ml on day of enrolment. Repeat dosing on day 5 in those not showing clinical improvement and still PCR positive.</p> <p>Plasma infusion timing: Mean duration of symptoms before enrolment was 10 days (6-15 IQR)</p>	<p>Study halted pre-maturely after 86 of the target 200 patients enrolled as DSMB had concerns about study design. 53 of the 66 patients tested (80%) patients already had anti-SARS-CoV-2 antibodies at enrolment. Median antibody titres by PRNT of the tested 56 the patients in the study as well as 115 donors screened by neutralizing antibody titres was comparable (1:160 vs. 1:160). N-Abs were detected in 46 with titre >1:20 in 44 of the 56 patients. No difference in mortality, hospital stay, day 15 disease severity in the two arms.</p> <p>Among 9 study-arm patients with day 7 N-Ab testing, administration of CP caused four-fold rise in neutralizing titres. No plasma-related AE or SAE was observed</p>

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S. No.	Study	Study design	Convallescent plasma characteristics	Results
10	Joyner <i>et al</i> ⁴⁸ (US) EAP COVID-19 Plasma Consortium) published as a pre-print on August 12, 2020	Study type: <i>Post hoc</i> analysis of CP recipients under a compassionate use programme with correlation of infused antibody titres with clinical outcomes. n=35,322 CP recipients Study population: High proportion of severely ill patients with 52.3 per cent in ICU and 27.5 per cent on mechanical ventilation Control arm: Nil Blinding: Nil Randomization: Nil	Donor characteristics: Symptom free for 14 days, rest details NA Donor antibody levels: Measured <i>post hoc</i> by Anti SARS-CoV-2 IgG CLIA and quantified by the S/CO ratio. Plasma dose: At least one CP unit infusion subjects infused within three days of diagnosis and 85 per cent within 10 days.	Seven days mortality in patients transfused within three days of diagnosis versus more than four days was 8.7 versus 11.9 ($P<0.001$). Similar findings seen in 30 days mortality. Dose response curve seen with dose of infused IgG titres. This analysis was performed on 3082 patients. Seven days mortality in patients who received CP with IgG titres >18.45 S/CO, between 4.62 and 18.45 S/CO and <4.62 S/CO was 8.9 per cent, 11.6 and 13.7 per cent, respectively ($P=0.048$) Similar findings also seen in 30 days mortality. Overall, the survivors in this cohort received higher volume of CP as compared with the non-survivors.
11	Maor <i>et al</i> ⁵⁶ (Israel) published on August 12, 2020	Study type: Prospective cohort study n=49 Study population: 22 per cent moderate, 78 per cent severely ill with 66 per cent mechanically ventilated. Blinding: Nil Randomization: Nil	Donor characteristics: required two negative nasopharyngeal swabs before donation and 14 days period after last negative swab. Donor antibody levels: Not known at the time of infusion. Antibody titres were determined by commercial ELISA kit manufactured by EuroImmun AG targeting the S1 domain of the spike protein as well as neutralizing antibody titres using PRNT assay. Plasma dose: Two doses of 200 ml CP given 24 h from each other Plasma infusion timing: Median 10 days from PCR positivity.	Median neutralizing antibody titres in CP was 1:160 (IQR 1:160-1:640). There was good correlation between the ELISA and the PRNT assay. In patients receiving CP with antibody level <4 units, 36.7 per cent improved by day 14, and in patients with CP >4, 68.4 per cent improved, a difference that was significant.
12	Salazar <i>et al</i> ⁷⁰ (Houston, USA) published on August 3, 2020	Study type: Single-arm intervention study n=316 Study population: Severe or life-threatening COVID. 12 on mechanical ventilator, 1 on ECMO. Control arm: Propensity score based 251 matched controls from the same institute were compared to 136 CP transfused patients. Blinding: Nil Randomization: Nil Other treatments: Steroids, azithromycin, tocilizumab, remdesivir	Donor characteristics: 18-65 yr old donors, 14 days asymptomatic with one PCR negative before donation. Donor antibody levels: ELISA for anti-SARS-CoV-2 antibody titre targeting Spike protein, developed in house was used. Titre of 1:1350 in this assay. corresponds to 1:160 in neutralization assay. Plasma dose: One to two units of CP administered. Second unit transfused if patient had clinical worsening as per pre-defined criteria and in patients with BMI >30 kg/m ² .	76 per cent patients received single unit CP. >90 per cent of patients received CP units with antibody titre > 1:1350. Significant reduction in mortality within 28 days in CP transfused group as compared to the matched controls. Difference was significant in CP recipients infused within 72 h of admission and with antibody titres >1:1350. This benefit also extended to multiple secondary outcomes. Comparison of this group with CP recipients after 72 h and also CP recipients with lower titres of antibodies yielded significant differences in mortality

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S. No.	Study	Study design	Convalescent plasma characteristics	Results
13	Avendano-Sola <i>et al</i> ⁵⁷ (14 centres across Spain) published as a pre-print on September 1, 2020	Study type: Open-label, multicentre randomized control trial n=81 (38 in CP arm, 43 in SOC arm). Study population: Moderately ill COVID-19 patients with either infiltrates on CXR or hypoxia on room air, within 12 days of onset of illness. Excluded patients with high-flow devices or ventilators Blinding: Nil Randomization: Yes Other treatments: Corticosteroids, remdesivir, tocilizumab, HCQ, lopinavir/ritonavir	Donor characteristics: Complicated with EU requirements for CP donation. Donor antibody levels: Donors had antibody levels >1.1. Neutralization assay was performed by a microneutralization assay using a pseudovirus-based assay. Neutralization assay was performed <i>post hoc</i> . Plasma dose: Single unit of 250 ml CP. Plasma infusion timing: Median duration of symptoms before enrolment was eight days (6-9 IQR)	0 versus 14 per cent patients progressed to death or mechanical ventilation in plasma and control arm. Mortality rates were 0 per cent and 9.3 per cent at day 15 in the plasma as well as control arm. These differences were not significant. The infused CP units had antibody titres of >1:80 with median titres of 1:292 (IQR 1:238-1:451). 49.4 per cent patients were positive for anti-SARS-CoV-2 antibodies at enrolment. 6 SAEs in CP arm, none were related to CP infusion.
14	Ibrahim <i>et al</i> ¹¹ (Connecticut, USA) published as a pre-print September 1, 2020	Study type: Open-label, single-arm, phase II trial n=38 Study population: Severely and critically ill patients Blinding: Nil Randomization: Nil	Donor characteristics: Not mentioned in detail. Donor antibody levels: Donors had antibody levels >1:320. Plasma dose: Two doses of 200 ml plasma infused 1-2 h apart Plasma infusion timing: Mean 12.6 days after symptom onset in severely ill patients and 23.1 days after symptom onset in critically ill group	Overall mortality of 37 per cent in the whole group. Mortality in severe group was 13 per cent versus in critical group was 55 per cent. There was a difference in secondary outcomes such as progression of ARDS and hospital stay in both the groups
15	Agarwal <i>et al</i> ⁹ (39 centres across India) published on October 22, 2020	Study type: Open-label, multicentre randomized control trial n=464 Study population: Moderately ill COVID-19 patients with PaO ₂ /FiO ₂ ratio between 200 and 300 Blinding: Nil Randomization: Yes Other treatments: Corticosteroids, remdesivir, tocilizumab, HCQ, lopinavir/ritonavir	Donor characteristics: Young donors, mean age 34 yr, with pre-dominantly mild illness with median of 41 days post-diagnosis of COVID-19. Donor antibody levels: No antibody quantification was done before use of CP. Quantification by neutralizing antibody titres using micro-neutralization test was performed in the archived plasma samples <i>post hoc</i> . Plasma dose: Two doses of 200 ml plasma infused 24 h apart. Plasma infusion timing: Median duration of symptoms before enrolment was 10 days (6-11 IQR)	CP was not associated with reduction in mortality (13.6% in intervention arm and 14.6% in control arm) or prevention of progression to severe disease. Overall, the quality of CP used was of a poorer antibody titre with median neutralizing antibody titres of 1:40 (IQR 1:30-1:80). CP use arm had benefits in earlier resolution of symptoms, decrease in oxygen requirement and virological clearance. Incidence of AE was 10 events in 235 patients and in 3 patients (1.3%) CP infusion was suspected to be associated with mortality. Subgroup analysis revealed no benefit of CP use in patients given infusion within three days of symptom onset or those given relatively high titre antibodies of >1:80. 83 per cent of study subjects already had detectable Nab titres in serum at the time of enrolment with median of 1:90 (IQR 1:30-1:240)

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S. No.	Study	Study design	Concomitant plasma characteristics	Results
16	Libster <i>et al</i> ² (Multiple centres in Argentina) published as a pre-print on January 6, 2021	Study type: Multicentre randomized double blind, placebo control trial n=160 Study Population: Elderly COVID patients aged >75 yr with mild disease. Blinding: Yes Randomization: Yes	Donor antibody levels: CP units with antibody titres >1:1000 by the COVIDAR ELISA assay. Plasma dose: Single unit of plasma, 250 ml infused. Plasma infusion timing: Within 72 h of diagnosis.	Study interrupted at 76% of recruitment due to logistical issues. 16% of CP and 31% of placebo arm experienced severe disease. There was a dose dependant response with outcomes better in patients with high titre CP use. There was a significantly increased antibody titres on day 2 in the treatment arm as compared with the control arm. Overall suggestive of beneficial effect in this cohort
17	Simonovich <i>et al</i> ³ (12 centres in Argentina) published on November 24, 2020	Study type: Open label, multicentre randomized placebo control trial n=333 (208 in CP arm, 105 in placebo arm) 2:1 Study Population: Mild to moderately ill COVID-19 patients (room air SpO ₂ <93%, or P/F ratio <300 or mSOFA ≥2) patients on mechanical ventilation, multiorgan failure excluded. Blinding: Yes Randomization: Yes; in 2:1 ratio Other treatments: Corticosteroids, anti-viral agents.	Donor antibody levels: Median titre of 1:3200 (IQR 1:800 to 1:3200) by the COVIDAR ELISA assay. The N-Ab titre was available for 125 CP units and the median titre was 1:300 (IQR 1:136 to 1:511). Plasma dose: Single unit of plasma, either from single donor or pooled from two to five donors. Median infusion volume 500 ml. Weight based correction of infused plasma volume was used. Plasma infusion timing: Median duration of symptoms to enrolment was 8 days (5-10 IQR)	No difference in mortality (10.96% in CP arm, 11.43% in control arm); No significant difference in clinical outcomes measured according to the WHO ordinal scale (OR 0.83 with 95% CI 0.52 to 1.35) at day 30. Among 215 patients with baseline antibody titres available, 46.5% were negative and median titre was 1:50. Five FNHTR reactions were noted in the CP group.
18	Ray <i>et al</i> ⁴ (Kolkata, India) published as a pre-print on November 28, 2020	Study type: Open label, single centre randomized placebo control trial n=80 Study Population: Moderate to severely ill COVID-19 patients Blinding: No Randomization: Yes. Other treatments: HCQ, Azithromycin, Ivermectin, Doxycycline, Corticosteroids, Tocilizumab, Remdesivir.	Donor antibody levels: Antibody levels tested by EuroImmun ELISA and a surrogate neutralizing antibody kit. A S/CO ratio >1.5 was taken for selecting donors of CP. Plasma dose: Two units of 200 ml each. Plasma infusion timing: Median duration of admission to enrolment was 4 days.	Significant decrease in the cytokine levels in patients receiving CP. However, there was no clinically significant difference in outcomes in mortality, duration of oxygen requirement or hospital stay. Subgroup analysis revealed reduction in mortality as well as duration of hypoxia in patients aged <67 years.

CP, convalescent plasma; AEs, adverse events; HCQ, hydroxychloroquine; TRALI, transfusion-related acute lung injury; TACO, transfusion-associated circulatory overload; HR, hazard ratio; ARDS, acute respiratory distress syndrome; IQR, interquartile range; SAE, serious adverse event; S/CO, signal to cut off ratio; CXR, chest X-ray; SAE, serious adverse events; CPAP, continuous positive airway pressure; MV, mechanical ventilation; VTE, venous thromboembolism; DSMB, Data and Safety Monitoring Board; PRNT, plaque reduction neutralization test; EU, European Union; SOC, standard of care; IL-6, interleukin-6; CXR, chest X-ray; FNHTR, febrile non hemolytic transfusion reaction

The largest experience of systematic use of CP in COVID-19 comes from the USA. Apart from the ongoing clinical trials and the compassionate use under the Emergency Investigational New Drug (eIND) license from FDA, CP has been made accessible to patients and clinicians under the EAP of the USFDA⁷⁵. Under this programme, more than 80,000 units of CP were infused, and the safety analysis of this programme was mentioned earlier^{56,57}. The group recently published their *post hoc* analysis after determining the NAb titres in 35,322 patients⁴⁸ (Table VI, row 10). The two main findings were that infusion of CP within three days of diagnosis compared to later showed benefit in seven-day mortality (8.7 vs. 11.9%, $P < 0.001$) and that there was a dose-response curve for mortality with respect to infused antibody titres. The patients who received CP units with low, medium and high antibody titres had a mortality of 13.7, 11.6, and 8.9 per cent. The antibody titres were not known at the time of the administration of CP. Hence, the arm with low-titre CP can be considered an internally blinded control arm⁷⁶. Similarly, Israel's compassionate CP use programme has also shown a dose-response curve between titres and mortality in their *post hoc* antibody titre analysis involving 49 patients. Several other single-arm studies have been published, a few with matched control arm with claims of CP use efficacy (Tables V and VI).

This evidence, although compelling, has not been replicated in the initial RCTs on CP. The first RCT on CP therapy in COVID-19 was an open-label RCT from seven centres in Wuhan³³ (Table VI, row 5). It was terminated early due to slow recruitment, with only 103 of the target 200 individuals enrolled. This trial employed high titre CP units and showed a higher rate of clinical improvement in severely ill patients but not in the critically ill. There was earlier symptom resolution and viral clearance in the CP arm. The limitations were the lost statistical power due to early termination, lower dosing than other studies and the long median time of >30 days from symptom onset to plasma infusion³³.

A multicentre open-label RCT from 14 centres from the Netherlands randomized 86 of the planned 462 patients and gave CP to 43³⁵ (Table VI, row 9). CP with median neutralizing titres 1:640 was infused. The cohort was predominantly with mild illness and admitted early in the course of disease with a median time from symptom onset of 10 days. While there was a trend towards mortality benefit in the CP arm (26

vs. 14%), there was no significant outcome difference between the two arms. Moreover, 80 per cent of the enrolled patients had anti-SARS-CoV-2 antibodies in their serum at baseline with titres comparable to those of the 115 convalescent donors. The titres of the infused CP units were higher than these baseline recipient titres, and more than a four-fold rise of titres within a week could be demonstrated amongst CP recipients. The RCT was halted for revision of design since the investigators reasoned that administering CP to already seropositive patients would confer no additional benefit³⁵.

Another RCT from Spain was halted prematurely due to a fall in recruitment³⁷ (Table VI, row 13). The data on 81 patients enrolled who received high titre CP with median titres of 1:292 (IQR 168-882) showed a mortality benefit at day 14 compared to the control arm; however, the number of events was very low.

Most recently, the ICMR-sponsored PLACID trial also published its findings⁴⁹ (Table VI, row 15). This open-label RCT could recruit its target, unlike the earlier RCTs and randomized 462 patients. There was no difference in mortality or progression to severe disease between the plasma and the control arm. A high proportion of participants had anti-SARS-CoV-2 antibodies at the time of enrolment, and their antibody titres were higher than those of CP donors. This trial replicated the real-world use of CP, wherein the antibody titres of CP were not known *a priori*. However, the median antibody titres in the infused CP units were significantly lower than the EAP or the Israel compassionate use data^{36,48}. The median NAb titres of infused plasma in this study were 1:40. Whether this reflects the heterogeneity of antibody responses in the Indian population versus the Western population remains to be seen. This experience established the need to have adequate antibody testing for CP done before its use in any trial setting.

Another important RCT on CP use was published in November 2020 by the PlasmAr group from Argentina⁷³ (Table VI, row 17). They enrolled 333 patients of mild to moderately severe COVID acute respiratory distress syndrome (ARDS). However, no mortality or difference in clinical outcome could be demonstrated between the two groups. The median titre of neutralizing antibody used in the study was 1:300, although the median time of administration was eight days, a relatively later time period in the course of disease. Only 39 patients were enrolled within

72 h of symptoms, a number too small to test the hypothesis of administration of CP within first three days of illness. Conversely another double blind RCT from Argentina by the INFANT-COVID-19 group conducted in elderly patients with mild COVID disease who were given high titre CP⁷². The results showed decreased progression to severe disease in patients who received CP and favoured the use of CP in this group (Table VI, row 16). In particular, all the recipients of CP received the infusion within 72 h of diagnosis. Another smaller RCT conducted in a single centre in Kolkata in 80 patients demonstrated a significant decrease in the hyperinflammatory cytokine response in patients with mild to moderate COVID ARDS⁷⁴ (Table VI, row 18). Although there was no demonstrable clinical benefit in terms of survival, duration of hypoxia or duration of hospital stay, there was some benefit noted in the subgroup of patients aged below 67 years⁷⁴.

CP therapy has been granted the Emergency Use Authorization in the USA on August 23, 2020⁴⁰. The high titre CP units in this authorization have been defined as having antibody titre with signal/cut-off ratio >12 ⁴⁰.

Is there a need for further randomized control trial for convalescent plasma use in COVID-19?

Until now, in the RCTs the infusion of high-quality CP in the requisite number of patients could not be done either due to poor recruitment or poor titres of antibodies. The two RCTs from Argentina^{72,73} which could infuse high antibody titre plasma to the target population showed contrasting results. While there was no benefit in infusion of plasma in moderate to severely ill patients in the PlasmAr study⁷³, there was benefit when it was given in mildly ill elderly patients within three days of diagnosis in INFANT-COVID-19 group⁷². While more than 100,000 units of CP has been used worldwide, there is still not unequivocal evidence for or against CP use. Therefore, there remains a need for further scientifically designed RCTs to answer these questions. There are hypotheses to suggest that even seropositive patients may derive the benefit of CP use by augmentation of natural antibody response, immunomodulatory actions and avoidance of cytokine storm⁷⁷. However, these aspects can be explored once the efficacy of CP's use has been established.

At present, there are over 100 registered trials worldwide investigating CP in COVID-19^{75,78,79}. Apart

from the recently published PLACID trial from India⁴⁹ including a large-scale trial called the PLATINA, which is an open-label RCT of CP use in severe COVID-19 illness, recruiting in 21 centres across Maharashtra (CTRI/2020/06/026123).

Convalescent plasma use in India

If proven beneficial in COVID-19, CP may be a promising treatment option for India. However, efforts will have to be made to make antibody testing available before any large-scale CP use programme in the country, after learning from the PLACID trial experience⁴⁹. Setting up a centralized antibody titre determination system under the aegis of a competent authority may help overcome such problems. Another operational requirement would be to ensure that patients receive CP units as soon as possible, ideally targeting infusions within three days of diagnosis. The scarcity of donors is another major problem. Creating a CP stockpile large enough to treat patients, provide prophylaxis for healthcare workers and provide enough raw material for producing purified products in the future requires a major public health initiative. Motivating eligible donors for multiple sessions of plasmapheresis is necessary to overcome these shortages.

Voluntary blood donation has always been a challenge in India⁸⁰, and these problems are compounded further for CP. Multiple barriers for CP donation exist among donors. The most common reasons for reluctance to donate include fear of visiting a healthcare facility during an epidemic, fear of waning of immunity and risk of reinfection due to CP donation. The imposition of lockdowns and curfews also leads to restricted mobility of donors to visit CP donation sites.

The Drug Controller General of India has approved the drug to be administered only under a trial protocol. The Ministry of Health and Family Welfare (MoHFW) guidelines for the treatment of COVID-19 lists CP as an off-label experimental therapy option⁸¹. These guidelines recommend using CP after the measurement of antibody titres by neutralization assays or anti-S-RBD IgG ELISA, which may not be possible in most centres of the country. For the design of clinical and research protocols guidelines issued by global regulatory bodies such as WHO⁸², International Society of Blood Transfusion (ISBT)^{52,83}, USFDA³⁸ and European Commission³⁹ may be useful.

Social and ethical implications of convalescent plasma in India

CP therapy for COVID-19 presents with its own unique social and ethical challenges. Although unproven in efficacy, there is a demand for CP therapy, especially in critically ill hospitalized patients.

Avoidance of monetary or other coercion is necessary to avoid the exploitation of CP donors. Plasma donation appeals should be only of pro-social altruistic nature and should be completely voluntary. It is necessary to ensure that available plasma is rationed in an unbiased and evidence-based manner. Documentation of outcomes should be mandatory. The formation of a

Table VII. Recommendations for various aspects of convalescent plasma (CP) therapy

Setting of use	CP use to be recommended only under well-designed RCT protocols designed keeping in mind the lessons learnt from the available data
CP donor	Recovered PCR proven COVID-19 patient. Should have had symptomatic illness with at least fever as one symptom. Preferable if has had moderate-to-severe illness. >14 days after resolution of symptoms. Within four months of symptom onset. No documentation of NP swab negativity necessary if >28 days symptom-free. High neutralizing antibody titre or its surrogate serological marker to be ensured before donation. Exact titre remains to be defined, but titre >1:160 should be targeted. CP donation after informed consent, ensuring lack of monetary or other coercion.
Antibody quantification of CP	Ideally with neutralizing antibody assay. In the absence of availability of neutralizing antibody titres, it is reasonable to screen with available serological assays for quantification of anti-SARS-CoV-2 antibodies.
Process of donation	Donation by plasmapheresis. Up to 15 per cent total blood volume per donation. Serial donations possible with gap of seven days between donations with monitoring of haemoglobin, total protein and albumin of the donor.
Ideal patient selection for CP	To be given under institute specific protocol with pre-defined inclusion, exclusion criteria and monitoring protocol. Administration with prophylactic intent or in mild illness to be prioritized over severe and critical illness. Preferably within 7-10 days of disease onset or three days of COVID-19 diagnosis. Preferably administered to seronegative patients. Detailed informed consent to be administered including experimental nature of illness, possible risks, lack of proven efficacy, lack of standardized antibody titre testing. ABO compatible plasma to be infused over 30-60 min with monitoring for infusion reactions.
Dosing of CP	No definite recommendations available based on current data.
RT, real-time; NP, nasopharyngeal swab	

Table VIII. Passive immunization in COVID-19 in India

Initiative	Current status/suggestions for future
Stock piling of CP in India for use in expected future seasonal rebounds of COVID-19	Initiatives such as plasma banks to be established. Such projects need to be set up nationwide. CP donor screening and harvesting should continue even after resolution of current epidemic situation. Serial donations by eligible donors should be encouraged.
Production of anti-SARS-CoV-2 hyper-immune globulin	Hyper-immune globulin production started in Japan and the USA ⁷⁹ . Indian pharmaceutical industry should gear up for the production of plasma products and indigenous hyper-immune globulin.
Production of anti-SARS-CoV-2 serum from non-human sources	Antibodies from animals such as horse ²¹ and llama ⁸⁴ have been shown to have <i>in vitro</i> efficacy against SARS-CoV-2. Exploration of this modality with laboratory as well as clinical studies is needed.
Development of anti-SARS-CoV-2 monoclonal antibodies	Multiple efforts are underway globally. Knowledge of the antibody isotype and target viral epitope which confers best protection can help design better and effective antibodies against SARS-CoV-2.

nationwide registry of CP use may go a long way to address these issues.

Conclusions

CP may be a promising and safe treatment option for COVID-19 and is feasible in the Indian setting. Its efficacy in this setting remains to be unequivocally established. Further research on donor selection, antibody cut-offs, precise indications of use and dosing is required before more widespread CP use becomes possible. Emerging evidence points out the necessity of measuring antibody titres in infused plasma units and selecting high titre units for infusion. The recommendations regarding various aspects of CP therapy are summarized in Table VII. The development of more potent modalities such as hyper-immune globulins would be the next step in enhancing passive immune transfer-based therapeutics for COVID-19 (Table VIII).

Acknowledgment: The graphics for the article were designed by huephoria (<http://huephoria.in/>).

Financial support & sponsorship: None.

Conflicts of Interest: None.

References

1. Worldometer. *Coronavirus: India*. Available from: <https://www.worldometers.info/coronavirus/country/india/>, accessed on January 19, 2021.
2. Bloch EM, Shoham S, Casadevall A, Sachais BS, Shaz B, Winters JL, *et al*. Deployment of convalescent plasma for the prevention and treatment of COVID-19. *J Clin Invest* 2020; *130* : 2757-65.
3. Casadevall A, Scharff MD. Serum therapy revisited: Animal models of infection and development of passive antibody therapy. *Antimicrob Agents Chemother* 1994; *38* : 1695-702.
4. Casadevall A, Dadachova E, Pirofski LA. Passive antibody therapy for infectious diseases. *Nat Rev Microbiol* 2004; *2* : 695-703.
5. Garraud O, Heshmati F, Pozzetto B, Lefrere F, Girot R, Saillol A, *et al*. Plasma therapy against infectious pathogens, as of yesterday, today and tomorrow. *Transfus Clin Biol* 2016; *23* : 39-44.
6. Zhou B, Zhong N, Guan Y. Treatment with convalescent plasma for influenza A (H5N1) infection. *N Engl J Med* 2007; *357* : 1450-1.
7. Hung IF, To KK, Lee CK, Lee KL, Chan K, Yan WW, *et al*. Convalescent plasma treatment reduced mortality in patients with severe pandemic influenza A (H1N1) 2009 virus infection. *Clin Infect Dis* 2011; *52* : 447-56.
8. Cheng Y, Wong R, Soo YO, Wong WS, Lee CK, Ng MH, *et al*. Use of convalescent plasma therapy in SARS patients in Hong Kong. *Eur J Clin Microbiol Infect Dis* 2005; *24* : 44-6.
9. Yeh KM, Chiueh TS, Siu LK, Lin JC, Chan PK, Peng MY, *et al*. Experience of using convalescent plasma for severe acute respiratory syndrome among healthcare workers in a Taiwan hospital. *J Antimicrob Chemother* 2005; *56* : 919-22.
10. Ko JH, Seok H, Cho SY, Ha YE, Baek JY, Kim SH, *et al*. Challenges of convalescent plasma infusion therapy in Middle East respiratory coronavirus infection: A single centre experience. *Antivir Ther* 2018; *23* : 617-22.
11. 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* : 1554-61.
12. Sahr F, Ansumana R, Massaquoi TA, Idriss BR, Sesay FR, Lamin JM, *et al*. Evaluation of convalescent whole blood for treating Ebola Virus Disease in Freetown, Sierra Leone. *J Infect* 2017; *74* : 302-9.
13. van Griensven J, Edwards T, Baize S; Ebola-Tx Consortium. Efficacy of convalescent plasma in relation to dose of Ebola virus antibodies. *N Engl J Med* 2016; *375* : 2307-9.
14. van Griensven J, Edwards T, de Lamballerie X, Semple MG, Gallian P, Baize S, *et al*. Evaluation of convalescent plasma for Ebola virus disease in guinea. *N Engl J Med* 2016; *374* : 33-42.
15. Maiztegui JI, Fernandez NJ, de Damilano AJ. Efficacy of immune plasma in treatment of Argentine haemorrhagic fever and association between treatment and a late neurological syndrome. *Lancet* 1979; *2* : 1216-7.
16. 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* : 80-90.
17. Devasenapathy N, Ye Z, Loeb M, Fang F, Najafabadi BT, Xiao Y, *et al*. Efficacy and safety of convalescent plasma for severe COVID-19 based on evidence in other severe respiratory viral infections: A systematic review and meta-analysis. *CMAJ* 2020; *192* : E745-55.
18. Rojas M, Rodríguez Y, Monsalve DM, Acosta-Ampudia Y, Camacho B, Gallo JE, *et al*. Convalescent plasma in COVID-19: Possible mechanisms of action. *Autoimmun Rev* 2020; *19* : 102554.
19. Marano G, Vaglio S, Pupella S, Facco G, Catalano L, Liunbruno GM, *et al*. Convalescent plasma: New evidence for an old therapeutic tool? *Blood Transfus* 2016; *14* : 152-7.
20. Siracusano G, Pastori C, Lopalco L. Humoral immune responses in COVID-19 patients: A window on the state of the art. *Front Immunol* 2020; *11* : 1049.
21. Zhou P, Yang XL, Wang XG, Hu B, Zhang L, Zhang W, *et al*. A pneumonia outbreak associated with a new coronavirus of probable bat origin. *Nature* 2020; *579* : 270-3.

22. Tian X, Li C, Huang A, Xia S, Lu S, Shi Z, *et al*. Potent binding of 2019 novel coronavirus spike protein by a SARS coronavirus-specific human monoclonal antibody. *Emerg Microbes Infect* 2020; 9 : 382-5.
23. Wu F WA, Liu M, Wang Q, Chen J, Xia S, *et al*. Neutralizing antibody responses to SARS-CoV-2 in a COVID-19 recovered patient cohort and their implications. *medRxiv* 2020. doi: 10.1101/2020033020047365.
24. Okba NM, Müller MA, Li W, Wang C, GeurtsvanKessel CH, Corman VM, *et al*. Severe acute respiratory syndrome coronavirus 2-specific antibody responses in coronavirus disease patients. *Emerg Infect Dis* 2020; 26 : 1478-88.
25. Suthar MS, Zimmerman MG, Kauffman RC, Mantus G, Linderman SL, Hudson WH, *et al*. Rapid generation of neutralizing antibody responses in COVID-19 patients. *Cell Rep Med* 2020; 1 : 100040.
26. Prompetchara E, Ketloy C, Palaga T. Immune responses in COVID-19 and potential vaccines: Lessons learned from SARS and MERS epidemic. *Asian Pac J Allergy Immunol* 2020; 38 : 1-9.
27. Zhao J, Yuan Q, Wang H, Liu W, Liao X, Su Y, *et al*. Antibody responses to SARS-CoV-2 in patients of novel coronavirus disease 2019. *Clin Infect Dis* 2020; 28 : ciaa344.
28. Guo L, Ren L, Yang S, Xiao M, Chang D, Yang F, *et al*. Profiling early humoral response to diagnose novel coronavirus disease (COVID-19). *Clin Infect Dis* 2020; 71 : 778-85.
29. Liu W, Fontanet A, Zhang PH, Zhan L, Xin ZT, Baril L, *et al*. Two-year prospective study of the humoral immune response of patients with severe acute respiratory syndrome. *J Infect Dis* 2006; 193 : 792-5.
30. Amanat F, Stadlbauer D, Strohmeier S, Nguyen TH, Chromikova V, McMahon M, *et al*. A serological assay to detect SARS-CoV-2 seroconversion in humans. *Nat Med* 2020; 26 : 1033-6.
31. Hegerova L, Gooley TA, Sweerus KA, Maree C, Bailey N, Bailey M, *et al*. Use of convalescent plasma in hospitalized patients with COVID-19: Case series. *Blood* 2020; 136 : 759-62.
32. GeurtsvanKessel CH, Okba NM, Igloi Z, Embregts CW, Laksono BM, Leijten L, *et al*. Towards the next phase: evaluation of serological assays for diagnostics and exposure assessment. *medRxiv* 2020. doi: 10.1101/2020.04.23.20077156.
33. Li L, Zhang W, Hu Y, Tong X, Zheng S, Yang J, *et al*. Effect of convalescent plasma therapy on time to clinical improvement in patients with severe and life-threatening COVID-19: A randomized clinical trial. *JAMA* 2020; 324 : 460-70.
34. Luchsinger LL, Ransegnola B, Jin D, Muecksch F, Weisblum Y, Bao W, *et al*. Serological assays estimate highly variable SARS-CoV-2 neutralizing antibody activity in recovered COVID19 patients. *J Clin Microbiol* 2020; 58 : e02005-20.
35. Gharbharan A, Jordans CC, GeurtsvanKessel C, den Hollander JG, Karim F, Mollema FP, *et al*. Convalescent plasma for COVID-19. A randomized clinical trial. *medRxiv* 2020. doi: 10.1101/2020.07.01.20139857.
36. Maor Y, Cohen D, Paran N, Israely T, Ezra V, Axelrod O, *et al*. Compassionate use of convalescent plasma for treatment of moderate and severe pneumonia in COVID-19 patients and association with IgG antibody levels in donated plasma. *EClinicalMedicine* 2020; 26 : 100525.
37. Avendano-Sola C, Ramos-Martinez A, Munez-Rubio E, Ruiz-Antoran B, Malo de Molina R, Torres F, *et al*. Convalescent plasma for COVID-19: A multicenter, randomized clinical trial. *medRxiv* 2020. doi: 10.1101/2020.08.26.20182444.
38. U.S. Food and Drug Administration. *Recommendations for Investigational COVID-19 Convalescent Plasma*. Available from: <https://www.fda.gov/vaccines-blood-biologics/investigational-new-drug-ind-or-device-exemption-ide-process-cber/recommendations-investigational-covid-19-convalescent-plasma>, accessed on October 18, 2020.
39. European Commission Directorate-General for Health and Food Safety. *An EU Programme of COVID-19 Convalescent Plasma Collection and Transfusion*. Available from: http://isbtweb.org/fileadmin/user_upload/Deirdre_Fehily_Belgium.pdf, accessed on October 18, 2020.
40. U.S. Food and Drug Administration. Clinical Memorandum. EUA 26382: Emergency Use Authorization (EUA) Request (original request 8/12/20; amended request 8/23/20). Product: COVID-19 Convalescent Plasma. Available from: <https://www.fda.gov/media/141480/download>, accessed on October 18, 2020.
41. Bloch EM, Goel R, Wendel S, Burnouf T, Al-Riyami AZ, Ang AL, *et al*. Guidance for the procurement of COVID-19 convalescent plasma: Differences between high- and low-middle-income countries. *Vox Sang* 2020; 10.1111/vox.12970.
42. Robbiani DF, Gaebler C, Muecksch F, Lorenzi JCC, Wang Z, Cho A, *et al*. Convergent antibody responses to SARS-CoV-2 in convalescent individuals. *Nature* 2020; 584 : 437-42.
43. Duan K, Liu B, Li C, Zhang H, Yu T, Qu J, *et al*. Effectiveness of convalescent plasma therapy in severe COVID-19 patients. *Proc Natl Acad Sci U S A* 2020; 117 : 9490-6.
44. Salazar E, Perez KK, Ashraf M, Chen J, Castillo B, Christensen PA, *et al*. Treatment of coronavirus disease 2019 (COVID-19) patients with convalescent plasma. *Am J Pathol* 2020; 190 : 1680-90.
45. Whitaker B. Inside a Convalescent Plasma Therapy Program Treating Coronavirus Patients. Available from: <https://www.cbsnews.com/news/coronavirus-antibodies-blood-plasma-therapy-60-minutes-2020-05-31/>, accessed on October 18, 2020.
46. Zhang B, Zhou X, Zhu C, Song Y, Feng F, Qiu Y, *et al*. Immune phenotyping based on the neutrophil-to-lymphocyte ratio and IgG level predicts disease severity and outcome for patients with COVID-19. *Front Mol Biosci* 2020; 7 : 157.
47. Long QX, Tang XJ, Shi QL, Li Q, Deng HJ, Yuan J, *et al*. Clinical and immunological assessment of asymptomatic SARS-CoV-2 infections. *Nat Med* 2020; 26 : 1200-4.
48. 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-month experience. *medRxiv* 2020. doi: 10.1101/2020.08.12.20169359.
49. Agarwal A, Mukherjee A, Kumar G, Chatterjee P, Bhatnagar T, Malhotra P, *et al.* Convalescent plasma in the management of moderate COVID-19 in adults in India: open label phase II multicentre randomised controlled trial (PLACID Trial). *BMJ* 2020; *371* : m3939. doi: 10.1136/bmj.m3939.
 50. Ministry of Health and Family Welfare, Government of India. *Drugs and Cosmetics Act 1940 and Rules 1945, Amended.* Available from: https://cdsco.gov.in/opencms/export/sites/CDSCO_WEB/Pdf-documents/acts_rules/2016DrugsandCosmeticsAct1940Rules1945.pdf, accessed on October 18, 2020.
 51. National Blood Transfusion Council. *Guidelines for Blood Donor selection and Blood Donor Referral.* Available from: <http://nbt.naco.gov.in/assets/resources/policy/Letter-reg-%20guidelines-for-blood-donor-selection&referral-2017.pdf>, accessed on October 18, 2020.
 52. Working Party of Global Blood Safety, International Society of Blood Transfusion. Points to Consider in the Preparation and Transfusion of COVID-19 Convalescent Plasma in Low-and Middle-Income Countries. Available from: http://isbtweb.org/fileadmin/user_upload/FINAL_Points_to_consider_in_the_preparation_of_COVID_convalescent_plasma_in_LMIC.pdf, accessed on May 9, 2020.
 53. Joint United Kingdom (UK) Blood Transfusion and Tissue Transplantation Services Professional Advisory Committee Guidelines for the Blood Transfusion Services 3.7: Volume of Donation. Available from: <https://www.transfusionguidelines.org/red-book/chapter-3-care-and-selection-of-whole-blood-and-component-donors-including-donors-of-pre-deposit-autologous-blood/3-7-volume-of-donation>, accessed on October 18, 2020.
 54. National AIDS Control Organisation, Ministry of Health and Family Welfare, Government of India. *Volume of plasma, Plasmapheresis, Apheresis. Standards for Blood banks and Blood Transfusion Services.* Available from: http://nbt.naco.gov.in/assets/resources/sop/Standards_for_Blood_Banks_and_Blood_Transfusion_Services.pdf, accessed on October 18, 2020.
 55. Working Party of Global Blood Safety, International Society of Blood Transfusion. Points to Consider in the Preparation and Transfusion of COVID-19 Convalescent Plasma. Available from: https://isbtweb.org/fileadmin/user_upload/Points_to_consider_in_the_preparation_of_COVID_convalescent_plasma_-_200331_ISBT_WP_GBS_Final.pdf, accessed on May 9, 2020.
 56. Joyner MJ, Wright RS, Fairweather D, Senefeld JW, Bruno KA, Klassen SA, *et al.* Early safety indicators of COVID-19 convalescent plasma in 5000 patients. *J Clin Invest* 2020; *130* : 4791-7.
 57. Joyner MJ, Bruno KA, Klassen SA, Kunze KL, Johnson PW, Lesser ER, *et al.* Safety update: COVID-19 convalescent plasma in 20,000 hospitalized patients. *Mayo Clin Proc* 2020; *95* : 1888-97.
 58. Tetro JA. Is COVID-19 receiving ADE from other coronaviruses? *Microbes Infect* 2020; *22* : 72-3.
 59. Katzelnick LC, Gresh L, Halloran ME, Mercado JC, Kuan G, Gordon A, *et al.* Antibody-dependent enhancement of severe dengue disease in humans. *Science* 2017; *358* : 929-32.
 60. Shen C, Wang Z, Zhao F, Yang Y, Li J, Yuan J, *et al.* Treatment of 5 critically ill patients with COVID-19 with convalescent plasma. *JAMA* 2020; *323* : 1582-9.
 61. Zhang B, Liu S, Tan T, Huang W, Dong Y, Chen L, *et al.* Treatment with convalescent plasma for critically ill patients with severe acute respiratory syndrome coronavirus 2 infection. *Chest* 2020; *158* : e9-13.
 62. Ahn JY, Sohn Y, Lee SH, Cho Y, Hyun JH, Baek YJ, *et al.* Use of convalescent plasma therapy in two COVID-19 patients with acute respiratory distress syndrome in Korea. *J Korean Med Sci* 2020; *35* : e149.
 63. Ye M, Fu D, Ren Y, Wang F, Wang D, Zhang F, *et al.* Treatment with convalescent plasma for COVID-19 patients in Wuhan, China. *J Med Virol* 2020; *92* : 1890-1901. .
 64. Zeng QL, Yu ZJ, Gou JJ, Li GM, Ma SH, Zhang GF, *et al.* Effect of convalescent plasma therapy on viral shedding and survival in patients with coronavirus disease 2019. *J Infect Dis* 2020; *222* : 38-43.
 65. Rajendran K, Krishnasamy N, Rangarajan J, Rathinam J, Natarajan M, Ramachandran A. Convalescent plasma transfusion for the treatment of COVID-19: Systematic review. *J Med Virol* 2020; *92* : 1475-83.
 66. Liu STH, Lin HM, Baine I, Wajnberg A, Gumprecht JP, Rahman F, *et al.* Convalescent plasma treatment of severe COVID-19: A propensity score-matched control study. *Nat Med* 2020; *26* : 1708-13.
 67. Perotti C, Baldanti F, Bruno R, Fante CD, Seminari E, Casari S, *et al.* Mortality reduction in 46 severe COVID-19 patients treated with hyperimmune plasma. A proof of concept single arm multicenter trial. *Haematologica* 2020; *105* : 2834-40.
 68. Hartman W, Hess AS, Connor JP. Hospitalized COVID-19 patients treated with convalescent plasma in a mid-size city in the Midwest. *medRxiv* 2020. doi: 10.1101/2020.06.19.20135830.
 69. Rasheed AM, Fatak DF, Hashim HA, Maulood MF, Kabah KK, Almusawi YA, *et al.* The therapeutic potential of convalescent plasma therapy on treating critically-ill COVID-19 patients residing in respiratory care units in hospitals in Baghdad, Iraq. *Infez Med* 2020; *28* : 357-66.
 70. Salazar E, Christensen PA, Graviss EA, Nguyen DT, Castillo B, Chen J, *et al.* Treatment of coronavirus disease 2019 patients with convalescent plasma reveals a signal of significantly decreased mortality. *Am J Pathol* 2020; *190* : 2290-303.
 71. Ibrahim D, Dulipsingh L, Zapatka L, Eadie R, Crowell R, Williams K, *et al.* Factors associated with good patient outcomes following convalescent plasma in COVID-19: A prospective phase II clinical trial. *Infect Dis Ther* 2020; *9* : 1-14.

72. Libster R, Pérez MG, Wappner D, Coviello S, Bianchi A, Braem V, *et al*. Early high-titer plasma therapy to prevent severe COVID-19 in older adults. *N Engl J Med* 2021. doi: 10.1056/NEJMoa2033700.
73. Simonovich VA, Burgos Pratz LD, Scibona P, Beruto MV, Vallone MG, Vázquez C, *et al*. A randomized trial of convalescent plasma in COVID-19 severe pneumonia. *N Engl J Med* 2020; NEJMoa2031304.
74. Ray Y, Paul SR, Bandopadhyay P, D’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. doi: 10.1101/2020.11.25.20237883.
75. Expanded Access to Convalescent Plasma for the Treatment of Patients with COVID-19. Available from: <https://www.uscovidplasma.org/pdf/COVID-19%20Plasma%20EAP.pdf>, accessed on October 18, 2020.
76. The Exposure-Control Study. Available from: https://ccpp19.org/healthcare_providers/index.html, accessed on October 18, 2020.
77. Casadevall A, Joyner MJ, Pirofski LA. SARS-CoV-2 viral load and antibody responses: The case for convalescent plasma therapy. *J Clin Invest* 2020; 130 : 5112-4.
78. National COVID-19 Convalescent Plasma Project: Clinical Trials; 2020. Available from: https://ccpp19.org/healthcare_providers/component_3/index.html, accessed on October 18, 2020.
79. Sheridan C. Convalescent serum lines up as first-choice treatment for coronavirus. *Nat Biotechnol* 2020; 38 : 655-8.
80. Marwaha N. Voluntary blood donation in India: Achievements, expectations and challenges. *Asian J Transfus Sci* 2015; 9 : S1-2.
81. Ministry of Health and Family Welfare, Government of India. *Clinical Management Protocol: COVID-19*. Available from: <https://www.mohfw.gov.in/pdf/ClinicalManagementProtocolforCOVID19.pdf>, accessed on October 18, 2020.
82. WHO Blood Regulators Network (2017) Position Paper on Use of Convalescent Plasma, Serum or Immune Globulin Concentrates as an Element in Response to an Emerging Virus; 2020. Available from: https://www.who.int/bloodproducts/brn/2017_BRN_PositionPaper_ConvalescentPlasma.pdf?ua=1, accessed on October 18, 2020.
83. Epstein J, Burnouf T. Points to consider in the preparation and transfusion of COVID-19 convalescent plasma. *Vox Sang* 2020; 115 : 485-7.
84. National Institutes of Health. Llama Antibody Engineered to Block Coronavirus; 2020. Available from: <https://www.nih.gov/news-events/nih-research-matters/llama-antibody-engineered-block-coronavirus>, accessed on October 18, 2020.

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