Clinical study using mesenchymal stem cells for the treatment of patients with severe COVID-19

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Abstract   **The coronavirus disease 2019 (COVID-19) caused by SARS-CoV-2 was identified in December 2019. The symptoms include fever, cough, dyspnea, early symptom of sputum, and acute respiratory distress syndrome (ARDS). Mesenchymal stem cell (MSC) therapy is the immediate treatment used for patients with severe cases of COVID-19. Herein, we describe two confirmed cases of COVID-19 in Wuhan to explore the role of MSC in the treatment of COVID-19. MSC transplantation increases the immune indicators (including CD4 and lymphocytes) and decreases the inflammation indicators (interleukin-6 and C-reactive protein). High-flow nasal cannula can be used as an initial support strategy for patients with ARDS. With MSC transplantation, the fraction of inspired O_2 (FiO_2) of the two patients gradually decreased while the oxygen saturation (SaO_2) and partial pressure of oxygen (PO_2) improved. Additionally, the patients' chest computed tomography showed that bilateral lung exudate lesions were adsorbed after MSC infusion. Results indicated that MSC transplantation provides clinical data on the treatment of COVID-19 and may serve as an alternative method for treating COVID-19, particularly in patients with ARDS.**

Keywords   coronavirus disease 2019 (COVID-19); mesenchymal stem cell; acute respiratory distress syndrome; stem cell therapeutics

Introduction

The coronavirus disease 2019 (COVID-19) caused by SARS-CoV-2 was identified in December 2019 [1–3] and is regarded as a highly infectious epidemic disease [4–6]. As of June 6, 2020, the Chinese National Health Commission reported 84 624 confirmed cases and over 4645 deaths. Current epidemiological investigations show that the exposure of many patients can be traced to live seafood and wildlife markets [7,8]. Therefore, the host of SARS-CoV-2 could be a wild animal, such as bats, pangolins, poultry, and snakes [9–11]. Although the current epidemic situation in China has been effectively alleviated, an increasing number of countries continue to experience COVID-19 outbreaks [12,13]. Although controlling and preventing epidemics have been an ongoing effort worldwide, the effective treatment of COVID-19 is currently a top priority.

As the outbreak of COVID-19 was sudden, an effective drug for eliminating SARS-CoV-2 has yet to become available, and multiple organ dysfunction in patients with secondary infections remains a serious problem. Several methods for treating COVID-19, including traditional Chinese medicine and western medicine, have been explored. However, the efforts to control acute respiratory distress syndrome (ARDS) through drugs have yet to yield expected results. An autopsy report indicated the cause of death as ARDS and respiratory failure due to viral infection [14]. Pathological sections also revealed that a

large number of inflammatory factors accumulated in the tissues of the deceased.

Mesenchymal stem cell (MSC)-based therapies have attracted much attention because of their powerful self-renewal capability and pluripotency [15,16]. Currently, MSCs have been widely used in cell therapy, including a large number of basic research and clinical trials [17–19]. Safety and effectiveness have been documented in many clinical trials, especially those dealing with immune-mediated inflammatory diseases, such as graft-versus-host disease and systemic lupus erythematosus [20,21]. One of the main functions of MSCs is immune regulation, which can alleviate the inflammatory response in the body through immunosuppression [22–24]. MSCs possess the ability to alleviate inflammatory response and can thus serve as a potential treatment for patients with COVID-19 [25]. Recently, researchers demonstrated no serious adverse events (including hypoxemia, cardiac arrhythmia, and ventricular tachycardia) after the transplantation of allogeneic MSCs in nine patients with ARDS [26].

Menstrual blood is a special source of MSCs, which were initially identified from menstrual blood in 2007 [27]. MSCs derived from menstrual blood are widely studied because of their abundant source, strong proliferation ability, low immunogenicity, and painless surgery without ethical controversy [28–31]. Thus, menstrual blood-derived MSCs have attracted increasing attention, and they have been broadly applied in basic medicine and clinical trials [32,33]. Our team has also demonstrated that menstrual blood-derived MSC transplantation is an alternative method for treating ARDS with influenza A (H7N9) infection and thereby reducing mortality relative to controls [34]. As revealed in our five-year follow-up, menstrual blood-derived MSC transplantation did not adversely affect the human body. Therefore, menstrual blood-derived MSC transplantation is also relatively safe, although supporting data remain lacking.

The current study is a clinical pilot study that uses menstrual blood-derived MSCs for the treatment of two patients with severe COVID-19 in Wuhan. The primary endpoints mainly include mortality, nucleic acid detection of SARS-CoV-2, length of hospital stay, and oxygenation indicator after MSC infusion. The secondary endpoints mainly include temperature, blood routine examination, liver function, immune indicator, and inflammation indicator. We found that the oxygenation indicators improved, the immune indicators increased, and a number of inflammation indicators decreased after MSC treatment. Chest computed tomography (CT) showed the adsorption of bilateral lung exudate lesions after MSC transplantation. The two case reports provide a promising way to combat COVID-19, especially in emergency and severe cases. As for the function of MSC transplantation in treating COVID-19, it still needs further verification with many clinical samples and comprehensive assessments.

Source and preparation of MSCs

The allogeneic, menstrual blood-derived MSCs were prepared from menstrual blood obtained from a healthy female donor (age = 28 years). The mononuclear cell fraction of the menstrual blood was enriched and tested for nucleated cells, differential, viability, flow cytometry, and sterility prior to seeding for culture. At 70% confluence, MSCs were lifted and passed at a low density into a cell factory. At 70%–80% confluence, the MSCs were washed, harvested, resuspended, and cryopreserved. Karyotyping/G-banding was normal according to a previous study [35]. The cryopreserved MSCs were shipped frozen to the clinical sites in a validated liquid nitrogen dry shipper equipped with a continuous temperature monitoring device. Upon receipt, the cellular product was inspected and stored in a controlled and continuously monitored liquid nitrogen storage tank. Prior to administration, the MSCs were thawed, washed to remove dimethyl sulfoxide, and resuspended in Plasma-Lyte A by the local cell therapy laboratory. The total volume of the MSC infusion was 100 mL regardless of dose. The percent viability of the infused MSCs was determined by trypan blue exclusion after the MSCs were thawed and prepared for infusion. The viability ranged from 90% to 95%. According to the immunophenotype analysis, the menstrual blood-derived MSCs did not express hematopoietic stem cell markers (e.g., CD34, CD45, and CD133) and HLA-DR, but they expressed classical MSC markers (e.g., CD29, CD73, CD90, and CD105) and other surface molecules (e.g., CD9, CD44, and HLA-ABC). The surface maker and three-line differentiation of the MSCs was conducted in accordance with previous studies [31,35,36].

Biological measurements

Laboratory tests for blood, oxygenation indicators, inflammation indicators, immune indicators, coagulation function, and nucleic acid detection of SARS-CoV-2 were carried out at the Medical Inspection Department of the Renmin Hospital of Wuhan University, Wuhan, China. The other medical information of the patients was retrieved from their medical records. The following factors that could be correlated with the clinical outcomes of the patients with COVID-19 were evaluated: (1) baseline characteristics with regard to age, clinical symptoms, and underlying conditions; (2) related data from laboratory tests and imaging examinations; (3) combined treatments by basic supporting drug, antiviral treatment, antibiotic therapy, glucocorticoid, mechanical ventilation,

extracorporeal membrane oxygenation, artificial liver support system, and continuous renal replacement therapy. The CONSORT diagram of this clinical trial is shown in Fig. 1.
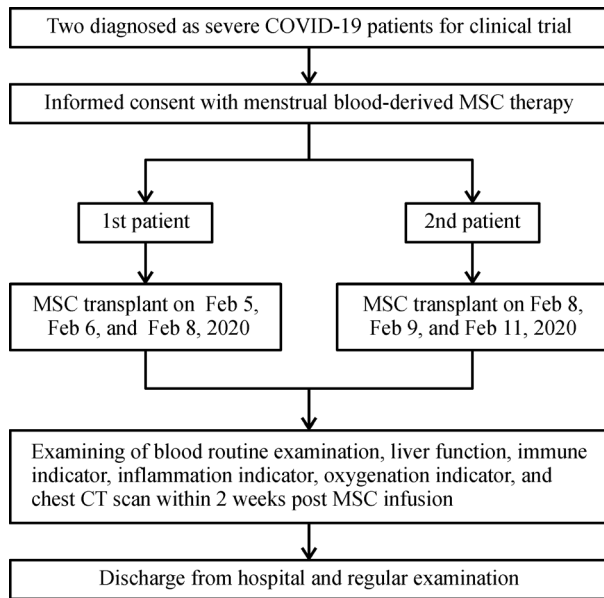

Fig. 1 Detailed CONSORT diagram for the treatment of two patients in our study. The two patients for clinical trial were diagnosed with severe COVID-19. Their informed consent for MSC therapy was obtained accordingly. The first patient received MSC transplant on February 5, February 6, and February 8, 2020. The second patient received MSC transplant on February 8, February 9, and February 11, 2020. They were then subjected to routine blood examination and tests for liver function, immune indicators, inflammation indicators, oxygenation indicators, and chest CT scan within 2 weeks post-MSC infusion. The two patients were then discharged from the hospital and regularly examined.

Treatment for the two patients

The allogeneic MSCs were obtained from a healthy female donor (age = 28 years), who was notified promptly and then subsequently signed a written informed consent. The patients received MSCs. The MSC administration in the patients with COVID-19 was conducted by volunteers, and the participants signed written informed consent forms for the multi-center and open-label clinical trial (No. ChiCTR2000029606). This study was approved by the Ethics Committee of Shulan (Hangzhou) Hospital, Hangzhou, China, and by the Ethics Committee of Renmin Hospital of Wuhan University (No. WDRY2020-K011), Wuhan, China.

Case 1

A 37-year-old woman presented with fever for 9 days and dyspnea for 4 days and was admitted to the Renmin Hospital of Wuhan University on January 31, 2020. The patient had a history of hypertension for 5 years and had been living in Wuhan for a long time. Physical examination showed fever with a temperature of 38.7 °C, breathing rate of 24 times per minute, and pulse of 118 times per minute. The oxygen saturation (SaO_2) was 98%, and the partial pressure of oxygen (PO_2) was 99 mmHg on the basis of 100% fraction of inspired O_2 (FiO_2). The results of the laboratory examinations (Table 1) showed increased leukocyte count (13.6×10^9/L), increased neutrophils (95.0%), decreased lymphocytes (4.1%), and decreased hemoglobin (72.0 g/L). On February 4, before MSC treatment, the inflammatory indicators indicated elevated C-reactive protein (CRP, 42.5 mg/L) and increased interleukin-6 (IL-6, 27.1 pg/mL). The patient was also tested for cellular immunity and humoral immunity (Table 1). The patient was negative for the following common respiratory pathogens: influenza A virus, mycoplasma pneumonia, respiratory syncytial virus, adenovirus, *Chlamydia pneumoniae*, *Legionella pneumophila*, parainfluenza virus, and influenza B virus. She was diagnosed with COVID-19 on the basis of the real-time reverse transcriptase polymerase chain reaction amplification of the viral RNA from a sputum sample.

During hospitalization, oseltamivir, arbidol hydrochloride, lowering of blood pressure, and other symptomatic supportive treatments were given to the patient, but the symptoms of fever and dyspnea did not significantly improve. The infusion was initiated using a standard blood filter tubing set, and the infusion rate was controlled by the investigator on the basis of droplet count. The patient received MSC transplantation through intravenous infusion three times on February 5, 6, and 8, 2020. The injection dose of MSCs was set to 1 million per kg body weight [34]. A multiple intravenous infusion of allogeneic MSCs was well tolerated in the patient with COVID-19.

The symptoms of fever and dyspnea improved (Table 1). SaO_2 was 97%, and PO_2 was 86 mmHg on the basis of 55% FiO_2 on February 10, 2020. The dynamic changes in laboratory examination are shown in Table 1. We found that after the MSC treatment, the lymphocytes increased, the inflammation indicators decreased, and the symptom of dyspnea improved (Table 1). The chest X-ray on February 1 and 4 indicated large, patchy, and high-density lesions in the bilateral lungs, and the costal diaphragm angle was not clear (Fig. 2). The X-ray on February 6 and 10 showed the absorption of the exudate lesions in the bilateral lungs (Fig. 2). The patient's nucleic acid test turned negative. A follow-up on the patient was conducted on February 17, 2020.

Table 1 Routine laboratory test values, pre- and post-admission of MSC in the 1st patient

Variables	Reference range	Jan 31	Feb 4	Feb 5 MSC treatment	Feb 6 MSC treatment	Feb 7	Feb 8 MSC treatment	Feb 9	Feb 10	Feb 11	Feb 14	Feb 17
Temperature (°C)	36.1–37	38.7	37.8	37	37	38	37.2	37.2	36.7	36.8	37.1	37.1
Routine blood examination												
LC ($\times 10^9$/L)	3.5–9.5	13.6	3.2	5	5.3	5.3	4.4	3.1	2.7	3.2	3.5	2.3
Lymphocytes (%)	20–50	4.1	16.9	25.4	13.2	11	13.8	15.4	23.4	21.2	28.9	41.6
Neutrophils (%)	40–75	95	80.3	71.8	84.1	86.7	83	79.2	68.2	69.5	63.2	47.6
Platelets ($\times 10^9$/L)	125–325	319	350	305	323	243	237	278	321	334	347	320
Hb (g/L)	115–150	72	85	87	84	79	77	76	77	81	87	89
Liver function												
ALB (g/L)	40–55	38	35.1	34.9	33.5	32.6	32	31.2	31.3	32.3	33.6	34.7
ALT (U/L)	7–40	18	23	27	25	21	18	19	18	20	25	26
AST (U/L)	13–35	20	22	26	26	19	17	18	16	23	25	22
Immune indicator												
CD4 (%)	33–58	47.7	N/A	55.2	51.5	N/A	N/A	54.9	58	N/A	N/A	N/A
CD8 (%)	13–39	18.1	N/A	16.3	18.5	N/A	N/A	16.5	17.1	N/A	N/A	N/A
IgG (g/L)	7–16	N/A	N/A	20	21.5	N/A	N/A	21.9	22	N/A	N/A	N/A
IgM (g/L)	0.4–2.3	N/A	N/A	2.4	2.3	N/A	N/A	1.8	1.8	N/A	N/A	N/A
IgA (g/L)	0.7–4	N/A	N/A	2.5	2.2	N/A	N/A	2	1.9	N/A	N/A	N/A
Inflammation indicator												
CRP (mg/L)	0–10	N/A	42.5	20.3	42.3	103.4	103.4	32.9	37.6	20.7	10.9	5
IL-6 (pg/mL)	<10	N/A	27.1	15.7	N/A	N/A	18	21.5	9.8	5.6	5	2.2
Oxygenation indicator												
FiO_2 (%)	<60	100	100	N/A	55	50	50	60	55	N/A	N/A	N/A
SaO_2 (%)	95–98	98	92	N/A	96	93	98	99	97	N/A	N/A	N/A
PO_2 (mmHg)	80–100	99	66	N/A	85	67	97	131	86	N/A	N/A	N/A

LC, leukocyte count; Hb, hemoglobin; ALB, albumin; ALT, alanine aminotransferase; AST, aspartate aminotransferases; IgG, Immunoglobulin G; IgM, immunoglobulin M; IgA, immunoglobulin A; CRP, C-reactive protein; IL-6: interleukin-6; FiO_2, fraction of inspired O_2; SaO_2, oxygen saturation; PO_2, partial pressure of oxygen; N/A, not available.
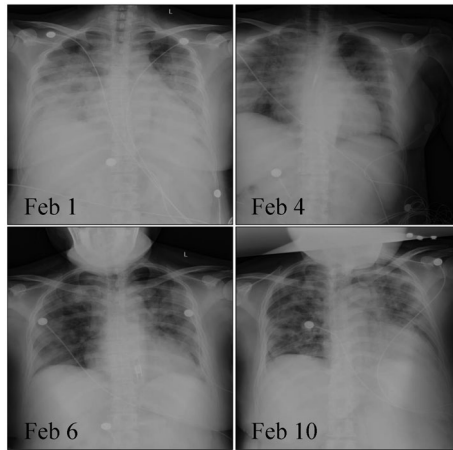
Fig. 2 Chest computerized tomography (CT) images of the first patient. The chest X-ray on February 1 and 4 indicated large, patchy, and high-density lesions in the bilateral lungs, and the costal diaphragm angle was not clear. The X-ray on February 6 and 10 showed the absorption of the exudate lesions in the bilateral lungs.

Case 2

A 71-year-old man living in Wuhan, China was diagnosed at Wuhan Seventh People's Hospital on January 28, 2020. The patient presented with fever for 20 days, dyspnea, cough for 10 days, and a body temperature of 39 °C. Then, he presented with dyspnea and cough for 4 days and was admitted to the Renmin Hospital of Wuhan University on February 7, 2020. Physical examination showed dyspnea and cough, a body temperature of 36.5 °C, breathing rate of 34 times per minute, and pulse of 96 times per minute. SaO_2 was 98%, and PO_2 was 99 mmHg on the basis of 80% FiO_2. The laboratory tests showed a decreased leukocyte count (2.6×10^9/L), increased neutrophils (81.3%), and decreased lymphocytes (10.5%) (Table 2). The inflammatory indicators showed elevated CRP (15.5 mg/L) and normal IL-6 (5.1 pg/mL). The real-time fluorescence polymerase chain reaction of the patient's sputum was positive for the SARS-CoV-2 nucleic acid. The chest X-ray indicated patchy and high-density shadows in the lower lung fields and the left middle lung (Fig. 3).

During hospitalization, ribavirin, arbidol hydrochloride, cefoperazone-sulbactam, lowering of blood pressure, and other symptomatic supportive treatments were given to the patient. At the same time, the patient received MSC treatment via intravenous infusion three times on February 8, 9, and 11, 2020. The injection dose of MSCs was determined as 1 million per kg body weight [34]. A multiple intravenous infusion of allogeneic MSCs was well tolerated in the patient with SARS-CoV-2-induced ARDS. SaO_2 was 99%, and PO_2 was 169 mmHg on the basis of 30% oxygen concentration on February 15, 2020. The

dynamic changes in the laboratory examination are shown in Table 2. We found that after the MSC treatment, the lymphocytes increased, the inflammation indicators, especially CRP, decreased, and FiO_2 declined (Table 2). Repeat chest X-ray showed the absorption of high-density exudate in the lower lung fields and left middle lung (Fig. 3). The expression of SARS-CoV-2 was negative according to the test of nucleic acid on February 16 and 19, 2020. A follow-up on the patient was conducted on February 22, 2020.

Discussion

SARS-CoV-2 infections are clinically similar to previous SARS-CoV and MERS-CoV infections [37,38]. The initial symptoms usually include fever, cough, shortness of breath, early symptom of sputum, and ARDS, which leads to lung injuries and pulmonary fibrosis [39,40]. Although about 20%–25% of people infected with MERS-CoV or SARS-CoV have diarrhea, patients with COVID-19 rarely have bowel symptoms [41,42]. This result implies that as long as the breathing difficulties of patients with COVID-19 are addressed quickly, their own immune system may recover on their own or with the aid of immunostimulants; then, they can successfully resist further virus invasion. However, in another study of 99 patients, chest pain, confusion, and nausea and vomiting were found, in addition to previous findings [43]. X-rays or chest CT images with unilateral or bilateral involvement revealed compatibility with viral pneumonia; moreover, bilateral multilobular and subsegmental consolidation areas were observed in the chest in the intensive care unit [1,44]. Further autopsy reports showed a large amount of sputum, which caused severe ARDS, while a pathological examination indicated a good deal of inflammatory factors in the lung tissue of the patient [14]. Therefore, effectively controlling the inflammatory factors would be a key to treating COVID-19. Early studies have reported that MSC transplantation may promote lung repair and regulate the process of inflammation to reduce fibrosis [45,46]. Thus, MSCs have a great potential in terms of their immunomodulatory effects and attenuation of inflammatory responses.

CRP is a typical acute phase serum protein, which rapidly rises in inflammatory response. CRP, especially tumor necrosis factor α (TNFα), IL-6, monocyte chemoattractant protein-1 (MCP-1), and IL-8 secreted by T cells, are highly sensitive biomarkers of inflammation and host response [47]. Several reports have focused on lymphopenia and high levels of CRP in COVID-19 patients [3,6,48]. In our study, we found that after the MSC treatment, the lymphocytes increased, the inflammation indicators (CRP and IL-6) decreased, and the symptom of dyspnea improved after MSC and antiviral treatment. High-flow nasal cannula (HFNC) can be used as an initial

Table 2 Routine laboratory test values, pre- and post-admission of MSC in the 2nd patient

Variables	Reference range	Feb 7	Feb 8 MSC treatment	Feb 9 MSC treatment	Feb 10	Feb 11 MSC treatment	Feb 12	Feb 13	Feb 15	Feb 17	Feb 19	Feb 22
Temperature (°C)	36.1–37	36.5	36.1	36.1	36.3	36.3	N/A	36.6	36.2	N/A	36.5	36.5
Routine blood examination												
LC ($\times 10^9$/L)	3.5–9.5	2.6	5.8	6.6	6.5	5.7	5.4	4.8	4.8	3	4.2	4
Lymphocytes (%)	20–50	10.5	10.4	10.8	11.8	9.3	15.4	24.6	23.3	31.2	28.7	35.5
Neutrophils (%)	40–75	81.3	73.4	82.8	82.7	83.8	75.2	74.8	62.6	53.9	60.8	53.5
Platelets ($\times 10^9$/L)	125–325	273	295	267	247	215	213	181	149	132	99	138
Hb (g/L)	115–150	132	125	126	127	132	133	128	133	123	122	114
Liver function												
ALB (g/L)	40–55	34.6	29.4	32.5	34.1	31.8	31.5	32.7	31.2	33	29.9	30.7
ALT (U/L)	7–40	106	80	60	45	39	45	71	109	88	126	102
AST (U/L)	13–35	73	40	25	23	20	29	48	72	46	65	43
Immune indicator												
CD4 (%)	33–58	N/A	N/A	27.2	N/A	N/A	27.3	N/A	37.9	N/A	44.3	46.4
CD8 (%)	13–39	N/A	N/A	19.7	N/A	N/A	18.4	N/A	16.6	N/A	17.9	16
IgG (g/L)	7–16	18.8	N/A	21.5	N/A	N/A	29.9	N/A	33.4	N/A	34.6	39.8
IgM (g/L)	0.4–2.3	0.61	N/A	0.62	N/A	N/A	0.75	N/A	0.89	N/A	0.91	0.94
IgA (g/L)	0.7–4.0	2.29	N/A	2.16	N/A	N/A	1.9	N/A	1.85	N/A	1.66	1.64
Inflammation indicator												
CRP (mg/L)	0–10	15.5	64.3	28.5	19.7	9.1	6.2	4.1	3.9	6.8	4.6	3.8
IL-6 (pg/mL)	<10	5.1	N/A	1.5	1.5	16.5	3.3	4	14.7	5.9	6.1	10.5
Oxygenation indicator												
FiO$_2$ (%)	<60	80	N/A	76	N/A	44	44	41	30	N/A	34	30
SaO$_2$ (%)	95–98	98	N/A	96	N/A	97	99	99	99	N/A	96	99
PO$_2$ (mmHg)	80–100	99	N/A	81	N/A	101	114	122	169	N/A	195	150

LC, leukocyte count; Hb, hemoglobin; ALB, albumin; ALT, alanine aminotransferase; AST, aspartate aminotransferases; IgG, immunoglobulin G; IgM, immunoglobulin M; IgA, immunoglobulin A; CRP, C-reactive protein; IL-6, interleukin-6; FiO$_2$, fraction of inspired O$_2$; SaO$_2$, oxygen saturation; PO$_2$, partial pressure of oxygen; N/A, not available.
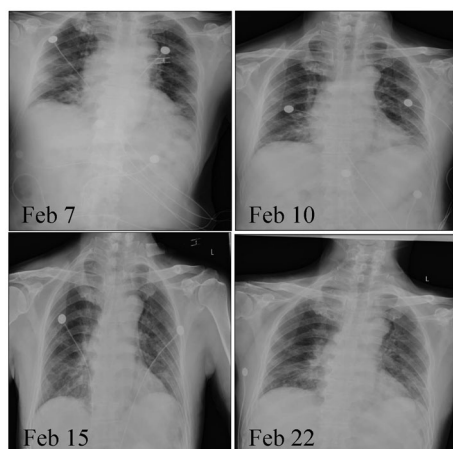
Fig. 3 Chest CT images of second patient. The chest X-ray on February 7 and 10 indicated patchy and high-density shadows in the lower lung fields and the left middle lung. Repeat chest X-ray on February 15 and 22 showed that high-density exudate was absorbed in the lower lung fields and the left middle lung.

support strategy for patients with ARDS. Both patients with symptoms of acute respiratory distress at admission were given HFNC over 50%. With the MSC and antiviral treatment, the FiO_2 of the two patients gradually decreased while SaO_2 and PO_2 returned to normal. Consistent with other studies, the early administration of MSCs significantly reduced the damage of the lung architecture and inflammatory cell infiltration [49]. Thus, MSCs may inhibit the inflammatory effect to relieve the symptoms of COVID-19.

In our previous report, 17 patients received MSC treatment for H7N9 infection [34]; 4 of these patients received a five-year follow-up and did not show evident adverse reactions. From the two reported cases in the current work, we also did not find obvious adverse reactions in our short-term clinical research. However, whether MSC infusion will cause infusion reaction or transient shortness of breath still requires further data and comprehensive research. Recently, Leng et al. published a clinical paper about MSC treatment for COVID-19 patients [48]; they used seven enrolled patients to investigate the inflammatory and immune function of adverse effects for 14 days post MSC transplantation. Then, they found that MSC could significantly remit the functional outcomes of the patients without observed adverse effects. Therefore, we need extensive clinical data to identify whether or not short-term adverse reactions occur after MSC administration. Currently, no evidence proves that MSC is associated with long-term adverse events. Zheng et al. proved no obvious toxicities or seriously adverse events after MSC infusion for 12 patients with moderate to severe ARDS in a single-center, randomized, double-blind, and placebo-controlled trial

[50]. Our previous study also showed that menstrual blood-derived MSCs exerted no obvious side effects according to a five-year follow-up of four MSC patients [34]. Although the tolerability and safety of MSC transplantation in patients lack long-term follow-up, it is still an imperative way to treat COVID-19, especially severe cases.

Zhang and Ma reported that the COVID-19 pandemic had a mild stressful impact on the mental health and quality of life of local Chinese residents in Liaoning Province [51]. Currently, no study has reported the quality of life of COVID-19 patients after hospital discharge. Therefore, we suggest that the quality of life of COVID-19 survivors can be studied in the future to assess the impact of the COVID-19 pandemic.

This clinical trial has several limitations. For example, only 2 patients received MSC treatment; the sample size is thus too small. Therefore, we can neither generalize our phase one experience nor draw conclusions about either the efficacy or long-term safety of MSCs as treatment for COVID-19. As a large proportion of patients live far from Hangzhou City (Wuhan City), the transportation of MSCs is a big issue. In this trial, although the function of MSCs offers a great perspective in treating COVID-19, extensive clinical data are still needed to support their use in future clinical medicine.

Summary

This work offers several clues about MSC-based therapy for combating COVID-19. First, we hope that other types of MSCs (such as menstrual blood-derived MSCs) can be preserved in advance in local cell banks for use in emergencies. Second, the variation tendencies of lymphocytes, CRP, and FiO_2 are the same for both patients in this work; these indicators may be reacted first and should be monitored for future clinical research using MSC therapy. Third, combined with our previous experience and this case study [34], multiple injections (about three times) are appropriate.

Along with our previous studies [34,52], the current work shows that MSCs can improve lung function through anti-inflammatory effects on injured lungs. Although the clinical research of menstrual blood-derived MSCs is still in its infancy, we believe that MSCs will be a promising tool for future clinical medicine.

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Compliance with ethics guidelines

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