ARTICLE

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Mesenchymal stromal cell therapy for COVID-19 acute respiratory distress syndrome: a double-blind randomised controlled trial

María E. Martínez-Muñoz[1,2,7], Concepción Payares-Herrera[2,3,7], Inés Lipperheide[4], Rosa Malo de Molina[5], Isabel Salcedo[1,2], Rosalía Alonso[2], Trinidad Martín-Donaire[1,2], Rocío Sánchez[1,2], Rocío Zafra[1,2], Miguel García-Berciano[1,2], Andrea Trisán-Alonso[5], Manuel Pérez-Torres[4], Antonio Ramos-Martínez[2,6], Piedad Ussetti[2,5], Juan J. Rubio[4], Cristina Avendaño-Solà[2,3] and Rafael F. Duarte[1,2]✉

Mesenchymal stromal cells (MSC) have immunomodulatory and tissue-regenerative properties and have shown promising results in acute respiratory distress syndrome (ARDS) of multiple causes, including COVID-19. We conducted a randomised (1:1), placebo-controlled, double-blind clinical trial to assess the efficacy and safety of one bone marrow-derived MSC infusion in twenty patients with moderate to severe ARDS caused by COVID-19. The primary endpoint (increase in PaO2/FiO2 ratio from baseline to day 7, MSC 83.3 versus placebo 57.6) was not statistically significant, although a clinical improvement at day 7 in the WHO scale was observed in MSC patients (5, 50% vs 0, 0%, p = 0.033). Median time to discontinuation of supplemental oxygen was also shorter in the experimental arm (14 versus 23 days, $p = 0.007$), resulting in a shorter hospital stay (17.5 versus 28 days, $p = 0.042$). No significant differences were observed for other efficacy or safety secondary endpoints. No infusion or treatment-related serious adverse events occurred during the one-year follow-up. This study did not meet the primary endpoint of PaO2/FiO2 increase by day 7, although it suggests that MSC are safe in COVID-19 ARDS and may accelerate patients' clinical recovery and hospital discharge. Larger studies are warranted to elucidate their role in ARDS and other inflammatory lung disorders.

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INTRODUCTION

Acute respiratory distress syndrome (ARDS) induced by SARS-CoV-2 viral pneumonia is the most relevant and severe complication of COVID-19 [1]. ARDS is characterised by a diffuse disruption of the alveolar-epithelial barrier, interstitial edema and inflammatory damage, that may lead to respiratory failure, intensive care unit (ICU) admission, mortality and morbidity with long-term disability in survivors [2, 3]. Despite changes in COVID-19 epidemiology and some improvements in the management of critical patients over time [4, 5], a clinical need remains for safe and novel evidence-based therapies to reduce the inflammatory organ damage that underlies ARDS and accelerate the recovery of functional lung tissue [6, 7].

Mesenchymal stromal cells (MSC) have immunomodulatory, tissue-regenerative and multi-lineage differentiation properties for which they are widely used in cellular therapy [8, 9]. MSC administration is safe, has a predominant pulmonary lodging following intravenous infusion [10] and has shown anti-inflammatory and tissue repairing effects, which may lead to decreased mortality in ARDS of multiple causes [11, 12], including COVID-19 [13]. Here, we report the results of efficacy and long-term safety of bone marrow-derived MSC advanced therapy in a double-blind, randomised, placebo-controlled clinical trial (RCT) in patients with moderate to severe COVID-19 ARDS.

MATERIAL AND METHODS

Trial design

This double-blind, placebo-controlled RCT (COVID-AT; EudraCT 2020-002193-27; NCT04615429) was conducted at Hospital Universitario Puerta de Hierro Majadahonda (HUPHM), Madrid, Spain. It aimed to evaluate the efficacy and safety of allogeneic MSC administration, compared to placebo,

[1]Department of Haematology and GMP Cellular Therapy Unit, Hospital Universitario Puerta de Hierro Majadahonda, Madrid, Spain. [2]Instituto de Investigación Sanitaria Puerta de Hierro Segovia Arana, Madrid, Spain. [3]Department of Clinical Pharmacology, Hospital Universitario Puerta de Hierro Majadahonda, Madrid, Spain. [4]Intensive Care Unit, Hospital Universitario Puerta de Hierro Majadahonda, Madrid, Spain. [5]Department of Pneumology, Hospital Universitario Puerta de Hierro Majadahonda, Madrid, Spain. [6]Department of Internal Medicine and Infectious Diseases, Hospital Universitario Puerta de Hierro Majadahonda, Madrid, Spain. [7]These authors contributed equally: María E. Martínez-Muñoz, Concepción Payares-Herrera. ✉email: rduarte.work@gmail.com

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in patients with moderate to severe ARDS caused by COVID-19. Randomisation sequence, with a 1:1 allocation to either MSC treatment ($n = 10$) or the control group ($n = 10$), was created using Sealed Enveloped software (Sealed Envelope Ltd. 2021, London, UK) and random assignment was performed through a centralized system using REDCap software. Full study protocol has been previously published [14]. The trial was conducted in compliance with the ethical principles of the Declaration of Helsinki and the Good Clinical Practice and the Good Manufacturing Practice guidelines. Regulatory and Ethical approvals were obtained from the Spanish Medicine Agency (AEMPS) and the Research Ethics Committee at HUPHM (approval number 82-20).

Participants

Patients with moderate to severe COVID-19 ARDS (pressure of arterial oxygen to fraction of inspired oxygen ratio, PaO2/FiO2, ≤200 mmHg) [15], were eligible for inclusion within the first 96 h from ARDS onset, and within the first 72 h following orotracheal intubation, if applicable. Informed consent was obtained from all patients or their representatives prior to inclusion (witnessed oral consent, documented in writing). Patients with imminent progression to death, end-stage conditions and those requiring extracorporeal membrane oxygenation (ECMO) or hemodialysis at the time of treatment administration were excluded. Full eligibility criteria are available in the published protocol [14].

Study treatments

Patients in the experimental group received a single intravenous infusion of MSC from healthy allogeneic donor bone marrow (20 mL with approximately 1×10^6 MSC/kg). MSC manufacture details in GMP conditions have been published previously [16, 17], are in accordance with ISCT definitions for MSC (multipotent, phenotypically compliant, plastic adherent cells) [18] and with the Investigational Medicinal Product Dossier (IMPD) approved by the AEMPS (reference code PEI-10-146). MSC were culture-expanded using platelet lysate, collected over 3–4 passages, and cryopreserved until use. The placebo was a single 20 mL dose of phosphate-buffered saline-based solution containing 7.5% dimethyl sulfoxide and 4% human serum albumin (identical to the experimental treatment, without MSC). Packaging, labeling, and distribution were carried out in the same manner for both treatment arms. Products were quickly thawed and infused using a 60mL-syringe and a 3-way connector in approximately 3–5 min within a maximum of 30 min from thawing.

Outcomes

The primary endpoint was the change in the PaO_2/FiO_2 ratio from baseline to day 7 after treatment administration. Key secondary endpoints were 7-day, 14-day, 28-day and 12-month mortality, clinical status according to the World Health Organization (WHO) 7-point ordinal scale (daily until day 14, and on day 28), time until clinical improvement, time to PaO_2/FiO_2 greater than 200 mmHg, duration of treatment with supplemental oxygen, hospitalization and ICU admission duration, incidence of new onset fibrosis, incidence of serious and Grade ≥3 adverse events (AEs) and laboratory markers of inflammation and disease severity (including absolute lymphocyte count, neutrophil to lymphocyte ratio, C-reactive protein, D-dimer, interleukin-6 and lactate dehydrogenase). Full blood counts, coagulation parameters and plasma levels of inflammatory markers were determined in all patients at baseline and days 2, 4, 7, 14 and 28 at HUPHM (Sysmex® XN-series Roche diagnostics, STAR Max® Stago and ADVIA® Chemistry XPT and Centaur XP Siemens). Comprehensive list of outcomes is available in the published protocol [14].

Statistical methods

To detect a difference of 40 units in the PaO_2/FiO_2 ratio change (SD 30), with a two-sided 5% significance level and a power of 80%, a sample size of 10 patients per group was estimated necessary. The main efficacy analysis was planned to be conducted when all patients had completed their 28-day follow-up after treatment, while for safety purposes all patients were followed for a year after treatment. Continuous variables are presented as mean and standard deviation and compared using the Student's T or Wilcoxon-Mann-Whitney test; time-to-event endpoints are presented as median and interquartile range (IQR; Q1, Q3); categorical variables are compared with the Chi-square test or the Fisher exact test. Values of p less than 0.05 are considered statistically significant. The statistical analysis was performed using the scientific software SAS® V9.4 and SAS® Enterprise Guide V7.15.

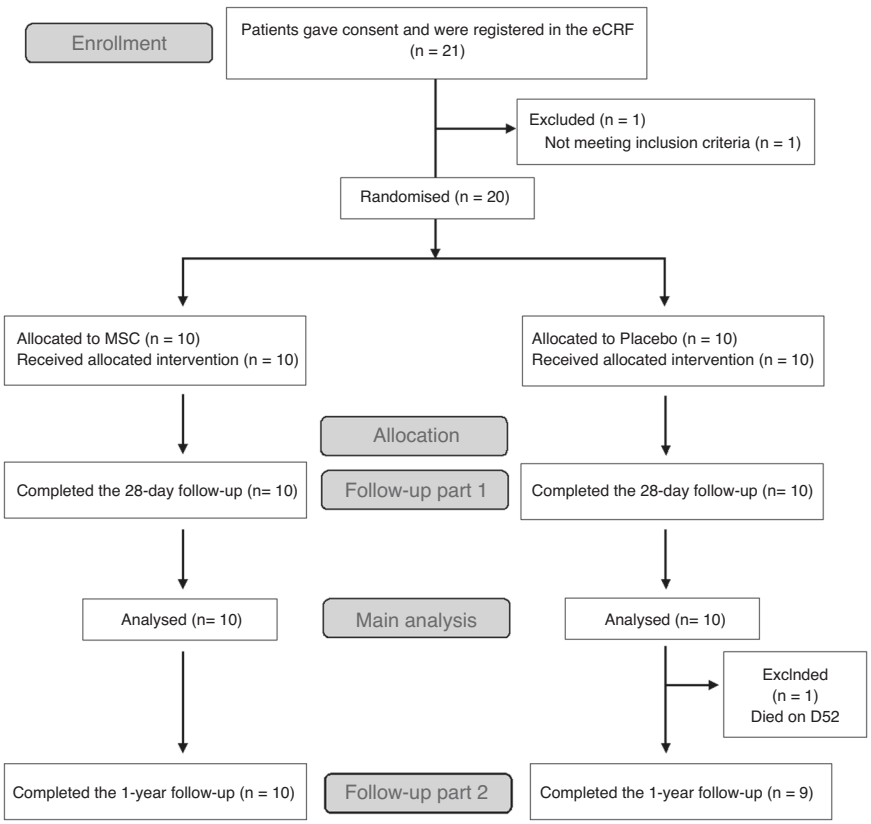

Fig. 1 CONSORT flow diagram COVID-AT study. eCRF electronic case report form, MSC mesenchymal stromal cells.

RESULTS

Recruitment and baseline characteristics

From October 1st to December 4th 2020, within the second wave of the COVID-19 pandemic in Spain, twenty-one patients were enrolled and twenty were randomised (Fig. 1; Table 1 and S1). Thirteen were men (65%). Median age was 63.5 years (range 46–77). Median time from onset of symptoms to randomisation was 11 days (IQR 9–13). The mean baseline PaO_2/FiO_2 ratio was 95.23 ± 39.11, seven patients were on invasive mechanical ventilation (35%; four in the control group and three in the treatment arm) and thirteen were receiving supplemental oxygen therapy via a high-flow nasal cannula (10, 50%), a reservoir mask (2, 10%) or continuous positive airway pressure (1, 5%). Patients were treated with the standards of care at the time, including corticosteroids and low molecular weight heparin in all cases, and tocilizumab in all but one. Only one patient received remdesivir. Subjects in this trial did not receive convalescent plasma or monoclonal antibodies. No statistical differences were observed in the distribution of baseline characteristics between the study arms.

MSC treatment

An average of $82.89 \pm 11.73 \times 10^6$ MSC ($0.99 \pm 0.06 \times 10^6$ per kilogram of body weight) were administered per patient. The post-thaw viability of the infused cells (trypan blue) was greater than 98% in all cases. Median time from thawing to the start of infusion was five minutes (IQR 4–5) and median duration of infusion was four minutes (IQR 4–5), all completed within 30 min after thawing, as planned.

Efficacy outcomes

The increase in PaO2/FiO2 ratio from baseline to day 7 after treatment was not statistically different between the study MSC group and the placebo arm (83.3 ± 67.4 vs 57.6 ± 40.6) in the placebo group ($p = 0.31$). Thus, the primary endpoint of this trial was not reached (Table 2). A greater proportion of subjects in the MSC arm showed a clinical improvement of at least one category of the WHO 7-point ordinal scale at day 7 (5 patients, 50%) than in the control arm (0 subjects, 0%; $p = 0.03$). Overall, by day 28, twelve out of 20 patients (60%), seven in the MSC arm and 5 in the control arm ($p = 0.65$), had a marked clinical improvement with discontinuation of oxygen supplementation (WHO ≤ 3). Such clinical improvement was faster in the MSC arm, with a shorter median time to discontinuation of supplemental oxygen therapy than in those treated with placebo (14 days, IQR 10–18 vs 23 days, IQR 19.5–25; $p < 0.01$). This resulted in a reduced duration of hospitalization in the MSC group (17.5 days [IQR 11–28] vs 28 days [IQR 26–28]; $p = 0.04$). Daily distribution during the initial 28-day

Table 1. Baseline demographic and clinical characteristics.

Characteristic	MSC (*N* = 10)	Placebo (*N* = 10)	All (*N* = 20)
Age — median (range), years	59.5 (47-77)	65.5 (46-75)	63.5 (46–77)
Male sex — no. (%)	5 (50)	8 (80)	13 (65)
Weight — mean ± SD, Kg	82.5 ± 16.0	94.3 ± 18.9	88.4 ± 18.1
Height — mean ± SD, cm[a]	164.2 ± 11.3	171.0 ± 8.1	167.8 ± 10.1
Body Mass Index — mean ± SD, $Kg/m^{2,}$ [a]	29.0 ± 3.1	32.0 ± 4.5	31.5 ± 3.8
Comorbidities — no. (%)			
- Diabetes mellitus	4 (40)	2 (20)	6 (30)
- Obesity	3 (30)	6 (60)	9 (45)
- Cardiovascular disorder (including HBP)	9 (90)	6 (60)	15 (75)
- Chronic lung disease	1 (10)	2 (20)	3 (15)
Concomitant treatments — no. (%)			
- Remdesivir	0 (0)	1 (10)	1 (5)
- Glucocorticoid therapy	10 (100)	10 (100)	20 (100)
- Tocilizumab	10 (100)	9 (90)	19 (95)
- Low molecular weight heparin	10 (100)	10 (100)	20 (100)
PaO2/FiO2 ratio at baseline — mean ± SD	99.5 ± 42.1	91.0 ± 37.6	95.3 ± 39.1
WHO Score (7 points) — no. (%)			
4: Hospitalized, requiring supplemental oxygen by mask or nasal prongs	1 (10)	1 (10)	2 (10)
5: Hospitalized, on non-invasive ventilation or high flow devices	6 (60)	5 (50)	11 (55)
6: Hospitalized, on invasive mechanical ventilation	3 (30)	4 (40)	7 (35)
Laboratory parameters — mean ± SD			
- Lymphocytes (10^3/microL)	0.88 ± 0.59	0.70 ± 0.55	0.79 ± 0.56
- Neutrophil-lymphocyte ratio	18.12 ± 22.46	14.95 ± 6.79	16.54 ± 16.23
- C-reactive protein (mg/L)	98.45 ± 70.46	95.64 ± 111.54	96.89 ± 92.91
- IL-6 (pg/mL)	307.95 ± 380.18	450.22 ± 555.46	375.34 ± 463.38
- D-dimer (ng/mL)	1480.00 ± 2444.40	1360.00 ± 279.68	1420.00 ± 1694.45
- LDH (U/L)	406.33 ± 128.89	356.20 ± 120.11	379.95 ± 123.51
Time from symptom onset to randomization — median (IQR), days	11.0 (9.0, 12.0)	11.5 (9.0, 16.0)	11.0 (9.0, 13.0)

COVID-19 coronavirus disease 2019, *HBP* high blood pressure, *IL-6* interleukin-6, *IQR* interquartile range, *LDH* lactate dehydrogenase, *MSC* mesenchymal stromal cells, *SD* standard deviation, *WHO* world health organization.
[a]Unavailable for one subject in the MSC group.

Table 2. Efficacy outcomes.

Clinical outcomes	MSC ($N = 10$)	Placebo ($N = 10$)	*P* value
Primary outcome			
- Change in the PaO2/FiO2 ratio from day 0 to day 7 — mean ± SD [a]	83.3 ± 67.4	57.6 ± 40.6	0.315
Secondary outcomes			
- Improvement of ≥1 category at WHO 7-point scale — no. (%)			
•At day 7	5 (50)	0 (0)	0.033
•At day 28	9 (90)	8 (80)	1.000
- Time to improvement of ≥1 category at WHO 7-point scale — median [IQR], days			
•All cases	10.5 [7–21]	14.0 [13–21]	0.516
•Only those improving ≥1 category, (no.)	10 [7–18] (9)	13.5 [12–16] (8)	0.352
- Patients that had oxygen therapy withdrawn by day 28 — no. (%)	7 (70)	5 (50)	0.650
- Time to discontinuation of oxygen therapy (WHO ≤3) — median [IQR], days			
•All cases	16 [11–28]	27 [23–28]	0.024
•Only those who discontinued, (no.)	14 [10–18] (7)	23 [20–25] (5)	0.007
- Proportion of patients that were discharged at day 28 — no. (%)	7 (70)	4 (40)	0.370
- Duration of Hospitalization — median [IQR], days	17.5 [11–28]	28 [26–28]	0.042
- Proportion of patients that required ICU admission — no. (%)	5 (50)	8 (80)	0.350
- Duration of ICU admission — median [IQR], days			
•All cases	3 [0-20]	15 [10–17]	0.341
•Only those admitted to ICU, (no.)	17 [9–28] (5)	16 [13–17] (8)	0.764
- Mortality at day 28 — no. (%)	0 (0)	1 (0)	-
- Mortality at 12 months — no. (%)	0 (0)	1 (10)	0.305
- New onset fibrosis at 12 months[b] — no. (%)	4 (50)[c,e]	1 (11.1)[d]	0.08

MSC mesenchymal stromal cells, *ICU* intensive care unit, *IQR* interquartile range, SD standard deviation, *WHO* world health organization.
[a]PaO2/FiO2 ratio values were obtained in supine position.
[b]Based on CT/X-ray imagine and pulmonary function tests.
[c]Data available for 8 of 10 patients.
[d]Data available for 9 of 10 patients.
[e]Patient 01–12 was experiencing an intercurrent event (radiation pneumonitis) at the 12-month visit. A conservative approach was undertaken, and the patient was included as ITT population. In a blinded review, the case was considered to meet the pre-established definition for new onset fibrosis.

period of the subjects' clinical status in both treatment groups is shown in Fig. 2. No statistically significant differences were detected in other efficacy outcomes (Table 2 and supplementary material).

At the end of the 12-month follow-up period, nineteen patients were alive (95%), and seventeen had data available to assess long-term fibrosis (9 in the experimental group and 8 in the placebo group; 85%). All patients had some degree of radiological sequelae in the 12-month imaging tests. Clinically, most cases were mild without a relevant impact (i.e., normal pulmonary function test results at the 12-month visit; 12, 71%) and only five had abnormal pulmonary function test results and new onset fibrosis (29%), with no statistical differences between treatment groups.

Safety outcomes

Eleven patients reported at least one serious or grade ≥3 AE, four patients in the MSC group and seven patients in the placebo group (Table 3). None of the AEs were considered related to the treatment. Regarding AEs of special interest (AESI), no infusion-related AEs were notified. Four patients, two in each treatment group, experienced infections, unrelated to the study treatment, and all patients recovered without sequelae. One patient was diagnosed with a thymoma five months after MSC treatment. This AESI was considered non-related to study treatment, and the patient recovered after surgery and local radiotherapy of the tumor. Only one patient died, in the control group, at day 52, due to complications of an intestinal perforation. After completing the planned 1-year study follow-up, patients continued their care according to clinical practise. No new SAEs, AESI or deaths had been reported up to two years after treatment.

Inflammatory and severity biomarkers

We observed a progressive decline in inflammatory indicators from day 0 to day 14 in both treatment arms (Fig. 3). The recovery of lymphocyte blood counts from baseline to day 14 was significantly higher in the MSC group than in the control arm (1.2 ± 0.9 versus $0.3 \pm 0.6 \times 10^3$/uL, $p = 0.039$). Thus, patients in the MSC group showed higher lymphocyte counts at day 14 (2.09 ± 1.24 versus $1.03 \pm 0.58 \times 10^3$/uL, $p = 0.04$). In addition, they also had lower C-reactive protein level (0.44 ± 0.36 versus 10.07 ± 17.04 mg/L, $p = 0.01$) and neutrophil-lymphocyte ratio (5.22 ± 6.40 versus 14.27 ± 12.04, $p = 0.03$) than patients in the control arm. No statistically significant differences were observed for these measurements at other timepoints, nor for the changes in other parameters such as D-dimer, interleukin-6, lactate dehydrogenase or ferritin.

DISCUSSION

This clinical trial shows encouraging overall outcomes in a patient population with moderate/severe ARDS. During the first 28-day period to assess efficacy from treatment administration no one died, and most patients improved in their respiratory status (85%) and discontinued supplemental oxygen therapy (60%). Beyond center and patient characteristics, these favorable general

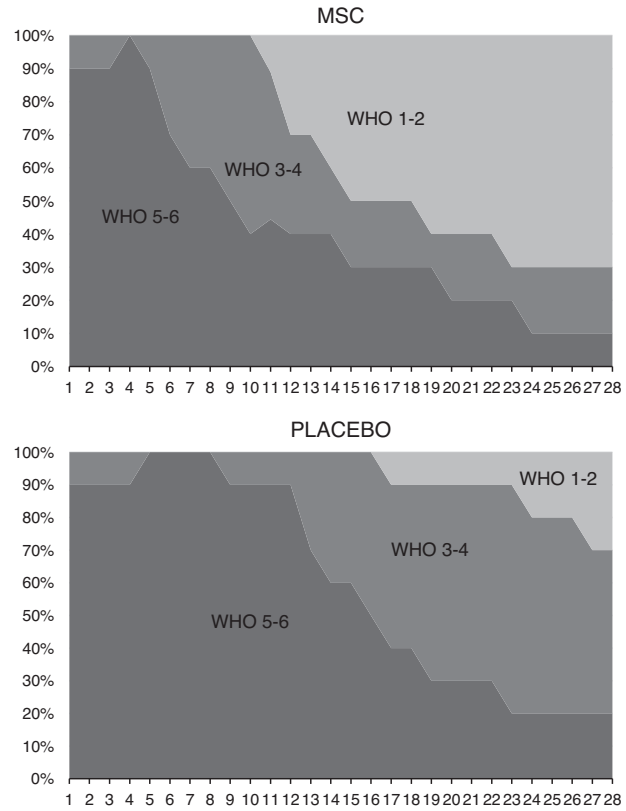

Fig. 2 Evolution of clinical status of patients treated with MSC as compared with placebo. Daily distribution of the clinical status during the initial 28-day efficacy period. Patients are divided in three categories based on the WHO ordinal scale: WHO 1–2 (not hospitalized, whether with or without limitations on activities), WHO 3–4 (hospitalized without supplemental oxygen or with oxygen through nasal prongs or mask), and WHO 5–6 (hospitalized requiring high-flow oxygen devices, non-invasive ventilation, invasive ventilation or ECMO). ECMO: Extracorporeal membrane oxygenation; WHO: world health organization; MSC: mesenchymal stromal cells.

outcomes likely reflect the fact that we carried out this trial during the second wave of the pandemic, at which point the management of COVID-19 ARDS was better established. The study also showed long-term safety of MSC infusion in moderate to severe SARS-CoV-2 ARDS patients.

In terms of efficacy, our double-blind placebo controlled RCT did not meet its primary endpoint, since the increase in PaO2/FiO2 ratio between day 0 and day 7 in patients treated with MSC versus those treated with placebo did not reach statistical significance. Our relatively small sample size might have limited the study's power to show differences between study groups. In particular, considering the rather favorable results in terms of hard outcomes for the whole series described above. The choice of the primary endpoint based on the improvement of the PaO2/FiO2 ratio is probably another limitation. Despite it being a robust test for diagnosis and initial assessment of severity of ARDS, its value to assess response to treatment over time in these patients is limited by multiple factors, including changes in patient position during management and at the time of testing, namely, the impact of pronation/supination on pulmonary circulation and oxygenation, as well as ventilator settings and PEEP [19, 20]. Furthermore, it requires an invasive arterial blood gas technique that is rarely performed in cases with favorable clinical course and outside the ICU. Others have also recently reported that the PaO2/FiO2 ratio may not be a good parameter to assess clinical response, particularly in ARDS patients with oxygen therapy ranging from masks and high-flow devices to invasive mechanical ventilation [21]. Despite not meeting the primary endpoint, more MSC patients improved in at least one category of the WHO 7-point scale as early as day 7 after treatment infusion (50% vs 0%), and they required approximately 10 days less of supplemental oxygen therapy and of hospital stay. Of note, we now know that the improvement in WHO ordinal scale has been proposed as one of the main outcome measurements of clinical response in these patients [22]. Inflammatory markers also improved in both groups, although lymphocyte counts increase from day 0 to day 14, and the neutrophil-lymphocyte ratio, an independent risk factor for in-hospital mortality in COVID-19 [23], were also significantly better in the MSC arm.

MSC have immunomodulatory, antimicrobial, and tissue-regenerative properties and have shown promising results in ARDS of multiple causes [24]. In the context of COVID-19, Leng et al. initially reported the interesting finding that MSC did not express angiotensin-converting enzyme 2 or serine protease TMPRSS2, which are required for SARS-CoV-2 cell entry [25]. Subsequently, case reports, small single-arm series and non-randomized comparative studies provided initial evidence of safety and potential efficacy of MSC in COVID-19 patients [26–29], followed by some RCT [21, 30–36], and several meta-analyses suggesting a reduced risk of mortality and improvement of secondary clinical outcomes in MSC treated patients with severe or critical COVID-19, with no safety issues [22, 37, 38]. All these studies also have limitations and comparability between them may not be clear, due to a wide variety of designs, particularly regarding the endpoints to assess efficacy, and the use of MSC from different sources (e.g., bone marrow, adipose tissue, and umbilical cord) and with different doses and timings. Most of the few RCT published used umbilical cord derived MSC, at higher total doses than in our study, at various time points from the onset of symptoms (1-45 days) and in patients with a wide range of disease severity. These RCT took place primarily during the outset of the pandemic, when mortality rates of COVID-19 were very high. Our trial offers the results of a single infusion of MSC derived from bone marrow, in a defined population of moderate/severe ARDS patients during the second wave in Spain, and with a very low mortality rate. Cases were recruited in a short period of only 9 weeks, which allowed for a homogenous disease management, despite rapidly changing treatment guidelines at the time. Although the primary PaO2/FiO2 endpoint was not met, positive results in some secondary clinical endpoints might be relevant in the current clinical scenario of improved overall outcomes or maybe even in different future threats. It may help future investigations set more adequate primary endpoints regarding ARDS monitoring. These data also contribute to the body evidence that is needed, as the meta-analyses have underscored, to elucidate the actual role of MSC therapy in ARDS and other inflammatory diseases, which is an aim that single small studies might not have enough power to address independently.

Regarding safety, in line with previous studies on the use of MSC in lung disorders, no relevant concerns appeared in our trial. No treatment-related AEs were reported. One single patient died in the placebo group with no relation to the study treatment. Radiological findings in the 12-month long-term evaluation were common, but with low clinical relevance. Five of 17 evaluable patients had new onset fibrosis at 12 months, four in the MSC group and one in the placebo group (Table 2). All but one had required ICU admission during their original ARDS care. One case of fibrosis in the MSC group was associated with radiation pneumonitis caused by radiotherapy of a thymoma (see above). Although no significant differences were observed between treatment arms, these findings highlight the importance of long-term follow-up in patients treated with advanced therapy medicinal products. Only two other RCT on the use of MSC in

Table 3. Safety outcomes.

MedDRA system organ class	Preferred term	MSC ($n = 10$)	Placebo ($n = 10$)	Total ($n = 20$)
Patients with at least one serious adverse event or grade 3–4 adverse event, n (%)		4 (40)	7 (70)	11 (55)
Description of Grade 3–4 Events, n (%)				
Cardiac disorders	Cardiac arrest	0 (0)	1 (10)[a]	1 (5)
	Cardiac infarction	1 (10)	0 (0)	1 (5)
Vascular disorders	Shock	0 (0)	1 (10)	1 (5)
Respiratory, thoracic, and mediastinal disorders	Pneumothorax	0 (0)	1 (10)	1 (5)
	Pulmonary embolism	1 (10)	2 (10)	3 (10)
Gastrointestinal disorders	Intestinal perforation	0 (0)	1 (10)	1 (5)
Infections	Bloodstream infection	1 (10)	1 (10)	2 (10)
	Complicated urinary tract infection	0 (0)	1 (10)	1 (5)
	Urosepsis	0 (0)	1 (10)	1 (5)
Renal disorders	Acute kidney failure	0 (0)	1 (10)	1 (5)
Neoplasm, benign, malignant, and unspecified	Thymoma	1 (10)	0 (0)	1 (5)

MedDRA Medical Dictionary for Regulatory Activities, *MSC* mesenchymal stromal cells.
[a]Due to an endotracheal tube occlusion by secretions; patient recovered with no sequelae.

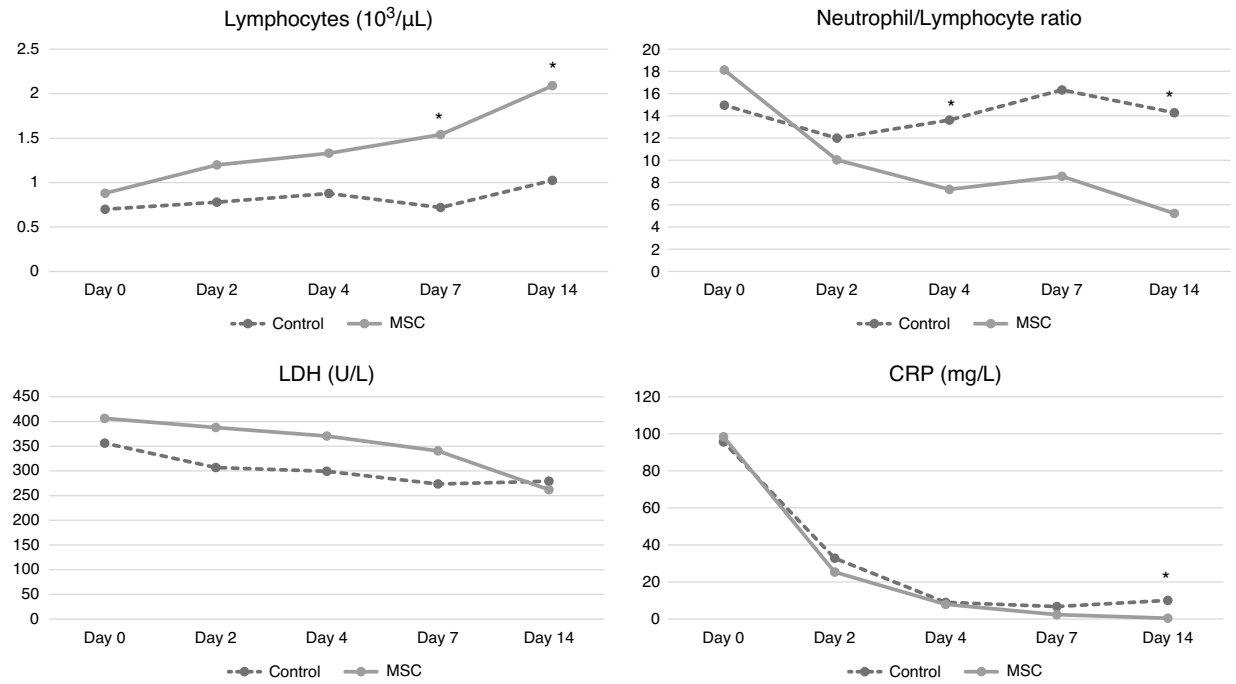

Fig. 3 Inflammatory and severity biomarkers. *$p < 0.05$. CRP C-reactive protein, LDH lactate dehydrogenase.

COVID-19 patients have reported long-term results thus far. Rebelatto et al. [35] found no differences in chest CT abnormalities between the study groups and no pulmonary fibrosis in the 4-month follow-up. In a larger RCT, only 10 of 86 patients had normal CT images at 12 months, but all of these had received MSC treatment [39]. Of note, patients in this study were included rather late after symptom onset (mean 45 days), which might have hindered the possibility to show greater long-term benefit of MSC therapy, despite the relatively large sample size.

Larger RCT and subsequent meta-analyses will enhance our ability to assess efficacy of this cellular therapy in COVID-19 and help elucidate relevant issues such as the most appropriate dosing schedule or MSC source, subgroups of patients that would benefit the most, the potential impact of this therapy in long-term pulmonary fibrosis, and how these findings may translate to patients with ARDS of other causes.

CONCLUSION

MSC therapy did not lead to a significant increase on PaO2/FiO2 by day 7 in this double-blind, placebo-controlled RCT. However, secondary endpoints suggest that MSC are safe, and that, even in a context of low mortality, patients treated with MSC may have a faster clinical recovery with a reduction in the duration of oxygen therapy and hospital length of stay. Further investigation is warranted to improve efficacy assessment of this therapy in ARDS patients of this or other causes, who have otherwise no specific treatment beyond corticosteroids and supportive care therapy.

DATA AVAILABILITY

The de-identified participant data will be available to other researchers, for scientific purposes, upon request to the corresponding author, with approval from the COVID-AT Steering Committee.

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AUTHOR CONTRIBUTIONS

MEMM, CPH, IL, RMM, IS, RA, CAS and RFD: Conception and design. MEMM, CPH, IS, and RA: Collection and/or assembly of data. MEMM, CPH, IS, RA, CAS and RFD: data analysis and interpretation. MEMM, CPH and RFD: manuscript writing. IL, RMM, ATA, MPT, AR, PU and JJR: Provision of study material or patients and clinical follow-up. TMD, RS, RZ, MGB: MSC manufacture. CAS: sponsor's regulatory responsibilities. IL and RFD: funding acquisition. All authors critically reviewed the manuscript and accepted the final version.

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COMPETING INTERESTS

The authors declare no competing interests.

ETHICS APPROVAL AND CONSENT TO PARTICIPATE

This trial adhered to the Declaration of Helsinki and received ethics approval with the title "Ensayo clínico doble ciego, aleatorizado, controlado, para evaluar la eficacia de las células mesenquimales estromales alogénicas en pacientes con síndrome de distrés respiratorio agudo debido a COVID-19 (COVID-AT)", on June 3rd 2020, from the local research ethics committee "Comité de Ética de la Investigación con medicamentos del HUPHM", with approval number 82-20. Informed consent was obtained prior to performing any study procedures from all patients or their representatives (witnessed oral consent, documented in writing).

ADDITIONAL INFORMATION

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Correspondence and requests for materials should be addressed to Rafael F. Duarte.

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