Cell Therapy for Lung Disease: Current Status and Future Prospects

Sara Rolandsson Enes1,2 · Daniel J. Weiss1

Abstract

Purpose of Review Mesenchymal stromal cell (MSC)–based therapies provide a platform for new therapeutic strategies in lung diseases. This review provides an overview of the current status of the field, along with some of the challenges ahead including better understanding of MSC actions in different lung diseases, personalized approaches to select patients most likely to benefit, and the growing problem of stem cell tourism.

Recent Findings A newly evolving concept suggests that MSCs shape their immunomodulatory actions depending on the environment they encounter. Furthermore, in some models, it appears that dying or dead cells may contribute to the therapeutic efficacy by activating the host response.

Summary Despite many pre-clinical studies demonstrating that MSCs can be used to treat lung disorders, clinical trials have failed to show improved outcome. Understanding the complex interaction between MSCs and the host microenvironment is likely to be an important area for enhancing the efficacy of MSC-based cell therapies.

Keywords Mesenchymal stromal cells · MSC · Cell therapy · Microenvironment · Stem cell tourism

Introduction

Respiratory diseases remain a significant cause of morbidity and mortality worldwide, and for many of the severe lung diseases including idiopathic pulmonary fibrosis (IPF), chronic obstructive pulmonary disease (COPD), cystic fibrosis (CF), bronchopulmonary dysplasia (BPD), and acute respiratory distress syndrome (ARDS) there is currently no cure. Therefore, extensive efforts continue for development of new therapeutic strategies. The promising pre-clinical results using mesenchymal stromal cell (MSC)–based cell therapy in experimental lung disease models (reviewed in [1, 2]), in combination with successful clinical trials in other diseases such as graft-versus-host-disease, have resulted in an increased interest and rapidly growing number of clinical trials using MSCs for a range of lung diseases and critical illnesses. MSCs are multipotent cells with immune-modulatory and regenerative properties, and in combination with their low or absent constitutive HLA class I and II expression, they are theoretically ideal candidates to be used in cell-based therapies, (reviewed in [3, 4]). In this review, we summarize the current status of the field of MSC therapies, highlight new discoveries about the role of the microenvironment in control of MSC therapeutic behaviors, and discuss the current challenges that need to be overcome in order to provide clinical benefit in patients with lung diseases.

MSC-Based Therapy in Lung Diseases: Where Do We Stand?

The enthusiasm for using MSCs as a cell-based therapy was raised by encouraging pre-clinical in vivo data leading to the first clinical trial using MSCs as therapy for hematologic malignancies by Lazarus et al. in 1995 [5] and has further translated into clinical trials for several different acute and chronic lung diseases. Searching on the ClinicalTrials.gov database for trials listed through November 27, 2019 using the keywords “lung diseases” and “mesenchymal stromal cell” or “stromal cell” or “mesenchymal stem cell”, or “pulmonary...
disease” and “mesenchymal stromal cell” or “stromal cell” or “mesenchymal stem cell” identified 77 human clinical trials, of which 68 are for patients with lung disease. So far, 19 of the studies have been completed with 11 of them having published results in the PubMed database, 3 are currently active trials, 22 are open and in the process of recruiting their first patient, 7 are not yet recruiting, 13 have unknown status, and 4 of the trials have been withdrawn or terminated [6] (Table 1). In this section, we will summarize and discuss some of the clinical studies that have been completed and highlight a few promising currently ongoing studies.

**Acute Respiratory Distress Syndrome**

Despite an increased understanding of the pathology and current advances in supportive care, morbidity and mortality remain high in patients with acute respiratory distress syndrome (ARDS) [7]. ARDS pathology is driven by an acute severe inflammatory response, and acknowledging that the general hypothesis is that MSCs mainly act as immunomodulatory cells via rapid-acting paracrine effects, ARDS could be an ideal target for MSC-based therapies. This assumption is further supported by pre-clinical studies demonstrating that MSC-based therapy is beneficial in experimental models of ARDS (reviewed in [8, 9]).

In 2014, MSC-based treatment entered clinical trials for ARDS patients. In this trial, allogeneic adipose-derived MSCs were administered to 12 ARDS patients (one single dose of MSCs (1×10^6 cells/kg) or saline IV) [10]. A subsequent phase 1 dose-escalation safety trial (START trial, NCT01775774) using a single dose of allogeneic bone marrow-derived MSCs at 1.0, 5.0, and 10.0×10^6 cells per kg of predicted bodyweight in 9 ARDS patients was conducted [11]. These two trials were early phase 1 studies with the primary outcome being safety and were both underpowered to detect significant differences in efficacy. Both trials showed that MSCs were well tolerated in ARDS patients. While no significant differences in efficacy were observed, in the START study changes observed in the Sequential Organ Failure Assessment (SOFA) score and Lung Injury score (LIS) with the high dose of 10.0×10^6 MSCs/kg were promising. This led to a randomized phase 2a safety trial (NCT02097641) in which patients with moderate to severe ARDS received a single dose of the higher concentration (10.0×10^6 MSCs/kg) of bone marrow–derived MSCs. Similar to the first trial, no safety issues were observed but no significant improvement in 28-day mortality was observed between the MSC treated group and the placebo group [12]. The authors reported that the patient group receiving the MSC treatment were more severely ill compared with the placebo group. However, there were no differences in SOFA between MSC- or placebo-treated groups in SOFA when assessed at either 3, 7, or 14 days. Also, an unexpected finding was that many of the MSC batches had a very low post-thaw viability at the time of injection, which was speculated to have contributed to the lack of efficiency. However, as discussed further below, a new evolving concept suggests that dead or dying MSCs might be beneficial. More encouraging results were observed in a case study describing two patients with severe refractory ARDS who each received systemic administration of allogeneic bone marrow-derived MSCs (2×10^6 MSCs/kg) on a compassionate use basis [13]. In contrast to previous studies, rapid significant clinical improvements occurred with both patients surviving and eventually being discharged from the hospital.

In a recent industry-sponsored trial, the company Athersys Inc. recently (May 2019) published a press release in which they announced positive results at 28 days of follow-up in their randomized, placebo-controlled, phase 1/2 ARDS (MUST-ARDS) trial using IV administration of an adult stem cell product named MultiStem® [14••]. MultiStem® is composed of multipotent adult progenitor cells (MAPCs) obtained from adult bone marrow and expanded in adherence cultures. The MAPCs share some but not all attributes of MSCs utilized in the START study of from adult bone marrow, but they claim that cells are not the same as MSC. The key efficacy endpoint was met with a decreased mortality rate in the MultiStem® group (4/20= 25%) compared with the placebo group (4/10= 40%). With these positive results, it will be exciting to learn the results of other ongoing clinical trials (Table 1).

**Bronchopulmonary Dysplasia**

Bronchopulmonary dysplasia (BPD) remains a major contributor to mortality and morbidity in infants born prematurely, and current strategies to prevent this disease have been only moderately successful. BPD is a multifactorial disease where none of the current treatment strategies has effectively decreased complications in BPD survivors. Over the past years, the interest in using MSC-based therapies to treat BPD has increased especially in response to findings in pre-clinical studies demonstrating positive benefits [15, 16]. Currently, there are several clinical trials for BPD registered as completed or active on ClinicalTrials.gov (Table 1). The first published trial using MSCs to treat infants with BPD was a phase 1, dose-escalation trial (NCT01297205) using umbilical cord blood–derived MSCs at a concentration of 1×10^7 or 2×10^7 cells per kg [17]. This study demonstrated that the treatment was well tolerated in patients with BPD and that the levels of IL-6, IL-8, MMP-9, TNF-α, and TGF-β in tracheal aspirates were significantly reduced compared with baseline values. A 2-year follow-up study of this trial (NCT01632475) was published in 2017 [18], revealing that although one infant in the MSC group died suffering from Enterobacter cloacae sepsis, the remaining 8 infants showed no sign of transplant-related adverse outcomes or tumorigenicity was observed [18].
| NCT number   | Status             | Conditions         | Phases | Enrollment | Study designs                                      |
|-------------|--------------------|--------------------|--------|------------|---------------------------------------------------|
| NCT00683722 | Completed          | COPD               | Phase 2| 62         | Randomized, parallel assignment, quadruple        |
| NCT01175655 | Completed          | BO                 | Phase 1| 10         | Single group assignment, open label               |
| NCT01297205 | Completed          | BPD                | Phase 1| 9          | Single group assignment, open label               |
| NCT01306513 | Completed          | Emphysema          | Phase 1| 10         | Single group assignment, open label               |
| NCT01385644 | Completed          | IPF                | Phase 1| 8          | Non-randomized, single group assignment, open label|
| NCT01775774 | Completed          | ARDS               | Phase 1| 9          | Single group assignment, open label               |
| NCT01828957 | Completed          | BPD                | Phase 2| 69         | Randomized, parallel assignment, quadruple        |
| NCT01872624 | Completed          | Emphysema          | NA     | 10         | Non-randomized, parallel assignment, open label   |
| NCT01919827 | Completed          | IPF                | Phase 1| 17         | Single group assignment, open label               |
| NCT01977131 | Completed          | Silicosis          | Phase 1/2| 10   | Single group assignment, single (participant)     |
| NCT02013700 | Completed          | IPF                | Phase 1| 9          | Randomized, parallel assignment, double           |
| NCT02023788 | Completed          | BPD                | Phase 1| 8          | Case-only, prospective                            |
| NCT02097641 | Completed          | ARDS               | Phase 2| 60         | Randomized, parallel assignment, triple           |
| NCT02277145 | Completed          | Post-radiotherapy pulmonary fibrosis | Phase 1 | 10 | Single group assignment, open label |
| NCT02381366 | Completed          | BPD                | Phase 1/2| 12   | Non-randomized, single group assignment, open label |
| NCT02594839 | Completed          | ILD                | Phase 1/2| 20   | Randomized, parallel assignment, open label       |
| NCT02625246 | Completed          | Bronchiectasis     | Phase 1| 6          | Non-randomized, single group assignment, open label |
| NCT02668068 | Completed          | Pneumoconiosis     | Phase 1| 80         | Randomized, parallel assignment, single (participant) |
| NCT02804945 | Completed          | ARDS               | Phase 2| 20         | Single group assignment, open label               |
| NCT01795950 | Terminated         | PAH                | Phase 1| 6          | Non-randomized, single group assignment, open label |
| NCT01632475 | Active, not recruiting | BPD             | Phase 1| 9          | Case-only, prospective                            |
| NCT02161744 | Active, not recruiting | COPD            | Phase 1| 9          | Single group assignment, open label               |
| NCT02192736 | Active, not recruiting | Asthma           | Phase 1/2| 20   | Single group assignment, open label               |
| NCT01897987 | Recruiting         | BPD                | Phase 2| 70         | Randomized, parallel assignment, triple           |
| NCT02181712 | Recruiting         | BO                 | Phase 1| 19         | Non-randomized, single group assignment, open label |
| NCT02348060 | Recruiting         | COPD               | 100    |            | Cohort, prospective                              |
| NCT02859415 | Recruiting         | Esophageal neoplasms | Phase 1/2| 100 | Non-randomized, sequential assignment, open label |
| NCT02866721 | Recruiting         | CF                 | Phase 1| 15         | Single group assignment, open label               |
| NCT02946658 | Recruiting         | Lung disease       | Phase 1/2| 100 | Non-randomized, parallel assignment, single (participant) |
| NCT03042143 | Recruiting         | ARDS               | Phase 1/2| 75   | Randomized, parallel assignment, quadruple        |
| NCT03131999 | Recruiting         | Asthma             | Phase 1| 6          | Non-randomized, sequential assignment, open label |
| NCT03378063 | Recruiting         | BPD                | Early Phase 1| 100 | Non-randomized, parallel assignment, open label |
| NCT03392467 | Recruiting         | Severe BPD         | Phase 2| 60         | Randomized, parallel assignment, quadruple        |
| NCT03558334 | Recruiting         | BPD                | Phase 1| 12         | Non-randomized, parallel assignment, open label   |
| NCT03608592 | Recruiting         | ARDS               | NA     | 26         | Single group assignment, open label               |
| NCT03631420 | Recruiting         | BPD                | Phase 1| 9          | Single group assignment, open label               |
| NCT03774537 | Recruiting         | BPD                | Phase 1/2| 20   | Non-randomized, parallel assignment, open label   |
| NCT03857841 | Recruiting         | BPD                | Phase 1| 18         | Randomized, sequential assignment, quadruple      |
| NCT03871330 | Recruiting         | BPD                | Phase 1| 30         | Single group assignment, open label               |
| NCT03929120 | Recruiting         | ILD                | Phase 1| 10         | Single group assignment, open label               |
| NCT04003857 | Recruiting         | BPD                | Phase 2| 60         | Randomized, parallel assignment, triple           |
| NCT04047810 | Recruiting         | COPD               | Phase 1| 15         | Single group assignment, open label               |
| NCT04055415 | Recruiting         | PH                 | Phase 1/2| 60   | Randomized, parallel assignment, double           |
| NCT04062136 | Recruiting         | BPD                | Phase 1| 10         | Single group assignment, open label               |
| NCT03909750 | Not yet recruiting | COPD               | Phase 1| 50         | Non-randomized, parallel assignment, single (participant) |
| NCT024444961 | Not yet recruiting | BPD                | Phase 1| 10         | Single group assignment, open label               |
| NCT number       | Status               | Conditions | Phases       | Enrollment | Study designs                                      |
|------------------|----------------------|------------|--------------|------------|---------------------------------------------------|
| NCT02985346      | Not yet recruiting   | IPH        | Early Phase 1| 100        | Non-randomized, parallel assignment, open label    |
| NCT03058068      | Not yet recruiting   | CF         | Phase 1      | 18         | Randomized, parallel assignment, double           |
| NCT03601416      | Not yet recruiting   | BPD        | Phase 2      | 57         | Randomized, parallel assignment, open label        |
| NCT03645525      | Not yet recruiting   | BPD        | Phase 1/2    | 180        | Randomized, parallel assignment, quadruple         |
| NCT03818854      | Not yet recruiting   | RDS        | Phase 2      | 120        | Randomized, parallel assignment, triple           |
| NCT04018729      | Not yet recruiting   | Severe COPD| Phase 2/3    | 34         | Randomized, parallel assignment, double           |
| NCT01207869      | Unknown status       | Severe BPD | Phase 1      | 10         | Randomized, parallel assignment, double           |
| NCT01373203      | Unknown status       | ALI        | NA           | 20         | Case-only                                         |
| NCT01758055      | Unknown status       | Emphysema  | Phase 1      | 12         | Single group assignment, open label               |
| NCT01902082      | Unknown status       | ARDS       | Phase 1      | 20         | Randomized, parallel assignment, triple           |
| NCT02112500      | Unknown status       | RDS        | Phase 2      | 10         | Single group assignment, open label               |
| NCT02135380      | Unknown status       | IPF        | Phase 1/2    | 60         | Randomized, parallel assignment, open label        |
| NCT02175303      | Unknown status       | ALI        | Phase 1/2    | 25         | Non-randomized, single group assignment, open label|
| NCT02215811      | Unknown status       | ARDS       | Phase 1      | 10         | Single group assignment, open label               |
| NCT02444455      | Unknown status       | ARDS       | Phase 1/2    | 20         | Single group assignment, open label               |
| NCT02543073      | Unknown status       | BO         | Phase 1/2    | 60         | Non-randomized, parallel assignment, open label    |
| NCT02645305      | Unknown status       | COPD       | Phase 1/2    | 20         | Single group assignment, open label               |
| NCT02749448      | Unknown status       | Pulmonary disease | Phase 1 | 10       | Single group assignment, open label               |
| NCT02790762      | Unknown status       | Pneumocociosis | Phase 1 | 10        | Single group assignment, open label               |
| NCT01559051      | Withdrawn            | COPD       | Phase 1/2    | 0          | Single group assignment, open label               |
| NCT01849159      | Withdrawn            | Emphysema  | Phase 1/2    | 0          | Randomized, parallel assignment, open label        |
| NCT02041000      | Withdrawn            | COPD       | NA           | 0          | Single group assignment, open label               |

*NCT* clinical trial registry number in ClinicalTrials.gov database, *BPD* bronchopulmonary dysplasia, *IPH* idiopathic pulmonary fibrosis, *ARDS* acute respiratory distress syndrome, *PAH* pulmonary arterial hypertension, *COPD* chronic obstructive pulmonary disease, *BO* bronchiolitis obliterans, *CF* cystic fibrosis, *ILD* interstitial lung disease, *PH* pulmonary hypertension, *IPH* idiopathic pulmonary hemosiderosis, *RDS* respiratory distress syndrome, *AUI* acute lung injury, *NA* not applicable
These studies will hopefully reveal if COPD is suitable for as active or recruiting participants with COPD (Table 1). Currently, there are four trials listed on ClinicalTrials.gov out the lung to the same extent as carbon monoxide gas. demonstrates that the third-party MSCs survive and diffuse through- as well as with the baseline FEV1 [27]

with MSC have been whether or not the cells survive in vivo after administration to patients, and whether they could access sites of injury. In a recently published phase 1 study using MSCs to treat COPD (NCT00683722) was performed in 2013 [24]. This investigation demonstrated the safety of using non-HLA-matched bone marrow–derived MSCs in an older patient population with moderate-to-severe COPD. Additional studies have since been performed using both systemic and direct airway delivery approaches and have demonstrated similar results of safety of MSC administration to COPD patients [25, 26, 27]. However, none of these studies has demonstrated efficacy for treating COPD. Concerns with MSC have been whether or not the cells survive in vivo after administration to patients, and whether they could access sites of injury. In a recently published phase 1 study (Australian clinical trials registry number 12614000731695), MSCs were radiolabeled with indium-111 prior infusion. As in previous trials, all COPD patients tolerated the MSC infusions well. Interestingly, within 30 min the pre-labeled cells were detected in the lungs where they remained detectable for 24 h. Moreover, the amount of positive labeling correlated with the diffusing capacity of the lung for carbon monoxide as well as with the baseline FEV1 [27]. This study demonstrates that the third-party MSCs survive and diffuse throughout the lung to the same extent as carbon monoxide gas. Currently, there are four trials listed on ClinicalTrials.gov as active or recruiting participants with COPD (Table 1). These studies will hopefully reveal if COPD is suitable for MSC-based therapy either as the main treatment or in combination with other relevant therapies.

Idiopathic Pulmonary Fibrosis

Idiopathic pulmonary fibrosis (IPF) is a life-threatening, progressive fibrotic lung disease of still poorly understood etiology. It has a patchy pathology with areas of massive deposition of extracellular matrix components, leading to reduced lung function. Currently, there are no curative treatments and in severe stages of the disease lung transplantation remain the only option [28]. MSC-based strategies have been extensively investigated in pre-clinical bleomycin-induced and other models of fibrotic lung injuries (reviewed in [29]). Despite the lack of convincing pre-clinical data on established lung fibrosis, early-phase clinical trials have been completed (Table 1). In 2013, Tzouvelekis et al. demonstrated in a prospective, non-randomized, no placebo-controlled phase 1 study that adipose-derived MSC were safe to give to IPF patients with no adverse events detected [30]. In a dose-escalation phase 1 study (NCT01385644), Chambers et al. demonstrated that it was safe to inject placenta-derived MSCs at either 1×10^6 or 2×10^6 cells/kg; however, this study was not powered to detect efficacy [31]. In 2017, Glassberg et al. published the results from the AETHER study (NCT02013700) where they used a single dose of bone marrow–derived MSCs at 20×10^6, 100×10^6, or 200×10^6 cells per infusion. This non-randomized and non-placebo controlled study included 9 patients, and while a few serious adverse events were reported, none was determined to be related to the MSC treatment. Similar to the other MSC trials, this study was not powered to detect efficacy, and no significant improvement in outcome was reported [32]. Taken together, clinical trials to date have shown safety and feasibility, but not benefit. Considering the general hypothesis that MSCs mainly act through rapid paracrine immunomodulatory mechanisms, advanced stages of IPF might not be the best suitable disease for MSC-based therapies.

Current Challenges and Hurdles in the Field that Needs to Be Overcome

MSC-based cell therapy for treatment of severe lung diseases has demonstrated promising results in experimental models; however, the lack of ability to translate these encouraging results into clinically relevant effects in patients has hampered the progression of the field. Despite an enormous interest in using MSCs for human clinical settings, the knowledge of MSC mechanisms of actions are limited and have predominantly been derived from pre-clinical studies [4, 33]. A growing literature suggests that the MSC phenotype and in vivo functions differ from the basic in vitro understanding. For
example, we and others have reported data suggesting that the in vivo mode of action changes depending on the inflammatory lung environment encountered [34•, 35•, 36••]. Moreover, MSCs exposed to the ARDS environment affects monocytes/macrophages by altering their phagocytic capacity and increasing anti-inflammatory M2 macrophage markers [35•]. In addition, Islam et al. recently demonstrated that the beneficial effect of MSCs was determined by the lung micro-environment. They further observed that high levels of both IL-6 and fibronectin in combination with low antioxidant capacity within the lung microenvironment resulted in poor outcome after MSC treatment in experimental lung injury [36••]. In addition, we recently demonstrated that MSCs stimulated with bronchoalveolar lavage fluid samples from patients with either ARDS or non-ARDS lung diseases exhibited both disease-specific and common MSC phenotypes [34•]. Taken together, the success of MSC-based clinical trials in lung diseases and other critical illnesses relies heavily on understanding the in vivo mode of action. Thus, a growing body of data suggests that the microenvironment within the diseased lung plays an important role in directing potential MSC therapeutic actions.

The generally accepted hypothesis has been that MSC therapeutic actions rely on the viability of the cells. However, this assumption has recently been challenged and recent studies suggest that dead or dying cells might improve the therapeutic effect by triggering host immune cells [37–40]. For example, administration of viable or heat-inactivated MSCs into an LPS induced lung injury mouse model evoked similar beneficial response [40]. More specifically, it is possible that systemically infused MSCs undergo apoptosis, which in turn activate the host immune system and lead to the immunomodulatory effects [37–40]. These are interesting data, however it is important to remember that these are xeno-transplantations (human into mouse), which may or may not contribute to these observations. The data on dead or dying MSCs in clinical trials are limited; nevertheless, a high portion of non-viable cells (85%) was reported in a recent multicenter double-blinded randomized trial of systemic bone marrow–derived MSCs in ARDS patients [12]. No efficacy was observed in this trial suggesting that non-viable MSCs may not have clinical benefit in ARDS. There are currently relative few publications investigating the effects of dead or dying MSCs, and this needs to be further studied.

**Current Challenges and Hurdles for MSC Manufacturing for Clinical Applications**

Despite an increasing number of early phase clinical trials using MSC for lung diseases, there is at present no established scientific consensus concerning the source of origin of MSCs, dosing strategies, therapeutic dose, and cell product manufacturing (Table 2). MSC functionality changes depending on culture conditions including the isolation process, initial seeding density, number of passages, and culture surface. Despite this knowledge, a recent survey provided by the MSC committee of the International Society of Cell and Gene Therapy revealed that the current practice for manufacturing MSCs for clinical applications differs significantly in many of the above mentioned aspects amongst different US academic centers [41]. Although bone marrow was reported as the primary source of MSCs (93.3% of the facilities), MSCs are also isolated from adipose tissue, umbilical cord, umbilical cord blood, and placental tissue. Not surprisingly, MSCs isolated from different sources have different phenotypes and secretome profiles [42–46]. Furthermore, the survey revealed that multiple different isolation strategies were utilized. For example, 46.7% of centers used enzymatic digestion, 26.7% used an automated system, and 13.3% used Ficoll gradient centrifugation or filtration. In addition, 93.3% reported plastic adherence as the main enrichment method, with only 6.7% reporting use of FACS sorting, immuno-depletion, or immune-enrichment strategies. Utilizing standard plastic culture dishes is the most frequent way of cultivating MSCs, however three-dimensional cell culture systems may narrow the gap between pre-clinical and clinical research. For example, culture dishes coated with extracellular matrix molecules such as fibronectin and collagen have been demonstrated to support MSC growth [47, 48]. In addition, changing the composition of the matrix proteins resulting in altered elasticity or stiffness has been demonstrated to alter MSC phenotype [49]. However, the majority of the centers (80%) report using 2D culture methods with only three centers reporting exclusive use of 3D methods (bioreactor, bags, or hollow fibers) to expand MSCs. Also, seeding density affects MSC expansion and function. Fewer than 50% of centers report that they use low-density (30%, 50–500 cells/cm²) or intermediate density (16.7%, 500–2500 cells/cm²) meaning that the majority of facilities use a high plating density (>2500 cells/cm²). The same wide variation in current practice in producing MSCs for clinical use has also been found in a similar survey performed on MSC manufacturing facilities in Europe [50].

In addition to the challenges discussed above, efforts to map out strategies to foresee which patient groups are most likely to respond to MSC-based treatments need to be made. Many of the experimental lung injury models used in preclinical studies have been optimized to detect maximum effects, and might therefore not reflect the truly in vivo situation (reviewed in [51]). A compelling amount of pre-clinical data suggest that MSCs can act by secretion of a spectrum of paracrine factors, and tracking studies of systemically administered MSCs have demonstrated that, following initial lodging of the majority of cells in the pulmonary vascular bed, the majority are cleared within a few days [1, 27•, 52]. It is therefore unlikely that MSC-based treatments can remodel.
| NCT number     | Conditions         | Cell source          | No. of cells                  | Route | Dosing                      | Comments                                                                 | PMID       |
|----------------|--------------------|----------------------|-------------------------------|-------|-----------------------------|--------------------------------------------------------------------------|------------|
| NCT00683722    | COPD               | BM-MSC (Prochymal)   | 100.0×10^6 cells/infusion     | IV    | 1 dose/month (total 4 months) | MSCs were produced under good manufacturing practice conditions (Therapeutic Goods Administration) Female donors age 17–30 years | 23,172,272 |
| NCT0175655     | BO                 | BM-MSC               | 2.0×10^6 cells/kg             | IV    | 2 doses/week (for 2 weeks)  | In combination with LVRS                                                | 28,186,707 |
| NCT01297205    | BPD                | UCB-MSC              | 1.0×10^7 or 2.0×10^7 cells/kg | IT    | Single dose                 | Transfected by a vector containing human HGF cDNA                        | 25,039,426 |
| NCT01306513    | Emphysema          | BM-MSC               | 1.0×10^6 cells/kg             | IV    | Single dose                 | Male donors age 24 and 25 years                                         | 25,529,339 |
| NCT01385644    | IPF                | Placenta-MSC         | 1.0×10^6 or 2.0×10^6 cells/kg | IV    | Single dose                 | After fully lavage of the localized lesions                              | 27,890,713 |
| NCT01775774    | ARDS               | BM-MSC               | 1.0×10^6, 5.0×10^5, or 1.0×10^8 cells/infusion | IV    | Single dose                 | One female and two male donors age 18–45 years                           | 30,455,077 |
| NCT01828957    | BPD                | UCB-MSC (Pneumostem) | 1.0×10^6 cells/kg             | IT    | Single dose                 | 5-year long-term follow-up study                                        | 28,186,686 |
| NCT01872624    | Emphysema          | BM-MSC               | 1.0×10^6 cells (in 30 ml saline) | EB    | In combination with EBV insertion |                                                                           | 26,400,297 |
| NCT01918827    | IPF                | BM-MSC               | 1.0×10^6 cells/kg             | IV    | 1 dose/week (for 3 weeks)   |                                                                           | 27,890,713 |
| NCT01977131    | Silicosis          | BM-MSC               | 2.0×10^5 cells/kg             | IV    | Single dose                 |                                                                           | 30,455,077 |
| NCT02013700    | ARDS               | BM-MSC               | 1.0×10^5, 1.0×10^4, or 1.0×10^8 cells/infusion | IV    | Single dose                 |                                                                           | 30,455,077 |
| NCT02023788    | BPD                | UCB-MSC (Pneumostem) | 1.0×10^6 or 2×10^6 cells/kg   | IT    | Single dose                 |                                                                           | 30,992,220 |
| NCT02097641    | ARDS               | BM-MSC               | 1.0×10^6 cells/kg             | IV    | Single dose                 |                                                                           | 30,992,220 |
| NCT02277145    | Pulmonary fibrosis | UC-MSC               | 1.0×10^6 cells/kg             | FB    | In combination with EBV insertion |                                                                           | 28,341,525 |
| NCT02381366    | BPD                | BM-MSC               | 1.0×10^6 cells/kg             | IT    | In combination with EBV insertion |                                                                           | 28,341,525 |
| NCT02594839    | ILD                | BM-MSC               | 1.0×10^6 cells/infusion       | IV    | 2 doses at interval of 7 days (every 3 months for 1 year) | | 28,341,525 |
| NCT02625246    | Bronchiectasis     | BM-MSC               | 2.0×10^6 or 1.0×10^7 cells    | IV    | Injected after whole-lung lavage |                                                                           | 28,341,525 |
| NCT02660068    | Pneumocystis       | BM-MSC               | 1.0×10^6 cells/kg             | IV    | Single dose                 |                                                                           | 28,341,525 |
| NCT02804945    | ARDS               | UC-MSC               | max. dose of 3×10^6 cells/kg  | IV    | Single dose                 |                                                                           | 28,341,525 |
| NCT01795950    | PAH                | Placenta-ASC         | 0.5×10^6, 1.0×10^6, or 2.0×10^6 cells/kg | IV    | Single dose                 |                                                                           | 28,341,525 |
| NCT01632475    | BPD                | UCB-MSC (Pneumostem) | 1.0×10^6 or 2×10^6 cells/kg   | IT    | Single dose                 |                                                                           | 28,341,525 |
| NCT02161744    | COPD               | Adipose-MSCs         | 1.0×10^6 cells/kg             | IV    | Single dose                 |                                                                           | 28,341,525 |
| NCT02192736    | Asthma             | MTF factors          | Autologous                    | IV    | In combination with EBV insertion |                                                                           | 28,341,525 |
| NCT02277145    | ILD                | UC-MSC               | 1.0×10^6 cells/kg             | IV    | In combination with EBV insertion |                                                                           | 28,341,525 |

| NCT clinical trial registry number in ClinicalTrial.gov database, BPD bronchopulmonary dysplasia, IPF idiopathic pulmonary fibrosis, ARDS acute respiratory distress syndrome, PAH pulmonary arterial hypertension, COPD chronic obstructive pulmonary disease, BO bronchiolitis obliterans, ILD interstitial lung disease, max maximal, BM bone marrow–derived, UC umbilical cord–derived, UCB umbilical cord blood–derived, ASC mesenchymal-like adherent stromal cells, MTF allogeneic mesenchymal trophic factors, No number, IV intravenous, IT intratracheal, IB intrabronchial, EB endobronchial, IN intra-nasally, FB fiberoptic bronchoscopy, LVRS lung volume reduction surgeries, EBV one-way endobronchial valve, HGF hepatocyte growth factor |
chronically injured and destroyed tissue such as found in advanced lung fibrosis or emphysema by differentiating into other cell types. Rather, it is more likely that MSC-based cell therapies will be more beneficial in diseases involving acute inflammation and infection such as ARDS and sepsis septic shock. Moreover, ARDS is a multifaceted and heterogeneous disorder and there may be very meaningful differences in the host environment that will affect the therapeutic activity of MSCs. As such, we need to find strategies for selecting patients most likely to respond to the treatment. Another way to improve the clinical outcome would be to design MSC potency assays. However, without knowing the in vivo mode of action makes it difficult to determine what this assay or assays should be [51, 53]. In addition, effort should also be made to optimize and unify outcome parameters measured in clinical trials for lung diseases in order to design successful clinical trials and to obtain maximum biologic and mechanistic information.

Stem Cell Tourism

In parallel with the progression of the field of cell-based therapies, a growing problem with commercial stem cell therapies has developed both in the USA and globally [54, 55]. In modern life with daily access to internet and social media platforms, desperate patients, families, and caregivers can easily be misled into participating in very expensive and unproven stem cell treatments, which are not covered by insurance. Importantly, many stem cell clinics fail to prove safety and do not fulfill recognized biological and medical standards, exposing patients to unnecessary risks. These unethical clinics not only harm patients and their families, they also have the potential to hamper the progression of scientifically rigorous potential stem cell therapies. Therefore, the FDA, the International Society for Cell and Gene Therapies (ISCT), the International Society for Stem Cell Research (ISSCR) and an increasing number of respiratory disease foundations have taken stances against these stem cell clinics [56–62]. In an attempt to increase the knowledge about these clinics to patients, families, and caregivers, the American Thoracic Society (ATS) Respiratory Cell and Molecular Biology Assembly Stem Cell Working Group posted online statements and several other related publications [58–62]. In addition, earlier this year Google announced that they are not any longer accepting advertising for “unproven or experimental medical techniques such as most stem cell therapy, cellular (non-stem) therapy, and gene therapy” [63].

Conclusions

MSC-based therapies for severe lung diseases such as ARDS, COPD, IPF, and BPD have demonstrated promising results in experimental lung models; however, this has not translated into significant improved clinical outcome in patients to date. Importantly, current clinical trials have all demonstrated that MSC-based therapies are safe for lung disease patients; however, no significant efficacy or improved lung function has currently been demonstrated except for one promising investigation in patients with ARDS [14]. There are several challenges ahead for this field including revealing the in vivo mechanism of action, selecting the patient group most likely to respond, development of relevant potency assays, and the accelerating problem with the unethical stem cell clinics. Despite these challenges, there has been significant progression in the field and hopefully ongoing clinical trials will bring positive and encouraging clinical data with improved outcomes and hope for the future.

Funding Information Open access funding provided by Lund University.

Compliance with Ethical Standards

Conflict of Interest The authors declare no conflicts of interest.

Human and Animal Rights and Informed Consent This article does not contain any studies with human or animal subjects performed by any of the authors.

Open Access This article is licensed under a Creative Commons Attribution 4.0 International License, which permits use, sharing, adaptation, distribution and reproduction in any medium or format, as long as you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons licence, and indicate if changes were made. The images or other third party material in this article are included in the article's Creative Commons licence, unless indicated otherwise in a credit line to the material. If material is not included in the article's Creative Commons licence and your intended use is not permitted by statutory regulation or exceeds the permitted use, you will need to obtain permission directly from the copyright holder. To view a copy of this licence, visit http://creativecommons.org/licenses/by/4.0/.

References

Papers of particular interest, published recently, have been highlighted as:

- Of importance
- Of major importance

1. Savukinas UB, Enes SR, Sjoland AA, Westergren-Thorsson G. Concise review: the bystander effect: Mesenchymal stem cell-mediated lung repair. Stem Cells. 2016;34(6):1437–44.
2. Ryan AL, Ikonomou L, Atarod S, Bolukbas DA, Collins J, Freishtat R, et al. Stem cells, cell therapies, and bioengineering in lung biology and diseases 2017. An Official American Thoracic Society Workshop Report. Am J Respir Cell Mol Biol. 2019;61(4):429–39.
36. Islam D, Huang Y, Fanelli V, Delsedime L, Wu S, Khang J, et al. Identification and quantification of microenvironment is crucial for effective mesenchymal stromal cell therapy in acute lung injury. Am J Respir Crit Care Med. 2019;199(10):1214–24. 

37. Weiss DJ, English K, Krasnodembskaya A, Isaza-Correa JM, Hawthorne J, Mahon BP. The necrobiology of mesenchymal stromal cells affects therapeutic efficacy. Front Immunol. 2019;10:1228.

38. de Witte SFH, Luk F, Sierra Parraga JM, Gargesha M, Merino A, Korevaar SS, et al. Immunomodulation by therapeutic mesenchymal stromal cells (MSC) is triggered through phagocytosis of MSC by mononuclear cells. Stem Cells. 2018;36(4):602–15.

39. Galleu A, Riffio-Vasquez Y, Trento C, Lomas C, Dolcetti L, Cheung TS, et al. Apoptosis in mesenchymal stromal cells induces in vivo recipient-mediated immunomodulation. Sci Transl Med. 2017;9(416).

40. Luk F, de Witte SF, Korevaar SS, Roemeling-van Rhijn M, Franquesa M, Strini T, et al. Inactivated mesenchymal stem cells maintain immunomodulatory capacity. Stem Cells Dev. 2016;25(18):1342–54.

41. Phinney DG, Galipeau J, C. Msc Committee Of The International Society Of T. Gene. Manufacturing mesenchymal stromal cells for clinical applications: A survey of Good Manufacturing Practices at U.S. academic centers. Cytotherapy. 2019;21(7):782–92.

42. Rollandsson Enes S, Ahman E, Palani A, Hallgren O, Bjerner L, Malmstrom A, et al. Quantitative proteomic characterization of lung-MSC and bone marrow-MSC using DIA-mass spectrometry. Sci Rep. 2017;7(1):9316.

43. Rollandsson Enes S, Andersson Sjoland A, Skog I, Hansson L, Larsson H, Le Blanc K, et al. MSC from fetal and adult lungs possess lung-specific properties compared to bone marrow-derived MSC. Stem Cells. 2016;34(6):29160.

44. Aires AO, Mendes-Pinheiro B, Teixeira FG, Anjo SI, Ribeiro-Samy S, Gomes ED, et al. Unveiling the differences of secretome of adult lung from studies of transplanted allografts. J Clin Invest. 2007;117(4):989–96.

45. Melief SM, Zwagtinga JJ, Fibbe WE, Roelofs H. Adipose tissue-derived multipotent stromal cells have a higher immunomodulatory capacity than their bone marrow-derived counterparts. Stem Cells Transl Med. 2013;2(6):455–63.

46. Salzig D, Leber J, Merkewitz K, Lange MC, Koster N, Czermak P. Attachment, growth, and detachment of human mesenchymal stem cells in a chemically defined medium. Stem Cells Int. 2016;2016:5246584.

47. Somaiah C, Kumar A, Mawrie D, Sharma A, Patil SD, Bhattacharyya J, et al. Collagen promotes higher adhesion, survival and proliferation of mesenchymal stem cells. PLoS One. 2015;10(12):e0145068.

48. Engler AJ, Sen S, Sweeney HL, Discher DE. Matrix elasticity directs stem cell lineage specification. Cell. 2006;126(4):677–89.

49. Trento C, Bernardo ME, Nagler A, Kuci S, Bornhauser M, Kohl U, et al. Manufacturing mesenchymal stromal cells for the treatment of graft-versus-host disease: a survey among centers affiliated with the European Society for Blood and Marrow Transplantation. Biol Blood Marrow Transplant. 2018;24(11):2365–70.

50. Galipeau J, Krampaer M. The challenge of defining mesenchymal stromal cell potency assays and their potential use as release criteria. Cytotherapy. 2015;17(2):125–7.

51. English K. Mechanisms of mesenchymal stem cell immunomodulation. Immunol Cell Biol. 2013;91(1):19–26.

52. Galipeau J, Krampaer M, Barrett J, Dazzi F, Deans RJ, DeBruijn J, et al. International Society for Cellular Therapy perspective on immune functional assays for mesenchymal stromal cells as potency release criteria for advanced phase clinical trials. Cytotherapy. 2016;18(2):151–9.

53. Dominici M, Nichols K, Srivastava A, Weiss DJ, Eldridge P, Cuenne N, et al. Positioning a scientific community on unproven cellular therapies: the 2015 International Society for Cellular Therapy Perspective. Cytotherapy. 2015;17(12):1663–6.

54. Dominici M, Nichols KM, Levine AD, Rasko JE, Forte M, O’Donnell L, et al. Science, ethics and communication remain essential for the success of cell-based therapies. Brain Circ. 2016;2(3):146–51.

55. Ikonomou L, Panoskalsis-Mortari A, Wagner DE, Freishtat RJ, Weiss DJ, C. American Thoracic Society Respiratory, et al. Unproven stem cell treatments for lung disease-An emerging public health problem. Am J Respir Crit Care Med. 2017;195(7):P13–4.

56. Marks P, Gottlieb S. Balancing safety and innovation for cell-based regenerative medicine. N Engl J Med. 2018;378(10):954–9.

57. ATS RCMB Stem Cell Working Group. Statement on unproven stem cell interventions for lung diseases. New York: American Thoracic Society; 2016. https://www.thoracic.org/members/assemblies/assemblies/rcmb/working-groups/stem-cell/resources/statement-on-unproven-stem-cell-interventions-for-lung-diseases.pdf. Accessed Dec 2019.

58. Weiss DJ, Turner L, Levine AD, Ikonomou L. Medical societies, patient education initiatives, public debate and marketing of unproven stem cell interventions. Cytotherapy. 2018;20(2):165–8.

59. Ikonomou L, Freishtat RJ, Wagner DE, Panoskalsis-Mortari A, Weiss DJ. The global emergence of unregulated stem cell treatments for respiratory diseases. Professional Societies Need to Act. Ann Am Thorac Soc. 2016;13(8):1205–7.

60. Ikonomou L, Wagner DE, Turner L, Weiss DJ. Translating basic research into safe and effective cell-based treatments for respiratory diseases. Ann Am Thorac Soc. 2019;16(6):657–68.

61. Wagner DE, Turner L, Panoskalsis-Mortari A, Weiss DJ, Ikonomou L. Co-opting of ClinicalTrials.gov by patient-funded studies. Lancet Respir Med. 2018;6(8):579–81.

62. A new policy on advertising for speculative and experimental medical treatments, 2019. https://support.google.com/google-ads/answer/9475042. Accessed Dec 2019.

Publisher’s Note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.