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Potential application of mesenchymal stromal cells as a new therapeutic approach in acute respiratory distress syndrome and pulmonary fibrosis

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Abstract

While the COVID-19 outbreak and its complications are still under investigation, post-inflammatory pulmonary fibrosis (PF) has already been described as a long-term sequela of acute respiratory distress syndrome (ARDS) secondary to SARS-CoV2 infection. However, therapeutical strategies for patients with ARDS and PF are still limited and do not significantly extend lifespan. So far, lung transplantation remains the only definitive treatment for end-stage PF. Over the last years, numerous preclinical and clinical studies have shown that allogeneic mesenchymal stromal cells (MSCs) might represent a promising therapeutical approach in several lung disorders, and their potential for ARDS treatment and PF prevention has been investigated during the COVID-19 pandemic. From April 2020 to April 2022, we treated six adult patients with moderate COVID-19-related ARDS in a late proliferative stage with up to two same-dose infusions of third-party allogeneic bone marrow-derived MSCs (BM-MSCs), administered intravenously 15 days apart. No major adverse events were registered. Four patients completed the treatment and reached ICU discharge, while two received only one dose of MSCs due to multiorgan dysfunction syndrome (MODS) and subsequent death. All four survivors showed improved gas exchanges (PaO2/FiO2 ratio > 200), contrary to the others. Furthermore, LDH trends after MSCs significantly differed between survivors and the deceased. Although further investigations and shared protocols are still needed, the safety of MSC therapy has been recurrently shown, and its potential in treating ARDS and preventing PF might represent a new therapeutic strategy.

Keywords Acute respiratory distress syndrome, ARDS, Pulmonary fibrosis, Mesenchymal stromal cells, COVID-19

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To the Editor

Pulmonary fibrosis (PF) is a relatively rare but severe condition characterized by reduced lung compliance and function. Despite having a multifactorial etiology, post-inflammatory PF can be the consequence of severe pulmonary infection and acute respiratory distress syndrome (ARDS). While the COVID-19 outbreak and its complications are still under investigation, ARDS-related PF has already been described as a long-term sequela. However, pharmacologic therapy has been largely ineffective for patients with ARDS, and management mainly focuses on supportive care measures. Likewise, PF has limited treatment options, as currently approved therapies do not significantly extend lifespan. So far, lung transplantation remains the only definitive treatment for end-stage PF, though this option is not always available and is associated with peri-operative high morbidity and mortality and poor long-term survival.

Table 1 Baseline demographic, clinical characteristics, and outcomes of the patients who developed acute respiratory distress syndrome (ARDS) SARS-CoV2-related and who have been enrolled in this study

	Survivors (*N* = 4)	Non-survivors (*N* = 2)	Total (*N* = 6)
Age, years (mean ± SD)	58.2 ± 13.9	71 ± 3.5	62.6 ± 12.9
Sex, N (%)			
Male	3	2	5 (83.33%)
Female	1	0	1 (16.77%)
Body mass index (median, range)	28.5 (26.7–33.9)	26.7 (25.8–27.7)	27.8 (25.8–33.9)
Charlson Comorbidity Index (median)	2	3.5	3
Comorbidities, N (%)			
- Tabagic habit	0 (0%)	0 (0%)	0 (0%)
- Type 2 diabetes mellitus (T2DM)	1 (25%)	0 (0%)	1 (16.66%)
- Arterial hypertension (AH)	2 (50%)	0 (0%)	2 (33.33%)
- Chronic obstructive pulmonary disease (COPD)	0 (0%)	0 (0%)	0 (0%)
- Immunosuppression	0 (0%)	0 (0%)	0 (0%)
SOFA score (median)	5.5	7	6.5
WHO ordinal scale score (median)	4	4	4
PaO_2/FiO_2 ratio (mean ± SD)	135 ± 36.9	89 ± 14.8	119.8 ± 37.6
Crs, cmH_2O (mean ± SD)	28.2 ± 4.7	27.5 ± 10.6	28 ± 6
Days in ICU (mean ± SD)	58.7 ± 30.6	30.5 ± 0.7	49.3 ± 27.8
Days of mechanical ventilation (mean ± SD)	28 ± 18.1	29.5 ± 0.7	28.5 ± 14.1
Steroid treatment, N (%)	100%	100%	100%
Antibiotic treatment, N (%)	100%	100%	100%
Antiviral treatment, N (%)	0%	0%	0%
Hyperimmune plasma treatment, N (%)	75%	50%	66.66%
WBC, 10^9/L (media ± SD)	17.0 ± 10.3	20.2 ± 10	18.7 ± 9.2
Lymphocytes, 10^9/L (mean ± SD)	4.2 ± 5.6	0.8 ± 0.2	3.1 ± 4.7
CRP, mg/dL (mean ± SD)	6.55 ± 4.1	4.8 ± 6.7	5.9 ± 4.4
PCT, ng/mL (mean ± SD)	0.3 ± 0.2	0.3 ± 0.0	0.3 ± 0.1
LDH, U/L (mean ± SD)	670 ± 281	370 ± 117	570 ± 272

SOFA sequential organ failure assessment; PaO_2 partial pressure (arterial) of oxygen; FiO_2 fraction of inspired oxygen; Crs compliance of the respiratory system; ICU intensive care unit; WBC white blood cells; CRP C-reactive protein; PCT procalcitonine; LDH lactate dehydrogenase

Over the last few years, numerous preclinical and clinical studies have shown that advanced therapy medicinal products (ATMP) based on allogeneic mesenchymal stromal cells (MSCs) might represent a promising therapeutical approach in several lung disorders [1–3]. Several data have described the potential of MSCs and their ability to migrate to a site of injury and guide tissue regeneration. Moreover, when intravenously administered, 50–80% of MSCs tend to localize in the lungs with a first-pass effect [4]. Furthermore, since MSCs do not express the two primary human receptors for host-pathogen interaction in SARS-CoV-2 infection [5], their potential for ARDS treatment and PF prevention has been exploited during the COVID-19 pandemic.

From April 2020 to April 2022, we treated six adult patients in mechanical ventilation for moderate COVID-19-related ARDS (median PaO_2/FiO_2 ratio 130, median Crs 26.5 cmH_2O) in a late proliferative stage with third-party allogeneic bone marrow-derived MSCs (BM-MSCs) on a compassionate use basis [6]. The work was approved by the local Ethics Committee, and conducted in accordance with the Declaration of Helsinki.

The patients (1 female and 5 males) had a median age of 65 years (44–76 year) and a median body mass index (BMI) of 27.8 [(25.8–33.9) IQR 3.03] and received up to two same-dose infusions (1×10^6/kg body weight) of BM-MSCs, administered intravenously 15 days apart. They were then monitored and considered for subsequent monthly BM-MSC infusions if signs of PF were observed. All subjects had already been treated with pronation cycles, myorelaxants, dexamethasone, and antibiotics according to international and national guidelines. Moreover, four patients had also received hyperimmune plasma for COVID-19 before BM-MSC's first infusion (median 15 days). The cohort's demographic and clinical characteristics are described in Table 1.

Four patients completed the treatment, while the remaining two received only one dose of MSCs due to a rapid deterioration in their clinical conditions and exitus after the onset of septic shock and multiorgan dysfunction syndrome (MODS). However, the other four patients were successfully discharged from the Intensive Care Unit (ICU) and are still alive at 1-year follow-up. In this regard, several studies have widely discussed mortality as a primary outcome after MSC therapy, but consistent results still need to be provided. However, a decreased

length of hospital stay might be a better indicator of efficacy since it entails improved clinical conditions and reduced mortality [7, 8].

Regarding clinical outcomes, all patients showed improved gas exchange after the first dose of MSCs (Fig. 1), but that was insufficient to hinder the disease progression in those with severe ARDS (P3 and P6). Nevertheless, the other individuals showed an increase in PaO_2/FiO_2 ratio > 200 either after the first (P1 and P4) or the second (P2 and P5) MSC infusion, thus evolving in mild ARDS. However, clear radiological signs of improvement were not detected after the last administration, though survivors showed moderate resolution of bilateral parenchymal damage or lack of deterioration, as reported in the literature [9]. Still, notable lung structure changes may take some time after MSC treatment. Finally, no patients required further monthly BM-MSC infusions.

From the laboratory's perspective, we observed an improvement in lymphocyte numbers in survivors, while patients who died still displayed lower levels after the last dose [10] (Fig. 2). Furthermore, lactate dehydrogenase (LDH) trends after MSCs differed between survivors and the deceased (Fig. 3). High LDH blood levels are a biomarker commonly associated with higher mortality and poor prognosis in several conditions, including ARDS [11]. Recent studies on COVID-19 patients documented a correlation between high levels of LDH and the severity of the disease and intra-hospital mortality [12]. Regarding inflammation markers, C-reactive protein (CRP) trends declined in all cases except one (P3) but did not show any substantial correlation with outcomes (Fig. 4). However, in the literature, the association of this parameter with MSC treatment is controversial since some studies reported no differences in CRP trends between cases and controls, while others documented inferior blood values in MSC recipients [13]. Moreover, some authors have suggested that steroids and hyperimmune plasma may mitigate the anti-inflammatory effect of MCSs [14]. Furthermore, despite initially not being considered subject to rejection, recent studies have indicated that MSCs may be influenced by the host's immune response, particularly in the inflammatory milieu and hypoxia [15].

The literature has extensively documented the pleiotropic anti-inflammatory and regenerative effects of MSCs, and this ATMP likely plays a significant role in ameliorating COVID-19 through immunomodulation. However, it is essential to mention that MSCs may have modest constitutive immune-modulating properties. Some studies have described the potential discrepancies in MSC phenotype induction when exposed to the host's pro-inflammatory mediator [16]. In patients with severe or critical COVID-19, high levels of pro-inflammatory cytokines during MSC treatment may induce a stronger phenotype, even without prior ex vivo priming, while in patients with mild or moderate COVID-19, the immune modulatory phenotype may be less effective. Moreover, age-related quantitative and qualitative changes in the immune system may affect the host's immune response to MSC treatment [17]. Furthermore, the interaction between allogenic and resident MSCs is still under investigation [18].

Finally, no major adverse events were registered after BM-MSC administration, including venous thromboembolism (VTE) or pulmonary embolism (PE). The

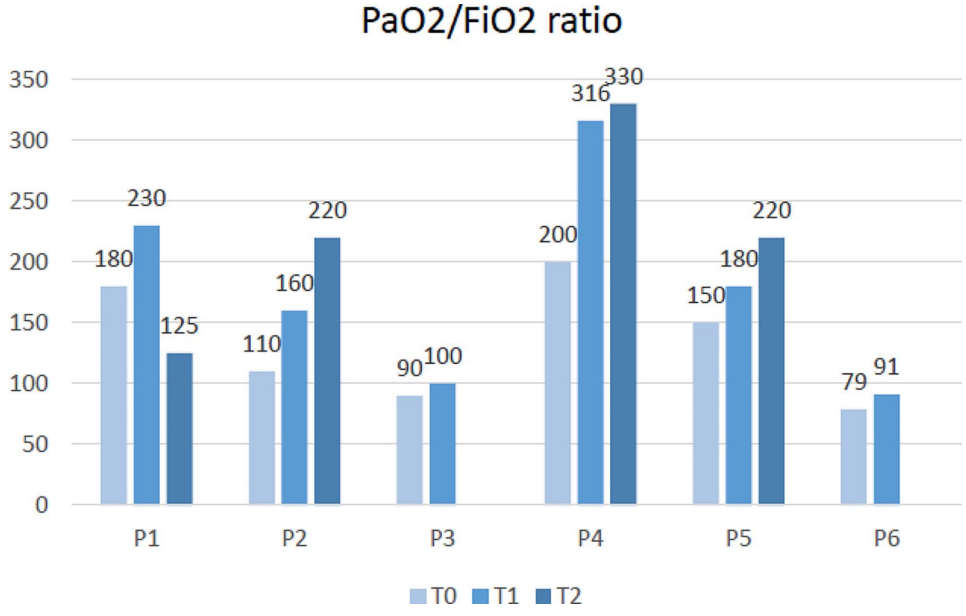

Fig. 1 Trends of PaO_2/FiO_2 ratio of each patient at the time of enrollment (T0), within 24 h after the infusion of the first (T1) and second (T2) dose of BM-MSCs, administrated 15 days apart PaO_2 partial pressure (arterial) of oxygen; FiO_2 fraction of inspired oxygen

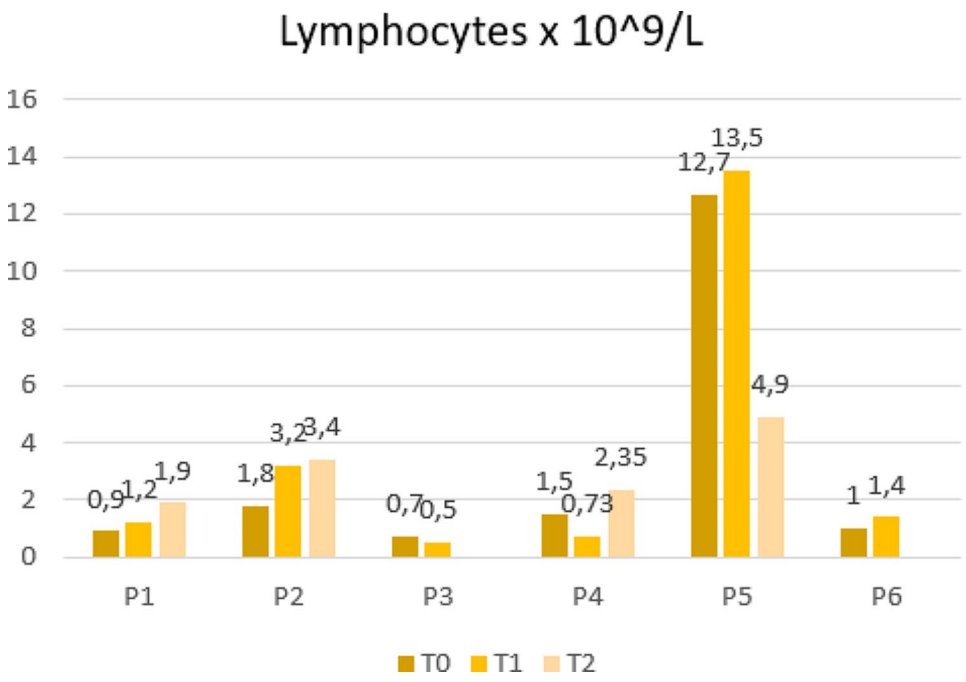

Fig. 2 Trends of lymphocytes' levels of each patient at the time of enrollment (T0), within 24 h after the infusion of the first (T1) and second (T2) dose of BM-MSCs, administrated 15 days apart

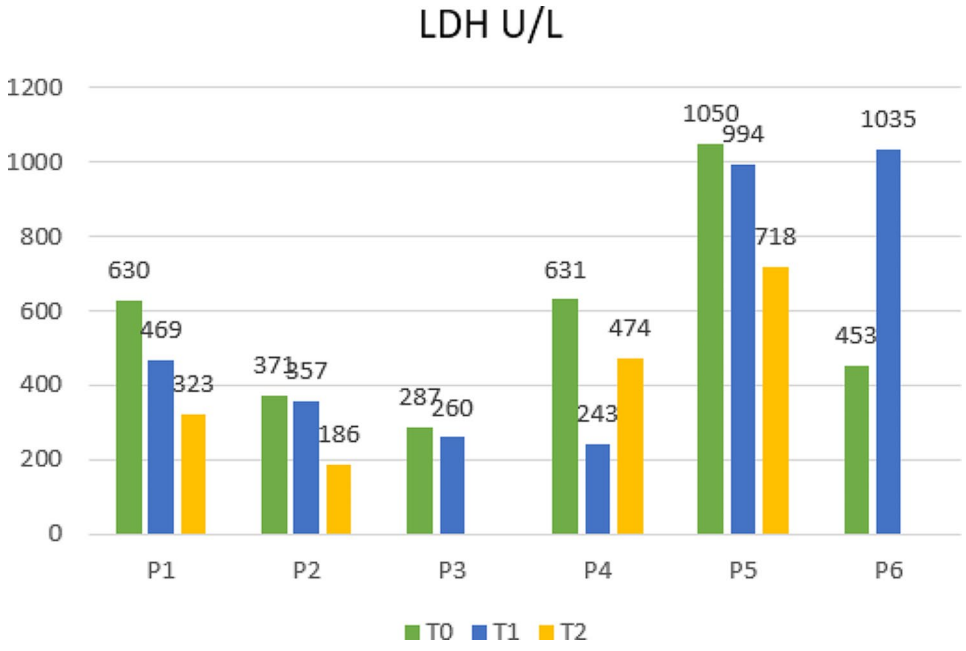

Fig. 3 Trends of lactate dehydrogenase (LDH) levels of each patient at the time of enrollment (T0), within 24 h after the infusion of the first (T1) and second (T2) dose of BM-MSCs, administrated 15 days apart

safety of MSC therapy in COVID-19 patients has already been documented in the literature. A recent randomized study of BM-MSCs versus placebo in early-onset COVID-19-related ARDS reported the absence of infusion-related toxicities and similar serious adverse events over 30 days between the enrolled groups [19]. Data from systematic reviews also described the lack of significant adverse effects after MSC therapy [13] or reported mild adverse events that resolved spontaneously or with minimal supportive treatment in all patients [14].

Although the results of MSC treatment in ARDS and PF are encouraging, additional information from controlled studies should be obtained regarding MSCs source, administration schedule, and dose to design a

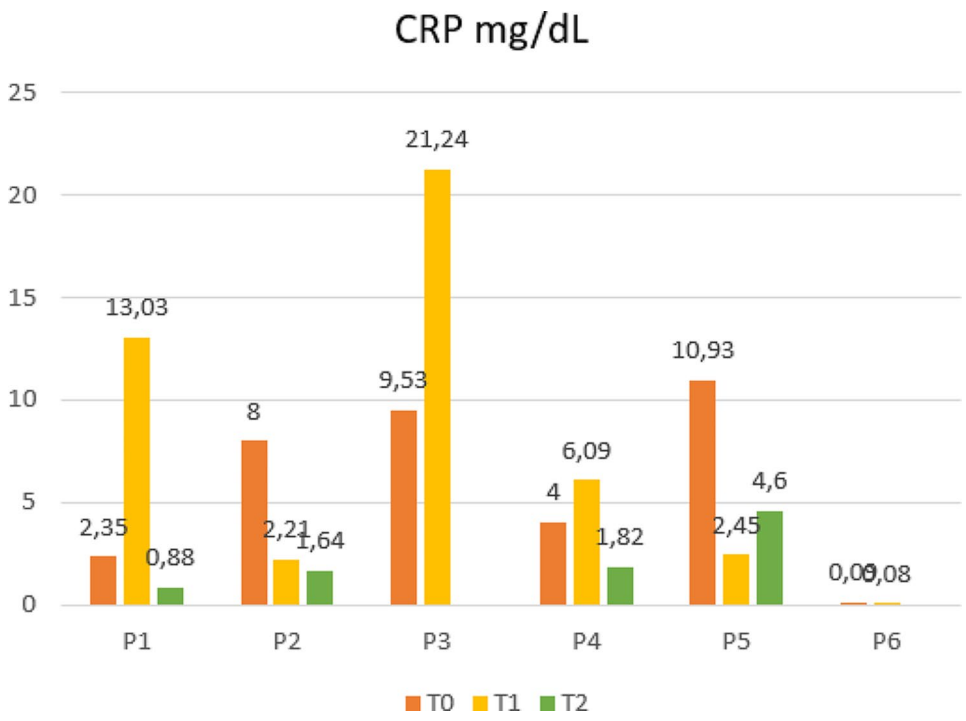

Fig. 4 Trends of C-reactive protein (CRP) levels of each patient at the time of enrollment (T0), within 24 h after the infusion of the first (T1) and second (T2) dose of BM-MSCs, administrated 15 days apart

shared clinical protocol for BM-MSC therapy in early chronic lung injury and prevention of PF secondary to post-infective ARDS. Moreover, a deeper understanding of the interactions between infused third-party allogeneic MSCs and lung-resident cellular populations, including resident MSCs, macrophages, and lymphocytes, might offer valuable insight into the pathogenesis of PF and provide novel therapeutic tools [18, 20, 21]. In conclusion, the dual nature of MSCs in developing and treating post-inflammatory fibrotic diseases represents an inviting challenge for research and might have pivotal applications in clinical settings.

Abbreviations

ARDS	acute respiratory distress syndrome
ATMP	advanced therapy medicinal products
BMI	body mass index
BM-MSCs	bone marrow-derived MSCs
COVID-19	coronavirus disease 19
CRP	C-reactive protein
ICU	intensive care unit
LDH	lactate dehydrogenase
MSCs	mesenchymal stromal cells
MODS	multiorgan dysfunction syndrome
PF	pulmonary fibrosis
PE	pulmonary embolism
SARS-CoV-2	severe acute respiratory syndrome coronavirus 2
VTE	venous thromboembolism

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Not applicable.

Author contributions
Gazzaniga G contributed to this work through analysis and interpretation of data and manuscript writing and editing.Voltini M, Carletti A, Lenta E, Meloni F, Briganti DF, Avanzini MA, Comoli P, and Belliato M contributed to data collection, revision, conceptualization, and supervision.

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Data availability
The datasets used and analysed during the current study are available from the corresponding author on reasonable request.

Declarations

Ethics approval and consent to participate
The work was approved by the local Ethics Committee, and conducted in accordance with the Declaration of Helsinki. Informed consent and consent for publication were acquired retrospectively from each patient or, in case of unconsciousness or death, a substitute (next of kin, when appropriate), according to local laws.

Competing interests
The authors declare no competing interests.

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References

1. Moreira A, Naqvi R, Hall K, Emukah C, Martinez J, Moreira A, Dittmar E, Zoretic S, Evans M, Moses D, Mustafa S. Effects of mesenchymal stromal cell-conditioned media on measures of lung structure and function: a systematic review and meta-analysis of preclinical studies. Stem Cell Res Ther. 2020;11(1):399. https://doi.org/10.1186/s13287-020-01900-7. PMID: 32933584; PMCID: PMC7493362.

2. Matthay MA, Calfee CS, Zhuo H, Thompson BT, Wilson JG, Levitt JE, Rogers AJ, Gotts JE, Wiener-Kronish JP, Bajwa EK, Donahoe MP, McVerry BJ, Ortiz LA, Exline M, Christman JW, Abbott J, Delucchi KL, Caballero L, McMillan M, McKenna DH, Liu KD. Treatment with allogeneic mesenchymal stromal cells for moderate to severe acute respiratory distress syndrome (START study): a randomised phase 2a safety trial. Lancet Respir Med. 2019;7(2):154–62. https://doi.org/10.1016/S2213-2600(18)30418-1. Epub 2018 Nov 16. PMID: 30455077; PMCID: PMC7597675.
3. Cheng W, Zeng Y, Wang D. Stem cell-based therapy for pulmonary fibrosis. Stem Cell Res Ther. 2022;13(1):492. https://doi.org/10.1186/s13287-022-03181-8. PMID: 36195893; PMCID: PMC9530416.
4. Ferrini E, Stellari FF, Franceschi V, Macchi F, Russo L, Murgia A, Grisendi G, Villetti G, Dominici M, Donofrio G. Persistency of mesenchymal Stromal/Stem cells in lungs. Front Cell Dev Biol. 2021;9:709225. https://doi.org/10.3389/fcell.2021.709225. PMID: 34336863; PMCID: PMC8322774.
5. Avanzini MA, Mura M, Percivalle E, Bastaroli F, Croce S, Valsecchi C, Lenta E, Nykjaer G, Cassaniti I, Bagnarino J, Baldanti F, Zecca M, Comoli P, Gnecchi M. Human mesenchymal stromal cells do not express ACE2 and TMPRSS2 and are not permissive to SARS-CoV-2 infection. Stem Cells Transl Med. 2021;10(4):636–42. https://doi.org/10.1002/sctm.20-0385. Epub 2021 Jan 26. PMID: 33188579; PMCID: PMC7753681.
6. Lenta E, Avanzini MA, Belliato M, Zecca M, Croce S, Valsecchi C, Moretta A, Comoli P. 2022. Acute respiratory distress syndrome secondary to Sars-COV-2 infection: treatment with mesenchymal stromal cells (mscs) to prevent pulmonary complications. Journal of Advanced Health Care. vol. 4, no. 2, Apr. 2022. https://doi.org/10.36017/jahc202242196.
7. Kirkham AM, Bailey AJM, Monaghan M, Shorr R, Lalu MM, Fergusson DA, Allan DS. Updated Living Systematic Review and Meta-analysis of controlled trials of mesenchymal stromal cells to treat COVID-19: a Framework for Accelerated Synthesis of Trial evidence for Rapid Approval-FASTER approval. Stem Cells Transl Med. 2022;11(7):675–87. https://doi.org/10.1093/stcltm/szac038. PMID: 35758400; PMCID: PMC9299509.
8. Horwitz LI, Garry K, Prete AM, Sharma S, Mendoza F, Kahan T, Karpel H, Duan E, Hochman KA, Weerahandi H. Six-Month outcomes in patients hospitalized with severe COVID-19. J Gen Intern Med. 2021;36(12):3772–7. https://doi.org/10.1007/s11606-021-07032-9. Epub 2021 Aug 5. PMID: 34355349; PMCID: PMC8341831.
9. Sadeghi B, Ringdén O, Gustafsson B, Castegren M. Mesenchymal stromal cells as treatment for acute respiratory distress syndrome. Case Reports following hematopoietic cell transplantation and a review. Front Immunol. 2022;13:963445. https://doi.org/10.3389/fimmu.2022.963445. PMID: 36426365; PMCID: PMC9680556.
10. Sengupta V, Sengupta S, Lazo A, Woods P, Nolan A, Bremer N. Exosomes derived from bone marrow mesenchymal stem cells as treatment for severe COVID-19. Stem Cells Dev. 2020;29(12):747–54. https://doi.org/10.1089/scd.2020.0080. Epub 2020 May 12. PMID: 32380908; PMCID: PMC7310206.
11. Terpstra ML, Aman J, van Nieuw Amerongen GP, Groeneveld AB. Plasma biomarkers for acute respiratory distress syndrome: a systematic review and meta-analysis*. Crit Care Med. 2014;42(3):691–700. https://doi.org/10.1097/01.ccm.0000435669.60811.24. PMID: 24158164.
12. Fialek B, Pruc M, Smereka J, Jas R, Rahnama-Hezavah M, Denegri A, Szarpak A, Jaguszewski MJ, Peacock FW, Szarpak L. Diagnostic value of lactate dehydrogenase in COVID-19: a systematic review and meta-analysis. Cardiol J. 2022;29(5):751–8. https://doi.org/10.5603/CJ.a2022.0056. Epub 2022 Jun 28. PMID: 35762075; PMCID: PMC9550334.
13. Taufiq H, Shaik Fakiruddin K, Muzaffar U, Lim MN, Rusli S, Kamaluddin NR, Esa E, Abdullah S. Systematic review and meta-analysis of mesenchymal stromal/stem cells as strategical means for the treatment of COVID-19. Ther Adv Respir Dis. 2023 Jan-Dec;17:17534666231158276. https://doi.org/10.1177/17534666231158276. PMID: 37128999; PMCID: PMC10140776.
14. Kirkham AM, Bailey AJM, Shorr R, Lalu MM, Fergusson DA, Allan DS. Systematic review and meta-analysis of randomized controlled trials of mesenchymal stromal cells to treat coronavirus disease 2019: is it too late? Cytotherapy. 2023;25(3):341–52. https://doi.org/10.1016/j.jcyt.2022.10.003. Epub 2022 Oct 13. PMID: 36333234; PMCID: PMC9556962.
15. Kirkham AM, Monaghan M, Bailey AJM, Shorr R, Lalu MM, Fergusson DA, Allan DS. Mesenchymal stem/stromal cell-based therapies for COVID-19: first iteration of a living systematic review and meta-analysis: MSCs and COVID-19. Cytotherapy. 2022;24(6):639–49. Epub 2022 Jan 31. PMID: 35219584; PMCID: PMC8802614.
16. Huang Q, Wu X, Zheng X, Luo S, Xu S, Weng J. Targeting inflammation and cytokine storm in COVID-19. Pharmacol Res. 2020;159:105051. https://doi.org/10.1016/j.phrs.2020.105051. Epub 2020 Jun 27. PMID: 32603772; PMCID: PMC7320704.
17. Bajaj V, Gadi N, Spihlman AP, Wu SC, Choi CH, Moulton VR. Aging, immunity, and COVID-19: how Age influences the host Immune Response to Coronavirus infections? Front Physiol. 2021;11:571416. https://doi.org/10.3389/fphys.2020.571416. PMID: 33510644; PMCID: PMC7835928.
18. Ciavarella C, Pasquinelli G. The dual nature of mesenchymal stem cells (MSCs): Yin and Yang of the inflammatory process, Update on Mesenchymal and Induced Pluripotent Stem cells. IntechOpen Apr. 2020;22. https://doi.org/10.5772/intechopen.82877.
19. Bowdish ME, Barkauskas CE, Overbey JR, Gottlieb RL, Osman K, Duggal A, Marks ME, Hupf J, Fernandes E, Leshnower BG, Golob JL, Iribarne A, Rassias AJ, Moquete EG, O'Sullivan K, Chang HL, Williams JB, Parnia S, Patel NC, Desai ND, Vekstein AM, Hollister BA, Possemato T, Romero C, Hou PC, Burke E, Hayes J, Grossman F, Itescu S, Gillinov M, Pagani FD, O'Gara PT, Mack MJ, Smith PK, Bagiella E, Moskowitz AJ, Gelijns AC. A randomized trial of mesenchymal stromal cells for moderate to severe Acute Respiratory Distress Syndrome from COVID-19. Am J Respir Crit Care Med. 2023;207(3):261–70. https://doi.org/10.1164/rccm.202201-0157OC. PMID: 36099435; PMCID: PMC9896641.
20. Qin L, Liu N, Bao CL, Yang DZ, Ma GX, Yi WH, Xiao GZ, Cao HL. Mesenchymal stem cells in fibrotic diseases-the two sides of the same coin. Acta Pharmacol Sin. 2023;44(2):268–87. https://doi.org/10.1038/s41401-022-00952-0. Epub 2022 Jul 27. PMID: 35896695; PMCID: PMC9326421.
21. Chen L, Qu J, Kalyani FS, Zhang Q, Fan L, Fang Y, Li Y, Xiang C. Mesenchymal stem cell-based treatments for COVID-19: status and future perspectives for clinical applications. Cell Mol Life Sci. 2022;79(3):142. https://doi.org/10.1007/s00018-021-04096-y. PMID: 35187617; PMCID: PMC8858603.

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